The Role of Dopamine and Acetylcholine as Modulators of Selective Attention and Response Speed

Universität Trier

Fachbereich Psychologie

Fach Allgemeine Psychologie und Methodenlehre

eingereichte Dissertation von

Dipl.-Psych. Katja Kerstin Schneider

1. Supervisor: Prof. Dr. Christian Frings, University of Trier

2. Supervisor: Prof. Dr. Jobst Meyer, University of Trier

February 5, 2015

Abstract

The principles of top-down and bottom-up processing are essential to cognitive psychology. At their broadest, most general definition, they denote that processing can be driven either by the salience of the stimulus input or by individual goals and strategies. Selective top-down attention, specifically, consists in the deliberate prioritizing of stimuli that are deemed goal-relevant, while selective bottom-up attention relies on the automatic allocation of attention to salient stimuli (Connor, Egeth, & Yantis, 2004; Schneider, Schote, Meyer, & Frings, 2014). Variations within neurotransmitter systems can modulate cognitive performance in a domain-specific fashion (Greenwood, Fossella, & Parasuraman, 2005). Noudoost and Moore (2011a) proposed that the influence of the dopaminergic neurotransmitter system on selective top-down attention might be greater than the influence of this system on selective bottom-up attention; likewise, they assumed that the cholinergic neurotransmitter system might be more important for selective bottom-up than top-down attention. To test this hypothesis, naturally occurring variations within the two neurotransmitter systems were assessed. Five polymorphisms were selected; two of the dopaminergic system (the *COMT* Val158Met polymorphism and the *DAT1* polymorphism) and three of the cholinergic system (the *CHRNA4* rs1044396 polymorphism, the *CHRNA5* rs3841324 polymorphism, and the *CHRNA5* rs16969968 polymorphism). It was tested whether these polymorphisms modulated the performance in tasks of selective top-down attention (a Stroop task and a Negative priming task) and in a task of selective bottom-up attention (a Posner-Cuing task). Indeed, the dopaminergic polymorphisms influenced selective top-down attention, but exerted no effects on bottom-up attention. This aligned with the hypothesis proposed by Noudoost and Moore (2011a). In contrast, the cholinergic polymorphisms were not found to modulate selective bottom-up attention. All of the three cholinergic polymorphisms, however, affected the general response speed in the Stroop task, Negative priming task, and Posner-Cuing task (irrespective of the attentional processing). In sum, the findings of this study provide strong indications that the dopaminergic system modulates selective top-down attention, while the cholinergic system is highly relevant for the general speed of information processing.

Acknowledgments

First and foremost, I would like to thank my supervisor, Prof. Dr. Christian Frings. I learned a lot from him about all parts of the scientific process, from the early conceptualizations of a study to the publication of the results. His approach to research is highly efficient, insightful, and methodologically rigorous, and feel lucky to have been able to learn from him. I didn't instantly acclimatize to the world of research (as my first attempts of writing a paper will attest to), but Christian's invaluable feedback and support on every step of the way have proven to be great motivators. I am more grateful than I can express for all the time he invested and all of his encouragements and guidance.

At the same time I would like to express my appreciation and gratitude towards Prof. Dr. Jobst Meyer, who has been an invaluable source of knowledge and who always had an open door and a friendly ear for me. I greatly enjoyed our discussions at meetings as well as the many pleasant department breakfasts on Fridays.

I also feel indebted to Dr. Andrea Schote-Frese, who hugely furthered my understanding of genetics and always had time for a friendly chat about scientific or non-scientific matters. Working with Andrea for the last few years has been highly pleasant.

The study that this thesis is based on was sampled over the span of two months in autumn 2012. It was a large and elaborate study and, not surprisingly, conducting it amounted to a trying time period. I would like thank the students who helped. I experienced all of them as highly reliable and well organized, and thus it was a pleasure to work with them: I'm thanking Nina Schmidt, Lisa Pramme, Alexander Kotz, Nora Fuentes, Mia Spank, and Milena Ortner for their efforts! After the study had been conducted, the genetic analyses were the next big step. In the lab, I would have been lost without the diligent help of Ulrike Winnikes and Andrea Schote-Frese. Both spent countless time showing me the ropes – I relied on their long-standing experience, dedication and unwavering patience. I will always be grateful for the chance to learn the genetic methodology in such a pleasant and productive atmosphere. I also would like to thank Nina Schmidt, who helped with the DNA isolation, and Jessica Levy, who helped with the analyses of the polymorphisms. Both were very highly reliable and fun to be around. In addition, I am very grateful to Lilian Hülße, who analyzed the *CHRNA5* polymorphisms alongside of Ulrike Winnikes as part of her master thesis; both of them also had the idea that gender might be an important factor in the analysis of these polymorphisms.

Having now been in Trier for nearly three years, I would like to thank all of my colleagues in the department, as our Mensa visits have been daily sources of entertainment and it's been immensely pleasant to work with each of them. I especially would like to thank Nadine Nett, with whom I have

been friends since the first days of studying psychology. My utmost thanks also go to the brave, determined souls who proof-read chapters of this thesis: Tobi Tempel, Lisa Pramme, and Frank Mast. I cannot thank you enough!

Last but not least, I would also like to thank my family, my friends and my partner, all of whom have been highly supportive and also reminded me not to lose the bigger picture out of sight. I am grateful to Anatina Trakowski, Sarah Frank, Nadine Nett, and Katharina Maiwald for the close friendships that withstood the test of time and distance. My brother provided me both with much-needed distractions and with insights into what he calls “the real working world” (though knowing consultants, I’m suspicious of anything he tries to promote). My father has never been easy to faze and I know I can always count on him; for that I will always be more grateful than I can express. My grandmothers, on the other hand, are quite easy to faze and weren’t happy to see me move so far way beyond Siegen’s borders. Still, they supported me in all the ways they could, as they have done all my life. For this I deeply thank them. I would also like to express my gratitude to Benjamin Kirchhoff, who abandoned the Rhineland and moved to Trier with me. I considered this a huge sacrifice. Naturally, he managed to feel at home long before I did and now has gone fully native. Ben has provided unfailing moral support for me; I consider it invaluable that at the end of the day, we can quip and joke about anything. As a downside, of course, our high baseline setting for happiness has now thoroughly spoiled me.

Especially and most of all, I would like to thank my mother, Gabriele Schneider, who has supported me unconditionally and whole-heartedly for all my life. There is no goal that she hasn’t me thought me capable of reaching, and knowing that has always been a driving factor for me.

This thesis is dedicated to her.

List of Publications

Several parts of this dissertation were published in peer-reviewed journals during the course of the doctorate:

1. Schneider, K. K., Schote, A. B., Meyer, J., & Frings, C. (2014). Genes of the Dopaminergic System Selectively Modulate Top-down but not Bottom-up Attention. *Cognitive, Affective, & Behavioral Neuroscience.* Advance online publication. doi: 10.3758/s13415-014-0320-9

2. Schneider, K. K., Schote, A. B., Meyer, J., Markett, S., Reuter, M. & Frings, C. (2015). Individual response speed is modulated by variants of the gene encoding the alpha 4 sub-unit of the nicotinic acetylcholine receptor (CHRNA4). *Behavioural Brain Research.* Advance online publication. doi: 10.1016/j.bbr.2015.01.041

3. Schneider, K. K., Hülße, L., Schote, A. B., Meyer, J., & Frings, C. (in press). Sex matters! Interactions of gender and polymorphisms of a cholinergic receptor gene (CHRNA5) modulate response speed. *NeuroReport.*

Contents

List of Figures

List of Tables

List of Abbreviations

Abbreviation	Meaning
ACh	acetylcholine
ADHD	Attention-deficit/hyperactivity disorder
ADHD-SR	ADHD Self-Report checklist
ANOVA	analysis of variance
BA	Brodmann area
bp	base pair
C	Celsius
cDNA	complementary DNA
CFQ	Cognitive Failure Questionnaire
CHRNA4	cholinergic receptor, nicotinic, alpha 4 (receptor subunit)
CHRNA4	cholinergic receptor, nicotinic, alpha 5 (gene)
CHRNA5	cholinergic receptor, nicotinic, alpha 5 (receptor subunit)
CHRNA5	cholinergic receptor, nicotinic, alpha 4 (gene)
CNS	central nervous system
COMT	catechol-O-methyl transferase (enzyme)
COMT	catechol-O-methyl transferase (gene)
cp.	compare
D1R	D1 receptor
D2R	D2 receptor
DA	dopamine
DAT	dopamine transporter (protein)
DAT1	dopamine transporter (gene)
DLPFC	dorsolateral prefrontal cortex
DNA	deoxyribonucleic acid
dNTP	deoxyribonucleotide triphosphate
EEG	electroencephalograpy
e.g.	*exempli gratia* (for example)
et al.	*et alii* (and others)
FEF	frontal eye fields
fMRI	functional magnetic resonance imaging

i.e.	*id est* (that is)
kb	kilobase
LIP	lateral intraparietal cortex
LPFC	lateral prefrontal cortex
MANOVA	multivariate analysis of variance
MB-COMT	the membrane-bound COMT form
µl	microliter
min	minute
mM	millimolar
mRNA	messenger ribonucleic acid
ms	milliseconds
nAChR	nicotinic acetylcholine receptor
ng	nanogram
no.	number
NP effect	negative priming effect
p.	page
PCR	polymerase chain reaction
PET	positron emission tomography
PFC	prefrontal cortex
pmol	picomole
PNS	peripheral nervous system
PP effect	positive priming effect
RT	reaction time
s	second (time unit)
S-COMT	the soluble COMT form
SD	standard deviation
SOA	stimulus-onset-asynchrony
vs.	versus
VTA	ventral tegmental area
WCST	Wisconsin Card Sorting Test

Chapter 1

Selective Attention

Without selective interest, experience is an utter chaos.

(William James, 1890)

Attention has been researched since the dawn of cognitive psychology at the end of the nineteenth century. One of the founding fathers of experimental psychology, William James, proclaimed that:

> *"Everyone knows what attention is. It is the taking possession by the mind, in clear and vivid form, of one out of what seem several simultaneously possible objects or trains of thought. Focalization, concentration, of consciousness are of its essence. It implies withdrawal from some things in order to deal effectively with others, and is a condition which has a real opposite in the confused, dazed, scatter-brained state which in French is called distraction, and Zerstreutheit in German" (1890, p. 256).*

This quote has remained remarkably timely and profound – and yet it is not possible to entirely agree with it. Decades of enduring research on attention have led to the highly complex picture that we nowadays have of this concept. There is no all-encompassing theory of attention, nor is there much hope to draft one soon. Attention appears to be an almost evasive concept; only at first glance is it easily pinpointed. Most theories on attention are of a midsized scope, aiming to dissect the cognitive mechanics of specific subprocesses on the basis of a small number of paradigms (Wentura & Frings, 2012). In this regard, the research discipline resembles a construction site with no shortage of bricks, yet no construction plan (Wentura & Frings, 2012). This chapter is intended to provide an overview of the construct of attention in general and the construct of selective attention specifically. In subchapter 1.1, basal functions of attention will be outlined. In subchapter 1.2, different types of attentional selection in the visual field will be highlighted. Both subchapter 1.1 and 1.2 are meant to provide a framework of how attention can be broadly defined and categorized, both in a general sense and in regard to visuospatial selection. Subchapter 1.3 will then be focused on the research history of selective attention, elaborating on the causal necessity and understanding of the concept. Subchapter 1.4 is aimed at the dissociation of selective top-down and bottom-up attention (as well as related constructs). Finally, ways of measuring selective top-down and bottom-up attention will be presented in subchapter 1.5 and 1.6, respectively.

1.1 Approaching attention through its functions

Attention can be approached by classifying the *purpose* of different types of attention. An early classification by William James (1980) will be presented along with a modern, up-to-date view on the function of attention (Wentura & Frings, 2012). *Selection*, *planning/controlling* and *monitoring* are commonly defined as basal functions of attention (this classification has recently been summarized by Wentura and Frings, 2012). Selection is typically described as a mechanism that privileges goal-relevant stimuli, enabling target-oriented, purposeful actions. When driving a car, for example, drivers have to select stimuli that are relevant for cautious driving. A sign that signifies a speed limit, roadway damage, a traffic jam or other potential obstacles is a highly relevant stimulus. Roadside billboards, on the other hand, can be ignored without regret. *Planning and controlling* are needed to prepare and execute intentional, non-routinized actions. This component of attention is also required when driving a car, especially for beginning drivers who cannot yet rely on many automatized routines. Lacking experience, these beginners might have to make the effort of recalling specific traffic rules, or of recalling how to handle some parts of the vehicle. Driving will be a lot more difficult if the attentional load is high, i.e. if music is playing in the background or if other passengers make conversation. In this case, cognitive resources have to be splitted and distributed – this as well is part of the component of planning/controlling. *Monitoring* refers to the constant observation of the own surroundings. A driver has to notice abrupt changes; a ball that rolls onto the street is a good indicator that a child might soon trail behind. Analogously, the eyes of deer and other wild animals reflect the headlights of a car. While the animal itself might largely be hidden from view, the perception of "glowing eyes" should suffice as a warning and cause the driver to slow down. As Wentura and Frings (2012) remark, *selection, planning/controlling* and *monitoring* are not independent of each other. In real-life situations, all three functions are often highly intertwined. A driver not only implements driving rules, but is also on the lookout for relevant street signs. At the same time, the surroundings are non-consciously screened for abrupt changes. This intertwinement means that the different functions can interfere with each other (possibly because they draw resources from the same pool). When driving in an unfamiliar city, for example, picking the right lane at a busy intersection will be effortful and rely on detecting relevant cues. Less resources will then be available to monitor the surroundings for abrupt changes.

1.2 Visuospatial selective attention

Another way of classifying attention is by distinguishing what it is directed at (cp. Wentura & Frings, 2012). In the visual domain, the allocation of resources to different locations or elements in

space has been extensively studied (Klein & Lawrence, 2012). Which elements in the visual field are preferentially processed? The last decades of research yielded several answers to this question, and one can argue that attention can be *location-based*, *object-based* and *dimension-based* – three different types of visuospatial selection that are not mutually exclusive.

Location-based attention. Until the 1970s, it was the dominant view that eye movements can be equated with shifts in attention (Ansorge & Leder, 2011), i.e. that the visual focus is equivalent to the attentional focus. At that point in time, however, contrary results were already about a century old. The German scientist Hermann von Helmholtz (1821-1894) is widely known for his achievements in physics, but also investigated phenomena in the fields of medicine and psychology. Helmholtz gathered evidence that the focus of spatial attention can be shifted independently of eye movements (for an overview, cp. Ansorge & Leder, 2011). He examined attentional shifts via an enclosed box with a spyhole. A newspaper page was attached to the inner wall of the box, opposing the spyhole. The box was dark and the page was only visible to the participants when the setup was briefly illuminated. The participants were asked to fixate the center of the back wall of the box. Simultaneously, they were asked to either *attend* to the center of the back wall (here, the attentional focus equaled the visual focus) or to *attend* to the upper end of the box (here, the attentional focus was not equal to the attentional focus). If they were asked to attend to the center, the participants were able to read sentences in the middle of the page; if they were asked to attend to the upper corner, they were able to read the headlines (but not vice versa). In this way, Helmholtz could demonstrate that the focus of attention can diverge from the visual point of fixation (Ansorge & Leder, 2011). Similar results have been reported by Posner (1980) to a more widespread recognition (unsurprisingly – given that more than a century had passed – he was able to employ more rigorous methods). Posner distinguished between *orienting* (the allocation of attention to a stimulus) and *detecting* (the conscious awareness of a stimulus). He proposed that orienting can be attributable either to sensory input (in this case, the locus of control is *external*) or to processes within an organism (in which case the locus of control is *central*). Furthermore, he distinguished between overt and covert shifts in orienting. As the name suggests, overt shifts in attention are accompanied by eye movements (and often also head movements), so that they are observable from the outside. The benefit of overt shifts of attention lies in the alignment of the visual focus with the spatial region of interest. The visual focus is the area of sharpest vision and corresponds to the fovea, the part of the retina with the highest rate of detail sensitivity (Klein & Lawrence, 2012). Covert shifts of attention, on the other hand, involve the spatial allocation of attention in the absence of head and eye movements (Posner, 1980). Covert attention is not as easily investigable as overt attention and has to be inferred through carefully designed experiments (Klein & Lawrence, 2012). To this end, Posner utilized a task that is now called the Posner-

Cuing paradigm or spatial cuing paradigm (see subchapter 1.6 for an elaboration on this task). In this task, participants are asked to fixate a central point of the setup at all times. Cues appear prior to the appearance of the target. Some of these cues are presented peripherally, i.e. they can appear to either side (and in some distance) of the fixation point, at the possible locations of the target. The nature of these cues can vary (Klein & Lawrence, 2012); common are changes in luminance which are perceived as "light flashes". Cues that correctly indicate a target location are termed *valid,* while cues that do not indicate the target location correctly are termed *invalid.* Posner (1980) reported that valid peripheral cues led to reaction time (RT) benefits even prior to eye movements. Like Helmholtz, he concluded that the focus of spatial attention is dissociable from the visual focus. Posner, Snyder and Davidson (1980) likened the focus of spatial attention to a spotlight; according to them, the efficiency of stimulus detection is enhanced in the beam of the spotlight, the spatial area of the visual field that is attended to (and that is in not to be equated with foveal vision). One of the most debatable assumptions about the spotlight is the idea that its beam is constant in size (Müsseler & Prinz, 2002). There are strong indications that the size of the attentional spotlight can be adapted for the task at hand (LaBerge, 1983). In the *zoom lens* model, spatial attention is likened to the lens of a camera, where the size of the attended region can be adapted in a trade-off with the resolution (Eriksen & St. James, 1986). Accounts of the preferential processing of attended locations in the visual field have been challenged (or rather: expanded) by studies on the processing of objects.

Object-based attention. As target stimuli, Duncan (1984) presented two superimposed objects to the participants in his experiments: a box that was slashed by a line. Both the line and the box could vary in two feature dimensions (in case of the box, this was its height and the side on which a gap appeared; in case of the line, this was the tilt and texture). Duncan compared the performance of participants when they had to indicate the presence of two features either in the same object or in different objects. The dependent measure of these judgments was accuracy (in this regard it is noteworthy that the stimuli were only briefly displayed and that the exposure time was individually adapted to avoid both mere chance-levels of detection and ceiling effects). Participants judged, for example, the way the line was tilted and textured (same object, two judgments) or the way the line was tilted and the box was sized (different objects, two judgments). While two judgments on the same object could be made without interference, two judgments on different objects interfered with each other and were associated with a decreased level of accuracy in the second-reported dimension. In other words, it appears that several targets on the same objects can be processed simultaneously while several targets on different objects cannot be processed simultaneously. In this regard, it is important to factor in the spatial distance between separate objects. While Duncan (1984) used two superimposed objects, Egly, Driver, and Rafal (1994) utilized a different setup. In their experiments,

two rectangles were displayed (one of each side of a fixation point). In each trial, a cue appeared at one end of one rectangle, i.e. in one of four possible locations. It was the participants' task to indicate the appearance of the target that appeared subsequently at one of the four possible locations. The participants reacted more slowly when the target was invalidly cued (this is an effect of location-based attention). Importantly, however, these RT costs were significantly smaller when the invalidly cued location was part of the object on which the target appeared later on. It was beneficial, in that regard, if the attention was directed towards the target-object. It is important to note that each cue/target location was equidistant. Spatial distance therefore cannot serve as an explanation for this result. In other studies, single-object benefits were noted even when target elements on a single object were farther apart than target elements on distinct objects (Behrmann, Zemel, & Moser, 1998; Lavie & Driver, 1996). This is a clear indicator that location is not the only factor that influences the allocation of attention.

Dimension-based attention. Müller and O'Grady (2000) adapted the judgment paradigm by Duncan (1984). The participants were, for instance, asked to judge two attributes of a single object and two attributes of two objects (one attribute belonging to each object). When two different attributes had to be judged, the attributes could belong either to the same domain (for example, judging color: value and saturation) or to different domains (for example: saturation and size). Müller and O'Grady noted that the participants were more accurate when they had to make two judgments for the same object compared to two judgments for separate objects. Again, this is in support of object-based attention. However, the participants were also more accurate when they had to make judgments *within one domain* compared to cross-domain judgments. This benefit of a single domain was evident even when two objects instead of one had to be judged. The authors conclude that attribute- or dimension-based processing operates independently and in addition to object-based processing.

Summary. In conclusion, visual selective attention can be directed towards locations (independent of present objects), towards objects (independent of their locations) or towards features of objects (independent of whether they belong to the same or to different objects). All three types of attention can be dissociated from each other[1]. In everyday life, it is common to attend to locations, objects and features simultaneously (or to switch between attending to them). Wentura and Frings (2012) used the example of driving a car, where not only specific locations have to be attended (the road ahead), but also specific objects (other cars) and specific features (brake lights). While all three

[1] Results from visual search tasks indicate that a single feature (or so-called pop-out target) can be detected almost without delay no matter where in the visual field it is located (Treisman & Gelade, 1980). Such results are in favor of a dissociation of location-based and feature-based processing.

types of attention are aimed at different target segments in the visual field (are *spatially selective* in different regards), it is important to note that location-based, object-based and dimension-based attention are seen as independent from the basal functions of attention defined in subchapter 1.1 (cp., Wentura & Frings, 2012).

1.3 An overview on the research on selective attention

Providing selection is one of the basal functions of attention (Wentura & Frings, 2012). Through selection, a subset of stimuli is prioritized in relation to others, and this privilege typically benefits goal-oriented actions. Johnston and Heinz (1978) stated that "[…] we cannot be fully conscious of all the inputs that continuously flood our processing systems, some selection of perceptual information is needed […]" (p. 2). Selection is usually interpreted to mean that somewhere along the line of information-processing, the wide range of sensory perceptions is trimmed and cropped, and results in a limited number of intermediary cognitive processes and behavioral outputs. There are three imminent question that naturally arise from this assumption – namely the location of selection, the necessity of selection and the nature of selection.

The locus of selective attention. If there are limits in information processing, so-called serial bottlenecks (Anderson, 2007), where are they located? It is assumed that bottlenecks regulate the transition from *parallel* to *serial* processing, from a stage of processing in which all stimuli are analyzed in a simultaneous, parallel fashion to a slower stage of processing in which one process must be completed before the next one begins (Eysenck & Keane, 2010). Psychologists have been debating for decades if bottlenecks occur early or late in the sequence information processing. Another fervent point of debate has been centered on the criteria of the filter, and on the further fate of non-selected stimuli. Of particular interest for this field of research has been the task of dichotic listening. In this task, participants listen to a stream of messages on the right ear and a stream of different messages on the left ear and are asked to repeat one of these streams out loud (Cherry 1953). At the end of the session, the participants are asked what they remember of the message they were *not* asked to listen to. Typically, they remember very little (Anderson, 2007). Cherry (1953) reported that a change in the speaker was noticeable if the gender changed and that speech could be distinguished from noise. On the other hand, the participants were unable to indicate the language of the unattended message or specific contents of it. In his filter theory, Broadbent (1958) tried to explain this phenomenon. He proposed a rigorous filtering mechanism in an early stage of processing and hypothesized that relevant stimuli are selected on the basis of physical characteristics. According to Broadbent (1958), the

attended ear is the physical cue for the filtering process. All information presented there is deemed relevant and passed on for serial processing, while information from the unattended ear is not processed further. This assumption was shaken by several reports (Gray & Wedderburn, 1960; Corteen & Wood, 1972; Corteen & Dunn, 1974), among them the finding that participants recognize their own name in the message presented to the "unattended" ear (Moray, 1959). Since recognition requires a semantic analysis of the word content, this opposes Broadbent's idea that only physical characteristics like the auditory canal, the voice level or the sound type determine the filtering process. Treisman (1960, 1964a, 1964b) extended Broadbent's theory by the hypothesis that an early filter serves to attenuate less relevant information without eliminating it completely. According to Treisman, every information can be subjected to a hierarchical series of analyses, and stimuli like the own name, which are individually important, have a low threshold for detection. Another theory entirely was devised by Deutsch and Deutsch (1963). They assumed that all registered stimuli are processed simultaneously, whether they are attended to or not. A message is selected when it is deemed relevant for determining and executing a response. In this case, it is entered into working memory and subjected to further analyses. The selection of a message can sometimes be at odds with the instructions in this task, if participants choose the word meaning as the significant criteria for a response (rather than the ear the message is presented at). In the theory by Deutsch and Deutsch (1963), bottlenecks arise only due to limits of the response system, late in the course of information processing. The theories of Broadbent (1958), Treisman (1960, 1964a; 1964b) and Deutsch and Deutsch (1963) are three prominent accounts which seek to explain limits in information processing. Specifically, they seek to explain if selection occurs at an early, sensory stage, or after semantic analyses have been completed and the stimulus has been fully interpreted. These accounts appear to be at opposing ends of the spectrum. However, they can also be elegantly combined when flexible filters are taken into consideration. As in the other theories, Johnston and Heinz (1978) assume a procession of subsequent analyses, beginning with the perceptual analyses of a stimulus. According to their theory, selection takes place as early as feasible. If a response-determining selection can be made early, on physical characteristics like choosing the "right ear", then it is made early. If higher analyses are necessary, selection takes place in a later stage of processing. This flexible onset of selection is highly convenient under the assumption that selective attention is increasingly required in the course of information processing, and that attention in turn requires and drains resources. An early selection would therefore be efficient and desirable.

The cause of selective attention. The views on why selection is necessary differ fundamentally. In many of the earlier landmark theories, selective attention was described as a consequence of an inherent *system limitation* of the brain (Allport, 1989), as a mechanism that regulates and organizes scarcity (Hommel, 2010). Citing Mesulam (1985), Allport (1993) stated that attentional processes

would be superfluous if the brain had the capacity to process every information equally. It is assumed that processes of selection serve to protect the mind from overexertion, similarly to how a riverbed is protected from overflowing by building a dam. The notion that attention itself is a resource limited in capacity – one that can only be sparsely allocated – is a related concept (Allport, 1993). This focus on scarcity and natural limits in the cognitive architecture is called the *limited-capacity view* (Ansorge & Leder, 2011). The filter theory by Broadbent (1958) and the attenuation theory by Treisman (1960; 1964a; 1964b) are based on this point of view. Both of these theories have another key assumption in common: a distinction between a stage of parallel processing and a subsequent stage of serial processing. This view has been encapsulated by Neisser (1967), whose account has been largely influential for several other theories in the field of visual processing and feature integration (Treisman & Gelade, 1980; Treisman, 1985; Wolfe, 1994). Neisser distinguished between a *preattentive and parallel* as well as an *attentive and serial* stage of processing. He assumed that it would be impossible to process the whole available visual input simultaneously and in a detailed fashion. Instead, he proposed that global operations act on the whole visual field in a largely parallel fashion, prior to the employ of attention. According to Neisser, this preattentive stage of processing does not provide fine patterns, emotional content or actual awareness of objects, but contributes to perception by segmenting the visual field. Preattentive operations amount to a mechanical recognition system that creates the context of properties on which attention can act. "Focal attention" (p. 86) is then seen as necessary for more far-reaching analyses, which lead to the actual identification of objects in the visual field. According to Neisser, focal attention can be only be allocated to parts of the visual field; it is costly to engage and heavily draws from cognitive resources. In sum, the preattentive stage is thought to be fast, parallel, and is meant to prepare the visual field for further processing. In contrast, the attentive stage is seen as slower, serial, spatially selective, and is meant to provide deeper analyses. Neisser calls this two-level analysis of visual input a *constructive act.* How an object is perceived is supposed to depend on the selection of relevant information, and relevancy can vary across individuals (which means that properties are not automatically or inevitably processed). In a play on the key statement of Gestalt psychology that *the sum is more than its parts*, Neisser adds that *the whole is prior to its parts* (p. 91). The view that systemic limitations are a feature of the mind has been incorporated into many theories and models, often under different terms (Hommel, 2010). The *selection-for-action* account is diametrically opposed to the *limited-capacity* view (Allport, 1987). Selective perception is here not seen as a "necessary evil", but rather as an accomplishment of action control (Ansorge & Leder, 2011). At any given time, only a limited number of actions can be executed by an organism. A hawk, for instance, can only target a single bird out of a flock of birds. A tree may hang full of fruits, but only one apple can be picked by one hand at one point in time. Selective processes are then needed to map the target-related aspects of perceptual input to the control

parameters of an action system (Allport, 1993). The late-selection theory by Deutsch and Deutsch (1963) can be reinterpreted as a selection-for-action account, since selection is here driven by the needs of the output system, not by an inherent limitation of the mind. In this theory, it is assumed that all sensory input information is fully analyzed and weighted, yet only the goal-relevant informant is encoded and used to determine a response (Müsseler & Prinz, 2002). Hommel (2010) even went several steps further and labelled attention a "by-product of action control" (p. 134). He considered whether systematic interactions between particular types of actions and particular perceptual dimensions are derived phylogenetically or ontogenetically – in any case subsuming attention completely under an action system. In sum, it is the key assumption of the limited-capacity view that inherent limitations of the mind are the reason why selective attention is needed. In contrast, it is the key assumption of the selection-for-action view that selective attention causes limits in capacity.

The nature of selective attention. "Attention" is an umbrella term, denoting many different processes and effects. Ansorge and Leder (2011) stated that attention does not explain why perception is selective, stressing that the term itself is of a descriptive character first and foremost. In fact, the term has also been labeled a "pseudo-explanation for the phenomena we still fail to understand" (Hommel, 2010, p. 121). In the same vein, the subcategory of selective attention has been defined in a multitude of different ways. For example, *selection* can be used to denote either a causal mechanism *or* the result of a causal mechanism (Allport, 1993; Ansorge & Leder, 2011). In bottleneck models, selective attention has been viewed as a processing structure in the stream of information processing (Moors & De Houwer, 2006), for example a filter. It has, however, also been viewed as a resource that is variably allocated to the different levels of information processing, a fluid source of energy instead of a static structure (Moors & De Houwer, 2006; Johnston & Heinz, 1978). Even if one assumes that selective attention is a mechanism, there is no consensus on how this mechanism operates. Treisman (1960; 1964a; 1965b) assumed that selective attention is a filter, which attenuates less relevant information. Houghton and Tipper (1994) assumed that relevant stimuli receive excitatory feedback while irrelevant stimuli receive inhibitory feedback – a model in which attenuation and amplification are both implicated. Accordingly, the term "selective attention" can be used either to denote the fast detection of relevant stimuli or the ignoring (and possibly inhibition) of irrelevant stimuli (Allport, 1993; Ansorge & Leder, 2011). The multiple uses of the term can convey fundamentally different (and sometimes mutually exclusive) concepts. This ambiguity was described as "dreadfully confusing" by Allport (1993). In so far it is important to use a consistent definition of the term, to use measurements that are as clear as possible – and to be aware which conclusions can and cannot be drawn from them.

Summary. While the importance of selection is unquestioned, it is not yet clear where selection is located in the stream of information processing, how mechanisms of selection operate and why

these mechanisms have emerged (i.e., if they originate in a capacity limit of the mind or not). Still, such differing views should not be misunderstood as painting a bleak picture – quite the opposite is the case. Selective attention as a construct is at the heart of many fruitful debates in cognitive psychology. In the context of this dissertation, the questions of *where* and *why* will not be further highlighted. The question *how* selective mechanisms operate, however, will be considered in more detail. Of interest here are the subtypes of selective top-down and bottom-up attention (for an elaboration, see subchapter 1.4 – 1.6), specifically in regard to their neurobiological correlates.

1.4 Two sides of a coin: Top-down and bottom-up attention

Selective attention has been introduced in regards to its locus, nature and origin, and multiple effects where this type of attention is likely involved have been overviewed. In the following subchapters, two specific types of selective attention will be highlighted: *top-down* and *bottom-up selective attention*. There are quite a few concepts that are closely related to this distinction; three of them will be explained in more detail.

Endogenous and Exogenous Attention. The discrimination between these types of orienting is attributable to Posner (1980), whose terminology was in part already introduced in subchapter 1.2. Through his spatial cuing paradigm, Posner noted that attention can be allocated independently of eye movements (covertly). Moreover, he observed that the processing of central or symbolic cues (which trigger endogenous attention) differs from the processing of peripheral cues (which trigger exogenous attention). Pashler, Johnston, and Ruthruff (2001) stated that Posner modeled the concept of exogenous attention explicitly on the concept of a reflex. This adheres to the notion that exogenous attention is characterized by a huge degree of automaticity, that the process of attention-drawing by salient peripheral stimuli is inevitable once it is initiated. Jonides (1981) tested the effects of central versus peripheral cues in a series of experiments. In these, it was the participants' task to indicate if the letter L or R was present in an array of eight letters that were placed on the circumference of an imaginary circle. Cues preceded the target display: arrowheads that pointed to the position of a letter. The cues were central if they were placed in the center of the imaginary circle. They were peripheral if they were placed on the circumference of the circle, next to a letter location. In the first experiment, Jonides tested whether a simultaneous working memory task interfered with the performance in the cuing task. To this end, participants saw up to seven randomly chosen digits before the beginning of a

trial. After the trial, they were asked to recall the string of digits. The validity effect[2] in trials with central cues decreased under an increasing memory load. In contrast, different memory loads had virtually no impact on the validity effect in trials with peripheral cues. The second experiment was identical to the first experiment, with the exception that the memory task was omitted. In this experiment, however, the participants were instructed on the validity of the cues. In the first subgroup, participants were informed that the likelihood of a correct indication of the target location was fairly low. Still, they were instructed to attend to the cues, as they were on average (supposedly) beneficial for their performance. In the second subgroup, the participants were informed that the cues were only randomly coupled to the targets and that it was best to ignore them altogether. The validity of both central and peripheral cues amounted to only 12.5 % in this experiment, so that it was most likely that the target did *not* appear at the location that the cue indicated. The participants that were asked to ignore the cues were on average 2 milliseconds (ms) slower in trials with valid central cues compared to trials with invalid central cues. In contrast, these participants were on average 98 ms faster in trials with valid peripheral cues compared to trials with invalid peripheral cues. Thus, the participants were able to ignore central cues, but unable to ignore peripheral cues; a RT difference between valid and invalid trials with peripheral cues still persisted. In the third experiment, the participants were informed about the probability of cue occurrence. The task was identical to the one used in experiment one (with the exclusion of the memory task). The first subgroup of participants was informed that central cues would appear on 80 % of the trials and peripheral cues on 20 % of the trials. The second subgroup was informed that peripheral cues would appear on 80 % of the trials and central cues on 20 % of the trials. In both groups, the participants were informed about the cue validity (which was 70 %). Jonides noted that the effectiveness of central cues was directly related to the expectations of the participants. In contrast, peripheral cues effectively modulated the performance even if they were not expected to occur frequently. In sum, Jonides (1981) concluded that peripheral cues did not heavily engage cognitive resources, and maintained their attention-capturing property even the cue validity was enormously low, if the participants were instructed to ignore them and if they were informed about their low occurrence. In addition, peripheral cues produced greater effects on RTs than central cues. For these reasons, Jonides (1981) concluded that there are two modes of attentional control. In part this may be due to the deeper encoding of central cues. Jonides specified that central cues, due to their symbolic nature, need to be interpreted, while peripheral cues appear at the target location (or close to it), so that the determination of their location is a sufficient analysis. Jonides speculated that the more extensive processing of central cues renders their impact more vulnerable to interfering processes. On a related note, Briand and Klein (1987) investigated whether the concept of Posner's "beam" (1980) can be seen as equivalent to the concept of Treisman's and Gelade's "glue" within the

[2] The validity effect is the mean RT difference between valid and invalid trials.

Feature Integration theory (1980). They did this by using a spatial cuing task with peripheral and central cues. The letter "R" served as target and had to be identified. The target could be distinguished from distractor letters either on basis of a single feature (feature search) or required a more extensive comparison (conjunction search). Referring to the Feature Integration theory, this means that a parallel search would be sufficient in the first case but not in the second. The exogenous orienting of attention, triggered by peripheral cues, interacted with the search type that was required. The effect of cuing was larger when peripheral cues were used in a conjunction-search task. The orienting of attention in relation to central cues was similar in both search conditions. This result was replicated by Briand (1998) and was interpreted to mean that Posner's "beam" and Treisman's and Gelade's "glue" are indeed one and the same process – i.e., the attention that is drawn by peripheral cues helps to implement the feature integration of objects. Reviewing these and other studies (Briand & Klein, 1987; Briand, 1998; Klein, 1994), Klein and Lawrence (2012) conclude that exogenous attention and endogenous attention do not differ in the mode of control over the same "beam" of attention (cp. Jonides 1981), but originate from separate sources. In conclusion, the orienting of attention can be triggered endogenously or exogenously (Berger, Henik, & Rafal, 2005). Differences between these cues lie, for example, in their varying proneness to modulation by cue validity, cue contingencies, and instructions on their processing – or in other words, in their degree of controllability. On a grand scale, everyday life can be seen as an interplay of competing demands on visual orienting, arising either from internal goals (endogenously) or external events (exogenously) (Berger at al., 2005).

Automatic and Non-automatic Processing. The concepts of endogenous and exogenous attention are closely related to the concept of automaticity – they are not wholly identical, but overlap in some parts. Jonides (1981), in fact, proposed that central and peripheral cues differ in the extent to which they draw attention automatically. What Posner (1980) had termed "exogenous control", Jonides termed "automatic control"; what Posner had termed "endogenous control", Jonides termed "voluntary control." Generally, it is assumed that automatic processes require little to no voluntary effort. It is also assumed that they are performed fast and in a parallel fashion, use up little attentional resources, are not consciously accessibly or consciously controllable, and are mostly relevant in the context of comparatively easy or highly practiced tasks. In addition, they are presumed to involve a low depth of cognitive processing. The opposite characteristics are assumed for non-automatic processes[3] (cp. Sternberg, 2008). Accordingly, it was frequently assumed that tasks can be performed

[3] It should be noted that many different terms are used to denote the opposite of automatic processes. Among those terms are the following: *controlled, effortful, conscious, attentional, deliberate* and *strategic* (Moors & De Houwer, 2006). Moors and De Houwer (2006) prefer the term "non-automatic" due to its neutrality.

in two modes of processing (Shiffrin & Schneider, 1977; Shiffrin, Dumais, & Schneider, 1981). However, this all-or-nothing point of view is something of an oversimplification and has been replaced by other concepts (Moors & De Houwer, 2006). Non-automatized processing can lead to automatized processing. Formerly novel tasks that required many resources to accomplish can be automatized with practice. Driving a car, for instance, becomes second nature, to the point that a driver can react to unforeseen events without employing conscious control over his actions. If another car swerves into the own lane on the freeway, a seasoned driver will have little difficulties in swerving as well to avoid a collision. Such a swift, automatic reaction can be highly problematic in itself, if the own car is then steered into another lane without checking for other vehicles first. In short, automatic processes have the advantage of speed, while controlled processes have the advantage of flexibility (Eysenck & Keane, 2010). Shiffrin and Schneider (1977) let participants judge whether a stimulus in a display-set matched stimuli of a previously learned memory-set. After 2100 trials, participants were much faster in identifying matches. When the task was reversed so that the memory set was now constituted of former distractors, performance suffered drastically. It took 2400 trials to reach the hit rate that had been reached after 1500 trials in the original test. It can be concluded that it takes practice to automatize actions and even more effort to alter an implemented, automatized routine. There are several problems with traditional approaches to automatic and non-automatic processing, among them the custom to use the terms *automatic* and *non-automatic* descriptively, with no explanation for the presumed transition from a slow, serial mode of processing to a fast and parallel one (Eysenck & Keane, 2010). Moors and De Houwer (2006) disentangled and analyzed the features that are usually subsumed under the concept of (non-)automaticity. They concluded that four features are left if overlapping portions are subtracted: *goal-relatedness, efficiency, consciousness,* and *speed.* The authors caution that these features are interrelated in a complex fashion, so that the presence or absence of one feature is no simple indicator for the presence or absence of other features. Logan (1985) proposed that practice changes these features independently of each other; that uniform time-courses cannot be expected. It is essential to consider how attention is related to these four features of automaticity (for the following paragraph, cp. Moors & De Houwer, 2006). *Goals* determine where attention is directed; this may extend not only to sensory input but also at the processes that operate on the sensory input ("processing goals"[4], p. 317). A process can be defined as *efficient* if it does not engage many attentional capacities (this corresponds to the notion that attention is a flexibly deployable resource). *Consciousness* has been closely linked to attention; the accessibility of memory content and perceptual experience is commonly seen as grounded on attentional processes (Cowan, 1995). Processing speed is linked to consciousness and in this way also linked to attention. Speed can

[4] These goals do not need to be conscious.

depend on many contextual factors and its relation to attention is therefore complex. Generally, well-practiced, unconscious actions are assumed to be quicker than conscious actions. On the other hand, non-practiced, conscious actions can benefit from a larger degree of attentional involvement and can then be quicker to execute. Of the four features, efficiency is most closely related to attention, as this concept is defined by the degree of its attentional engagement. Traditionally, it was assumed that automatic processes place no strain on attentional capacity. This notion cannot be upheld anymore (Eysenck & Keane, 2010; Kahneman & Chajczyk, 1983). Moors and De Houwer (2006) are in favor of a relative concept of automaticity, in which a process can be judged as *more automatic* or *less automatic* than another process, based on a subjective criterion. This approach differs not only from the strict all-or-nothing view of automaticity, but also from the gradual view of automaticity. According to the gradual view, automaticity can be specified according to its position on a continuum; the poles of this continuum signify the absolute extreme of automaticity and non-automaticity (Moors & De Houwer, 2006). The problem here is that such a continuum is difficult to quantify objectively, and moreover that it is questionable whether the extreme poles are even hypothetically possible (Moors & De Houwer, 2006). As a solution then, Moors and De Houwer propose a subjective criterion which serves as a standard of comparison. For example, one could describe a process as more automatic than it was before practice, or as more automatic than another process.

Top-down and Bottom-up Attention: Intersections and Differences to Related Concepts. The principles of top-down and bottom-up processing are essential to cognitive psychology. At their broadest, most general definition, they denote that processing can be driven by stimulus inputs or by factors within an individual, like past experiences and expectations (Eysenck & Keane, 2010). The terms *top-down* and *bottom-up* do not refer to attention exclusively. They can be applied to virtually every facet of cognition (Sternberg, 2008) and indicate the interaction of two opposite-directed forces; ultimately signifying the interaction between events occurring in an individual and events occurring in the environmental surroundings. Pashler and colleagues (2001) stated that it is critical to tease apart the principles that govern this interaction if one wants to understand *any* type of human behavior. In regard to attention, Sarter, Givens, and Bruno (2001) defined the terms as:

> *"'Top-down' or 'bottom-up' regulation of attentional processes represent conceptual principles rather than referring to anatomical systems, such as ascending and descending projections. 'Top-down' processes describe knowledge-driven mechanisms designed to enhance the neuronal processing of relevant sensory input, to facilitate the discrimination between signal and 'noise' or distractors, and to bias the subject toward particular locations in which signals may appear. […] Such a 'top-down' biasing of attentional performance contrasts with 'bottom-up' perspectives that describe attentional functions as driven mainly by the characteristics of the target stimulus and its sensory context. 'Bottom up' perspectives attempt to explain a subject's ability to detect targets and target-triggered attentional processing largely by the sensory salience of the targets, and their ability to trigger attentional processing by recruiting 'higher' cortical areas in a bottom-up manner (e.g., from the processing of a visual target in the primary visual cortex to temporal regions for object identification and to parietal regions for location). Importantly, 'top-down' and 'bottom-up' processes represent overlapping organizational principles rather than dichotomous constructs, and in most situations, top-down and bottom-up processes interact to optimize attentional performance" (pp. 147.-148).*

Selective top-down attention, specifically, is defined as a bias on grounds of cognitive long-term strategies or short-term goals (Connor, Egeth, & Yantis, 2004), resulting in the deliberate prioritizing of stimuli that are deemed goal-relevant (Schneider, Schote, Meyer, & Frings, 2014). Selective bottom-up attention, on the other hand, consists in filtering the sensory input for salient features, subsequently resulting in attentional shifts towards salient stimuli (Connor et al., 2004; Schneider et al., 2014). In contrast to bottom-up attention, top-down attention is assumed to be under voluntary control (Pashler et al., 2001). Human behavior can be imagined as lying on a continuum of top-down and bottom-up processes (Pashler et al., 2001); though perhaps for scientific practice, a relative approach is more suited than a gradual one. Even though the terms "top-down" and "bottom-up" were not used by Posner (1980), they were clearly implied when he described the driving forces behind the orienting of attention. Posner stated attention could be aligned either due to "a search plan internal to the organism" or due to "stimulus input" (p. 5); the literal translation of the word "endogenous" means *proceeding from within*, the literal translation of the word "exogenous" means *proceeding from outside*. In this way, Posner's concepts of exogenous and endogenous orienting clearly overlap with the top-down/bottom-up concept. Conversely, the orienting of attention constitutes only a minor portion of the cognitive processes that can be described in terms of top-down and bottom-up forces. To use a closely related example in the visual domain, attention is not always directed to a specific location, but can also travel randomly and then come across goal-relevant or especially salient stimuli (Moors & De Houwer, 2006; Fox, Russo, Bowles, & Dutton, 2001). Posner's modes of orienting are not

only related to the top/down principle, but also to (non-)automaticity. Observing differences in the processing of central and peripheral cues, Jonides (1981) assumed that exogenous control equaled a more automatic control, while endogenous control equaled a more voluntary control. This assumption that exogenous orienting is faster, more efficient, less conscious and less goal-related can certainly be shared. Generally, the concept of the exogenous and endogenous control of orienting benefits from its clear, precise definition. In comparison, (non-)automaticity is a much broader concept that encompasses not only different modes of orienting, but a huge array of processes in all sensory domains. The relationship between (non-)automaticity and top-down/bottom-up attention can almost be condensed to one key feature: voluntariness or goal-relatedness. Bottom-up shifts of attention are frequently describes as *involuntary*, top-down shifts of attention are frequently described as *voluntary* (Connor et al., 2004; Theeuwes, 2010). In the first case, control lies outside the individual; in the second case, control lies inside the individual. Theeuwes (2010) stated that "[top-down] selection is completely volitional: at any time, a person can choose at will from the environment what to select" (p. 77). Moors and De Houwer (2006) considered purely stimulus-driven processes to be unintentional and unrelated to proximal or remote goals. In contrast, top-down attention can then be defined as more goal-related, or as an "active volitional process" (Theeuwes, 2010, p. 77). *Active* implies awareness. It is noteworthy, then, that goal-relatedness does not fully equal volitional control; Moors and De Houwer (2006) state that goals can be unconscious as well. In sum – trying to disentangle the different concepts previously introduced – it can be stated that the exogenous and endogenous orienting of attention can be largely subsumed under the concepts of top-down/bottom-up attention and (non-)automaticity, but not vice versa. Top-down/bottom-up attention and (non-)automaticity partially overlap, but should not be equated. There is, however, a component of *less automaticity* to top-down attention. For visual selection, Theeuwes (2010) concluded that attention be allocated either voluntarily and in accordance with an individual's goals, or can be allocated to salient stimuli regardless of their value for the current goal-set. This process has been termed *attentional capture*. For instance, it can be said that peripheral cues in a Posner-Cuing task *capture* attention. Theeuwes (2010) described this process as a "passive automatic way" (p. 77), implicating that the process is not consciously initiated and executed. It has been a point of much debate what contributes to stimulus salience, i.e. what properties actually make a stimulus stand out from its context and lead to attentional capture (Pashler et al., 2001). There are opposing theories on the role and impact of selective top-down and bottom-up attention, respectively. Folk and colleagues (1992) designed an experiment in which the color and the abrupt onset of both targets and distractors was varied. They found that distractors only capture attention when their properties overlap with those of the target – i.e., color distractors are salient in the context of color targets, and abrupt-onset distractors are salient in the context of abrupt-onset targets, but not vice versa (also cp. Pashler et al., 2001). This finding could be replicated with

three different dimensions as well (Folk, Remington, & Wright, 1994). These results indicate that top-down processes might dominate bottom-up processes. It was proposed that bottom-up processes are not hard-wired – i.e., that involuntary shifts of attention are not triggered like a reflex at any salient input (Folk et al., 1992; Pashler et al., 2001). In conditions of spatial uncertainty, these processes are assumed to respond selectively to those properties that can potentially predict the location of a target (Folk et al., 1992). The authors termed the involuntary capture of attention within the frame set by top-down goals *Contingent Attentional Capture* (Folk et al., 1992; Folk et al., 1994; Remington, Folk, & McLean, 2001). It is important to note that involuntary shifts of attention are not negated in this model – rather, their sphere of impact is redefined. Folk and colleagues (1992) compared this to the programming of a software. According to situation-specific goals, a code is compiled. This code specifies the internal control settings of the software. It determines which stimuli properties will lead to shifts in attention. Once the program is running, however, those stimuli that capture attention do not do so in a voluntary, controllable way. In other words, the configuration of the control setting occurs *offline*, while the automatic allocation of attention occurs *online* (Pashler et al., 2001). The *Contingent Attentional Capture* account favors a dominant role of top-down control. In contrast, a dominant role for bottom-up attention has been proposed by Theeuwes (2010). He assumed that there is an *attentional window* in the visual field; this window can be adapted in size (attention can be more or less spread-out). Theeuwes assumed that preattentive processes take place exclusively within this window and provide computational analyses of salience. In alignment with these, the object with the highest salience (the object that is most unlike its context) is then selected for further processing. This selection is supposed to be spatial and completely subsumed under bottom-up control; top-down goal sets do not factor into this process. Once it has been selected, the object can be identified. Before selection, it is salient; after selection, the object can be identified as something bright red on a green background[5]. When the object has been identified, it is matched to internal goals. If it is goal-relevant, a response is made. If it is not relevant, top-down attention facilitates a quick disengagement of attention and inhibits this location. The approach of Theeuwes (2010) is similar to the *Contingent Attentional Capture* approach insofar as analyses take place only in a pre-set frame that can be voluntarily modulated – but: the attentional window denotes only the spatial distribution of attention and is not related to internal goal-sets. The initial wave of analyses take place without top-down involvement; it is presumed that the spatial selection of an object is first and foremost driven by bottom-up processes. In contrast, it is assumed by the *Contingent Attentional Capture* approach that salience itself is preset in a top-down fashion. In sum, visual selection is attributable to top-down and

[5] Theeuwes assumed that top-down knowledge cannot affect the preattentive processing stream because dimensions are not identified at this stage of processing (the categories here are “like” and “unlike”, not “red” and “green”).

bottom-up processes to varying degrees. Some accounts favor a dominant role of top-down attention; others favor a dominant role of bottom-up attention.

1.5 Measuring selective top-down attention

The Stroop task. This task is one of the most well-established paradigms in cognitive psychology, no doubt due to the fact that the *Stroop effect* or *congruency effect* obtained here is both large and robust (MacLeod, 1991) and serves to illuminate the processing of interference and response conflicts. The task is named after its inventor, John Ridley Stroop, who reported a series of three experiments in 1935. Stroop let participants read two sheets of words with a hundred words each. One of the sheets contained words (red, blue, green, brown, purple) that were printed in matching (congruent) or non-matching (incongruent) colors. The other sheet contained the same sequence of words but was printed in black. In the first experiment, it was the participants' task to read the sheets as quickly as possible. A comparison of the overall time needed for each sheet indicated no effect of interference between the word content and the word color. On average, the participants needed 43 seconds (s) to complete the sheet with color words and 41 s to complete the sheet with black words. In the second experiment, it was the participants' task to name the print color as quickly as possible. As before, they were given a sheet of color words printed in different colors. This time, the second sheet did not feature words but an equal number of colored squares. The color had to be indicated here as well. On average, the participants needed 110 s to complete the sheet with colored words and 63 s to complete the sheet with colored squares. While no test of significance was applied by Stroop, he reported that the difference between both conditions equaled 4.35 standard deviation units in terms of the square-color-naming performance. So far, the results can be summed up to mean that participants experienced little trouble in reading words, no matter what color they were printed in. On the other hand, they did experience difficulties when stating the print color of a word with color content (e.g., "blue" printed in red). This phenomenon is now known as the *Stroop effect.* In his third experiment, Stroop examined the effect of practice and found that after participants were on average 34 % faster in the color-naming condition on the last day of practice. However, as MacLeod (1991) notes, the interpretation of this result is rendered difficult due to methodological problems. Modern versions of the Stroop task differ from the original variants in several ways. Usually, the task is now computerized and the RT is measured in relation to each stimulus, not in relation to whole lists. Instead of verbally stating the word color, manual reactions are often required of the participants (e.g. by pressing keys on the keyboards). The task has also been modified more drastically (MacLeod & Dunbar, 1988; Flowers, Warner, & Polansky, 1979; for an overview, see again MacLeod, 1991). Nevertheless,

the phenomenon of *interference* is still at the core of the Stroop task. Interference refers to incongruent stimuli in which the color does not match the content. These stimuli typically lead to slower RTs and more errors if the color has to be indicated (MacLeod, 1991), the reason for this effect being that the processing of the feature dimension content *interferes* with the feature dimension color. One process (reading) hinders and obstructs another process (color naming). A popular explanation of this effect has been made with the automaticity account (MacLeod, 1991), which states that reading is such a well-practiced action that its execution is automatic and cannot be suppressed. Naming a color, on the other hand, is a skill that is rarely needed and therefore much less automatic. As stands to reason, the role of automaticity for the Stroop effect can by examined by training participants in this skill. MacLeod and Dunbar (1988) did exactly that and reported that a less practiced action became more automatic and then even interfered with the more practiced action (in a manner of speaking, this can be seen as a reversal of Stroop's first experiment). In conclusion, the Stroop task is ideally suited to examine selective top-down attention, since one specific feature (color) has to be selected against others and because in doing so, a less practiced process has to be shielded against a reasonably more practiced process. As color is only more goal-relevant because it was instructed as such, this relies on top-down control.

The Negative Priming task. This task is closely related to the Stroop task. While task-relevant features and task-irrelevant features are part of the same object in the Stroop task, different stimuli are usually used as targets and distractors in the Negative Priming task. In a typical sequence of this task, a *prime* trial is succeeded by a *probe* trial; in both cases, participants attend to a target and try to ignore additional stimuli that require no response (Fox, 1995). Neill (1997), for example, used a setup in which a string of three letters was presented in the prime and probe trial. Participants had to indicate which letter was presented at the location in the middle (target identification task). The two flanking letters were always identical to each other and required no response. A repetition of the target from prime to probe usually results in a *positive priming* (PP) effect, i.e. a facilitated response to the target in the probe trial (Fox, 1995). This facilitated processing can be manifested in shortened RTs and smaller error scores. In contrast, the *negative priming* (NP) effect denotes a slowdown in RTs and a possible increase in errors if the distractor of the prime trial becomes the probe target (May, Kane, & Hasher, 1995). The NP effect shows that the ignoring of an object interferes with its subsequent processing as response-relevant. A large variety of different NP tasks have been introduced, covering a plethora of research questions (for an overview, see Fox, 1995, and May, Kane, & Hasher, 1995). Tipper (1992) proposed that dual processes are at work in selective attention: excitatory processes aimed *towards* a target and inhibitory process aimed *away* from distractors. According to Houghton and Tipper (1994; 1996), the NP effect is caused by the interference of these processes. More precisely,

Houghton and Tipper assume that perceptual input receives excitatory input upon registering and is matched against an internal template. The input can then be identified as a target and receive internal support, i.e. further activation, or be identified as a distractor and receive inhibitory feedback. In this context, the authors speak of the "excitatory foregrounding of targets and inhibitory backgrounding on nontargets" (Houghton & Tipper, 1996, p. 26). When the distractor stimulus is not displayed anymore, its internal representation still receives inhibitory feedback but stops to receive the excitatory feedback derived from its visual displaying. In this case, Houghton and Tipper assume that the stimulus becomes suppressed below background levels of activation. They term this an *inhibitory rebound.* Rebounds are assumed to be very transient phenomena. When a previous distractor is displayed as a target in this time interval, however, it is need of a larger activation than a wholly novel input would be. This "overcoming" of the inhibitory rebound is assumed to be at the core of the NP effect (Houghton & Tipper, 1996). Another theory of the NP effect has been formulated with the so-called Episodic Retrieval account (Neill, 1997; Mayr & Buchner, 2007). In sum, the Negative Priming task is well suited to measure selective attention, since a target has to be selected among non-targets here. At the same time, the task taps top-down attention. As in the Stroop task, the relevancy of the target is based on (arbitrary) instructions. In the case of distractor-to-target trials, what is arguably the dominant response has to be overridden and another response enforced instead (reacting instead ignoring).

1.6 Measuring selective bottom-up attention

The Posner-Cuing task. This task has become a staple in the research on covert orienting, both for exogenous and endogenous attention (Posner, 1980; Posner, Nissen, & Ogden, 1978; Klein & Lawrence, 2012; Wenzel, 2012). In this task, participants are instructed to keep a fixation cross fixated at all times; this is crucially important to avoid overt shifts in attention. Typically, two boxes are located next to the fixation cross, one on each side. The target is preceded by a cue that validly or invalidly indicates the target position. Arrows are used as central cues; they replace the fixation point for a short interval and point to one of the boxes. Peripheral cues are salient on a sensory level (e.g., changes in luminance, which are perceived as a bright flash) and are typically presented at a location where the target can appear. The target appears in one of these boxes and is sometimes presented alongside with a distractor (that appears in the other box). Most often, participants are asked to indicate the appearance of the target (detection task). In newer studies, target identification – for example in regard to color or form – is also required more frequently (Wenzel, 2012). Responses are typically facilitated in valid trials and slowed in invalid trials. These effects are termed *benefits* and *costs*, respectively. The average RT difference between invalid and valid trials results in the *Posner effect* or

validity effect, which is greater when peripheral cues are used (Jonides, 1981). The time courses of facilitation and inhibition differ characteristically from central to peripheral cues (Müller & Rabbitt, 1989). Various modulations of the Posner-Cuing task have already been introduced in subchapter 1.2 and 1.4. In a Posner-Cuing task with peripheral cues, Posner and Cohen (1984) manipulated the stimulus-onset-asynchrony (SOA), the time interval between the onset of the cue and the onset of the target. As before, valid cues facilitated the subsequent response to the target at short SOAs. If the SOAs exceeded 200 – 300 ms, however, valid cues led to RT costs (cp. Klein, 2000). This effect is termed *inhibition of return,* and is interpreted as biasing orienting away from already examined objects, and thus encouraging orienting towards novel objects (Klein, 2000). In sum, the Posner-Cuing task is well suited to illustrate spatial selectivity and the exogenous variant (in which peripheral cues are utilized) is ideal to measure selective bottom-up attention.

The Dot Probe task. This task can be seen as an affective variant of the Posner-Cuing task (Wenzel, 2012) and measures selective attention to emotionally salient stimuli (MacLeod, Mathews, & Tata, 1986; Koster, Crombez, Verschuere, & De Houwer, 2004). In the first experiment in which this task was utilized (MacLeod et al., 1986), the sequence of events was the following: two words were presented in each trial, one above the other. One word was always threat-related while the other word was always neutral. In a third of the trials, a dot appeared at one of the word locations. It was the participants' task to read the upper word aloud and to react to the appearance of the dot by pressing a button. MacLeod and colleagues (1986) reported that participants with a generalized anxiety disorder were faster to detect the dot when it replaced a threat word instead of a neutral word. While this task was first developed to investigate attentional biases in the context of psychological disorders, it has been adapted and used in a broader context since then (Wenzel, 2012). The simultaneous presentation of two cue stimuli – for example words, but often also pictures – is still at the core of the task (Wenzel, 2012). The participants are then asked to indicate the presence of a subsequently shown target, most often a dot or a circle. The required response to it can vary; participants can be asked to indicate the presence of the target irrespective of its location (detection task), or to indicate the position of the target (probe location task) or to identify one of several targets, i.e. the type of target (probe classification task) (Wenzel, 2012; Mogg & Bradley, 1999). It is not common anymore that word cues are read aloud, or that a specific target location is highlighted (as it was the case in the original experiment, see MacLeod et al., 1986). A typical result in the Dot Probe task is the faster response to a target that was preceded by emotional cues (Wenzel, 2012). This effect is especially noticeable if disorder-specific cues are used in a sample of participants with that disorder (Wenzel, 2012). The relative effect of different types of cues was tested in a study by Brosch and colleagues (2011). In this study, participants fixated a cross which was then replaced by an arrow (a central cue). The arrow

would either point to a box on its left or on its right side. Afterwards, the arrow was replaced by a fixation cross and two peripheral stimuli were presented simultaneously. In each box, a picture of was shown (one always neutral and one always fearful, the latter being the peripheral emotional cue). At the same time, the frame of one of the boxes was thickened. This was perceived as a bright flash by the participants (the peripheral neutral cue). Both the central cues and the peripheral neutral cues that were used in this study correspond to the stimuli that Posner (1980) utilized to study endogenous and exogenous attention. The peripheral emotional cues that were used are typical Dot Probe stimuli. While the central cues were valid in 70 % of the trials, the peripheral exogenous and emotional cues were valid in 50 % of the trials. All three cue types were orthogonally manipulated, so that their effects could be dissociated. Brosch and colleagues (2011) reported that the cues had additive effects in the condition with a SOA of 100 ms. The RTs decreased linearly with the number of valid cues, being shortest when all cues were valid at the same time, i.e., when all cues correctly indicated the location of the target. In the long SOA condition (800 ms), only the central cues had an effect on the RTs. The participants were faster if the arrow correctly indicated the target location. The authors concluded that both peripheral types of cues operate in a similar fashion and cause effects that are of a rapid, reflexive, quickly decomposing nature. Since their execution did not mutually interfere with each other, it can be assumed that separate mechanisms must be at work behind them. This interpretation was strengthened by results from an electroencephalography experiment in the same study (Brosch et al., 2011), where the temporal loci of the effects of both cue types were disentangled. Both cue types were valid in 50 % of the trials and thus had no predictive value. Nevertheless, participants proved to be faster if the peripheral cues correctly indicated the target location – proving that they attended to them. In sum, it can be concluded that both types of cues trigger stimulus-driven bottom-up processes (Brosch et al., 2011). Since these processes are not identical, the Dot Probe task has an additional value in relation to the Posner-Cuing task, tapping into emotional processing.

Summary. Both the Stroop task and the Negative Priming task are well suited to measure selective top-down attention. In their most typical forms, the task are non-spatial, i.e. participants are not asked to attend to multiple areas of a display but rather to one location (which means that the overt or covert orienting of attention cannot be examined with these tasks). Usually, target identification is required of the participants. The attentional control is assumed to be more top-down driven, as it is voluntarily directed towards the target. In contrast, both the Posner-Cuing task and the Dot Probe task can be used to measure selective bottom-up processes. Both tasks measure spatial selectivity and tap covert attentional processes. In case of the Posner-Cuing task, target detection is most often required of the participants. In case of the Dot Probe task, the required responses vary. The attentional control is assumed to be more bottom-up driven in both tasks, as it is not voluntarily directed towards the target and is dependent on the salience of the stimulus material.

Chapter 2

The Dopaminergic and Cholinergic Neurotransmitter Systems

The brain is a world consisting of a number of unexplored continents and great stretches of unknown territory.

(Santiago Ramón y Cajal)

In the previous chapter, the complex psychological concept of attention was introduced. Traditionally, psychologists have been fond of "box models" to explain cognitive processes. Broadbent's filter model (1958), for example, has often been visualized as a box model. The sensory input that reaches the receptive organs can be symbolized by *arrows,* which are directed at a box entitled *selective filter*; the input that passes the filtering process at this stage can be forwarded to another box entitled *limited capacity decision channel*, and so on (cp. Treisman, 1964a; 1964b). Usually, the proceedings within a box are relatively well described, but the transfer of information from one box to another remains as nebulous as the neurobiological correlates of the entire sequence of processing. Of course, cognitive processes do not take place in a vacuum, but in the biological medium of the central nervous system (CNS). For decades, research disciplines focused on the mind and research disciplines focused on the brain have – in a manner of speaking – lived parallel lives. This changed significantly in the 1990s with the rise of new methods that made the inner workings of the brain more accessible. Prominent among those are imaging techniques and methods in the field of molecular genetics. Brain-centered and mind-centered research disciplines have now grown considerably closer and the vibrant field of *Cognitive neuroscience* attests to that. This chapter will be focused on ways in which neurotransmitter modulate cognitive processes, both in regard to attention and in regard to response speed. Neurotransmitters are sometimes also called *messenger substances,* and this is exactly what they are – the carriers of information within the CNS. Neurotransmitter molecules are released from the axon terminal of one neuron and bind to receptors on another neuron, thus enabling communication between neighboring cells (von Bohlen und Halbach & Dermietzel, 2006). These points of contacts between neurons are termed synapses. Importantly, neurotransmitters are unevenly distributed within the different brain structures. It has been reported that variations within neurotransmitter systems (concerning, for example, neurotransmitter concentrations) can modulate cognitive performance in a domain-specific fashion (Greenwood, Fossella, & Parasuraman, 2005). Examining variants of neurotransmitter-relevant genes is one way of accessing naturally occurring variations within neurotransmitter systems. This method is easily applicable in samples of human participants, while other methods – like the administration of agonists or single-unit recordings – can be too invasive and risky to allow their application. For those interested in the link between specific genes and cognitive performance, the current field of literature yields several obstructions. Studies with a focus on neurobehavioral molecular genetics have been conducted increasingly since the mid-1990s, but many of the older studies rely on small samples and are thus underpowered to detect the effects of individual genes. In addition, many studies in the field of molecular genetics are aimed to dissect the cognitive performance in healthy participants and in participants with specific diseases or disorders. For instance, the genetic basis of the Attention-deficit/hyperactivity disorder (ADHD) is comparatively well researched, while less studies are aimed at

the examination of selective attention in healthy participants. This chapter will be focused on the importance of two neurotransmitters systems for selective attention: dopamine (DA) and acetylcholine (ACh). While the main focus of this dissertation is the moleculargenetic dismantling of top-down and bottom-up selective attention, a secondary aim is the examination of response speed differences on the basis of variations within the cholinergic system (cp. subchapter 2.6). Consequently, this thesis is focused on the influence of DA-inactivating proteins on selective attention and on the influence of the nicotinic receptor class on attention as well as response speed. In subchapter 2.1, the pathways of the dopaminergic system will be highlighted. In subchapter 2.2, the metabolism of DA in terms of synthesis and degradation will be detailed. In subchapter 2.3, the effect of the catabolic enzyme catechol-O-methyl transferase on cognition will be discussed, specifically in regard to a genetic variation in its expressing gene, the *COMT* Val158Met polymorphism. Analogously, the dopamine transporter and the effect of one of its genetic variations will be discussed in subchapter 2.4. The pathways of the cholinergic system will be highlighted in subchapter 2.5. The nicotinic acetylcholine receptor will be introduced in subchapter 2.6, along with three of its genetic variations. In sum, this chapter is intended to outline important characteristics of the dopaminergic system and the cholinergic system and serves to highlight the genetic variations that are the focus of this thesis.

2.1 Dopaminergic pathways

The absolute amount of dopaminergic neurons in the brain is relatively low, yet DA neurons project to wide-spread regions of the brain and are intricately intertwined with other neurotransmitter systems (Schandry, 2006). A single dopaminergic neuron in the midbrain, for example, forms up to 100000 varicosities, each one establishing synaptic contacts with other neurons (von Bohlen und Halbach & Dermietzel, 2006). The connectivity of DA midbrain neurons thus well exceeds the connectivity of average neurons, which are linked to about 10 000 other neurons (Kosslyn & Rosenberg, 2001). Three to four major DA pathways in the brain are differentiated: the nigrostriatal pathway, the mesocortical and mesolimbic pathway (often also referred to as the mesocorticolimbic pathway), and the tuberoinfundibular pathway (see figure 2.1.).

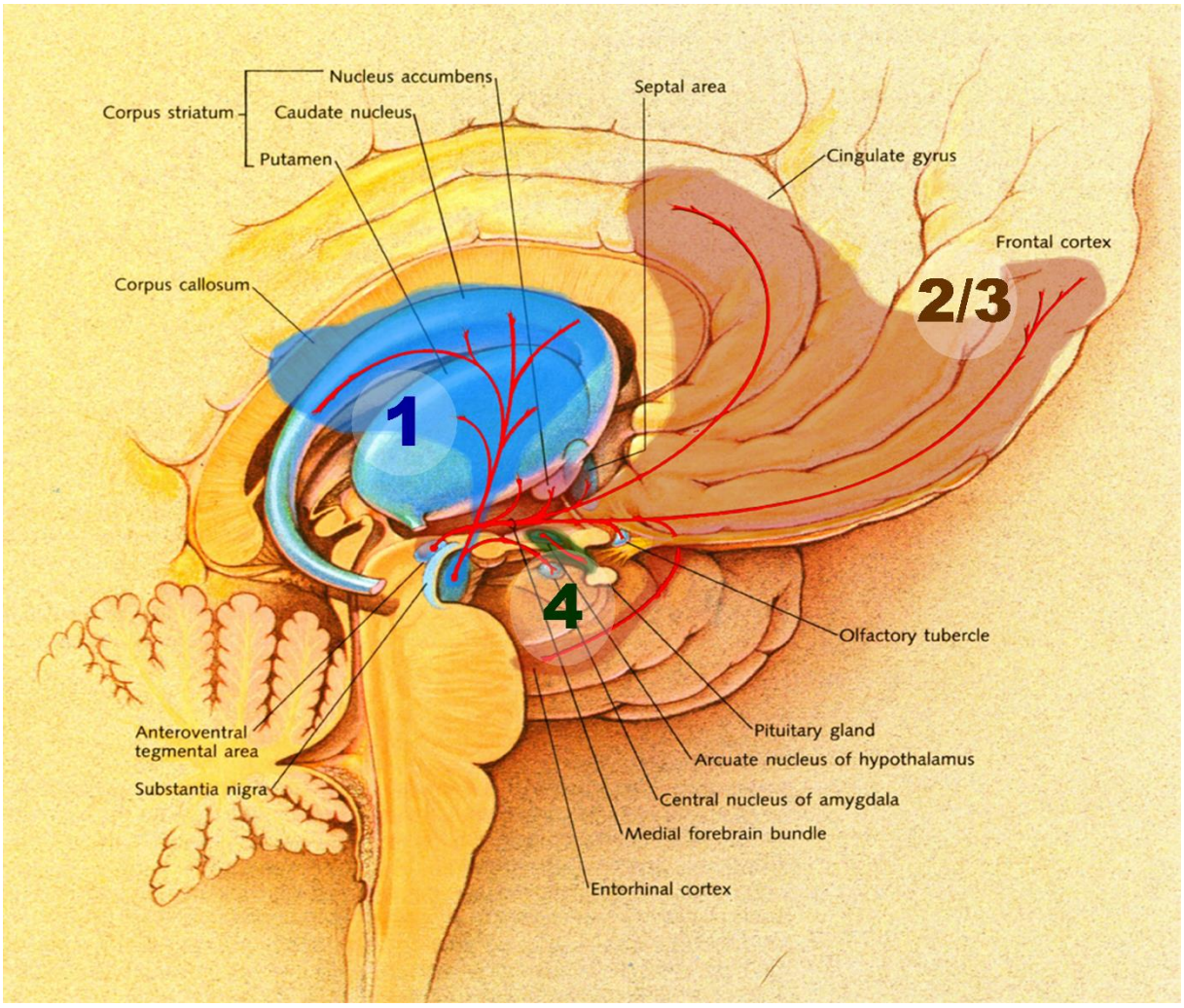


Figure 2.1.: The four major projection tracts of the dopaminergic system (figure taken and adapted from Snyder, 1999). (1) The nigrostriatal pathway. (2/3) The mesolimbic and mesocortical pathway. (4) The tuberoinfundibular pathway.

The nigrostriatal pathway originates in the population of dopaminergic neurons in the substantia nigra (Deumens, Blokland, & Prickaerts, 2002). These neurons project to the dorsal striatum, which constitutes an important part of the basal ganglia[6] (Deumens et al., 2002). The nigrostriatal pathway is involved in the regulation of motor function, as is evident from the deficits that patients of Parkinson's disease display. This neurodegenerative disorder is characterized by the degeneration of dopaminergic neurons in the substantia nigra and results in motoric deficits, among them tremor, rigidity, and akinesia (Deumens et al., 2002; Trepel, 2008). One of the most important dopaminergic areas in the brain is the ventral tegmental area (VTA), a midbrain cluster of dopaminergic cell bodies

[6] The basal ganglia is a subcortical structure of the telencephalon. In general, the striatum and the globus pallidus are seen as the core structures of the basal ganglia (Trepel, 2008). The striatum, in turn, can be distinguished into a dorsal part (consisting of the nucleus caudatus and putamen) and a ventral part (consisting of the nucleus accumbens and parts of the tuberculum olfactorium) (Deumens et al., 2002; Trepel, 2008).

that is located in close vicinity to the substantia nigra (von Bohlen und Halbach & Dermietzel, 2006). This cell group is the source of both the mesolimbic and mesocortical pathway. The projections of the VTA to the septal nuclei, the amygdala, and the ventral striatum are counted as the mesolimbic pathway, while the projections to the frontal cortex, the cingulate cortex, and the entorhinal cortex are counted as the mesocortical pathway (Riederer & Laux, 2010). The mesolimbic pathway plays a role in exerting reinforcing properties; it is a target site for psychostimulant drugs like cocaine or metamphetamine, which cause sensations of euphoria and heightened alertness, amongst other symptoms (Pierce & Kumaresan, 2006). The mesolimbic pathway has been described as a "generalized approach-seeking system that […] allow[s] organisms to generate efficient goal-directed activities in response to a large number of positive and negative incentives" (Ikemoto & Panksepp, 1999, p. 32). This pathway mediates learning-related processes in the prefrontal cortex (PFC) by providing prediction error signals that indicate when a reward fails to appear or appears at a different time than expected (Miller & Cohen, 2001). The mesocortical pathway is of great importance to cognitive functioning. An altered rate of DA transmission in the PFC has been linked to the occurrence of cognitive deficits (Goldman-Rakic, Muly, & Williams, 2000). In general, prefrontal dopaminergic activity has been associated with working memory performance (Goldman-Rakic et al., 2000; Brozowski, Brown, Rosvold, & Goldman, 1979), rule acquisition (Glickstein, DeSteno, Hof, & Schmauss, 2005), and cognitive flexibility as well as decision-making (Floresco & Magyar, 2006), to name just a few cognitive parameters that appear to be modulated by DA. Floresco and Magyar (2006) concluded in their review that the DA projections to the PFC crucially mediate *executive functions*, which is an umbrella term for control processes that regulate a variety of subprocesses through the updating of working memory, shifting between task sets and the inhibition of inappropriate responses (thus, "executive functions" are understood as high-order regulation mechanisms of cognition, cp. Miyake et al., 2000). Of the major pathways, the tuberoinfundibular pathway is the most regionally limited one (consisting only of short projections within the hypothalamus). Hypothalamic DA acts as release inhibiting factor for prolactin (Gudelsky, 1981), which is a stress-sensitive hormone and a mediator of the hormonal changes during puberty and pregnancy, but more generally also important for the libido (Schandry, 2006). In sum, DA is a wide-spread neurotransmitter in the brain and involved in the regulation of many different functions. The mesolimbic and mesocortical pathway are the most important routes by which the dopaminergic system modulates cognitive functioning and regulates affective states.

2.2 The DA metabolism (synthesis and degradation)

DA, norepinephrine, and epinephrine are catecholamine neurotransmitters: chemically characterized by their catechol compound and side-chain amines. The precursor for the biosynthesis of catecholamines is the non-essential amino acid tyrosine (Kuhar, Couceyro, & Lambert, 1999). Tyrosine is either supplied by food or synthesized in the body via the tyrosine hydroxylase enzyme. The high concentration of endogenous tyrosine leads to a virtual saturation of tyrosine hydroxylase, making this enzyme the rate-limiting factor of the catecholamine biosynthesis (Kuhar et al., 1999). Tyrosine is converted into L-DOPA by the tyrosine hydroxylase enzyme, while DA is formed efficiently through the decarboxylation of L-DOPA by the DOPA decarboxylase enzyme (Kuhar et al., 1999). The synthesis of DA takes place in the presynaptic terminals of dopaminergic neurons; DA is here formed in the cytosol and then transported into presynaptic vesicles, to hinder the degradation of the molecules by the enzyme monoamine oxidase (von Bohlen und Halbach & Dermietzel, 2006). An increased supply of cerebral L-DOPA will lead to an increased DA synthesis (von Bohlen und Halbach & Dermietzel, 2006). Unlike its precursors, DA cannot cross the blood-brain barrier (von Bohlen und Halbach & Dermietzel, 2006). Dopaminergic neurons are characterized by their large number of varicosities both in the dendrites and in the preterminal region of the axon collaterals (von Bohlen und Halbach & Dermietzel, 2006), amounting to complex interconnections. Once released from the presynaptic terminal into the synaptic cleft, DA molecules can bind to DA receptors and to some β-adrenergic receptors (Purves et al., 2001). There are two main types of DA receptors: the D1-like family (composed of the D1 receptor and D5 receptor) and the D2-like family (composed of the D2, D3, and D4 receptor) (Riederer, Eckert, Thome, & Müller, 2010). The receptors differ in their distribution, subcellular locations and in the chain of processes that is initiated by their activation (Riederer et al., 2010). DA can be inactivated and degraded in different ways. In general, DA-signaling is stopped when the DA molecules are removed from the synaptic cleft; this either leads to their recycling or degradation (Meiser, Weindl, & Hiller, 2013). The removal is achieved by specific reuptake mechanisms (von Bohlen und Halbach & Dermietzel, 2006). In addition, DA can also be rapidly returned into the presynaptic terminal via the dopamine transporter (DAT) (Giros, Jaber, Jones, Wightman, & Caron, 1996), a membrane-spanning protein that is located on presynaptic axon terminals and dendrites (Nirenberg, Vaughan, Uhl, Kuhar, & Pickel, 1996). DAT leads to the reuptake of DA, but does not degrade DA itself. In contrast, monoamino oxidase enzymes lead to the oxidative deamination of DA (Youdim, Edmondson, & Tipton, 2006), thus also degrading the neurotransmitter (Tunbridge, Harrison, & Weinberger, 2006). DA can also be inactivated and degraded by the catechol-O-methyl transferase (COMT) enzyme. COMT catalyzes the transfer of a methyl group from S-adenosyl-L methionine to DA, thus creating 3-methoxythyramine (Matsumoto et al., 2003), which is then further degraded into

homovanillic acid (for an overview on the DA catabolism, see Tunbridge et al, 2006). COMT and DAT are differently distributed throughout the dopaminergic centers of the brain. Overall, COMT plays a more important role for the inactivation of DA in the PFC, while DAT plays a more important role for the striatal inactivation of DA (Tunbridge et al., 2006). The effects of COMT and DAT will further be highlighted in the following two subchapters.

2.3 Catechol-O-methyl transferase

COMT was first described by Axelrod and Tomchick (1958). Among the substrates of COMT are not only catecholamines, but also catecholestrogens, intermediates of the melanin metabolism, ascorbic acid, and xenobiotic catechols (Lundström et al., 1995). Drugs with a catechol structure are also inactivated by COMT – like L-DOPA, which is used as a medication in Parkinson's disease, or methyldopa, which is used to treat hypertension (Lundström et al., 1995; Weinshilboum, Otterness, & Szumlanski, 1999). Two isoforms of the COMT enzyme have been detected: a soluble, cytoplasmatic form (abbreviated to S-COMT) and a membrane-bound form[7] (abbreviated to MB-COMT) (Craddock, Owen, & O'Donovan, 2006). The two isoforms are not equally distributed throughout the body. In most tissues, S-COMT is the predominant isoform. In the brain however, MB-COMT predominates. Here, MB-COMT constitutes about 70 % of the produced COMT enzymes (Tenhunen et al., 1994). Through *in situ* hybridization, it could be determined that COMT messenger ribonucleic acid (mRNA) is expressed in all layers of the PFC as well as in the striatum (Matsumoto et al., 2003). In rats, COMT is also expressed in the hippocampus (Matsumoto et al., 2003; Rivett, Francis, & Roth, 1983). Yet, the levels of COMT expression differ vastly between different brain areas. While COMT mRNA is expressed in abundance in the PFC, it is comparatively scarcer in the striatum (Matsumoto et al., 2003). In contrast, DAT is expressed at high levels in the striatum but only at low levels in the PFC (Sesack, Hawrylak, Matus, Guido & Levey, 1998; Lewis et al., 2001). Male COMT-deficient knockout mice displayed a 2- to 3-fold DA increase in the frontal cortex (Gogos et al., 1998)[8]. MB-COMT is characterized by a much higher affinity for DA than S-COMT (Matsumoto et al., 2003). This is only consequential, as the PFC appears to be the main center of COMT activity in the brain and as the DA concentrations in this region are much lower than, for example, in the striatum (Matsumoto et al.,

[7] There is no consensus where MB-COMT is located on a subcellular level. Tunbridge and colleagues (2006) outlined different findings, concluding that MB-COMT is likely located on postsynaptic neurons. Yet, it remains unclear whether MB-COMT enzymes are located on the plasma membrane of the cell (protruding either into the cytoplasm or synapse), or located on intracellular membranes like those of the mitochondrion or rough endoplasmic reticulum (Tunbridge et al., 2006).

[8] Gogos and colleagues (1998) did not detect a similar increase of DA for female knockout mice, which suggests gender-specific effects of DA regulation.

2003). The MB-COMT enzyme is well suited to regulate the low physiological concentrations of DA that are usually found in the PFC (Matsumoto et al., 2003). The chromosomal location of the *COMT*[9] gene has been mapped to 22q11.1→q11.2 (Grossman, Emanuel, & Budarf, 1992). The gene contains six exons, of which two are non-coding (Tenhunen et al., 1994). Tenhunen and colleagues (1994) reported that two transcripts are expressed from the *COMT* gene: a shorter transcript of 1.3 kilobase (kb) transcript and a longer transcript of 1.5 kb. They also detected two distinct promoters on the gene (P1 and P2). While the expression of the short manuscript is regulated by P1, the expression of the long manuscript is regulated by P2 (Tenhunen et al., 1994). S-COMT can be translated both from the short and from the long manuscript (Tenhunen et al., 1994); this dual functionality is implemented by a 50-amino acid extension of the reading frame (Bertocci et al., 1991; Lundström et al., 1991). Tenhunen and colleagues (1994) stated that the activity of both promoters is highly variable and dependent on the tissue in which the gene is expressed. They stated that P1 and P2 can be equally active in some tissues (like the liver), while not being equally active in others (the activity of P2 is much higher in the brain). The relative activity of the promoters is probably regulated by tissue-specific transcription factors (Tenhunen et al., 1994).

The COMT Val158Met (rs4680) polymorphism. The *COMT* Val158Met polymorphism is a single nucleotide polymorphism. This functional G→A transition is located at codon 158 of the *COMT* gene and results in the substitution of valine (Val) by methionine (Met) (Lachman et al., 1996). The two alleles are referred to as the Val allele and the Met allele. The Val allele is the ancestral allele (Palmatier et al., 2004). In regard to cognitive functioning, this polymorphism is one of the most extensively studied variants. It is an especially interesting variant due to its effect on the thermostability of the COMT protein. Chen and colleagues (2004) studied differences in the level of COMT activity in postmortem tissues of the dorsolateral PFC (DLPFC). At 37°C, the COMT activity in the DLPFC was significantly modulated by the *COMT* Val158Met polymorphism. In European-Americans, the level of COMT activity was 50 % higher in Val homozygotes than in Met homozygotes and 12 % higher in Val/Met heterozygotes than in Met homozygotes (see figure 2.2). The difference between the COMT activity of Val homozygotes and Val/Met heterozygotes was three times the difference between the COMT activity of Val/Met heterozygotes and Met heterozygotes. Thus, the COMT activity in heterozygotes was intermediate, but not equidistant in relation to the levels displayed by homozygotes. While previous reports of the trimodal distribution of COMT activity (Floderus, Ross, &

[9] Please note that the Catechol-o-methyl transferase enzyme is abbreviated to COMT, while the Catechol-o-methyl transferase gene is abbreviated to *COMT.* The same distinction will be made for all other polymorphisms that are discussed here. The non-cursive abbreviations will refer to the product that is expressed by the gene, while the cursive abbreviation will refer to the gene itself.

Wetterberg, 1981) have been supported by these newer results, it is evident that the *COMT* Val158Met genotype groups cannot be treated as equidistant groups.

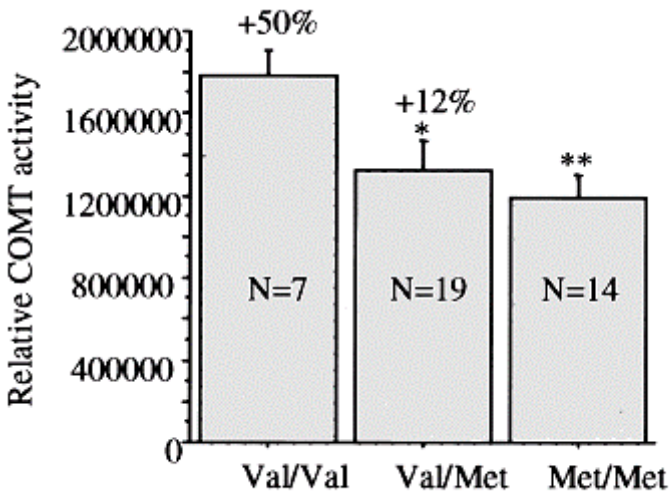


Figure 2.2.: The relative COMT activity in postmortem DLPFC tissues in a sample of European Americans (figure taken and adapted from Chen et al., 2004). Differences from Val/Val are indicated at a significance level of $p < .05$ by one asterisk and at a significance level of $p < .001$ by two asterisks. The level of COMT activity of carriers of the Val/Met genotype and of the Met/Met genotype differed significantly from the level of COMT activity of carriers of the Val/Val genotype.

Chen and colleagues (2004) also tested the effect of gender on COMT activity levels and reported that females had an overall 17 % lower level of COMT activity in the DLPFC tissue, independent of the particular genotypes. Compared to carriers of at least one Met allele, carriers of the Val/Val genotype displayed a worse performance in the Wisconsin Card Sorting Test (WCST), which is associated with the PFC function and measures executive cognition (but not general intelligence) (Egan et al., 2001). About 4 % of the variance in the WCST performances was explained by the *COMT* genotype. In addition, Egan and colleagues (2001) also tested whether the frontal activation between carriers of different *COMT* genotypes differed. They did this by analyzing the prefrontal activity during an n-back working memory task, utilizing functional magnetic resonance imaging (fMRI). Generally, a higher prefrontal activation has been interpreted to reflect a higher neural inefficiency, or in other words, a reduced signal-to-noise ratio (also see Servan-Schreiber, Printz, & Cohen, 1990; Egan et al., 2001, Mattay et al., 2003). Egan and colleagues (2001) found that the Met allele load was associated with a more efficient PFC response; namely, carriers of the Met/Met genotype displayed the lowest activations in the DLPFC and the cingulate cortex, while carriers of the Val/Met genotype displayed

intermediate activations and carriers of the Val/Val genotype displayed the highest activations. The link between the *COMT* Val158Met polymorphism and the PFC signal-to-noise ratio was further supported by a meta-analysis spanning twenty studies (Mier, Kirsch, & Meyer-Lindenberg, 2009). The authors found no indication for a publication bias (and only a non-significant trend for an association between the publication year and the reported effect sizes[10]). Mier and colleagues (2009) reported that Met allele carriers displayed a lower prefrontal activation during tasks of executive cognition (effect size: d = 0.92). The possible relation between a heightened level of prefrontal DA and an increase in prefrontal efficiency is termed the *efficiency hypothesis* (Dickinson & Elvevåg, 2009). Dickinson and Elvevåg (2009) also found the hypothesis to be supported by empirical findings. They conclude that the larger amount of prefrontal DA in Met allele carriers seems to lead to a "reduced signal variability and improved signal-to-noise" as well as a "sharper peak signal" (p. 81). Mattay and colleagues (2003) provided new insights into the link between the *COMT* Val158Met polymorphism and the PFC function by studying the effect of amphetamine – which increases the dopaminergic neurotransmission – on the performance in a WCST task and an n-back task. The n-back task was administered during an fMRI session. The study was conducted in a double-blind fashion and had a crossover design, i.e. each participant consumed both amphetamine and a placebo at different test sessions. In the WCST task, carriers of the Val/Val genotype displayed an improved performance under the influence of amphetamine, while carriers of the Met/Met genotype displayed a decreased performance under the influence of amphetamine. At all levels of working memory load in the n-back task, carriers of the Val/Val genotype displayed an improved performance under the influence of amphetamine (in terms of RT). This effect was accompanied by a more efficient signal-to-noise ratio in the PFC. At the highest working memory load in the n-back task, carriers of the Met/Met genotype displayed a decreased performance under the influence of amphetamine (in terms of accuracy and RT). This effect as was accompanied by a less efficient signal-to-noise ratio in the PFC. Mattay and colleagues (2003) interpreted these findings as the result of an inverted-U DA response curve between the amount of DA transmission and the efficiency of the PFC function (see figure 2.3). In other words, they assumed that a medium range of DA activity is optimal for PFC function, while low or high amounts of DA activity have adverse effects (this is called the *inverted-U curve* hypothesis, also cp. Goldman-Rakic et al., 2000). More specifically, they assumed that carriers of the Val/Val genotype are located on the up slope of the curve (low within the range of normal DA activity), while carriers of the Met/Met genotype are positioned closer to the peak of the curve (higher within the range of normal

[10] There was a trend between earlier publication dates and larger effect sizes. A possible explanation of this pattern is that relatively new findings were reported in the earliest studies; it was likely easier to publish these findings when the effect sizes were larger. As the link between the *COMT* Val158Met polymorphism and cognitive performance became more well-known, it probably became easier to publish studies with smaller effect sizes.

DA activity). The authors interpreted their results to mean that amphetamine and high working memory loads (both of which are thought to increase the DA transmission) shift the respective positions of Val/Val and Met/Met carriers on the curve. Val/Val carriers are presumably shifted more towards the optimal range of DA activity while Met/Met are shifted more down-slope on the curve, towards a less optimal range of DA activity.

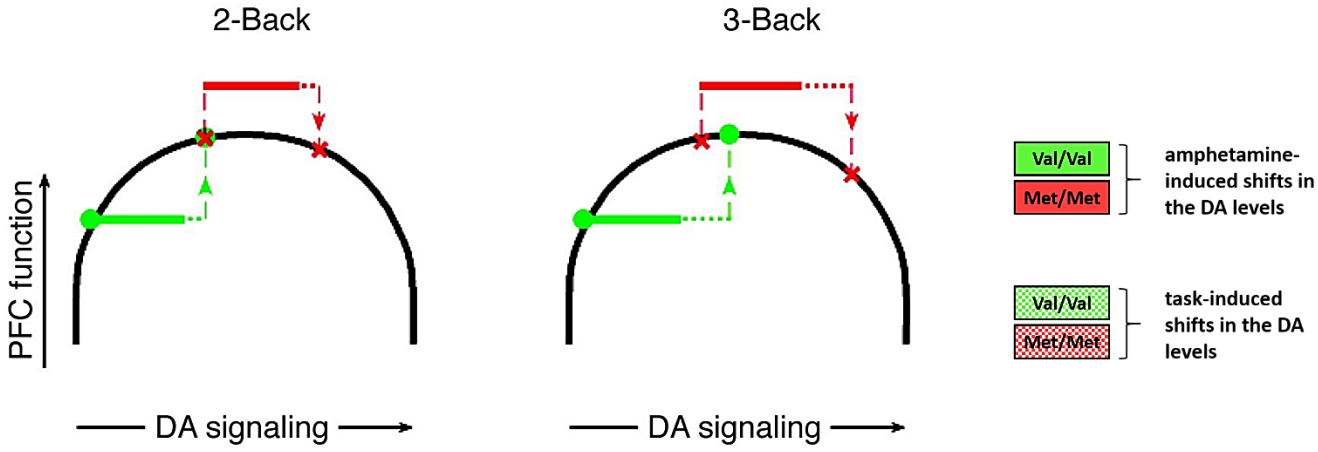


Figure 2.3.: The inverted-U model which relates the PFC function to the rate of DA signaling (figure taken and adapted from Mattay et al., 2003). The graphic depicts the assumed position of Val and Met homozygotes on the inverted-U curve under normal conditions and under conditions of an increased DA transmission (due to the consumption of amphetamine and varying working memory loads).

Similar links between dopaminergic modulations and cognitive performances have also been observed in male rats that were treated with the COMT-inhibitor tolcapone (Tunbridge, Bannerman, Sharp, & Harrison, 2004). Compared to control rats, these rats performed better in a task of set-shifting[11]. As noted by Chen and colleagues (2004), rats do not express either Val or Met in the 158th codon of the COMT protein, but Leucine, which leads to an even higher level of COMT activity than Val. Tunbridge and colleagues could demonstrate that an artificial increase in the DA levels in mice led to similar behavioral benefits as those displayed by human Met carriers under normal conditions. Interestingly, Mier and colleagues (2009) noted diametrically opposed effects of the *COMT* Val158Met polymorphism on the prefrontal activation during executive cognition tasks and emotional tasks. For

[11] The task requires the attentional focus to be shifted from one dimension of a stimulus to another and is, in rats, dependent on the medial PFC (Tunbridge et al., 2004).

Met allele carriers, the prefrontal activation was lower (presumably more efficient) during executive tasks, but higher (presumably less efficient) during emotional tasks (the effect sizes were *d* = 0.92 and *d* = -1.0, respectively). The opposite pattern was noted for carriers of the Val allele. This is a clear indicator that the *COMT* Val158Met polymorphism exerts pleiotropic behavioral effects, i.e. modulates multiple phenotypes. This also means that a distinction between a "beneficial allele" and a "disadvantageous allele" is too simplistic and not feasible, the effects of the polymorphism depending on the context. Mier and colleagues (2009) suggested that each allele "confers an environment-specific selective advantage" and that "the reciprocal effect […] represents a tradeoff between the cognitive efficiency and emotional resiliency that maintains each allele in the population" (p. 919).

2.4 The dopamine transporter

The dopamine transporter (DAT) is a membrane-spanning glycoprotein that regulates the reuptake of extracellular DA (von Bohlen und Halbach & Dermietzel, 2006). DAT is located in some distance to the active synaptic zone (von Bohlen und Halbach & Dermietzel, 2006). The extracellular clearance of DA required 1 s in wild-type mice, but 100 s in mice homozygous for a disrupted DAT gene (Giros et al., 1996). No effects of cocaine or D-amphetamine were detectable in these mice – which suggests that the usual target site of these drugs had been eliminated (also cp. Pierce & Kumaresan, 2006). While COMT is essential for the DA catabolism in the PFC, DAT is essential for the DA catabolism in the striatum. The gene that encodes the DAT is referred to as *SLC6A3* or *DAT1* (the latter term will be used here). The gene has been mapped to chromosome 5p15.3 and is characterized by a 40 base pair (bp) *variable number tandem repeat* polymorphism in the 3' untranslated region (Vandenbergh et al., 1992). This polymorphism does not directly influence the protein product due to its location on the untranslated region. However – while only approximately 1.5 % of the DNA codes for proteins – the other components of DNA are also relevant for the gene expression (Mignone, Gissi, Liuni, & Pesole, 2002). Untranslated regions are essential for the post-transcriptional regulation of the gene expression (for example, the mRNA stability, transport, subcellular localiziation, and translation efficiency) (Mignone et al., 2002). The *DAT1* polymorphism varies between 3 to 13 number repeats. The 10-repeat allele is the most common allele in Middle-European populations, followed by the 9-repeat allele (Kang, Palmatier, & Kidd, 1999). The *DAT1* polymorphism was shown to modulate the DAT density *in vitro*. In saturation binding assays and immunoblots, the DAT binding site density was about 50 % higher for the *DAT1* 10-repeat variant than for the 9-repeat variant (VanNess, Owens, & Kilts, 2005). Methylphenidate is a common medication for the treatment of ADHD. This drug effectively blocks the DAT; under the usual therapeutical doses, the consumption of methylphenidate is estimated to occupy

more than 50 % of the transporters (Volkow et al., 1998). Adults with ADHD who did not have a history of medication had lower concentrations of DAT in the left caudate and left nucleus accumbens than control participants (Volkow et al., 2007). For these reasons, the *DAT1* polymorphism was an obvious starting point for examining the link between ADHD and specific genetic variations. In a sample of adult participants with ADHD, the disorder has been linked to the 9-repeat/6-repeat genotype of the *DAT1* polymorphism (Franke et al., 2008). This relatively uncommon genotype was present in 6.5 % of the controls and in 11.7 % of the participants with ADHD. Brown and colleagues (2011) found that the 9-repeat allele was significantly more common in adult participants with ADHD compared to controls. On the other hand, the 10-repeat allele was associated with a reduced volume of the caudate in boys with ADHD (Durston et al., 2005) and with hypoactivation in the anterior cingulate cortex, cerebellar vermis and lateral PFC in a sample of adults (Brown et al., 2010). Given the inconsistent findings[12], it cannot be recommended to use the *DAT1*-related aberrant functioning in ADHD to draw conclusions on the relation between *DAT1* and cognitive functioning in healthy participants. Expanding previous findings (Fossella et al., 2002), Rueda and colleagues (2005) examined the effect of the *DAT1* polymorphism on attention in a sample of six year old children. The children were tasked with a variant of the Eriksen Flanker task, which is similar to the Stroop task. In the task, five fish were presented in a horizontal line; the direction in which the middle fish pointed had to be indicated. In incongruent trials, the center fish was pointed in a different direction than the other fishes. As in the Stroop task, a conflict measure can be derived by subtracting the RTs of the incongruent condition from the RTs in the congruent condition; the measure reflects the ability to resolve conflict and taps selective top-down attention. Homozygous carriers of the 10-repeat allele displayed a better performance in the Eriksen flanker task. They were both overall faster and less affected by incongruent stimuli. While 10-repeat allele heterozygotes were about 200 ms slower in incongruent trials, 10-repeat allele homozygotes were only 8 ms slower[13]. In addition to these behavioral measures, the 10-repeat homozygotes also achieved a significantly higher score in the temperamental factor “effortful control”, which was measured through the Children’s Behavior Questionnaire. This factor reflects attentional self-regulation and has been linked to the frontal lobe function (Rothbart, Ahadi, Hershey, & Fisher, 2001). Bertolino and colleagues (2006) proposed two possible routes by which the *DAT1* polymorphism affects the signal-to-noise ratio in the PFC and so modulates the performance in PFC-dependent tasks. For one, *DAT1* could have immediate effects in the PFC, since DAT is present in this area in in low concentrations. The different variants of *DAT1* are known to modulate the protein expression, so that they could contribute to the absolute DA availability in a similar fashion to the *COMT* Val158Met

[12] This heterogeneity might be due to sample differences in regard to age, gender, medication, different ADHD diagnoses, the severity and duration of ADHD, comorbidities and other not yet known, possibly interacting factors.

[13] The overall high RTs in this task were attributable to the young age of the participants.

polymorphism. The prefrontal DA availability might be comparatively decreased in the presence of one or two 10-repeat alleles. It stands to reason that direct prefrontal effects of *DAT1* cannot reach the magnitude of the effects of the *COMT* Val158Met polymorphism, since COMT is the primary catabolic agent in this brain area. Thus, direct PFC effects should only play a minor role. It is more likely that modulations occur primarily through the striatum. Decreased levels of striatal DA, as most likely the case in the presence of one or two 10-repeat alleles, might increase the signal-to-noise ratio in the PFC (Bertolino et al., 2006). In sum, according to Bertolino and colleagues (2006), the prefrontal signal-to-noise ratio can either be directly or indirectly influenced by the *DAT1* polymorphism. Both routes do not need to be mutually exclusive.

Summary. The *COMT* Val158Met polymorphism is a genetic variant on the gene that encodes COMT. This polymorphism has been found to modulate the thermostability of the COMT protein. The Met allele decreases the thermostability, which in turn increases in the prefrontal availability of DA. This allele has consistently been associated with better performances in a wide array of PFC-dependent cognitive tasks. The *DAT1* polymorphism is a genetic variant on the gene that encodes the DAT. The 10-repeat allele has been linked to a 50 % increase in the binding site density for DAT. This allele was linked to better performances in a Stroop-like task.

2.5 Cholinergic pathways

ACh acts as a neurotransmitter for up to 15 % of all neurons of the human nervous system (Snyder, 1999). Given this number, it is not surprising that ACh was the first neurotransmitter whose function became partly known. The pharmacologist Otto Loewi (1921) discovered that nerve signals do not only rely on electrical stimulation, but also have a chemical component; that nerves secrete a substance that can be isolated and used to stimulate other nerves. Today, it is known that the effects of ACh in the peripheral nervous system (PNS) are even more numerous than indicated by Loewi's experiments: ACh is the main neurotransmitter for nerves which act on voluntary muscles, glands, and a variety of peripheral organs (Snyder, 1999). Three major cholinergic subsystems can be distinguished in the brain (Dani & Bertrand, 2007). The first one originates from cholinergic neurons in the tegmentum and innervates the caudal pons, the brain stem, the thalamus and the dopaminergic areas in the midbrain (Dani & Bertrand, 2007). The second system originates from an array of basal forebrain nuclei and innervates the cortex and the hippocampus (Dani & Bertrand, 2007). The third system arises from cholinergic interneurons in the striatum and innervates both the striatum and the olfactory tubercle (Dani & Bertrand, 2007). Almost every area of the brain is cholinergically innervated. These

innervations are usually relatively diffuse and sparse; the third system is an exception, as it provides very rich and regionally limited innervations (Dani & Bertrand, 2007).

2.6 The nicotinic acetylcholine receptor

There are two major groups of ACh receptors: nicotinic and muscarinic receptors (von Bohlen und Halbach & Dermietzel, 2006). The nicotinic ACh receptor (nAChR) will be discussed in more detail in this paragraph (for an overview, also see Dani & Bertrand, 2007). This receptor was named after nicotine, since nicotine molecules bind to it as well (Purves et al., 2001). Nicotine molecules reach about 80 % of the efficacy of ACh molecules in terms of the receptor activation (Nelson, Kuryatov, Choi, Zhou, & Lindstrom, 2003). NAChRs are ligand-gated ion channels: after being stimulated, these receptors change their confirmation and build a channel that enables selective ions to pass (Schandry, 2006). A large number of different nAChR subtypes exist. These subtypes are diversely distributed (Lukas et al., 1999). Broadly speaking, muscle and neuronal subtypes of this receptor can be distinguished (Colquhoun, Shelley, Hatton, Unwin, & Sivilotti, 2003). The most common nAChR in the brain is the α4β2 subtype (Greenwood, Parasuraman, & Espeseth, 2012), which is composed of two α4 subunits and two β2 subunits. While this receptor is frequently spelled in the abbreviated form of "*α4β2*", its composition is better illustrated by using a spelling of "$(\alpha4\beta2)_2$", which indicates that the receptor is composed of two α4 subunits and two β2 subunits (as well as another, not further specified subunit). The $(\alpha4\beta2)_2$ subtype is the main high-affinity nAChR in the brain (Greenwood et al., 2012), which means that comparatively lower ligand concentrations are sufficient to occupy its binding sites (Greenwood et al., 2012). The binding sites are created in pockets between an adjacent α4 and β2 subunit (Dani & Bertrand, 2007). The fifth subunit of the $(\alpha4\beta2)_2$ nAChR is not involved in the forming of binding sites. For this reason, this subunit is labelled an "accessory" subunit (Kuryatov, Onksen, & Lindstrom, 2008). The role of accessory subunits is a modulating one. Depending on the brain area, about 10 – 40 % of the $(\alpha4\beta2)_2$ nAChRs have an accessory α5 subunit (Kuryatov et al., 2008; Mao, Perry, Yasuda, Wolfe, & Kellar, 2008). Accessory α5 subunits provide a greater sensitivity for the nAChR activation than α4 subunits (Kuryatov et al., 2008). Of special interest in this thesis are both the α4 and the α5 subunit of the nAChR. While the α4 subunit can either manifest as a binding-relevant subunit or as an accessory unit, the α5 subunit only serves as an accessory unit (Kuryatov et al., 2008). In this thesis, polymorphisms of the cholinergic system will not only be examined in regard to selective attention, but also in regard to response speed. Response speed (or interchangeably, RT) is the interval between the appearance of a sensory stimulus and the subsequent behavioral response to it. This measure is indicative of the mental speed of processing. Most RT tasks are comparatively simple and

will be completed in a range of several hundred milliseconds. Individual differences still occur, however, and have even been associated with estimates of intelligence (Sheppard & Vernon, 2008). The cholinergic system has been extensively studied in regard to response speed, often through the application of the nAChR agonist nicotine. Nicotine consumption led to beneficial RT effects in a range of different tasks (Hahn, Shoaib, & Stolerman, 2002; Blondel, Sanger, & Moser, 2000; Wesnes & Warburton, 1984; Hindmarch, Kerr, & Sherrwood, 1990). The performance in a Posner-Cuing task, for example, was measured both in humans and monkeys in relation to the nicotine consumption (Witte, Davidson, & Marrocco, 1997). Under the influence of nicotine, both groups displayed significantly decreased RTs. Given these findings, it is evident that nAChR genes are ideal starting points to examine ACh modulations of response speed. The *CHRNA4* gene, which encodes the α4 subunit of the nAChR, has been mapped to chromosome 20q13.2→q13.3 (Steinlein et al., 1994). The gene spans about 17 kb and contains six exons (Steinlein, Weiland, Stoodt, & Propping, 1996). The *CHRNA5* gene, which encodes the alpha 5 subunit of the nACh receptor, has been mapped to 15q24 (National Library of Medicine, 2014). The gene spans about 25 kb and contains six exons (Duga et al., 2001). Three variants of the *CHRNA4* and *CHRNA5* genes will be studied in regard to selective attention and response speed.

The CHRNA4 rs1044396 polymorphism. This polymorphism is characterized by synonomous C→T transition at codon 1545 of the *CHRNA4* gene. Codons with either alelle encode the amino acid Serin (Steinlein et al., 1997; Feng et al., 2004). It is not yet established whether the polymorphism itself is functional. Given its link to a variety of neurocognitive phenotypes, at the very least a linkage disequilibrium to non-synonomous variants in the regulatory or promotor region of the gene can be expected, however (Markett, Montag, & Reuter, 2011). Winterer and colleagues (2011)[14] injected complementary DNA (cDNA) into *Xenopus* oocytes (a genus of clawed frogs), leading to the expression of human $(\alpha4\beta2)_2$ nAChRs. They then measured the receptor responses under different concentrations of ACh and found that more nAChRs were in a high-affinity state in the presence of the rs1044396 C allele. Consequently, it has been hypothesized that C allele homozygotes have the highest amount of nAChRs in a high-affinity state while T allele homozygotes have the least amount of nAChRs in a high-affinity state (Greenwood et al., 2012). This hypothesis is still highly speculative at the moment. On a cognitive level, the *CHRNA4* rs1044396 polymorphism has been studied mainly in regard to visuospatial attention. Effects of the *CHRNA4* rs1044396 polymorphism on covert attention (Parasuraman, Greenwood, Kumar, & Fossella, 2005; Espeseth et al., 2006) and overt attention (Greenwood et al., 2005, Espeseth et al., 2010), visual as well as auditory attention (Espeseth,

[14] As Winterer's study was presented at a conference and is not available in print, see Greenwood et al., 2012, for a presentation and discussion of these results.

Endestad, Rootwelt, & Reinvang, 2007), selective attention (Parasuraman et al., 2005; Espeseth et al., 2006) and divided attention (Espeseth et al., 2010) have been reported. In their overview article, Greenwood and colleagues (2012) have proposed three assumptions about the link between the nAChR system and attention. First, they assumed that the $(\alpha4\beta2)_2$ nAChR subtype is of special importance in the tempoparietal junction, where the parietal lobe meets the temporal lobe. Second, they assumed that the T allele facilitates the processing of targets within the focus of attention, but slows the redirecting of attention outside the focus of attention[15]. Third, Greenwood and colleagues assumed that this effect is possibly mediated the affinity status of the nAChR receptor. They argued that T allele homozygotes "chronically maintain a more constricted attentional focus", because they have "more α4β2 nicotinic receptors in a low affinity state in the [tempoparietal junction]" (p. 1336). This assumption is still very speculative, however. While there is considerable evidence that response speed is linked to the nAChR, this measure is rarely the sole factor that researchers are interested in. In many studies, effects on response speed are reported in regard to specific experimental conditions, but not in regard to the average response speed across all conditions. Greenwood and colleagues (2005), however, reported that the average response speed in a cued visual search task increased linearly with the C allele dosage. The RTs were lowest in T homozygotes and highest in C homozygotes. This finding provides the first tentative indication that the *CHRNA4* rs1044396 polymorphism might modulate response speed.

The CHRNA5 rs3841324 polymorphism and CHRNA5 rs16969968 polymorphism. These polymorphisms are variations of the gene that encodes the acetylcholine receptor alpha 5 subunit (*CHRNA5*). Since a nearly complete linkage disequilibrium between them has been noted (r^2 = 0.389; *D'* = 0.955) (Wei et al., 2011), these polymorphisms will be discussed together. The effects of both polymorphisms are not yet well explored, especially in regard to cognitive measures. The *CHRNA5 rs3841324* polymorphism is an insertion/deletion length polymorphism within the promoter of the *CHRNA5* gene (Wang et al., 2009). In contrast to the short allele (S allele), the long allele (L allele) contains a 22 bp insertion. Analyzing postmortem brain tissues in a sample of European-Americans, Wang and colleagues (2009) noted that heterozygotes showed significantly lower levels of *CHRNA5* mRNA expression than S allele homozygotes. The expression levels of L allele homozygotes reached approximately 40 % of the expression level of S allele homozygotes. The difference between heterozygotes and L allele homozygotes, however, was not significant. The polymorphism covers several transcription factor binding sites, among them the binding site for SP-1, which stimulates the

[15] This assumption is rooted in a location-based understanding of visuospatial attention and alludes to concepts such as the spotlight or zoom lens metaphor (Posner, 1980, Eriksen & St. James, 1986).

transcription process (Berrettini & Doyle, 2012). In the S allele – the deletion variant – those binding sites are lost. In the WCST, carriers of the L allele made fewer perseverative errors after smoking, displaying an increased cognitive flexibility (Zhang, Kranzler, Poling, & Gelernter, 2010). The *CHRNA5* rs16969968 polymorphism is a missense mutation and located within exon 5 of the *CHRNA5* gene, in the central area of the second intracellular loop (Wang et al., 2009; Online Mendelian Inheritance in Man, OMIM, 2014, no. 118505). This G→A base transition in codon 398 of the *CHRNA5* protein causes an amino acid change from aspartic acid to asparagine. The G allele is the ancestral allele (OMIM, 2014, no. 118505). The aspartic acid residue that results from this allele is highly conserved across a variety of species (Bierut et al., 2008; Berrettini & Doyle, 2012). The response of nAChRs to an agonist was measured in human embryonic kidney cells (Bierut et al., 2008). The maximal response of the nAChRs that contained the G allele was more than two times higher than the maximal response of the nAChRs that contained the A allele. Thus, the rs16969968 polymorphism appears to influence the responsiveness of the α5-containing nAChR. So far, the polymorphism has been mainly studied as one of multiple risk locations for nicotine dependency (Saccone et al., 2007; Bierut et al., 2008; Sherva et al., 2008; Janes, Smoller, & David, 2012) and has been found to modulate the risk for developing lung cancer (Wei et al., 2011). Winterer and colleagues (2010) reported that the rs16969968 polymorphism was associated with a verbal and a performance subscale of the revised Wechsler Adult Intelligence Scale. Furthermore, the polymorphism was related to an aggregated performance measure composed of the performance in an n-back task and the Continuous Performance Test (the latter of which primarily measures sustained attention and vigilance). Wei and colleagues (2011) studied the effect of the diplotypes of the *CHRNA5* rs3841324 polymorphism and *CHRNA5* rs16969968 polymorphism on the risk of developing lung cancer. They found that the diplotype that was composed of both *CHRNA5* rs3841324 S alleles and both *CHRNA5* rs16969968 G alleles (S/S_G/G) had a protective effect for women but not for men. This finding indicates potential gender-based differences in the effects of both polymorphisms.

Summary. The *CHRNA4* rs1044396 polymorphism is a genetic variant of the gene that encodes the α4 subunit of the nAChR. The T allele of this polymorphism has been linked to a lower number of nAChRs in a high-affinity state. Moreover, it has been speculated that the T allele facilitates the processing of targets within the focus of attention (Greenwood et al., 2012). The *CHRNA5* rs3841324 polymorphism and *CHRNA5* rs16969968 polymorphism are variants of the gene that encodes the α5 subunit of the nAChR. The *CHRNA5* rs3841324 polymorphism been linked to differences in the *CHRNA*5 mRNA expression levels; specifically, S allele homozygotes displayed a higher level of expression. The *CHRNA5* rs16969968 polymorphism has been associated with the responsiveness of the nAChRs. Receptors that contained the G allele were more responsive towards an agonist.

Chapter 3

The Neurobiological Foundations of Top-down and Bottom-up Attention

Everything existing in the universe is the fruit of chance and necessity.

(Democritus, 400 BC)

Selective top-down and bottom-up attention are complementary forms of attentional control and rely either on internal goals or on the salience of external stimuli. What are the neural foundations of these governing principles? Both the dopaminergic and the cholinergic system have consistently been linked to selective attention, but likely exert different influences on top-down and bottom-up attention. This chapter is intended to outline the hypothesis that the dopaminergic system is more important for the modulation of selective top-down attention, while the cholinergic system is more important for the modulation of selective bottom-up attention. First, evidence for this dissociation will be introduced (subchapter 3.1). Then, the neural background of top-down attention will be detailed – and here specifically the importance of the PFC and affiliated areas (subchapter 3.2). Subsequently, it will be detailed why the Stroop task and Negative priming task (on the behavioral side) and the *COMT* Val158Met polymorphism and the *DAT1* polymorphism (on the moleculargenetic side) tap selective top-down processes that originate from these areas (subchapter 3.3). The neural foundation of selective bottom-up attention will be discussed in the same fashion. First, the importance of the sensory cortices and affiliated areas will be detailed (subchapter 3.4). Subsequently, it will be discussed why the Posner-Cuing and Dot Probe task (on the behavioral side) and the *CHRNA4* rs1044396, *CHRNA5* rs3841324 and *CHRNA5* rs16969968 polymorphisms (on the moleculargenetic side) tap processes that originate from these areas (subchapter 3.5).

3.1. Dismantling selective top-down and bottom-up Attention

The overarching hypothesis is based on the works of Behrad Noudoost and Tirin Moore (Noudoost & Moore, 2011a; 2011b; Squire, Noudoost, Schafer, & Moore, 2013; Clark & Moore, 2014). Noudoost and Moore have mainly studied the neural basis of selective top-down and bottom-up attention by examining animal models, specifically in Rhesus monkeys (macaques, or *Macaca mulatta*). Their method of choice is the *in vivo* recording of the electrophysiological response of single neurons. These responses can, for example, be studied in dependence of the microiontophoretic application of antagonists or agonists. In their review article on top-down and bottom-up attention (2011a), they stated that DA and ACh are the neurotransmitters that have been linked to attentional control most frequently. This, however, does not necessarily mean that DA and ACh are equally involved in the control of selective top-down and bottom-up attention. Specifically, Noudoost and Moore (2011a) stated:

"…one possibility is that [the DA and ACh systems] contribute differently to different forms of attention. Evidence to date suggests, for example, that ACh may serve a more unique role in bottom-up attention than it does in top-down attention, whereas the reverse may be true for DA. Studies of the neural correlates of attention have thus far yielded evidence of dissociable underlying neural circuits of these two varieties of attention, and it may turn out that the modulatory effects within those circuits differentially depend on DA and Ach" (p. 589).

Buschman and Miller (2007) conducted a groundbreaking study that dismantled components of selective top-down and bottom-up attention on a neuronal level. They studied the performance of Rhesus monkeys (*Macaca mulatta*) in a visual search task. It was the monkey's task to locate the target in an array of four stimuli (which were colored, tilted lines; see figure 3.1). There were two conditions: the conjunction condition (where the search was more difficult) and the pop-out condition (where the search was easier). In the conjunction condition, the target shared properties with the distractors. The target was a left-tilted green line, for example, while the distractors were also tilted in the same direction (but were of a different color) or were of the same color (but were tilted differently). In contrast, the target selection was much easier in the pop-out condition. Here, the target differed from the distractors in *both* color and orientation and was thus highly salient. The target was a left-tilted green line, for example, while the distractors were violet and right-tilted. The conjunction condition relies strongly on selective, visuospatial top-down attention, while the pop-out condition relies strongly on selective, visuospatial bottom-up attention.

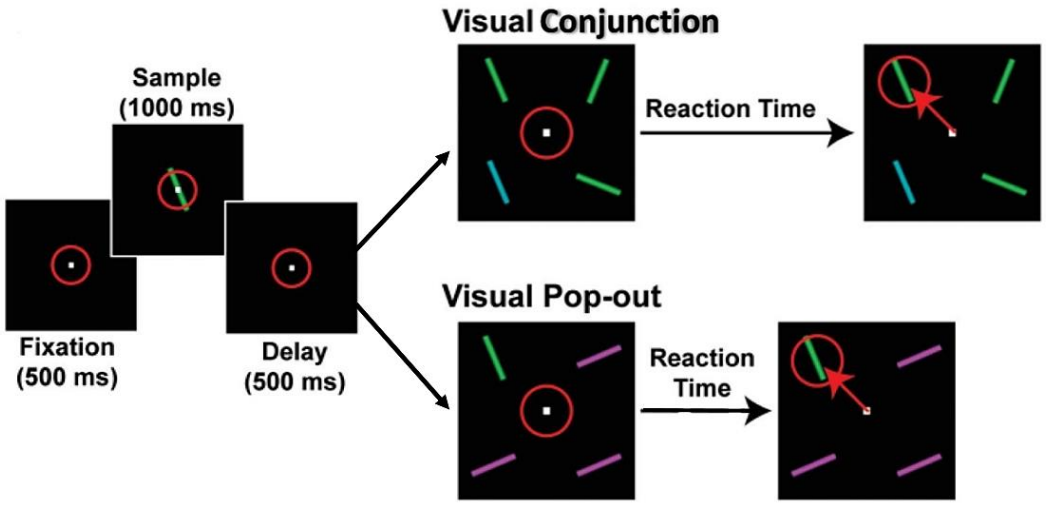


Figure 3.1.: The sequence of events in the conjunction and pop-out condition of the visual search task (figure taken and adapted from Buschman & Miller, 2007). The eye position throughout the trials is indicated by the red circle. The sample indicates the target that has to be located (in

this example a slightly left-tilted, green line). In the conjunction condition, the target shares properties with the distractors. In the pop-out condition, the target shares no properties with the distractors and is thus highly salient. Responses tend to be significantly faster than in the pop-out condition. Buschman and Miller used the conjunction condition to measure selective top-down attention and the pop-out condition to measure selective bottom-up attention.

Buschman and Miller recorded the activity of 802 neurons in three brain areas across 24 sessions. Those brain areas were the lateral prefrontal cortex (LPFC), the frontal eye fields (FEF) and the lateral intraparietal cortex (LIP). The FEF (Brodmann area, 8; short *BA 8*) – which are either subsumed under the PFC or counted as an adjacted area (Miller & Cohen, 2001) – are important for the control of visual attention (Squire et al., 2013). Buschman and Miller analyzed when each individual neuron first reflected the location of the target (the moment when it displayed selectivity for the target). This measure enabled the temporal dissociation of the responses in the different areas dependent on the conditions of visual search task. In the conjunction condition (top-down attention), the frontal neurons (LPFC and FEF) displayed selectivity first. About a third of the recorded frontal neurons showed selectivity before the saccade towards the target had occurred. In contrast, only 14 % of the LIP neurons displayed selectivity before the saccade. The early frontal populations showed selectivity 50 ms (FEF) and 40 ms (LPFC) prior to the saccade, whereas the early LIP neurons only showed significant selectivity for the target location 32 ms *after* the saccade. In the pop-out condition (bottom-up attention), the early populations of the LIP neurons displayed selectivity for the target 170 ms prior to the saccade, while the early LPFC neurons showed selectivity 120 ms prior to the saccade and the early FEF neurons 35 ms prior to the saccade. When the LPFC neurons first showed selectivity, a quarter of the LIP neurons had already begun encoding the target location. While the frontal neurons responded faster to the target in the top-down condition, the parietal neurons responded faster to the targets in the bottom-up condition. It is notable that LIP neurons reacted faster to bottom-up targets than the frontal neurons to top-down targets. That is only to be expected, however, since the enforcement of instructions usually takes longer than a primarily salience-based selection. In sum, the results of the study suggest that selective top-down attention first originates in the PFC (and the FEF), whereas selective bottom-up attention originates in the parietal, sensory cortex (cp. Buschman & Miller, 2007). This study provides a basis for the hypothesis that selective top-down attention is more dependent on DA and selective bottom-up attention more dependent on ACh than vice versa. The reasoning for this assumption will be outlined in the next paragraphs.

3.2. The neural foundation of selective top-down attention

The effects of prefrontal DA on selective top-down attention are most likely established via the D1 receptors, or short *D1Rs* (Goldman-Rakic et al., 2000; Goldman-Rakic, Castner, Svensson, Siever, & Williams, 2004). These receptors are so widely distributed in the PFC that approximately every fourth prefrontal neuron expresses D1Rs (Noudoost & Moore, 2011b). The effect of this receptor subtype on the processing of visual signals was tested in three Rhesus monkeys *(Macaca mulatta)* through the application of SCH23390, a selective D1R antagonist (Noudoost & Moore, 2011b, 2011a). The monkeys fixated a display in which two stimuli appeared at varying onsets (see figure 3.2). The task was a *free-choice saccade* task (which means that the monkeys could freely choose which stimuli they fixated). It is generally expected that both stimuli are fixated with an equal probability (across all trials). Noudoost and Moore (2011b) infused a specific population of FEF neurons with a D1R antagonist. The receptive fields of the affected FEF neurons were mapped to *one* of the targets that appeared in the display. When the D1R antagonist was administered to the FEF neurons, stimuli that appeared in the receptive fields of the FEF neurons were selected more frequently (Noudoost & Moore, 2011a). Thus, the blockade of D1Rs had a beneficial effect on target selection. In another task, the Rhesus monkeys fixated a central point of the display while the stimuli (tilted bars) appeared at other locations. Some of the stimuli were mapped not only to the receptive fields of the FEF neurons, but also to the receptive fields of V4 neurons[16]. The activity of these neurons was measured through single-unit recording. Through this experimental design, top-down control signals from the PFC to the visual cortex could be captured and their effect on the processing of visual stimuli be assessed. The administration of the D1R antagonist to the FEF neurons led to enhanced responses of the V4 neurons towards stimuli that appeared in their receptive fields (see figure 3.2). This enhancement manifested itself in three ways: through an increased firing rate, through an increased selectivity for the orientation of the target and through a reduced response variability across the trials (Noudoost & Moore, 2011a). These enhancement effects were present both when the monkeys' only task was the fixation of the central point and when the monkeys were trained to covertly attend to the stimuli.

[16] The visual cortex consists of five visual areas (V1 – V5). V1 is the also termed the primary visual cortex or the striate cortex, while the other areas make up the extrastriate visual cortex.

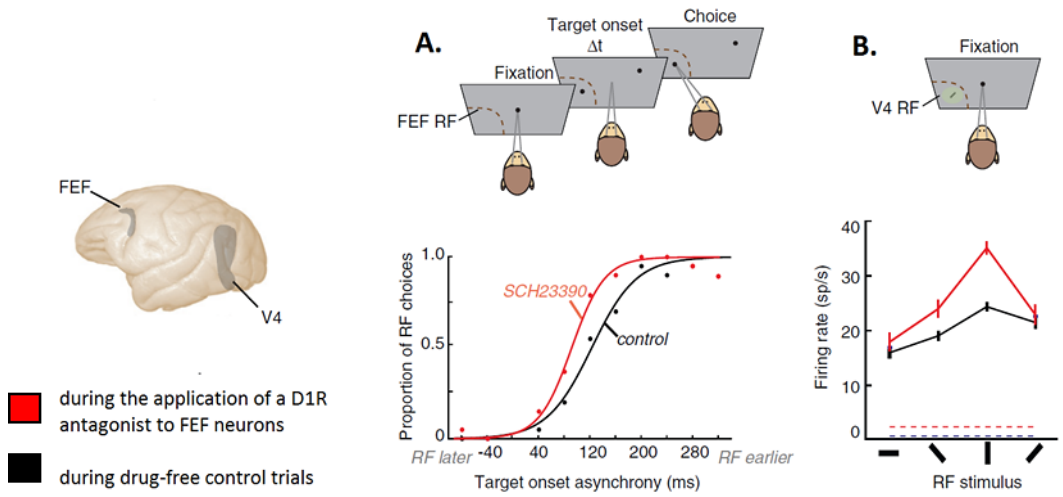


Figure 3.2.: The D1-mediated influence of the FEF on saccadic stimuli selection (figure taken and adapted from Noudoost and Moore, 2011a). (A) The monkeys fixated one of two stimuli during a trial of the free-choice saccade task. It is of interest whether the manipulation of the D1Rs in the PFC altered the tendency of the monkeys to make a saccade towards a specific stimulus. The graph depicts how often a saccade was made towards the receptive field stimuli depending on the temporal onset asynchronies of the stimuli appearance and depending on the dopaminergic manipulation (Noudoost & Moore, 2011a). The more leftward the curve is shifted, the more often a saccade was made towards stimuli within the receptive field. When the D1 antagonist was administered to the FEF neurons, the saccade was more often made towards the stimuli within the receptive field. (B) The magnitude of the activity of V4 neurons in monkeys that fixated a central point while stimuli appeared in other parts of the display. The V4 responses were greatest when a stimulus in their receptive field had the 'correct' orientation and when a D1R antagonist had been administered to FEF neurons that shared the same receptive fields (red line). Under the attenuation of the D1R activity in the FEF, the responses of corresponding V4 neurons in the extrastriate cortex were increased.

In sum, the blockade of D1Rs in the FEF led to beneficial effects on the saccadic target selection and increased the response magnitude of neurons in the visual cortex both during a task of covert attention and in the absence of an attentional task. In the latter case, the attenuation of the prefrontal D1R activity even led to attention-like effects in the V4 neurons (Noudoost & Moore, 2011a). This indicates that the dopaminergic activity in the PFC guides the processing of stimuli in the sensory cortices. Noudoost and Moore (2011a, p. 587) stated that "attentional control is achieved in part by PFC modulation of signals within sensory cortices" and elaborated that this route of top-down control might rely on prefrontal D1Rs to a large degree. However, it may be too simplistic to conclude that a *blockade* of D1Rs is universally beneficial for the transfer of top-down signals. Findings from working

memory tasks[17] show that both low doses of D1R antagonists and low doses of D1R agonists can exert positive effects (Clark & Noudoost, 2014). The key to understanding these adverse effects might be the proposed inverted-U curve relation between the amount of DA transmission and the efficiency of PFC function (Goldman-Rakic et al., 2000; Mattay et al., 2003). Intermediate levels of DA exert the most beneficial effects, according to this hypothesis; the effect of agonists and antagonists then depends on the dopaminergic baseline tone, which differs individually and is also susceptible to factors such as stress (Clark & Noudoost, 2014). In the light of the invasive nature of single-unit recordings and the mircoiontophoretic application of drugs, stress in the subjects is an important factor to consider. All of the reviewed findings are based on the study of non-human primates, which (on average) display a lower prefrontal DA tonus than humans (Palmatier et al., 2004). In humans, carriers of the Met/Met genotype of the *COMT* Val158Met polymorphism might be positioned at a more optimal region of the inverted-U curve of PFC function and DA signaling (Mattay et al., 2003; see subchapter 2.3). Here, a high prefrontal DA baseline tonus appears to be beneficial for the performance in PFC-dependent tasks (Dickinson & Elvevåg, 2009). It can be speculated that a comparatively *greater* activation of D1Rs in the PFC might lead to beneficial effects in humans. This line of argumentation will be continued in subchapter 7.2.

3.3. Measures and modulators of selective top-down attention

As discussed in subchapter 1.5, both the Stroop task and the Negative priming task are frequently used to tap selective top-down attention. What are the neural correlates of these tasks? If the findings of Buschman and Miller (2007) are to be generalized beyond visual search paradigms, the performance in these tasks should be associated with the function of the PFC. A modified Stroop task was used by MacDonald and colleagues (2000) to dismantle the neural correlates of the Stroop components. Before each trial, the participants were informed whether they had to read a word or indicate the color of a word (color-naming is the usually required response in Stroop tasks). The stimuli were presented after a delay to allow a greater temporal dissociation between the instruction phase and the response phase. During the execution of the task, the neural activity of the participants was accessed via an fMRI scanner. A double dissociation was noted by MacDonald and colleagues. The activity in the DLPFC (BA 9) was significantly increased in the color-naming condition, but not in the reading condition. The authors interpreted this finding to reflect the heightened need for top-down control in the color-naming condition. It is likely more effortful to maintain the instruction in this

[17] The performance in working memory tasks is hugely reliant on the PFC (Zaehle, Sandmann, Thorne, Jäncke, & Herrmann, 2011).

condition, since reading is a well routinized action, but color-naming is not. In contrast, the activity in the PFC-adjacent anterior cingulate cortex (BA 24, BA 32) was significantly increased in the incongruent, but not congruent trials of the color-naming condition. The incongruent trials of this condition are characterized by the highest degree of conflict (since a less automatic response has to be enforced against a more automatic response). All in all, the results suggest that the main role of the DLPFC lies in the task preparation (namely, the "representing and maintaining task demands" and "strategic control processes", p. 1836-1837), while the main role of the anterior cingulate cortex lies in the resolution of conflict. The neural correlates of components within the Negative priming task have been reviewed by Frings and colleagues (2014). The reviewed studies include a variety of different Negative priming tasks and will not be discussed in detail here. The role of the DLPFC, however, is particularly noteworthy (see figure 3.3). This region appears to be most consistently activated when the ignored repetition and control conditions of the Negative priming task are compared. In the ignored repetition, the prime distractor becomes the probe target, whereas prime and probe stimuli do not overlap in the control condition. The difference between these conditions reflects the typical Negative priming interference. The heightened activation of the DLPFC could reflect the more effortful maintenance of the task instructions in the ignored repetition, where the response-requiring target is constituted by the previously ignored distractor.

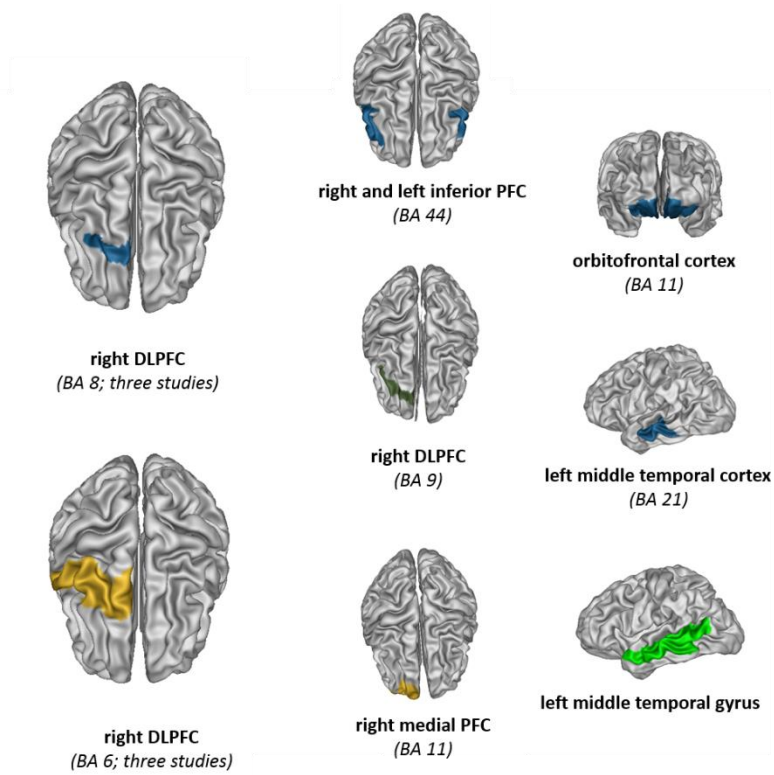


Figure 3.3.: Regions of activation in ignored repetition trials of the Negative priming task compared to control condition trials of the Negative priming task (figure taken and

adapted from Frings, Schneider, & Fox, 2014). While the prime distractor becomes the probe target in the ignored repetition, prime and probe stimuli do not overlap in the control conditions. The activated regions reflect the typical interference in Negative priming tasks.

In sum, areas of the PFC (especially the DLPFC) have indeed been associated with the performance in tasks of selective top-down attention. This supports the notion that the findings of Buschman and Miller (2007) can indeed be generalized beyond visual search tasks, consolidating the relevance of the PFC for this type of attention. The performance in the Stroop and Negative priming tasks will be utilized as dependent measures in this thesis, while genetic variants of the dopaminergic and cholinergic system will be assessed as possible modulators. Given the widespread associations between the *COMT* Val158Met polymorphism and the performances in PFC-dependent tasks (see subchapter 2.3), it only seems consequential that this polymorphism would modulate the performance in both the Stroop and the Negative priming task. Effects of the *DAT1* polymorphism on the dopaminergic metabolism in the PFC are most likely exerted indirectly, through the effects of this polymorphism in the striatum (see subchapter 2.4). In this regard it is of interest that striatal atrophy in early-stage patients of Huntington's disease was associated with a reduced performance in the Stroop task, the WCST and the Tower of Hanoi (Peinemann et al., 2005) – all three tasks are labelled as *executive* tasks and are established as tapping frontal lobe function, such as shifting between task sets or resolving interference (Miyake et al., 2000). In contrast, DA-depletion in the striatum did not affect the performance of rats in an adapted Posner-Cuing task, thus not influencing selective bottom-up attention (Ward & Brown, 1996). In sum, it is expected that both the *COMT* Val158Met polymorphism and the *DAT1* polymorphism modulate selective top-down attention. It is hypothesized that these variants have an effect on the performance in the Stroop task and Negative priming task. Given their particular distribution, it is not expected these variants modulate the performance in tasks of selective bottom-up attention.

3.4. The neural foundation of selective bottom-up attention

The findings from Buschman and Miller (2007) suggest that the LIP area is crucial for selective bottom-up processing. This area is part of the parietal lobe. Noudoost and Moore (2011a) speak more broadly of the "posterior areas" that modulate bottom-up processing. They include the sensory cortices in this term and thus do not focus solely on the parietal lobe. What is the role of ACh in these

brain areas? It has been noted that stimulations of the cholinergic basal forebrain nuclei can strengthen signals in the sensory cortices (Noudoost & Moore, 2011a). Increases in ACh transmission have also been associated with improved selective attention (Noudoost & Moore, 2011a). Herrero and colleagues (2008; also cp. Noudoost & Moore, 2011a) studied the effect of ACh by administering a task of covert selective attention to Rhesus monkeys *(Macaca mulatta)*. It was the monkey's task to fixate a central point at all times (see figure 3.4). A cue (a circle) appeared in one of two locations. The cue indicated which location had to be covertly attended by the monkey. After the cues disappeared, two stimuli were simultaneously presented. After 500 – 800 ms, a bright patch appeared in one of the stimuli. The monkeys had to release the bar they held when the patch appeared on the stimulus in the previously cued location (in this case, the stimulus became the target). When the patch appeared on the stimulus in the non-cued location, no response was required (this stimulus became the target). One of the possible stimulus locations was mapped to the receptive field of neurons in the primary visual (V1); the activity of these neurons was recorded. The stimuli were of different lengths and so differed in how well they mapped to the receptive fields. Herrero and colleagues examined how covert attention and the administration of ACh modulated the responses of V1 neurons to stimuli that appeared in their receptive field.

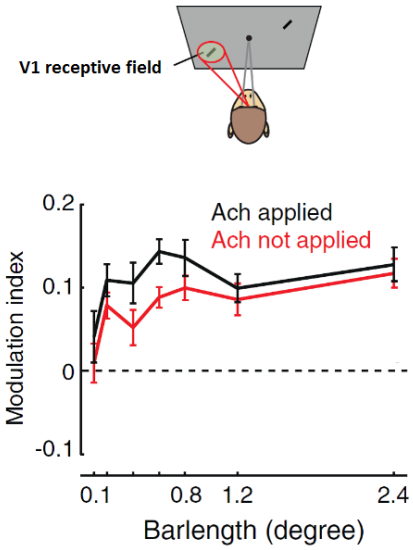


Figure 3.4.: The response of V1 neurons to stimuli of different lengths with *(black)* or without *(red)* the application of ACh (figure taken and adapted from Noudoost & Moore, 2011; cp. Herrero et al., 2008). The barlength indicates the size of the stimuli. Higher positive values of the modulation index indicate greater responses of the V1 neurons when attention was directed within the receptive field instead of outside of the receptive field. The results indicate that a heightened ACh transmission strengthens the effect of attention. Under the application of ACh, V1 neurons show a greater response to stimuli that appear in their receptive field.

There were three main results. First, the V1 neurons reacted more strongly when the stimuli were optimally mapped to their receptive fields. Second, the V1 reacted more strongly when attention was directed towards stimuli in their receptive fields. Third, the V1 neurons reacted more strongly when ACh was additionally administered (see figure 3.4). Both covert attention and ACh appear to determine the responses of the V1 neurons; the largest responses being observable in the presence of both. The design of this study can be slightly criticized in so far as components of selective top-down attention (which are instruction-related and reward-related) were not easily dissociable from components of selective bottom-up attention (which were manifested in the luminance change of stimuli, i.e. the patches). In a Posner-Cuing task, responses are required both in valid and invalid trials. The comparison of invalid trials to neutral and valid trials enables the measurement of bottom-up attention. In the task used by Herrero and colleagues, the monkeys *only* had to react when stimuli were validly cued. So while this study provides crucial evidence that ACh modulates the response strength of V1 neurons, it is unclear whether top-down or bottom-up attention was modulated here. This problem has been examined in Barn owls in regard to the cholinergic nucleus *isthmi pars parvocellularis* (IPC), which is termed the *parabigeminall nucleus* in mammals. The IPC is closely connected to the superior colliculus[18]. Importantly, this connection is "precise, reciprocal, and topographic" and leads to the innervation of "essentially all" layers and columns of the superior colliculus (Maczko, Knudsen, & Knudsen, 2006; p, 12799). It was reported that the IPC forms maps of visual stimuli as well as maps for auditory stimuli (Maczko et al., 2006). Since the IPC modulates the sensitivity of neurons in the superior colliculus (Maczko et al., 2006) and the superior colliculus is important both for the direction of gaze and the selection of stimuli, this could mean that the IPC helps to form a *salience map* in the superior colliculus (Asadollahi, Mysore, & Knudsen, 2010). A salience map is an internal, spatially precise representation of the saliences of the stimuli in the visual field (or other modalities). Asadollahi and colleagues recorded the response of IPC neurons in Barn owls. They tested the responses of these neurons to a variety of stimulus features – such as direction, size, orientation, visual contrast and speed of motion. They found that IPC neurons were sensitive to "changes in contrast, loom speed, translation speed and sound level, with most units increasing their firing rates with increasing stimulus strength" (p. 2573). In addition, the neurons reacted "almost invariantly" to stimuli as long as they were mapped to their receptive fields and more salient than a competing distractor stimulus (also cp. Noudoost & Moore, 2011a; see figure 3.5). Thus, the IPC neurons did not respond to the *absolute* intensity of the stimuli, but to the *relative* saliencies[19].

[18] The superior colliculus is also commonly referred to as the *optic tectum.* This term is misleading, since the structure serves multisensory processing and is not limited to the visual modality (Illing, 1996).

[19] Noudoost and Moore (2011a) stated that "the magnitude of response decreases sharply at the boundary where the relative salience of the [receptive field] stimulus falls below that of a stimulus outside of the [receptive field]" (p. 586). It seems that the IPC neurons almost react in a binary, *all-or-nothing* fashion.

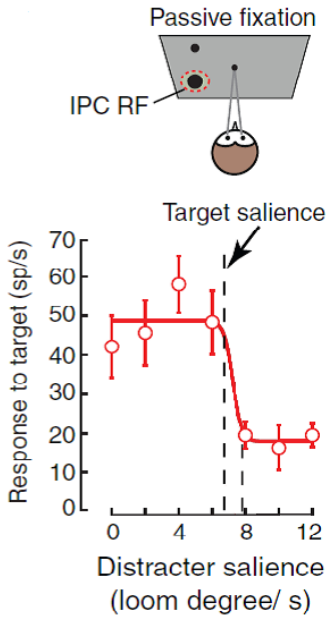


Figure 3.5.: The response of IPC neurons to a stimulus in their receptive field in dependence on the salience of a competing stimulus (figure taken and adapted from Noudoost & Moore, 2011; based on Asadollahi et al., 2010). The owl fixates a central point. Two stimuli – expanding dots – are presented simultaneously. One of these dots is mapped to the receptive field of the recorded IPC neurons while the other stimulus appears outside of their receptive fields. The response of the IPC neurons to the stimulus in their receptive field is measured in spikes per second. The salience of the distractor stimulus is indicated by the degree of the expansion per second. Accordingly, this graphic depicts the response of IPC neurons to stimuli in their receptive field in dependence on the speed with which a competing distractor stimulus expands. The IPC neurons either respond strongly (when the receptive field stimulus is more salient than the distractor stimulus) other they respond weakly (when the receptive field stimulus is less salient than the distractor stimulus). This switch-response (black arrow) occurs at an expansion of approximately 6° per second.

The properties of the IPC neurons indicate that the IPC is ideally suited to form a salience map of the visual or auditory field, and possibly also other modalities (Asadollahi, et al., 2010). Not only do these neurons respond to relative salience; they also enhance signaling when the relative saliences are too similar. The result of these computations is projected to the superior colliculus (Asadollahi, et al., 2010). Salience, of course, is the core concept of bottom-up attention and a contributing factor to target selection. The term “salience map” originates from the field of visual search. Noudoost and Moore (2011a) did not interpret the connection between the IPC and the superior colliculus in the context of a psychological theory, though the Guided Search theory (Wolfe, 1994), for example, is both well-established and offers an additional point of view in its theoretical conceptualizations.

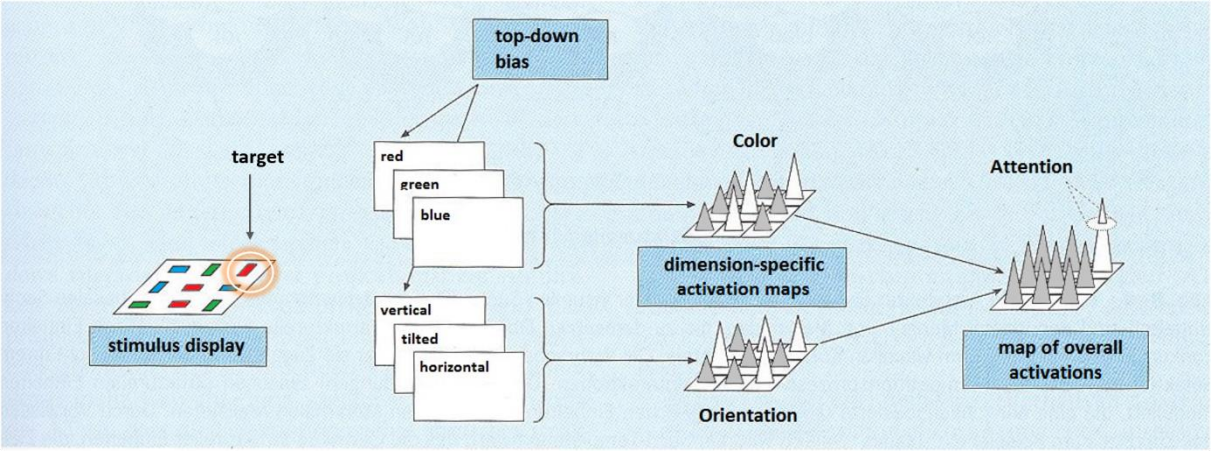


Figure 3.6.: Visual search according to the Guided Search theory (Cave & Wolfe, 1990; figure taken and adapted from Müsseler & Prinz, 2008). In this theory, both bottom-up and top-down mechanisms are presumed to guide the allocation of attention to different locations. In this example, the vertical red bar is the target stimulus that has to be detected among distractors. Activation maps are computed for specific dimensions. It is assumed that both the orientation (vertical) and the color (red) are positively biased through a top-down mechanism. In the color map, red stimuli are activated most. In the orientation map, vertical stimuli are activated most. The dimension-specific activation maps are subsequently condensed into an overall map of the activations. Attention is allocated to the location with the highest activation on the overall map of activations (in this case the target).

In visual search tasks, participants have to identify a target amongst an array of distractor stimuli. Targets that differ from the distractors in one feature can easily be found (they "pop out"), but targets that share features with distractors take longer to identify. In the Guided search theory (Wolfe, 1994; Wolfe & Gancarz, 1997; Wolfe, 2007; Müsseler & Prinz, 2008), it is assumed that both bottom-up and top-down mechanisms determine which part of a visual field is selected for attentional processing (see figure 3.6). According to this theory, different features in the visual field are first processed separately and in a fast, parallel fashion (cp. Wolfe, 1994, for the following paragraph). The visual field is internally represented through maps. These maps are dimension-specific; separate for color, orientation, and so on. Locations on these maps are differentially activated and so help to determine which locations should be selected for further attentional processing. Both bottom-up and top-down mechanisms contribute to the activation of different locations on the map. Wolfe describes bottom-up activation as based on the differences between the item in one location and the items in the surrounding locations. If an item in one location is particularly salient in one dimension, this location will be more strongly activated on the corresponding feature map. For example: if a red bar is surrounded by blue bars, the location of the red bar will be more strongly activated on the dimension map for color. In this

way, the salience of individual items determines the activation values of locations on the dimension-specific maps. In contrast, top-down activation is not based on salience, but on the task instructions and goals. The task might for example consist in selecting the vertically oriented item. If one bar is vertically oriented while the other bars are horizontally oriented or tilted, the location of the vertical bar will be more strongly activated on the dimension map for orientation. Top-down mechanisms influence activations based on pre-existing knowledge. The selection of a target often relies not only on one dimension, but several dimensions. If the target is a red, vertical bar, the maps of color and orientation are the ones that are crucial to determine a correct response. The activations on these maps are determined both by bottom-up and top-down influences. How are different maps integrated? Wolfe assumes that the dimension-specific activations are computed into an overall map of activations. This map contains the added-up, weighted activation values of all relevant dimension-specific maps. The location of the red, vertical bar should be most strongly activated on the overall activation map[20]. This location should then be selected for further attentional processing (which would lead to the participant finding the target). The Guided Search theory relies on the assumption of location-based attention and is an expansion of the Feature Integration theory (Treisman & Gelade, 1980; Treisman, 1985). The Guided Search theory is highly compatible with findings from animal studies, where the role of the IPC and the superior colliculus has been highlighted. Asadollahi and colleagues (2010) hypothesized that the IPC forms a *salience map* (by responding to the "relative strength of competing stimuli", p. 893). Notably, the IPC is the recipient of direct top-down projections from the gaze control area that is located in the owl's forebrain (Asadollahi, et al., 2010). It seems likely that the IPC is not simply a structure that helps to form salience maps, but possibly the neuroanatomical correlate of Wolfe's dimension-specific activation maps. The IPC is topographically organized, responds to visual and auditory saliences (which reflects the bottom-up component) and is also receptive to top-down influences. Wolfe's overall map of activations might be formed either in the IPC or (more likely) in the superior colliculus, which exhibits the signs of forming a corresponding topographical map and receives projections from the IPC. As the IPC (or in mammals, the parabigeminal nucleus[21]) is a cholinergic nucleus, ACh seems to play an important role in modulating visuospatial bottom-up attention. In their review article, Dutta and Gutfreund (2014) stated that the superior colliculus is crucial both for overt and covert attentional shifts to the most salient stimulus; the computations that are generated here are transferred to a wide range of other areas for further

[20] Wolfe's (1994) model explicitely assumes the existence of computational errors, so the target location does not *necessarily* become the most activated location on the overall map of activations. The example is simplistic in that regard.

[21] While the role of the IPC is relatively well understood, the role of the parabigeminal nucleus in humans has been the object of far less studies. A similar link between this nucleus and the superior colliculus can be assumed, but more studies are needed to corroborate this assumption (Krauzlis, Lovejoy, & Zénon, 2013).

processing. Interestingly, they stated that the superior colliculus is “homolog in all vertebrate species” and “one of the most phylogenetically conservative structures in the brain” (p. 3). This indicates that the processes that take place here are universally important in vertebrates – which would certainly apply to salience-based target selection.

3.5. Measures and modulators of selective bottom-up attention

As discussed in subchapter 1.6, both the Posner-Cuing task and the Dot Probe task are frequently used to tap selective bottom-up attention. What are the neural correlates of these tasks? Buschmann and Miller (2007) suggested that bottom-up selection first arises in the intraparietal cortex and adjacent regions. An important role of the superior colliculus can also be expected for salience-driven selection (Asadollahi, et al., 2010; Dutta & Gutfreund, 2014). In positron emission tomography (PET) studies of spatial cuing tasks, exogenous and endogenous attention were found to activate similar, but not identical regions in both the parietal and frontal cortex (Corbetta, 1998). Hopfinger and West (2006) recorded event-related potentials and found that exogenous attention modulated “the late phase of the extrastriate-cortex-generated P1 component” (p. 774); this was the earliest stage at which attention modulated the sequence of information processing. Salmi and colleagues (2009) argued for “largely overlapping” networks of both types of attention, finding activation via fMRI in the superior parietal, temporopariatal and frontal areas, while Hahn and colleagues (2006) argued for “largely dissociated networks”. They found the tempoparietal junction (amongst other areas) especially activated by stimulus-driven attention. The performance in the Dot Probe task has been associated with the intraparietal cortex, the visual cortex (both the V1 and the extrastriate areas) and other areas such as the amygdala or the ventrolateral prefrontal cortex (Wenzel, 2012). Dot Probe tasks are often relatively diverse, especially in regard to the utilized stimulus material. The neural correlates reflect this diversity. In sum, areas of the intraparietal cortex have indeed been associated with the performance in the Posner-Cuing task and Dot Probe task, as suggested by Buschman and Miller (2007). The involvement of frontal areas has also been highlighted, however. Notably, PET and fMRI sessions do not provide the same spatial and temporal resolution as single-unit recordings (which are precise on an ms-level), so that these findings cannot fully help to dissociate the role of these brain areas for top-down and bottom-up attention (or in other words, these reports cannot determine *when* the different populations of neurons started to process the target). Witte and colleagues (1997) conducted a pharmacological study. The performance of two female Rhesus monkeys in a Posner-Cuing task was observed dependent on the administration of either saline or nicotine (Witte et al., 1997). While the smallest doses of nicotine had no effect, intermediate and high doses led to reduced

Posner effects (i.e., to a reduction of the average RT difference between valid and invalid trials). These reduced Posner effects were caused by an increase in the response speed in invalid trials. In the same study, the Posner-Cuing task was also administered to human participants, who were either smokers or non-smokers. Smokers consumed one cigarette of their usual brand immediately prior to testing. They displayed significantly smaller Posner effects than non-smokers (see figure 3.7). This effect was driven by the higher response speed of smokers in invalid trials.

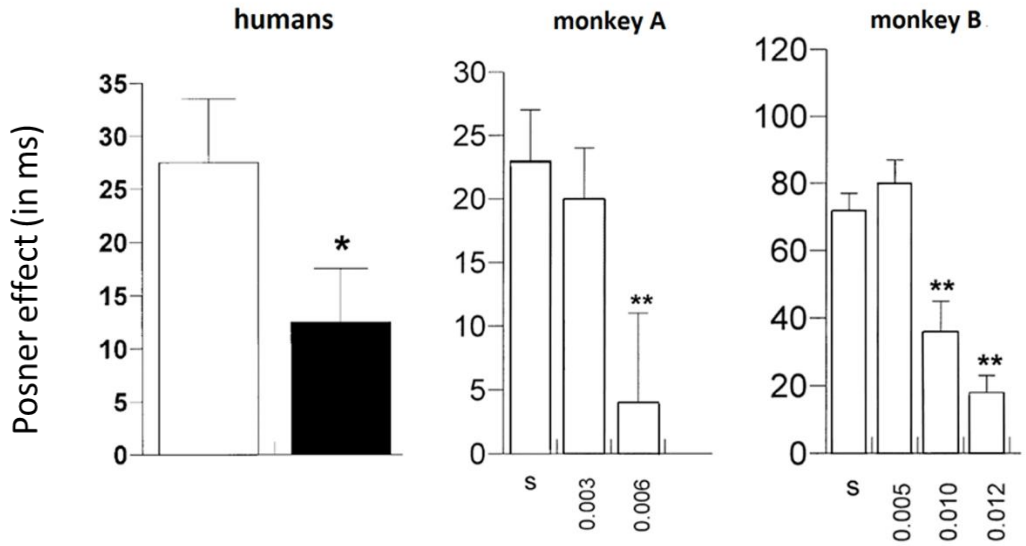


Figure 3.7.: The Posner effects in a Posner-Cuing task in humans and monkeys in dependence of nicotine intake (figure taken and adapted from Witte et al., 1997). Human smokers (black bar) displayed significantly reduced Posner effects compared to non-smokers (white bar). For the monkeys, *s* indicates saline (placebo) trials; the nicotine doses are given in milligram per kilogram bodyweight. Monkey A received lower doses of nicotine than monkey B. Witte and colleagues (1997) pretested which doses of nicotine were similarly effective for both monkeys and found that monkey A responded more sensitively to nicotine. Monkey A displayed a significantly reduced Posner effect at the highest nicotine dose. Monkey B displayed a significantly reduced Posner effect at the intermediate and highest doses of nicotine.

The equivalent effects of nicotine in humans and monkeys suggest that a heightened cholinergic transmission (as achieved via the agonist) lessens the impact of the validity of the Posner stimuli. Under the influence of the agonist, human smokers and nicotine-consuming monkeys were less affected by highly salient cues in invalid trials. These results indicate not only that the cholinergic system is involved in modulating visuospatial bottom-up attention, but also that specifically the nAChRs play an important role. Notably, the superior colliculus is one of the areas in the brain with the highest density of nAChRs (Clarke, Schwartz, Paul, Pert, & Pert, 1985). NAChRs are also present in sensory cortices like the V1, where they increase the contrast sensitivity and responsiveness of the thalamorecipient neurons in

layer IV (Noudoost & Moore, 2011a). These neurons are the "primary recipient of sensory input from the thalamus" and are responsible for distributing this information for additional processing (Xu, Jeong, & Rudy, 2013, p. 155). It stands to reason that naturally occurring variants within the nAChRs might modulate the processing of salience and thus impact on bottom-up attention. In this thesis, three polymorphisms in genes of the nAChR will be examined as possible modulators of selective bottom-up attention. The *CHRNA4* rs1044396 polymorphism has been linked to the numbers of nAChRs in a high-affinity state; the *CHRNA5 rs3841324* polymorphism has been associated with the level of *CHRNA5* mRNA expression and the *CHRNA5* rs16969968 polymorphism has been reported to influence the responsiveness of nAChRs towards an agonist (cp. subchapter 2.6). Due to their biological relevance within the cholinergic system, it is expected that these polymorphisms will modulate the performance in the Posner-Cuing task and in the Dot Probe task. At the same time, it is expected that these variants will *not* influence the performance in the Stroop task or Negative priming task to the same extent, as these tasks primarily tap selective top-down attention.

Summary. Selective top-down attention and bottom-up attention originate in different areas of the brain. The PFC (including the FEF) seems to be particularly relevant for top-down attention. The dopaminergic system might crucially modulate top-down attention through the D1Rs. DA is assumed to influence the PFC function via an inverted-U response curve, with intermediate levels of DA exerting the most beneficial effects. It is expected that two variants of the dopaminergic system (the *COMT* Val158Met polymorphism and the *DAT1* polymorphism) modulate the performance in the Stroop and Negative priming task. Selective bottom-up attention has been associated with the IPC, the superior colliculus and the sensory cortices. The cholinergic IPC might be responsible for forming dimension-specific activation maps and might help to form the overall activation maps in the superior colliculus. It is expected that three variants of the cholinergic system (the *CHRNA4* rs1044396, *CHRNA5 rs3841324,* and the *CHRNA5* rs16969968 polymorphisms) modulate the performance in in the Posner-Cuing and Dot Probe task, which measure selective bottom-up attention. The study design is beneficial in terms of convergent and discriminant validity, allowing the representation of both the dopaminergic system and cholinergic system by multiple variants. Noudoost and Moore (2011a, p. 589) asked for "more rigorous behavioral and psychophysical paradigms", stressing the importance of "clearly isolate[ing] the particular varieties of attention observable in human subjects". The employed tasks in this thesis provide clear-cut behavioral phenotypes, which aligns with the demand of Noudoost and Moore.

Green and colleagues (2008, p.710) also stated:

> *"…even the most precise molecular-genetic data cannot be useful if the phenotypes are not well defined. Thus, cognitive-neurogenetic studies are only as good as their ability to measure mental phenotypes validly and specifically; clear psychological theory and rigorous psychometrics are essential. In particular, describing and parsing the components of psychological functions will require well-developed behavioural tasks to ensure that the components that are being investigated are the ones that are actually being measured."*

This thesis might help to dismantle selective top-down and bottom-up attention on a molecular biological level, and might expand findings that so far have been observed in animals. The genetic approach enables the examination of naturally occurring variations within the DA and ACh neurotransmitter systems in human participants.

Chapter 4

The Objective of the Thesis

The primary objective of this thesis is the examination of the influences of the dopaminergic and cholinergic neurotransmitter systems on selective top-down and bottom-up attention. Based on the studies summarized in chapters two and three, the following assumptions are drawn: It is expected that the influence of the dopaminergic neurotransmitter system on selective top-down attention is greater than the influence of this system on selective bottom-up attention. Thus, it is assumed that the *COMT* Val158Met polymorphism and the *DAT1* polymorphism impact on the performance in the Stroop task and in the Negative priming task, but not (to the same extent) on the performance in the Posner-Cuing task and in the Dot Probe task. Specifically, it is expected that Met allele carriers of the *COMT* Val158Met polymorphism and carriers of the 10-repeat allele of the *DAT1* polymorphism display a better performance in both tasks. In alignment with the differing levels of COMT activity reported by Chen and colleagues (2004), the sample will be subdivided into Val allele homozygotes and carriers of the Met allele (Val/Val vs. Met+). Gender will be considered as a factor in all analyses of this polymorphism, since gender-based differences haven been reported repeatedly (Xie, Ho, & Ramsden, 1999; Chen et al., 2004; Tunbridge et al., 2006). Conversely, it is also expected that the influence of the cholinergic neurotransmitter system on selective bottom-up attention is greater than the influence of this system on selective top-down attention. It is assumed that the *CHRNA4* rs1044396 polymorphism, the *CHRNA5* rs3841324, and the *CHRNA5* rs16969968 polymorphism impact on the performance in the Posner-Cuing task and the Dot Probe task, but not (to the same extent) on the performance in the Stroop task and Negative priming task. Specifically, it is expected that carriers of the T allele of the *CHRNA4* rs1044396 polymorphism display a better performance in the Posner-Cuing task and the Dot Probe task. In regard to the *CHRNA5* rs3841324 polymorphism and the *CHRNA5* rs16969968 polymorphism, the analysis will be exploratory and no prior assumptions about their alleles will be drawn. The analyses of these polymorphisms will be based on binary groups. The rs3841324 polymorphism will be analyzed on basis of comparisons between homozygous S allele carriers and L allele carriers (S/S vs. L+), since Wang and colleagues (2009) reported significant differences in *CHRNA5* mRNA expression between S allele homozygotes and L/S heterozygotes, but not between L/S heterozygotes and L allele homozygotes. The *CHRNA5* rs16969968 polymorphism will be analyzed on basis of comparisons between the homozygous G allele group and the group of A allele carriers (G/G vs. A+), since the A allele was singled out as the risk allele on a behavioral level (Bierut et al., 2008). Gender will be considered as a factor in these analyses and the diplotypes of both polymorphisms will be examined as well (cp. Wei et al., 2011). Altogether, a double dissociation between the polymorphisms of the dopaminergic system and the polymorphism of the cholinergic system is expected in regard to selective top-down and bottom-up attention. In the following chapters, this assumption will be referred to as the *dissociation hypothesis* (cp. Noudoost & Moore, 2011a).

The secondary objective of this thesis is the study of cholinergic modulations of response speed. It is expected that the three cholinergic polymorphisms modulate response speed in the Stroop task, Negative priming task, Posner-Cuing task, and Dot Probe task (or in other words, irrespective of the required type of attentional selection). For the *CHRNA4* rs1044396 polymorphism, it is expected that the average RTs in all four tasks increase linearly with the C allele dosage (Greenwood et al., 2005). In regard to *CHRNA5* rs3841324 polymorphism and *CHRNA5* rs16969968 polymorphism, the scarcity of preceding studies allows only explanatory analyses. At this point, no assumption can be made which alleles of these polymorphisms might have beneficial effects on response speed. Yet again, the analyses of both *CHRNA5* polymorphisms will be based on binary groups. In the case of the rs3841324 polymorphism, the groups will be composed of S allele homozygotes and carriers of the L allele (S/S vs. L+). In the case of the rs16969968 polymorphism, the groups will be composed of G allele homozygotes and A allele carriers (G/G vs. A+). Gender will be considered as a factor in these analyses and the diplotypes of both polymorphisms will be examined as well.

Chapter 5

Methods

This chapter provides detailed information about the different components of the study. First, the composition of the sample will be explained in more detail (subchapter 5.1). Then, the protocol of the study will be outlined (subchapter 5.2). In subchapter 5.3, the materials and apparatus of the employed tasks will be described, and in subchapter 5.4, the procedure of the tasks will be specified. Details on the pen-and-paper control measures will be reported in subchapter 5.5, while the details of the genetic analyses will be given in subchapter 5.6.

5.1. Participants

182 participants from the University of Trier gave written consent. They were given the opportunity to get excluded from the study at any time if desired. Their median age was 21 years (ranging from 18 to 57 years) with 71.9 % of the participants being female. At the beginning of the experimental session, they were informed about the purpose of the study. Yet, references to the specific hypotheses were not given. The participants were encouraged to show their best performance in the experiments. As an incentive, they were informed that the best four participants (in regard to response speed and precision) would receive a gift certificate worth 50 €. Regardless of their performance, the participants received course credit for their participation in the study. All of the participants had normal or corrected-to-normal vision. The participants' medication status and dyschromatopsia were checked by self-report. Other control question were aimed at the consumption of nicotine, the level of physical activity and fitness, the level and type of education, the handedness, preexisting symptoms of ADHD and the ability to touch-type[22]. Ethnic data was not collected, as the risk of population stratification was low in this sample[23]. The experimental protocol was approved by the ethics committee of the University of Trier and was conducted in accordance to the latest revision of the Declaration of Helsinki.

[22] Touch-typing is the ability to use a keyboard without constant visual aid. It was assessed whether the participants were able to touch-type with the (German) standard system for finger placement and positioning.

[23] Recently, Steffens and colleagues (2006) have reported on the low extent of genetic heterogeneity in German samples. They compared three samples (with more than *n* = 700 participants each) that were drawn in southern Germany, northern Germany and north-east Germany. The F_{ST} value (as a coancestry coefficient) neither differed between the northern and southern sample nor between the northern and north-east sample. There was a small, but significant F_{ST} difference between the southern and north-east sample, which were geographically the widest apart. The author stated that the F_{ST} values between the most distant samples were "2-4 times lower than the F_{ST} value between Germans and other Germanic populations (Dutch, Danish, English, Austrian, Swiss, Belgians)" (p.27). The differences between their samples accounted for "far less than 5% of the total variance" (p. 26) and were too small to serve as a foundation for the prediction of population membership. As this study was conducted in Trier, situated in middle Germany, population stratification was not expected.

5.2. Study protocol

Up to three participants took part in the study at the same time (for a visualization of the study protocol, see figure 5.1). It took about two hours to participate in the study. During this time, the participants could drink as much water as they liked but were not allowed to eat. The experimental session started with briefing the participants. After they were fully informed and had given their consent to participate in the study, each of them were seated in soundproof chambers. They were asked to answer a set of demographic questions presented on the computer. Before the experiments started, it was ensured that all participants were placed 60 cm in front of the screen (to this end, chin rests were used). Each task was fully computerized. The instructions were given on screen. A task was only started when all participants had finished the previous task. The Stroop task, Negative priming task and Posner-Cuing task were conducted in a randomized order. To eliminate the possibility of carry-over effects, the Dot Probe task was always conducted last[24]. After completing the experiments, the room was changed and the participants were asked to fill out three paper-and-pencil questionnaires. The questionnaires were handed out in a randomized order. These questionnaires were the ADHD Self-Report checklist (ADHD-SR) (Rösler et al., 2004), the Cognitive Failure Questionnaire (*CFQ;* Broadbent, Cooper, FitzGerald, & Parkes, 1982; Lumb, 1995) and the Action Control scale (Kuhl, 1990). Of the three questionnaires, only the ADHD-SR questionnaire is a component of this thesis and will be introduced in detail. After the questionnaires were filled out, the revised D2 Test was administered to the participants (Brickenkamp, Schmidt-Atzert, & Liepmann, 2010). The test instructions were first given by the experimenter. The participants then worked on a practice sheet to acquaint themselves with assignment. When they had completed the sheet correctly, the test was started. Due to the length of the study, the participants were given the opportunity of taking breaks between the various parts. At the end of the experimental session, saliva was sampled from the participants and they were thanked for their participation in the study.

[24] The Dot Probe task was the only task in which affective stimuli were used. These stimuli were pictures of positive or negative valence and had high arousal values (additionally, 'neutral' pictures of intermediate valence and low arousal values were used).

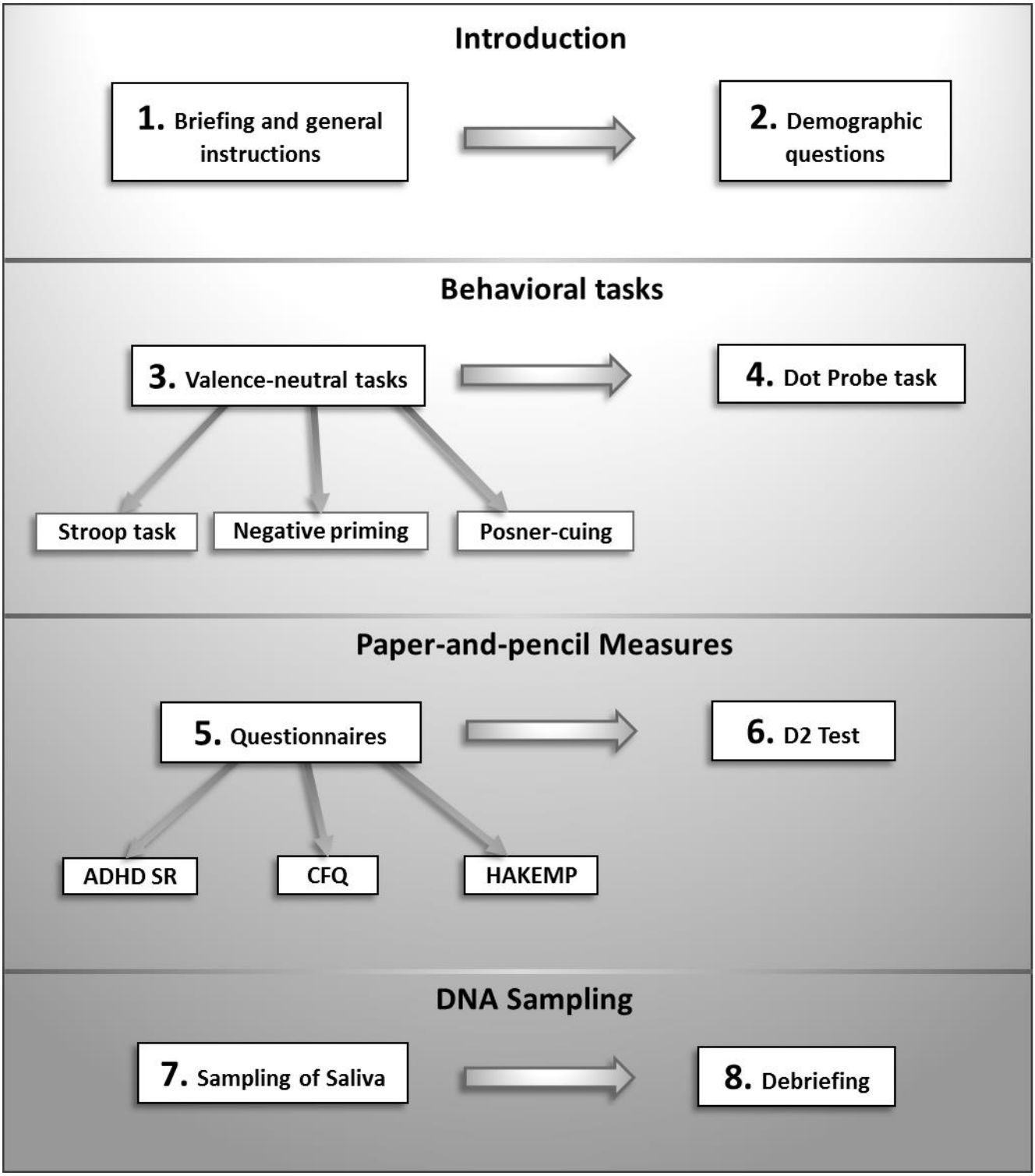


Figure 5.1.: The study protocol. The participants were first briefed (1) and then answered a set of computerized questions (2). Afterwards, the experimental tasks were administered to them – first the valence-neutral tasks in a randomized order (3) and then the Dot Probe task (4). Next, the questionnaires were handed out in a randomized order (5) and the D2 test was administered (6). As the last step, saliva was sampled from the participants (7) and they were debriefed (8).

5.3. Materials and apparatus

The experiments were conducted using the E-Prime software (version 2.0; Psychology Software Tools, Pittsburgh, PA, USA) and were carried out on standard PCs and monitors. Responses were made on QWERTZ-type keyboards. For the Stroop task the stimuli were the words "red" and "blue" in the font type Calibri, either presented in red or blue font color. Each letter was about 0.57° high and 0.48° wide. The background was white. In the Negative priming task, the stimuli were the letters "D", "F", "K" and "J" in the font type Calibri, presented black on a white background. Each letter was about 0.69° high and up to 0.48° wide. In the Posner-Cuing task, the stimuli were the letter "T" and the sign "+" in the font type Calibri, presented in black font color. Each sign measured about 0.57° x 0.48°, and was presented 2.39° lateral to the central fixation marker. The background was grey and the whole stimuli setup measured about 7.15° in width and 1.43° in height. In the Dot Probe task, the target stimulus was a white dot that measured about 0.29° in width and height. The cue stimuli were pictures that measured about 6.20° in width and 3.82° in height. There were 20 neutral, 20 positive, and 20 negative pictures. These pictures were selected from a normed database, the International Affective Picture System (Lang, Bradley, & Cuthbert, 2008). The pictures in this database are rated for affective valence and arousal on a scale of 1 – 9. The neutral stimuli chosen for this study had an overall valence rating of *M* = 4.97 (*SD* = 0.37; ranging from 4.23 to 5.59) and an overall arousal rating of *M* = 2.66 (*SD* = 0.53; ranging from 1.72 to 3.66). The positive stimuli had an overall valence rating of *M* = 7.61 (*SD* = 0.43; ranging from 6.65 to 8.34) and an overall arousal rating of *M* = 5.64 (*SD* = 0.89; ranging from 4.11 to 7.31). The negative stimuli had an overall valence rating of *M* = 2.37 (*SD* = 0.55; ranging from 1.62 to 3.79) and an overall arousal rating of *M* = 6.46 (*SD* = 0.72; ranging from 4.57 to 7.35). The background was grey and the whole stimuli setup measured about 15.94° in width and 3.82° in height.

5.4. Procedure

The Stroop task. The participants were instructed to place the left index finger on the key C of the computer keyboard and the right index finger on the key M of the computer keyboard. It was their task to indicate the color of a word (either blue or red) while ignoring the word content (either blue or red). They did this correctly by pressing C for words colored in blue and M for words colored in red. When all participants had confirmed to the experimenter that they understood the instructions, a block of 24 practice trials was started. If no further questions remained, the experimental trials were started subsequently. A typical Stroop trial consisted of the following events (see figure 5.2): A blank screen was shown for 1000 ms. A fixation marker ("+") then appeared at the center of the screen for 500 ms.

The stimulus was then presented until the participants responded to it. If the response had been wrong or too slow (RT > 2000 ms), a feedback display was inserted for 1500 ms before the experiment resumed with the next trial. The Stroop task consisted of 72 trials, 36 of which were congruent (the word color equaled the word content) and 36 of which were incongruent (the word color did not equal the word content). The RT differences between the congruent and incongruent condition typically result in the *congruency effect* or *Stroop effect*, i.e. the usually slowed and more error-prone responses to stimuli of interference.

The Negative Priming task. The participants were instructed to place the middle and index fingers of both hands on the keys D, F, J, and K of the keyboard. It was their task was to indicate the middle letter of a three-letter-string. They did this correctly by pressing the corresponding key (either D, F, J or K). When all participants had confirmed to the experimenter that they understood the instructions, a block of 24 practice trials was started. If no further questions remained, the experimental trials were started subsequently. A typical negative priming trial consisted of the following events (see figure 5.2): The participants started the trial by pressing the spacebar. A fixation marker ("+") appeared at the center of the screen for 400 ms. The prime display was then presented until the participants responded to the stimuli. If the response had been wrong or too slow (RT > 2000 ms), a feedback display was inserted for 1500 ms. Next, a blank screen was shown for 200 ms. Afterwards, a fixation marker was presented for 400 ms. The probe display then appeared until the participants responded. As before, a feedback display was inserted for 1500 ms if the response had been wrong or too slow (RT > 2000 ms). The experiment then resumed with the next trial, which the participants started themselves by pressing the space bar. After half of the trials, the participants had the option to take a short break. The Negative priming task consisted of 144 trials. Of those, 48 belonged to the attended repetition condition, where the prime target was repeated as the probe target. A further 48 trials belonged to the control condition, where prime and probe stimuli were completely distinct. Another 48 trials belonged to the ignored repetition condition, where the prime distractor was repeated as the probe target. The RT differences between the attended repetition and control condition usually result in a positive priming effect (short *PP effect*), i.e. the usually faster and less error-prone responses after a prime target is repeated as a probe target. The RT differences between the ignored repetition and control condition typically result in the NP effect[25], i.e. the slowed and more error-prone responses after a prime distractor is repeated as a probe target.

[25] Attending to an object makes a subsequent response to the same object easier, while ignoring an object makes a subsequent response to the same object more difficult (Fox, 1995). In this regard, the PP effect and NP effect seem to be mirror each other perfectly (hence the respective terms). This conclusion is somewhat misleading, however, since the underlying processes are not necessarily identical (Fox, 1995; May et al., 1995; Frings et al., 2014).

The Posner-Cuing task. The participants were instructed to place a finger on the key B of the computer keyboard. It was their task to indicate the appearance of the target letter T by pressing the key B. When all participants had confirmed to the experimenter that they understood the instructions, a block of 24 practice trials was started. If no further questions remained, the experimental trials were started subsequently. A typical trial consisted of the following events (cp. figure 5.3): A fixation marker ("*") appeared at the center of the screen for 1500 ms. A bright cue then illuminated one of the two boxes for 50 ms. The fixation marker reappeared for 100 ms. The target letter T was then presented in one of the boxes while the other box depicted a + symbol. The stimuli remained on screen until the participants responded. If the response had been wrong (hitting a wrong key or reacting in the catch trial condition) or too slow (RT > 2000 ms) a feedback display was inserted for 1000 ms. The Posner-Cuing task consisted of 360 trials, with 150 being valid (both cue and target were presented left in 75 cases and right in 75 cases) and another 150 being invalid (the cue was presented left in 75 cases and right in 75 cases, while the target was presented on the corresponding alternative position). Further 30 trials were neutral (the cues were presented on both positions) and 30 trials were catch trials (two + symbols were presented, but no target). Since valid and invalid trials occurred with an equal frequency, this task was an *exogenous* Posner-Cuing task. The RT differences between the valid and invalid condition typically result in the *validity effect* or *Posner effect*, i.e. the usually slowed responses after the wrong location has been cued.

The Dot Probe task. The participants were instructed to place the middle finger and the index finger of their dominant hand on the keys G and H of the computer keyboard. It was their task to indicate the location of a dot by pressing the key G when the dot appeared at the left side of the fixation marker and by pressing the key H when the dot appeared at the right side of the fixation marker. When all participants had confirmed to the experimenter that they had understood the instructions, a block of 10 practice trials was started. If no further questions remained, the experimental trials were started subsequently. A typical trial consisted of the following events (cp. figure 5.3): A fixation marker ("*") appeared at the center of the screen for 1000 ms. The picture pair was then presented for 500 ms. The target dot subsequently appeared either left or right and remained on screen until the participants responded. If the response had been wrong or too slow (RT > 2000 ms) a feedback display was inserted for 1000 ms before the next trial started. The Dot Probe task consisted of 110 trials. There were 40 trials with a neutral-positive picture pair and 40 trials with a neutral-negative picture pair. In these trials, the affective picture and the target dot appeared equally often at each location[26]. Half of the 40 trials were valid and half were invalid. The affective picture and the target appeared at the same location in valid trials and at opposite locations in invalid trials. The RT differences between the valid

[26] Subsequently, the 40 trials could be divided into four subcategories: (1) *affective picture right, target left,* (2) *affective picture left, target right,* (3) *affective picture right, target right,* (4) *affective picture left, target right.*

and invalid condition typically result in the *Dot Probe effect*, i.e. the typically slowed responses after the target appears at the location of a neutral picture stimulus. Additionally, there were 10 trials with a wholly neutral picture pair, 10 trials with a wholly positive picture pair, and 10 trials with a wholly negative picture pair. In these trials, the dot appeared at each location with an equal frequency. These trials were implemented to assess the effects of the valence of the stimulus material. In all trials, the picture stimuli were randomly selected out of the three respective pools of neutral, positive, and negative picture.

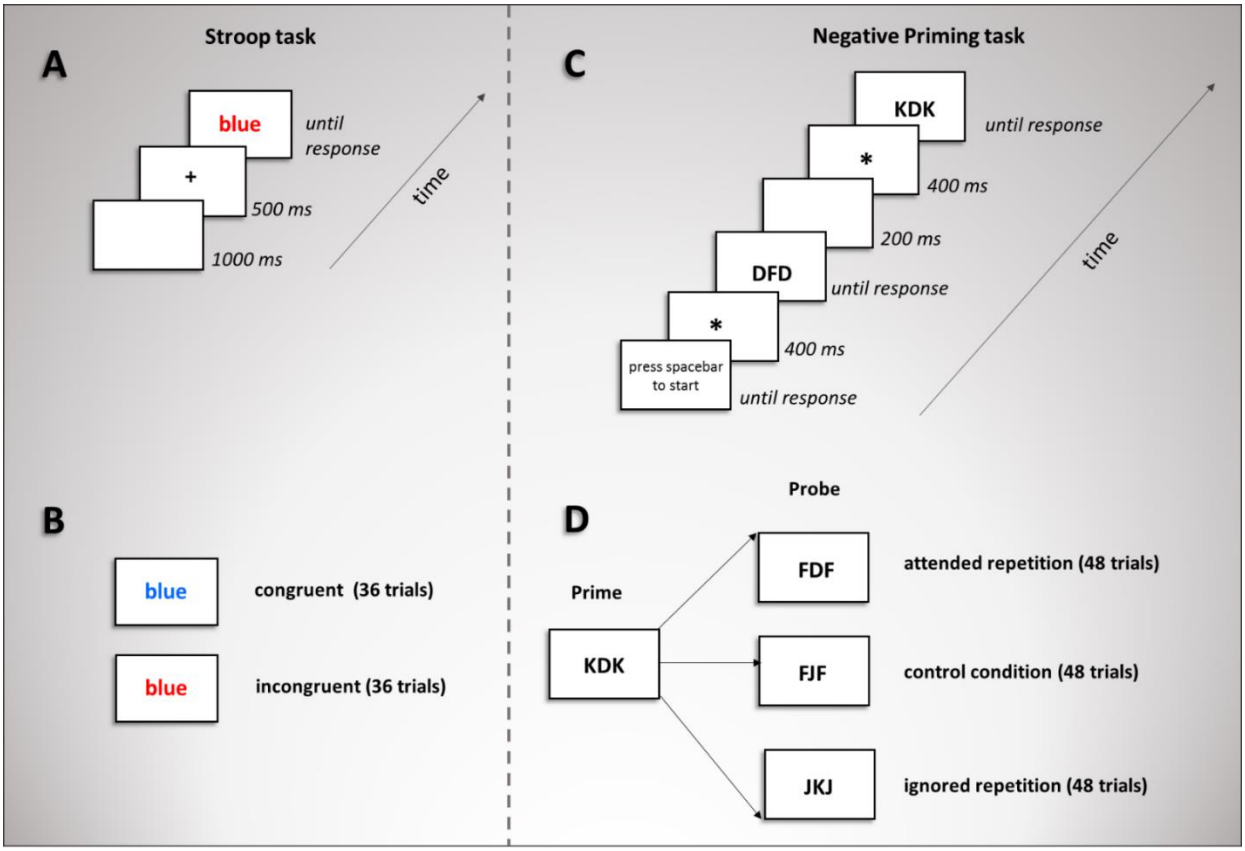


Figure 5.2.: Sequence of events (A) and conditions (B) in the Stroop task; sequence of events (C) and conditions (D) in the Negative priming task. The stimuli are not drawn to scale.

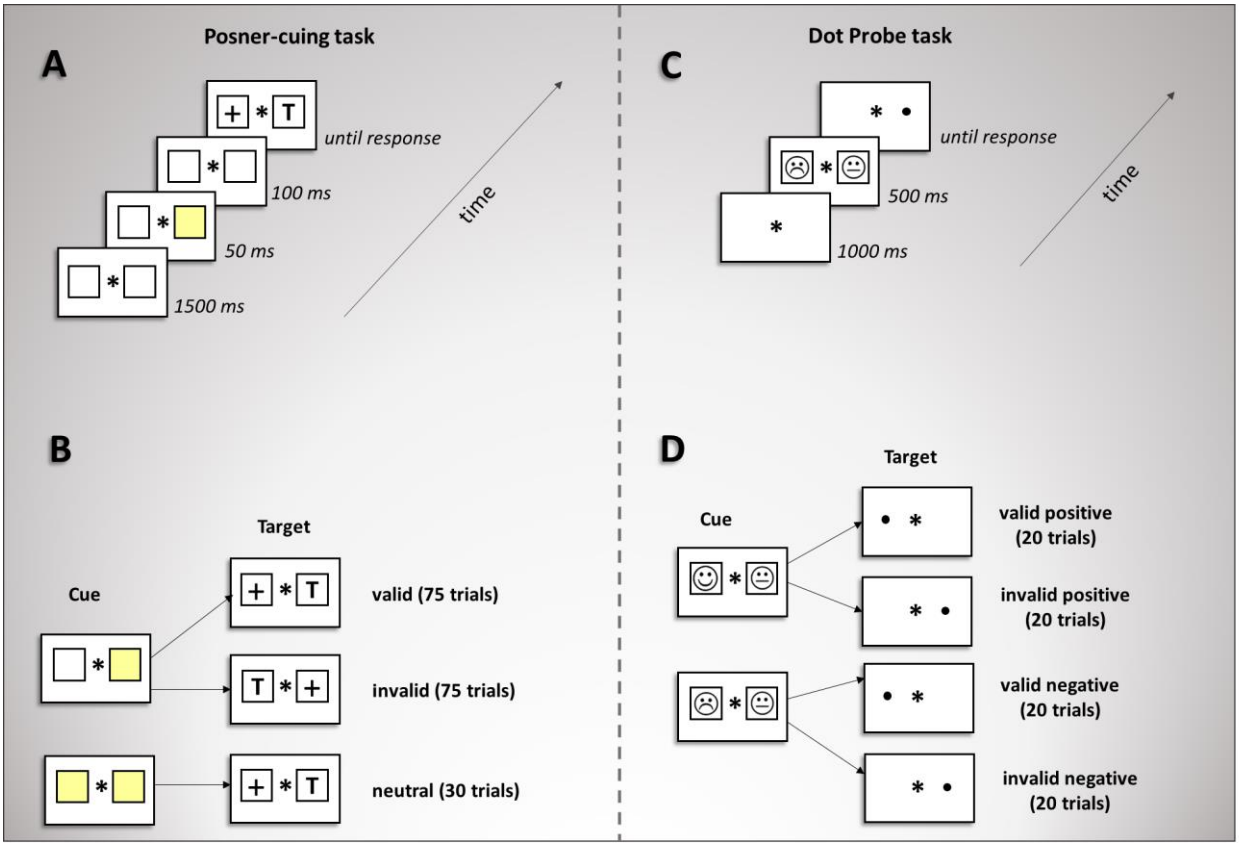


Figure 5.3.: Sequence of events (A) and conditions (B) in the Posner-Cuing task; sequence of events (C) and conditions (D) in the Dot Probe task. The stimuli are not drawn to scale. For the Posner-Cuing task, the yellow squares indicate the bright "light flash" cue (a salient change in luminance). The catch trials of the Posner-Cuing task are not depicted in the figure. For the Dot Probe task, the smiley graphics are visualizations of the cues but to not resemble the actual photographs that were used. Note that in each of the depicted conditions, target and cue appeared equally often on either side. The valence-consistent Dot Probe trials are not depicted in the figure (picture pairs that were either positive-positive, negative-negative, neutral-neutral)

5.5. Control measures

All participants were screened for ADHD. Their general attentional capacity was also measured. These control analyses were relevant to examine to which degree the results of this study were attributable to the general level of performance or to a common attentional disorder. An adult self-report ADHD checklist was administered to screen for any ADHD symptomatology in the participants (see appendix A1). The ADHD-SR checklist (Rösler et al., 2004) consists of 22 items on a four-point Likert-scale and corresponds to the diagnostic criteria of the DSM-IV and ICD-10. The coefficient of retest-reliability is $r \geq .70$ and thus surpasses the threshold for diagnostic measures (Stieglitz, 2000). A positive answer value on an item (score > 0) is registered as one point. The cut-off value for the

diagnosis of ADHD is defined at 10 points by Rösler and colleagues (2004). Of these, six points need to be derived from the items 1-9, three points from the items 10-14 and at least one point from the items 15-18. The revised D2 Test (Brickenkamp et al., 2010) is a cancellation test normed for ages 9 – 60 years. It provides a highly reliable measure of the general concentration and attention capacities; the coefficients of Cronbach's alpha and the retest reliability surpass $r \geq .85$. The test consists of a single paper sheet with 14 rows of letters (*d's* and *p's*) which each are surrounded with up to four short dashes**.** Each row consists of 57 items, resulting in 798 total items. It was the participants' task to cross out any d's with two dashes (targets) while d's with more or less dashes as well as p's (distractors) were to be ignored. The participants had a time limit of 20 s per row. They were encouraged to be as fast and precise as possible. The two main measures derived from the D2 Test are the concentration performance and the error score. The concentration performance is a standardized measure that reflects the absolute number of detected targets minus the number of omission and commission errors, thus reflecting both speed and accuracy. The error score is a standardized measure that indicates the percentage of processed items that were processed incorrectly (either due to omission or commission errors).

5.6. Genetic analyses

DNA was obtained from buccal cells followed by a mouthwash with Listerine (Qiagen Gentra Puregene Buccal Cell Kit, Hilden, Germany). All genotyping data were called by two independent individuals.

The COMT Val158Met polymorphism. A polymerase chain reaction (PCR) was performed in 50 µl reactions with a total DNA concentration of 100 ng, 2 mM $MgCl_2$, 10 pmol of each primer, 0.2 mM dNTPs, 5 % DMSO and 1.25 U Hot Star *Taq* Polymerase (Qiagen). The following primers were used in the PCR: *COMT* forward (5'- ACTGTGGCTACTCAGCTGTG -3') and *COMT* reverse (5'- CCTTTTTCCAGGTCTGACAA -3'). Thermal cycling was carried out using the following conditions: initial step 5 min at 95 °C followed by 40 cycles at 94 °C for 30 s, 55.1 °C for 30, 72 °C for 30 s and a final extension phase at 72 °C for 7 min. PCR products were separated on a 2 % agarose gel and visualized with ethidium bromide under UV light. Restriction analysis of the *COMT* Val158Met polymorphisms was performed with *Hsp92II* (Promega) and resulted in the three genotypes Val/Val, Val/Met and Met/Met. *COMT* Val158 Met primer sequences and fragment lengths were previously described (Tenhunen et al., 1994).

The DAT1 polymorphism. A PCR was performed in 50 µl reactions with a total DNA concentration of 100 ng, 1.5 mM MgCl2, 10 pmol of each primer, 0.2 mM dNTPs, and 1.25 U Hot Star *Taq* Polymerase (Qiagen). The following primers were used in the PCR: *DAT1* forward (5'- AGCTCAGGCTACTGCCACTC -3') and *DAT1* reverse (5'- AAAAAGCCATTCGCAAACAT - 3'). Thermal cycling was carried out using the following conditions: initial step 5 min at 95 °C followed by 40 cycles at 94 °C for 30 s, 53.6 °C for 45 s, 72 °C for 30 s and a final extension phase at 72 °C for 7 min. PCR products were separated on a 2 % agarose gel and visualized with ethidium bromide under UV light. The *DAT1* PCR products resulted in fragments of 640 bp (11-repeat), 600 bp (10-repeat), 560 bp (9-repeat), 520 bp (8-repeat). *DAT1* primer sequences and fragment length were previously described (Vandenbergh et al., 1992).

The CHRNA4 rs1044396 polymorphism. A PCR was performed in 50 µl reactions with a total DNA concentration of 100 ng, 1.5 mM $MgCl_2$, 10 pmol of each primer, 0.2 mM d'NTPs, and 1.25 U Hot Star *Taq* Polymerase (Qiagen). The following primers were used in the PCR: *CHRNA4* forward (5'–TCTCGCAACACCCACTC–3') and *CHRNA4* reverse (5'–GTCTGTGTCTTCGGCCTTCA–3'). Thermal cycling was carried out using the following conditions: initial step 5 min at 94 °C followed by 40 cycles at 94 °C for 30 s, 65.3 °C for 30 s, 72 °C for 30 s and a final extension phase at 72 °C for 7 min. PCR products were separated on a 2 % agarose gel and visualized with ethidium bromide under UV light. Restriction analysis of the *CHRNA4* rs1044396 polymorphism was performed with *HhaI* (Promega). Digest products were separated on a 2.5 % agarose gel and visualized with ethidium bromide under UV light. The T allele resulted in bands of 152 and 138 bps, while the C allele resulted in bands of 152, 105 and 33 bps. Accordingly, the restriction analysis resulted in the three genotypes T/T, C/T and C/C.

The CHRNA5 rs3841324 polymorphism. A PCR was performed in 50 µl reactions with a total DNA concentration of 100 ng, 1.0 mM $MgCl_2$, 10 pmol of each primer, 0.2 mM dNTPs, 5 % DMSO and 1.25 U Hot Star *Taq* Polymerase (Qiagen). The following primers were used in the PCR: rs3841324 forward (5'- GCTAGGAGCAGACAGGGTTG-3') and rs3841324 reverse (5'- GAGACAAAACGAGGGCAGAC -3'). Thermal cycling was carried out using the following conditions: initial step 5 min at 94 °C followed by 40 cycles at 94 °C for 30 s, 61°C 30 s, 94 °C for 30 s, and a final extension phase at 72 °C for 7 min. PCR products were separated on a 2 % agarose gel and visualized with ethidium bromide under UV light. The analysis of the *CHRNA5* rs3841324 polymorphism resulted in the three genotypes L/L, L/S and S/S.

The CHRNA5 rs16969968 polymorphism. A PCR was performed in 50 µl reactions with a total DNA concentration of 100 ng, 1.5 mM $MgCl_2$, 10 pmol of each primer, 0.2 mM dNTPs, and 1.25 U Hot Star *Taq* Polymerase (Qiagen). The following primers were used in the PCR: rs16969968 forward (5'- CCA AAC TGC TTT GCA TGA GA- 3') and rs16969968 reverse (5'- CCC ATT TTG CAG GTG CTT TA -3').

Thermal cycling was carried out using the following conditions: initial step 5 min at 94 °C followed by 40 cycles at 94 °C for 30 s, 58.1 °C for 30 s, 94 °C for 30 s, and a final extension phase at 72 °C for 7 min. PCR products were separated on a 1.5 % agarose gel and visualized with ethidium bromide under UV light. Restriction analyses of the *CHRNA5* rs16969968 products with T*aqI* resulted in fragments of 109 bp and 249 bp for the G allele while the A allele was not digested into different fragments.

Chapter 6

The Analyses of Attention Effects

This chapter is focused on the main hypothesis of this dissertation. It was hypothesized that DA and ACh modulate selective attention differently – specifically, that the selected dopaminergic polymorphisms influence the performance in tasks of selective top-down attention, while the selected cholinergic polymorphisms influence the performance in tasks of selective bottom-up attention (but not vice versa). First, the results of the genotyping analyses will be reported (subchapter 6.1). The analyses of the behavioral tasks will be reported subsequently (subchapter 6.2.). Afterwards, the effects of the dopaminergic and cholinergic polymorphisms on selective attention will be tested (subchapter 6.3. and 6.4, respectively). Control analyses in regard to the general attentional capacity and potential attentional deficits will be reported last (subchapter 6.5).

6.1. Genotyping

The genotype frequencies of the dopaminergic polymorphisms are depicted in table 6.1 and 6.2. The genotype frequencies of *COMT* Val158Met polymorphism did not deviate from the Hardy–Weinberg equilibrium (χ^2 = 0.93, df = 1, p > 0.05), and there were no differences in the genotype distributions between male and female participants (χ^2 = 1.80, df = 2, p > 0.05). The genotype frequencies of *DAT1* did not deviate from the Hardy–Weinberg equilibrium either (χ^2 = 0.52, df = 1, p > 0.05). There were also no differences in the genotype distributions between both groups of gender (χ^2 = 2.68 , df = 2, p > 0.05). Less common *DAT1* genotypes were detected in a minority of participants (n = 3). These were not further analyzed due to the statistical limitations. The *COMT* Val158Met polymorphism was not correlated with the *DAT1* polymorphism (r = .034, p = .654), indicating that their alleles did not co-occur above chance levels.

Table 6.1. Genotype frequencies of the *COMT* Val158Met polymorphism (in *n*)

Val/Val	Val/Met	Met/Met
46	83	50

Table 6.2. Genotype frequencies of the *DAT1* polymorphism (in *n*)

9/9	9/10	10/10
10	71	94

The genotype frequencies of the selected cholinergic polymorphisms are depicted in table 6.3 – 6.5. The genotype frequencies of the *CHRNA4 rs1044396* polymorphism did not deviate from the Hardy–Weinberg equilibrium (χ^2 = 2.96, df = 1, p > .05). However, differences in the genotype distributions between male and female participants were noted (χ^2 = 7.01, df = 2, p = .03). As a result,

gender was included as a covariant in the main analyses. The genotype frequencies of *CHRNA5* rs3841324 did not deviate from the Hardy–Weinberg equilibrium (χ^2 = 0.19, df = 1, p > 0.05). There were no differences in the genotype distributions between both gender groups (χ^2 = 0.65, df = 2, p > 0.05). The genotype frequencies of the *CHRNA5* rs16969968 polymorphism did not deviate from the Hardy–Weinberg equilibrium either (χ^2 = 1.67, df = 1, p > 0.05). There were also no differences in the genotype distributions between both groups of gender (χ^2 = 0.35, df = 2, p > 0.05).

Table 6.3. Genotype frequencies of the *CHRNA4 rs1044396* polymorphism (in *n*)

T/T	T/C	C/C
58	66	33

Table 6.4. Genotype frequencies of the *CHRNA5* rs3841324 polymorphism (in *n*)

L/L	L/S	S/S
65	84	31

Table 6.5. Genotype frequencies of the *CHRNA5* rs16969968 polymorphism (in *n*)

G/G	G/A	A/A
80	74	26

The *CHRNA4 rs1044396* polymorphism was neither correlated with the *CHRNA5* rs3841324 polymorphism (r = -.040, p = .623) nor with the *CHRNA5* rs16969968 polymorphism (r = -.068, p = .398). On the other hand, the *CHRNA5* rs3841324 polymorphism and the *CHRNA5* rs16969968 polymorphism were correlated (r = -.582, p < .001). A high linkage disequilibrium between both polymorphisms was observed in this study (r^2 = .34; D' = 0.97), indicating an above chance level co-occurrence of their alleles. The diplotype frequencies of these polymorphisms are depicted in table 6.6.

Table 6.6. Diplotype frequencies of the *CHRNA5* rs3841324 polymorphism and *CHRNA5* rs16969968 polymorphism

		CHRNA5 rs3841324		
		L/L	L/S	S/S
	G/G	14	35	31
CHRNA5	G/A	26	48	0
rs16969968	A/A	25	1	0

6.2. Task analyses

Stroop task. For the RT analyses, only trials with correct responses were considered (the error rate in the congruent condition was 1.18 %, while the error rate in the incongruent condition was 2.92 %). Responses were considered valid if they were neither too fast nor above a nonparametric outlier criterion of the RT distribution of the whole sample (Tukey, 1977). RTs that were more than 1.5 interquartile ranges above the third quartile of the RT distribution of the whole sample in each grouping condition were excluded from the analysis, as were RTs that were shorter than 150 ms. Due to these constraints, 2.05 % of all trials were discarded. Correct RTs were submitted into a repeated-measures analysis of variance (ANOVA) with congruency (congruent vs. incongruent) as factor. Here – as in all subsequent repeated-measures ANOVAs – Pillai's trace was used as a criterion. The effect of the congruency was significant [F (1, 180) = 36.78, p < .001, η_p^2 =.170]. Reactions were faster when stimuli were free of conflict, i.e. when the word color equaled the word content. On average, the participants displayed a response speed of M = 412 ms (SD = 74 ms) in the congruent condition and M = 430 ms (SD = 96 ms) in the incongruent condition (see figure 6.1); accordingly, the Stroop effect was M = 18 ms (SD = 40 ms). The error rates were also analyzed. They were submitted into a repeated-measures ANOVA with congruency (congruent vs. incongruent) as factor. There was a significant effect of the congruency [F (1, 180) = 19.83, p < .001, η_p^2 =.099]. On average, the participants made M = 0.71 (SD = 1.2) errors in the congruent condition and M = 1.23 (SD = 1.4) errors in the incongruent condition. Overall, the reactions were both faster and less error-prone in the congruent condition compared to the incongruent condition.

Negative priming. For the RT analyses, only trials with correct responses were considered (the error rate in the attended repetition condition was 0.35 %, the error rate in the control condition was

1.08 %, and the error rate in the ignored repetition condition was 1.23 %). Responses were considered valid if they were neither too fast nor above a nonparametric outlier criterion of the RT distribution of the whole sample (Tukey, 1977). RTs that were more than 1.5 interquartile ranges above the third quartile of the RT distribution of the whole sample in each grouping condition were excluded from the analysis, as were RTs that were shorter than 150 ms. Due to these constraints, 0.89 % of all trials were discarded. Correct RTs were submitted into a repeated-measures ANOVA with condition (attended repetition vs. control condition vs. ignored repetition) as factor. The effect of the condition was significant [F (2, 180) = 411.65, $p < .001$, $\eta_p^2 = .82$]. The participants displayed a response speed of *M* = 559 ms (*SD* = 70) in the attended repetition, *M* = 727 ms (*SD* = 119) in the control condition, and *M* = 732 ms (*SD* = 114) in the ignored repetition (see figure 6.1). Accordingly, the PP effect was *M* = 168 ms (*SD* = 85 ms), while the NP effect was *M* = -5 ms (*SD* = 36 ms). Of the *n* = 182 participants, *n* = 103 participants displayed a NP effect, while *n* = 79 participants did not. It was assessed whether the ability to touch-type influenced the performance in the Negative priming task. Correct RTs were submitted into a repeated-measures ANOVA with condition (attended repetition vs. control condition vs. ignored repetition) as factor and touch-typing as a covariant. There was a significant interaction between the task condition and the ability to touch-type [F (2, 175) = 8.81, $p < .001$, $\eta_p^2 = .09$][27]. Next, the error rates were also submitted into a repeated-measures ANOVA with condition (attended repetition vs. control condition vs. ignored repetition) as factor. Again, the effect of the condition was significant [F (2, 180) = 109.0, $p < .001$, $\eta_p^2 = .55$]. The participants made *M* = 0.56 (*SD* = 0.9) errors in the attended repetition, *M* = 2.53 (*SD* = 2.3) errors in the control condition, and *M* = 2.77 (*SD* = 2.3) errors in the ignored repetition. Overall, the participants were fastest and made the least errors in the attended repetition,

[27] All in all, *n* = 46 participants indicated that they were able to touch-type, while *n* = 132 indicated that they were not able to do so. The participants who were able to touch-type (in the following referred to as touch-typers) were faster in the Negative priming task. Across all conditions, they displayed an average response speed of *M* = 614 ms (*SD* = 77 ms); in contrast, the other participants displayed an average response speed of *M* = 694 ms (*SD* = 92 ms). The occurrence of the NP effect was correlated with the ability to touch-type ($r = .205$, $p = .006$). While 74 % of the touch-typers displayed the NP effect, only 51 % of the participants who were unable to touch-type displayed the NP effect. Correct RTs were submitted into a repeated-measures ANOVA with the condition (control condition vs. ignored repetition) as factor. The NP effect was not present if the touch-typers were fully excluded from the analysis of the sample [F (1, 131) = < 1, $p = .735$, $\eta_p^2 = .001$]. In contrast, the NP effect was significant at the two-tailed level if *only* the subset of touch-typing participants was considered [F (1, 45) = 10.54, $p = .002$, $\eta_p^2 = .19$]. While the touch-typers displayed an average NP effect of *M* = -14 ms (*SD* = 30 ms), the participants who were unable to touch-type displayed an average NP effect of *M* = - 1 ms (*SD* = 37 ms). Next, the correct RTs were submitted into a repeated-measures ANOVA with the condition (attended repetition vs. control condition) as factor. The PP effect was still present if the touch-typing participants were excluded from the analysis [F (1, 131) = 603.6, $p < .001$ $\eta_p^2 = .822$]. If only the touch-typing participants were analyzed, the PP effect was also observed [F (1, 45) = 125.5, $p < .001$ $\eta_p^2 = .736$]. While the touch-typers displayed an average PP effect of *M* = 126 ms (*SD* = 76 ms), the participants who were unable to touch-type displayed an average PP effect of *M* = 184 ms (*SD* = 84 ms). Overall, the touch-typers displayed smaller PP effects and larger NP effects than those participants who were unable to touch-type. The latter group seemed to benefit to a larger extent from the attended repetition, where it was not necessary to change the response key. These findings will be discussed in more detail in chapter 10. Control analyses for the Negative priming task will be reported in chapter 6.5.

while they were slowest and made the most errors in the ignored repetition. A large percentage of participants (43.4 %) did not display a NP effect at all.

Posner-Cuing task. For the RT analyses, only trials with correct reactions were considered (the error rate in the valid condition was 1.95 %, the error rate in the neutral condition was 1.50 % and the error rate in the invalid condition was 1.83 %). RTs that were more than 1.5 interquartile ranges above the third quartile of the RT distribution of the whole sample in each grouping condition (Tukey, 1977) were excluded from the analysis, as were RTs that were shorter than 150 ms. Due to these constraints, altogether 1.86 % of all trials were discarded. Correct RTs were submitted into a repeated-measures ANOVA with the cue validity as factor (valid vs. neutral vs. invalid). This effect was significant [F (2, 179) = 20.55, $p < .001$, $\eta_p^2 = .187$]. The participants displayed a response speed of M = 385 ms (SD = 46 ms) in the valid condition, M = 387 ms (SD = 48 ms) in the neutral condition, and M = 390 ms (SD = 46 ms) in the invalid condition (see figure 6.2). As expected, the participants were fastest in the valid condition and slowest in the invalid condition. Accordingly, the Posner effect was M = 5 ms (SD = 10 ms). The error rates were not analyzed, since the Posner-Cuing task was a detection task that required only one single response-key; as a result the overall sum of incorrect responses across all participants was only n = 4 (an incorrect response would have been hitting a wrong key on the keyboard).

Dot Probe task. For the RT analyses, only trials with correct reactions were considered (the error rate in the negative-valid condition was 1.68 %, the error rate in the negative-invalid condition was 0.99 %, the error rate in the positive-valid condition was 0.99 %, and the error rate in the positive-invalid condition was 0.77 %). RTs that were more than 1.5 interquartile ranges above the third quartile of the RT distribution of the whole sample in each grouping condition (Tukey, 1977) were excluded from the analysis, as were RTs that were shorter than 150 ms. Due to these constraints, altogether 1.11 % of all trials were discarded. Correct RTs were submitted into a repeated-measures ANOVA with condition (negative-valid vs. negative-invalid vs. positive-valid vs. positive-invalid) as factor. The effect of the condition was significant [F (3, 179) = 16.18, $p < .001$, $\eta_p^2 = .213$]. The participants displayed a response speed of M = 416 ms (SD = 64 ms) in the negative-valid condition, M = 411 ms (SD = 67 ms) in the negative-invalid condition, M = 411 ms (SD = 64 ms) in the positive-valid condition, and M = 403 ms (SD = 59 ms) in the positive-invalid condition (see figure 6.2). The error rates were also analyzed and submitted into a repeated-measures ANOVA with condition (negative-valid vs. negative-invalid vs. positive-valid vs. positive-invalid) as factor. Again, the effect of the condition was significant [F (3, 179) = 3.45, $p = .018$, $\eta_p^2 = .055$]. The participants made M = 0.19 (SD = 0.4) errors in the negative-valid condition, M = 0.16 (SD = 0.4) errors in the negative-invalid condition, M = 0.18 (SD = 0.5) errors in the positive-valid condition, and M = 0.10 (SD = 0.4) errors in the positive-invalid condition. In sum, the participants were faster and made less errors in the invalid conditions compared to the respective valid

ones. As the typical Dot Probe effect is reflected in the faster, less error-prone responses to targets that replace emotional stimuli (i.e., the faster and less-error prone responses in the *valid* conditions), this means that the Dot Probe effect was neither apparent in the RTs nor in the error rates. In fact, the observed pattern was the opposite of the one that had been expected. The lack of the Dot Probe effect may have been due to specific task or sample characteristics; originally developed to study anxiety disorders, the largest Dot Probe effects have been reported for samples of participants with types of anxiety disorders or phobias (Wenzel, 2012). The presence of psychiatric disorders was not assessed in this study, however. The lack of a clear Dot Probe effect rendered all further analyses of the task moot. For this reason, the Dot Probe task was not further studied in relation to the dopaminergic and cholinergic polymorphisms.

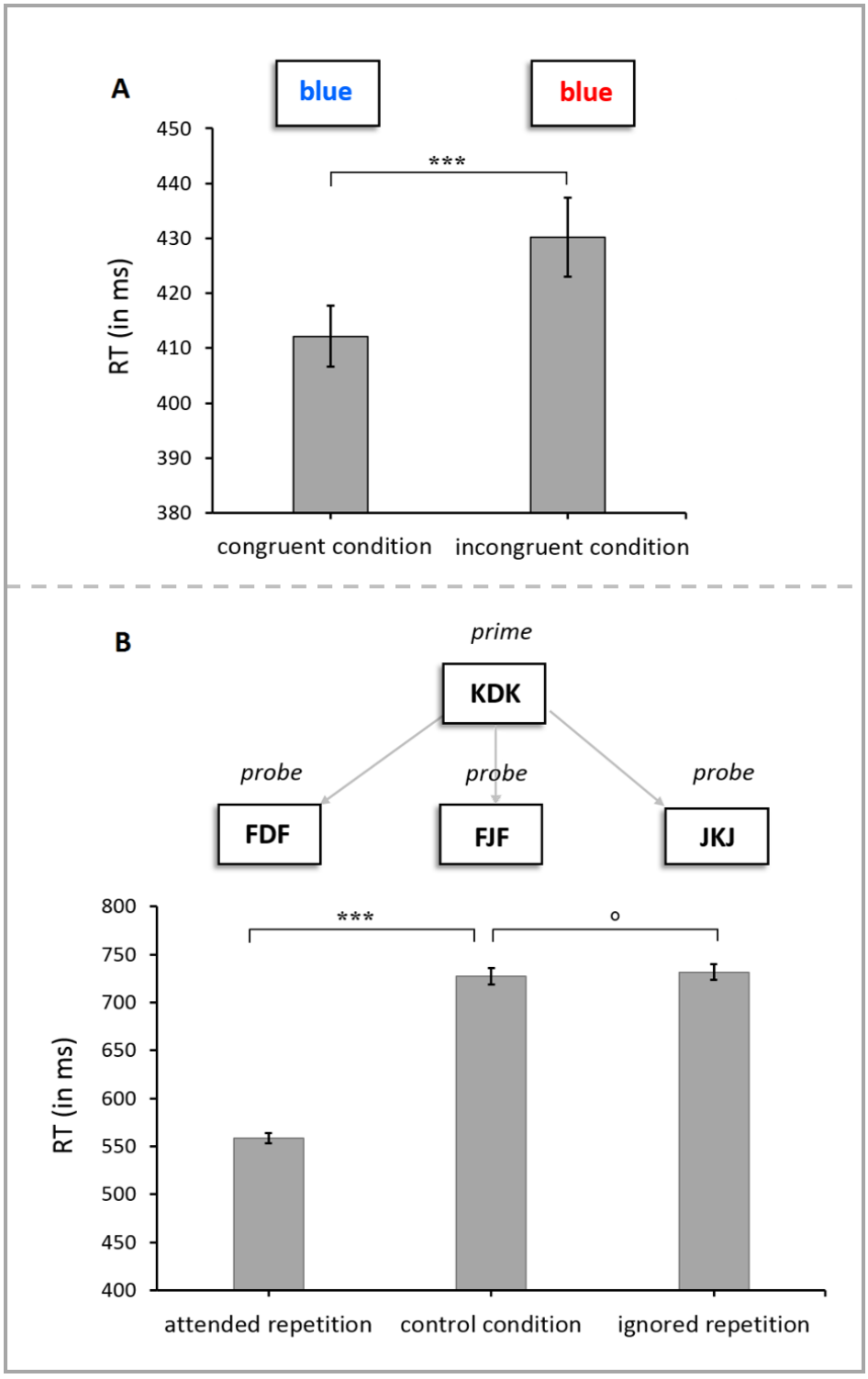


Figure 6.1.: RTs as a function of the experimental condition in the Stroop task (A) and the Negative priming task (B). The stimuli are not drawn to scale. The depicted trials are examples. In the Stroop task, the participants were faster in the congruent condition than in the incongruent condition. The Stroop effect is reflected in the difference between the congruent and incongruent condition (*M* = 18 ms, *SD* = 40 ms). In the Negative priming task, the participants were fastest in the attended repetition and slowest in the ignored repetition. While the PP effect was on average *M* = 168 ms (*SD* = 85 ms), the NP effect was *M* = - 5 ms (*SD* = 36 ms) and significant only at the one-tailed level. Error bars depict the standard error of the mean. *Notes:* ° $p < 0.10$, *** $p < 0.001$.

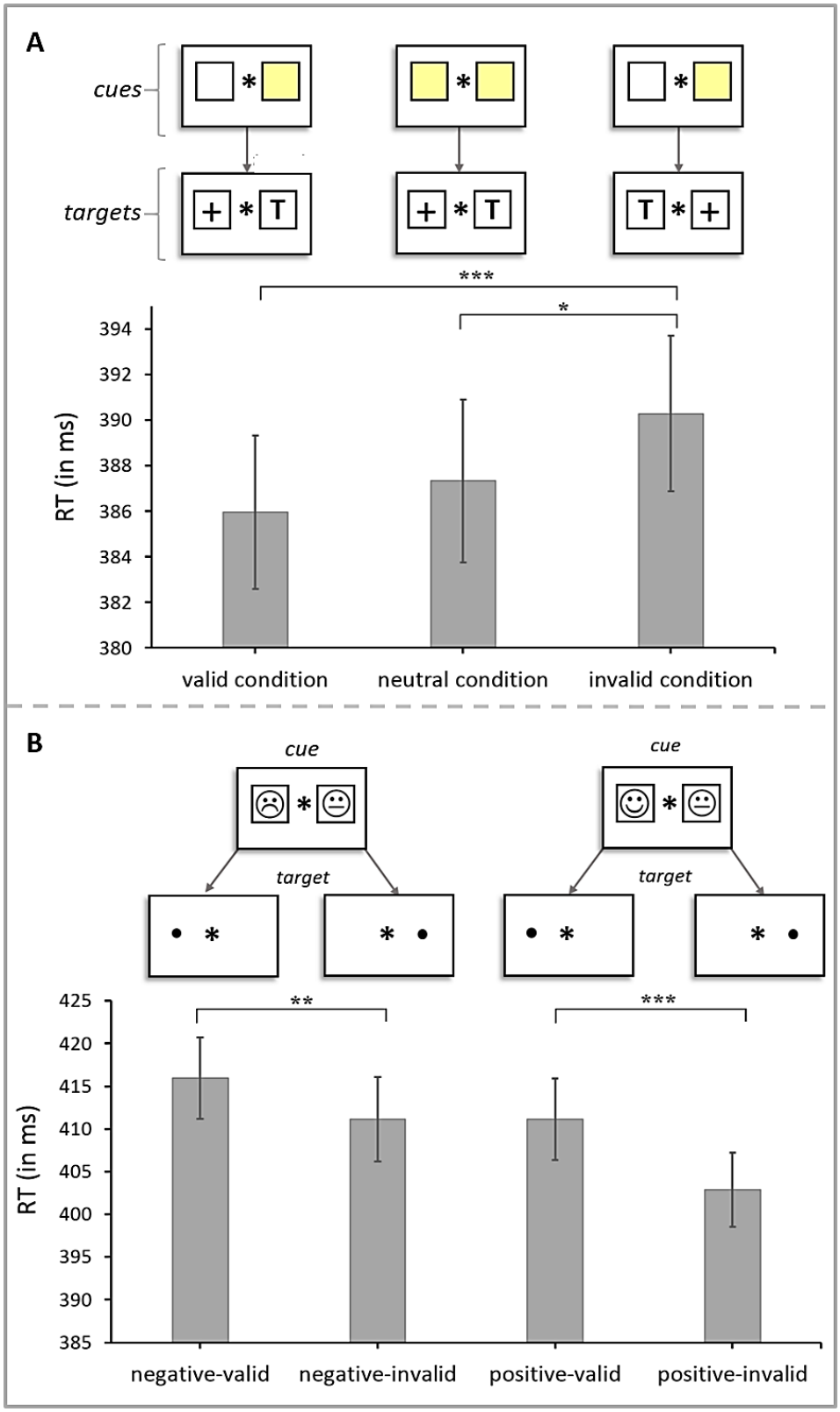


Figure 6.2.: RTs as a function of the experimental condition in the Posner-Cuing task (A) and the Dot Probe task (B). The stimuli are not drawn to scale. The depicted trials are examples. In the Posner-Cuing task, the participants were fastest in the valid condition and slowest in the invalid condition. The Posner effect is reflected in the difference between the valid and invalid

condition (*M* = 5 ms, *SD* = 85 ms). In the Dot Probe task, the participants were faster and made less errors in the invalid conditions compared to the respective valid ones. The typical Dot Probe effect (faster and less-error prone responses in the valid conditions) was not apparent in this study. As the obtained data was not easily interpretable, the task was excluded from all further analyses. Error bars depict the standard error of the mean. *Notes:* * p < 0.05, ** p < 0.01, *** p < 0.001.

6.3. Attentional analyses of the dopaminergic polymorphisms

The effect of the COMT Val158Met polymorphism on measures of selective attention. It was expected that carriers of the Met allele displayed a better performance in the tasks that tap selective top-down attention (the Stroop task and Negative priming task), but not in a task that taps into selective bottom-up attention (the Posner-Cuing task). The participants were grouped into Val allele homozygotes and carriers of the Met allele (Val/Val vs. Met+). Gender was considered as a factor in all analyses of this polymorphism. The gender frequencies of the *COMT* Val158Met polymorphism are depicted in table A2.

The Stroop-RTs were submitted into a 2 (congruency: congruent vs. incongruent) x 2 (*COMT* Val158Met: Val/Val vs. Met+) repeated-measures ANOVA with gender as a covariant. The interaction of condition and genotype was significant [F (1, 170) = 5.50, p = .020, η_p^2 = .031]. While both Val homozygotes and carriers of the Met allele were generally slower in the incongruent condition compared to the congruent condition, homozygotes for the Val allele were disproportionately slower in the incongruent condition (resulting in a Stroop effect of *M* = 22 ms for them and a Stroop effect of *M* = 13 ms for carriers of the Met allele). Thus, as hypothesized, carriers of the low-activity Met allele outperformed Val homozygotes in the Stroop task. Val homozygotes seemed to experience slightly larger difficulties in processing conflicting Stroop stimuli (see figure 6.3). Next, the Negative priming-RTs were submitted into a 3 (condition: attended repetition vs. control condition vs. ignored repetition) x 2 (*COMT* Val158Met: Val/Val vs. Met+) repeated-measures ANOVA with gender as a covariant. The interaction effect of condition and genotype was not significant [F (2, 170) < 1, p = .426, η_p^2 = .009]. Last, the RTs of the Posner-Cuing task were submitted into a 3 (condition: valid vs. neutral vs. invalid) x 2 (*COMT* Val158Met: Val/Val vs. Met+) repeated-measures ANOVA with gender as a covariant. The interaction effect of cue validity and genotype was not significant [F (2, 170) < 1, p = .982, η_p^2 = .009]. The power for detecting even a small effect (e.g., f = 0.10) was about 1-β = 0.80 given a sample size of n = 182 and an alpha-level of α = 0.05.

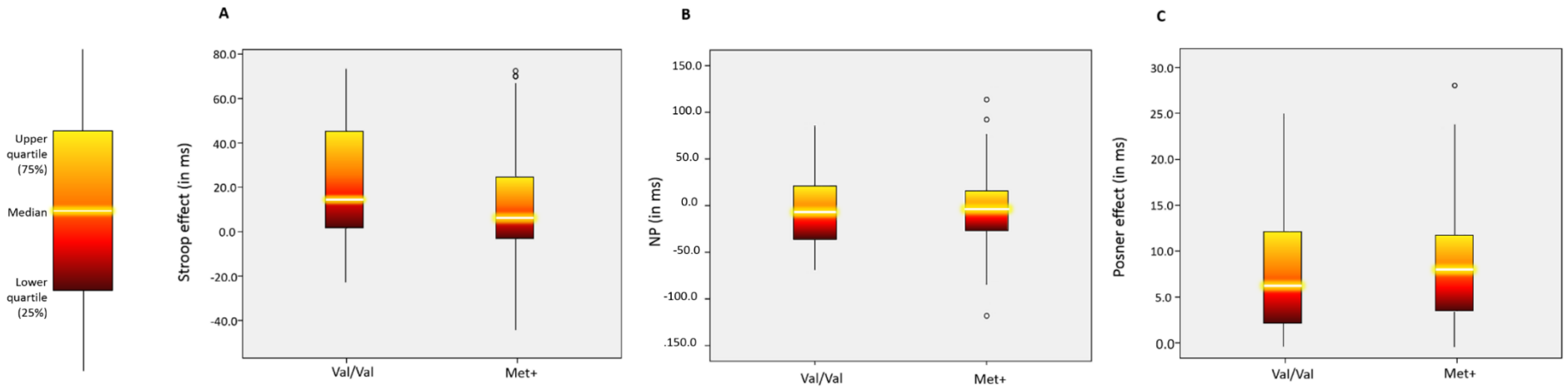


Figure 6.3.: Distribution of the Stroop effect RTs (A), NP effect RTs (B) and Posner effect RTs (C) as a function of the *COMT* Val158Met genotype. Carriers of the Met allele displayed a better performance in the Stroop task (by displaying significantly smaller Stroop effects). The performance in the Negative priming task and Posner-Cuing task was not significantly modulated by the *COMT* Val158Met genotype as a factor.

The effect of the DAT1 polymorphism on measures of selective attention. It was hypothesized that carriers of the 10-repeat allele of the *DAT1* polymorphism would display a better performance in the tasks that tap selective top-down attention (the Stroop task and Negative priming task), but not in the task that taps selective bottom-up attention (the Posner-Cuing task).

The Stroop-RTs were submitted into a 2 (congruency: congruent vs. incongruent) x 3 (*DAT1*: 9/9 vs. 9/10 vs. 10/10) repeated-measures ANOVA. The interaction effect of condition and genotype was significant [F (2, 171) = 9.31, p < .001, η_p^2 = .098]. As hypothesized, carriers of at least one 10-repeat allele exhibited a better performance in a task of top-down selection than participants homozygous for the 9-repeat allele. Both participants homozygous for the 9-repeat allele and 10-repeat allele carriers were slower in the incongruent condition than the congruent condition. 9-repeat homozygotes, however, were both generally slower in both conditions and displayed a larger Stroop effect than carriers of the 10-repeat allele. The resulting Stroop effects were M = 37 ms for homozygous carriers of the 9-repeat allele, M = 13 ms for carriers of the 9-repeat and 10-repeat allele and M = 14 ms for homozygous carriers of the 10-repeat allele. Homozygous 9-repeat allele carriers showed a significantly enlarged Stroop effect compared to the other genotype groups (Helmert contrast, p < .001) while the Stroop effects of heterozygous and homozygous 10-repeat allele carriers were not significantly different (Helmert contrast, p = .791). Next, the Negative priming-RTs were submitted into a 3 (condition: attended repetition vs. control condition vs. ignored repetition) x 3 (*DAT1*: 9/9 vs. 9/10 vs. 10/10) repeated-measures ANOVA. The interaction effect of condition and genotype was not significant [F (4, 344) < 1, p = .832, η_p^2 = .004]. Subsequently, the Cuing-RTs were submitted into a 3 (cue validity: valid vs. neutral vs. invalid) x 3 (*DAT1*: 9/9 vs. 9/10 vs. 10/10) repeated-measures ANOVA. The interaction effect of cue validity and genotype was not significant [F (4, 344) = 1.08, p = .344, η_p^2 = .012]. The distribution of the task effects in dependence on the *DAT1* polymorphism are depicted in figure 6.4. Again, the power for detecting even a small effect (e.g., f = 0.10) was about 1-β = 0.80 given a sample size of n = 182 and an alpha-level of α = 0.05.

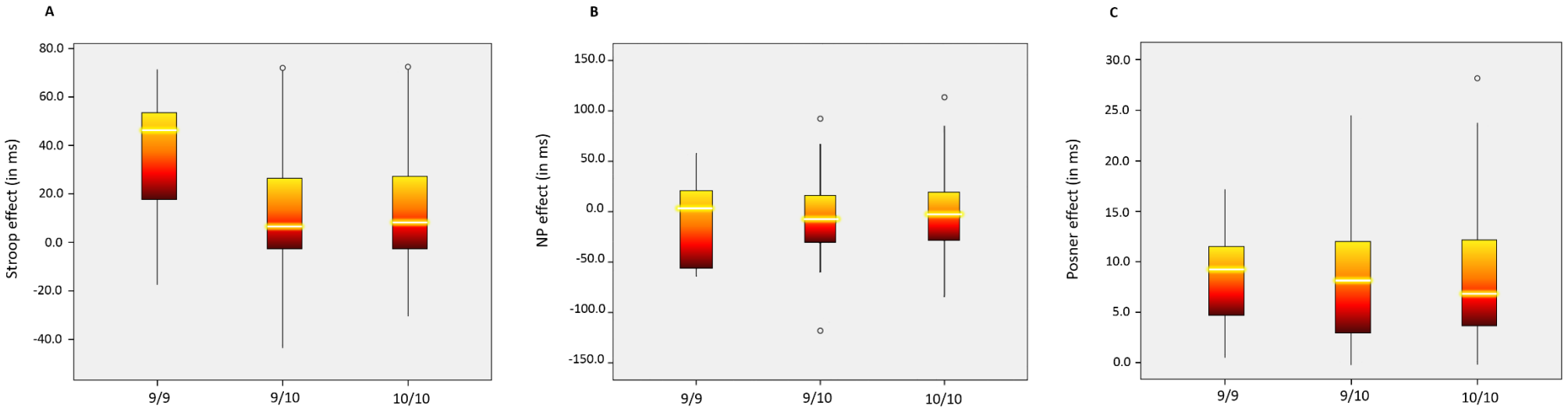


Figure 6.4.: Distribution of the Stroop effect RTs (A), NP effect RTs (B) and Posner effect RTs (C) as a function of the *DAT1* polymorphism. Carriers of the 10-repeat allele displayed a better performance in the Stroop task (by displaying on average significantly smaller Stroop effects). The performances in the Negative priming task and Posner-Cuing task were not significantly modulated by the *DAT1* polymorphism.

Summary. As hypothesized, carriers of the Met allele of the *COMT* Val158Met polymorphism and carriers of the 10-repeat allele of the *DAT1* polymorphism displayed a better performance in the Stroop task; the improved performance was reflected in smaller Stroop effects. Also as hypothesized, neither the *COMT* Val158Met nor the *DAT1* polymorphism was found to modulate the performance in the Posner-Cuing task. The findings in regard to the Negative priming task did not align with the previously drawn hypotheses, as neither the *COMT* Val158Met polymorphism nor the *DAT1* polymorphism was found to modulate the performance in this task.

6.4. Attentional analyses of the cholinergic polymorphisms

The effect of CHRNA4 rs1044396 polymorphism on measures of selective attention. It was hypothesized that the *CHRNA4* rs1044396 polymorphism would modulate the performance in a task that taps into selective bottom-up attention (the Posner-Cuing task) but not in tasks that tap into selective top-down attention (the Stroop task and Negative priming task). Specifically, it was hypothesized that carriers of the T allele would show a better performance in the Posner-Cuing task than carriers of the C allele.

The Stroop-RTs were submitted into a 2 (congruency: congruent vs. incongruent) x 3 (*CHRNA4*: T/T vs. C/T vs. C/C) repeated-measures ANOVA with gender as a covariant. The interaction between the condition and the *CHRNA4* genotype was not significant [F (2, 148) < 1, p = .805, η_p^2 = .001]. The Negative priming-RTs were submitted into a 3 (condition: attended repetition vs. control condition vs. ignored repetition) x 3 (*CHRNA4*: T/T vs. C/T vs. C/C) repeated-measures ANOVA with gender as a covariant. Again, the interaction effect between the condition and the *CHRNA4* genotype was not significant [F (4, 298) = 1.18, p = .316, η_p^2 = .016]. The Cuing-RTs were also submitted into a 3 (cue validity: valid vs. neutral vs. invalid) x 3 (*CHRNA4*: T/T vs. C/T vs. C/C) ANOVA with gender as a covariant. Yet again, the interaction effect of cue validity and genotype was not significant [F (4, 298) < 1, p = .842, η_p^2 = .005]. In sum, there were no indications that the *CHRNA4* rs1044396 polymorphism modulated the attentional performance in the Stroop task, Negative priming task, or Posner-Cuing task. The task effects as a function of the *CHRNA4* rs1044396 polymorphism are depicted in figure 6.5. The power for detecting a small effect (e.g., f = 0.10) was about 1-β = 0.70 given a sample size of n = 157 and an alpha-level of α = 0.05.

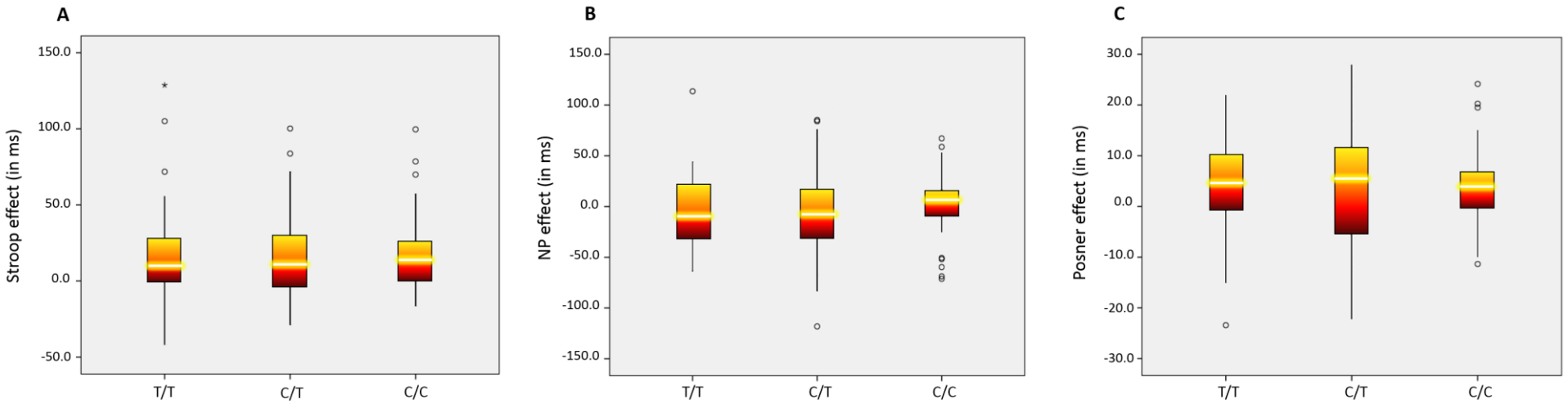


Figure 6.5.: Distribution of the Stroop effect RTs (A), NP effect RTs (B) and Posner effect RTs (C) as a function of the *CHRNA4* rs1044396 polymorphism. The performances in all three tasks were not significantly modulated by this polymorphism.

The effect of CHRNA5 rs3841324 polymorphism and CHRNA5 rs16969968 polymorphism on measures of selective attention. The selected *CHRNA5* polymorphisms were analyzed in an exploratory fashion; no prior assumptions about the individual effect of their alleles were drawn. Overall, it was expected that they would modulate the performance in a task that taps selective bottom-up attention (the Posner-Cuing task) but not in tasks that tap selective top-down attention (the Stroop task and Negative priming task). The rs3841324 polymorphism was analyzed on basis of comparisons between homozygous S allele carriers and L allele carriers (cp. chapter 2 and 4, as well as Wang et al., 2009). The rs16969968 polymorphism was analyzed on basis of comparisons between homozygous G allele carriers and A allele carriers (cp. chapter 2 and 4, as well as Bierut et al., 2008). First, possible main effects of the *CHRNA5* polymorphism were examined. Afterwards, the polymorphisms were analyzed with gender as a covariant. Due to the high linkage disequilibrium between the polymorphisms, their diplotypes were also analyzed.

Analyses without gender as a covariant. First, the *CHRNA5* rs3841324 and *CHRNA5* rs16969968 polymorphisms were analyzed as sole factors, without considering gender as a covariant. The Stroop-RTs were submitted into a 2 (condition: congruent vs. incongruent) x 2 (*CHRNA5* rs3841324: S/S vs. L+) repeated-measures ANOVA. The interaction between the condition and the *CHRNA5* rs3841324 genotype was not significant [F (1, 177) < 1, p = .986, η_p^2 < .001]. Next, the Negative priming-RTs were submitted into a 3 (condition: attended repetition vs. control condition vs. ignored repetition) x 2 (*CHRNA5* rs3841324: S/S vs. L+) repeated-measures ANOVA. Again, the interaction effect between the condition and the *CHRNA5* rs3841324 genotype was not significant [F (2, 177) < 1, p = .769, η_p^2 = .003]. The Cuing-RTs were also submitted into a 3 (condition: valid vs. neutral vs. invalid) x 2 (*CHRNA5* rs3841324: S/S vs. L+) repeated-measures ANOVA. The interaction effect of condition and genotype was not significant [F (2, 177) < 1, p = .572, η_p^2 = .006]. Next, the Stroop-RTs were submitted into a 2 (condition: congruent vs. incongruent) x 2 (*CHRNA5* rs16969968*:* G/G vs. A+) repeated-measures ANOVA. The interaction between the condition and the *CHRNA5* rs16969968 genotype was not significant [F (1, 177) < 1, p = .388, η_p^2 = .004]. The Negative priming-RTs were also submitted into a 3 (condition: attended repetition vs. control condition vs. ignored repetition) x 2 (*CHRNA5* rs16969968*:* G/G vs. A+) repeated-measures ANOVA. Again, the interaction effect between the condition and the *CHRNA5* rs16969968 genotype was not significant [F (2, 177) < 1, p = .949, η_p^2 = .001]. Last, the Cuing-RTs were submitted into a 3 (condition: valid vs. neutral vs. invalid) x 2 (*CHRNA5* rs16969968*:* G/G vs. A+) repeated-measures ANOVA. Yet again, the interaction effect of condition and genotype was not significant [F (2, 177) < 1, p = .406, η_p^2 = .010]. In sum, there were no indications that the *CHRNA5* rs3841324 polymorphism and the *CHRNA5* rs16969968 polymorphism modulated the performance in either of the three behavioral tasks. Given a sample size of n = 182 and an alpha-level of $\alpha = 0.05$, the power for detecting small effects (e.g., $f = 0.10$) was about $1-\beta = 0.80$.

Analyses with gender as a covariant. The gender frequencies of the selected *CHRNA5* polymorphisms are depicted in Table 6.7.

Table 6.7. Gender frequencies of the *CHRNA5* rs3841324 polymorphism and *CHRNA5* rs16969968 polymorphism

	CHRNA5 rs3841324			*CHRNA5* rs16969968		
	L/L	*L/S*	*S/S*	*G/G*	*G/A*	*A/A*
female	47	59	20	56	52	18
male	16	24	10	23	20	7

The Stroop-RTs were submitted into a 2 (condition: congruent vs. incongruent) x 2 (*CHRNA5* rs3841324: S/S vs. L+) repeated-measures ANOVA with gender as a covariant. The interaction between the condition, gender and the *CHRNA5* rs3841324 genotype was significant [F (1, 171) = 4.89, p = .028, η_p^2 = .028]. While male carriers of the S/S genotype displayed a Stroop effect of M = 2 ms (SD = 17 ms), male carriers of at least one L allele displayed a Stroop effect of M = 28 ms (SD = 65 ms). In contrast, female carriers of the S/S genotype displayed a Stroop effect of M = 27 ms (SD = 31 ms), while female carriers of at least one L allele displayed a Stroop effect of M = 14 ms (SD = 30 ms). The average Stroop effects per genotype group are depicted in figure 6.6**.** Next, the Negative priming-RTs were submitted into a 3 (condition: attended repetition vs. control condition vs. ignored repetition) x 2 (*CHRNA5* rs3841324: S/S vs. L+) repeated-measures ANOVA with gender as a covariant. The interaction effect between the condition, gender and the *CHRNA5* rs3841324 genotype was not significant [F (2, 171) < 1, p = .900, η_p^2 = .001]. The Cuing-RTs were also submitted into a 3 (condition: valid vs. neutral vs. invalid) x 2 (*CHRNA5* rs3841324: S/S vs. L+) repeated-measures ANOVA with gender as a covariant. The interaction effect of the condition, gender and genotype was not significant [F (2, 171) < 1, p = .698, η_p^2 = .004]. The Stroop-RTs were submitted into a 2 (condition: congruent vs. incongruent) x 2 (*CHRNA5* rs16969968*:* G/G vs. A+) repeated-measures ANOVA with gender as a covariant. The interaction between the condition, gender and the *CHRNA5* rs16969968 genotype was not significant, but showed a trends towards significance [F (1, 171) = 3.38, p = .068, η_p^2 = .019]. While male carriers of the G/G genotype displayed a Stroop effect of M = 16 ms (SD = 25 ms), male carriers of at least one A allele displayed a Stroop effect of M = 28 ms (SD = 78 ms). In contrast, female carriers of the G/G genotype

displayed a Stroop effect of *M* = 23 ms (*SD* = 33 ms), while female carriers of at least one A allele displayed a Stroop effect of *M* = 11 ms (*SD* = 27 ms). The average Stroop effects as a function of the genotype group are depicted in figure 6.6. Next, the Negative priming-RTs were submitted into a 3 (condition: attended repetition vs. control condition vs. ignored repetition) x 2 (*CHRNA5* rs16969968*:* G/G vs. A+) repeated-measures ANOVA with gender as a covariant. The interaction effect between the condition, gender and the *CHRNA5* rs16969968 genotype was not significant [F (2, 171) < 1, p = .737, η_p^2 = .004]. Subsequently, the Cuing-RTs were submitted into a 3 (condition: valid vs. neutral vs. invalid) x 2 (*CHRNA5* rs16969968*:* G/G vs. A+) repeated-measures ANOVA with gender as a covariant. The interaction effect of condition, gender and genotype was not significant [F (2, 171) < 1, p = .401, η_p^2 = .011].

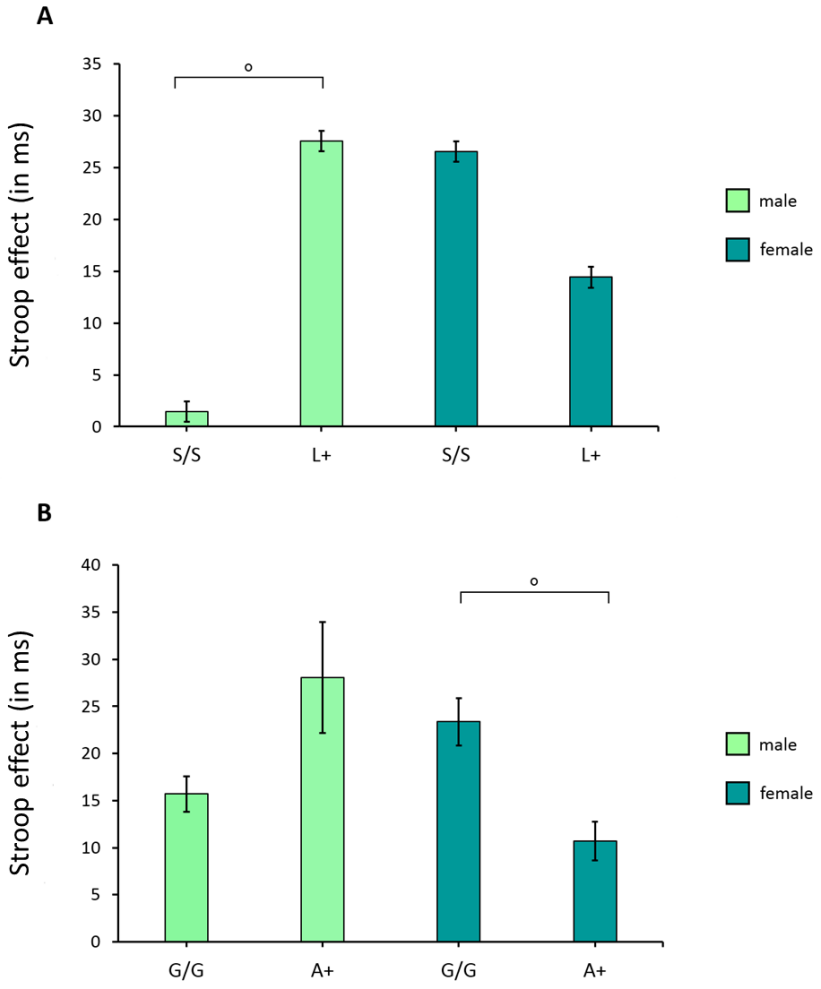


Figure 6.6.: Stroop effect RTs as a function of gender and of the *CHRNA5* rs3841324 polymorphism (A) and as a function of gender and the *CHRNA5* rs16969968 polymorphism (B). The effects of both polymorphisms were diametrically opposed in men and women. Male carriers of the S/S genotype of the *CHRNA5* rs3841324 polymorphism and of the G/G genotype of the *CHRNA5* rs16969968 polymorphism displayed smaller Stroop effects than their male counterparts, while the female carriers of the S/S genotype and G/G genotype displayed larger Stroop effects than their female counterparts. Error bars depict the standard error of the mean. *Notes:* ° p < 0.10.

Diplotype Analyses. As a last step, the diplotypes of the rs3841324 and rs16969968 polymorphisms were analyzed (for the diplotype frequencies, see table 6.6). The diplotypes were analyzed on basis of comparisons between carriers of the S/S genotype and G/G genotype (S/S_G/G+) and those who did not simultaneously carry *both* genotypes (S/S_G/G-). The Stroop-RTs were submitted into a 2 (condition: congruent vs. incongruent) x 2 (*CHRNA5*: S/S_G/G+ vs. S/S_G/G-) repeated-measures ANOVA with gender as a covariant. There was a significant interaction effect between the condition, gender, and the diplotype [F (1, 167) = 4.38, p = .029, η_p^2 = .028]. While male carriers of the S/S_G/G+ diplotype displayed a Stroop effect of M = 2 ms (SD = 17 ms), male carriers of other diplotypes displayed a Stroop effect of M = 28 ms (SD = 67 ms). In contrast, female carriers of the S/S_G/G+ diplotype displayed a Stroop effect of M = 27 ms (SD = 31 ms), while female carriers of other diplotypes displayed a Stroop effect of M = 15 ms (SD = 30 ms). Next, the Negative priming-RTs were submitted into a 3 (condition: attended repetition vs. control condition vs. ignored repetition) x 2 *CHRNA5*: S/S_G/G+ vs. S/S_G/G-) repeated-measures ANOVA with gender as a covariant. There was no significant interaction between the condition, gender, and the diplotype [F (2, 167) < 1, p = .908, η_p^2 = .001]. The Cuing-RTs were also submitted into a 3 (condition: valid vs. neutral vs. invalid) x 2 (*CHRNA5*: S/S_G/G+ vs. S/S_G/G-) repeated-measures ANOVA with gender as a covariant. Again, there was no significant interaction between the condition, gender, and the diplotype [F (2, 167) < 1, p = .697, η_p^2 = .004].

Summary. The *CHRNA4* rs1044396 polymorphism did not modulate the performance in the Stroop task, Negative priming task or Posner-Cuing task. This finding did not align with the previous hypothesis, since it had been proposed that this polymorphism would impact on the performance in the Posner-Cuing task (which taps selective bottom-up attention). The *CHRNA5* rs3841324 polymorphism and *CHRNA5* rs16969968 polymorphism did not modulate the performance in the Negative priming task or Posner-Cuing task, but exerted an influence on the performance in the Stroop task when gender was considered as a covariant. The effects of both polymorphisms were diametrically opposed in men and women. Male carriers of the S/S genotype of the *CHRNA5* rs3841324 polymorphism and of the G/G genotype of the *CHRNA5* rs16969968 polymorphism displayed smaller Stroop effects than their male counterparts, while the female carriers of the S/S genotype and G/G genotype displayed larger Stroop effects than their female counterparts. Again, these findings did not align with the previous hypotheses, since it had been proposed that the selected *CHRNA5* polymorphisms would impact on the performance in the Posner-Cuing task (which taps selective bottom-up attention), but not on the performance in tasks that primarily measure selective top-down attention.

6.5. Analyses of the control measures

As a last step, control analyses were conducted. It was assessed whether the presence of any ADHD symptomatology or the general attentional capacity influenced the obtained results.

ADHD-SR checklist. All of the participants (*n* = 182) filled out the ADHD screening questionnaire. The average score of all participants was *M* = 9.36 (*SD* = 3.68). The median was at *M* = 9 with a range of 0 to 18 points. The majority of participants (*n* = 166) was diagnosed negatively for ADHD, while *n* = 16 participants were tested positive for ADHD. The average score of the participants not diagnosed with ADHD tendencies was *M* = 8.79 (*SD* = 3.31). The average score of the participants diagnosed with ADHD tendencies was *M* = 15.31 (*SD* = 1.62). During the query segment of the study, only one participant stated that he/she had previously been diagnosed with ADHD. This participant was also positively diagnosed via the ADHD SR.

D2 test. All of the participants (*n* = 182) participated in the D2 test. Due to procedural errors, *n* = 4 tests could not be taken into consideration. Due to excessive error rates larger than 25%, a further *n* = 12 test results could not be taken into consideration. The average score of the concentration performance was *M* = 186.13 (*SD* = 38.08), the average error rate was 8.15 % (*SD* = 6.31).

Control and correlational analyses. The main tests of the dopaminergic and cholinergic polymorphisms were reanalyzed to assess the effect of the ADHD score and the D2 performance on the obtained results. To this end, the ADHD diagnosis and the D2 concentration performance measure were added as covariants to the analyses. It was tested whether the results remained unaltered – i.e. whether they remained significant or non-significant – if those measures were included. This was the case for all analyses (data not shown here as these were only peripheral analyses). In sum, neither the presence of ADHD symptomatology nor the general attentional capacity modulated the effects of the dopaminergic and cholinergic polymorphisms on measures of selective attention. The Negative priming task also underwent a specific reanalysis. Since the NP effect had not been displayed by about 43 % of the participants, the influence of the five polymorphisms on the performance in the Negative priming task was reanalyzed specifically for the subset of participants who displayed the NP effect. Several factors must have led to the low occurrence rate of the NP effect (cp. chapter 6.2). The reanalyses were undertaken to assess the influence of the polymorphisms on the performance in the Negative priming task in the absence of those noise-generating factors. However, there were also no significant interactions between the Negative priming task conditions and the genotypes in the subset of those participants who displayed the NP effect (data not shown here as these were only peripheral analyses). For the sake of completeness, the correlations between the behavioral, gene, and questionnaire data are reported (see table 6.8).

The Stroop effect negatively correlated with the concentration performance in the D2 test. This indicates that higher performance levels in the Stroop task are reflected in a measure of concentration and attentional capacities, with better performing participants being less affected by the congruency of Stroop stimuli and by the target/distractor distinction in the D2 test. RT costs in the Posner-Cuing Task were also negatively correlated with the concentration performance. This as well is an indicator that better performing participants were both less affected by the target/distractor distinction in the D2 test and by the validity of the Posner-Cuing stimuli. Participants with lower scores in concentration performance appeared to be more reliant on the prediction value of the Posner cue. If the cue was invalid and highlighted the wrong target location, they were slower to orient themselves to the correct target location. RT benefits in the Posner-Cuing task, on the other hand, were positively correlated with the error score in the D2 test. Participants who benefitted to a larger extent from cues highlighting the right target location were prone to make more mistakes in the D2 test. Again, this indicates an association between a larger dependence on the cue validity in the Posner-Cuing task and an inferior performance in the D2 test. In the same vein, the PP effect of the Negative priming task was correlated with the error score in the D2 test. None of the experimental measures were directly correlated to the ADHD diagnosis derived from the ADHD-SR scale. However, the error score in the D2 test was positively correlated with the ADHD diagnosis. Participants with a higher error percentage in the D2 test were more likely to have a positive ADHD diagnosis. In fact, participants not diagnosed with ADHD had an average D2 error score of 9.3 %, while participants diagnosed with ADHD had an average D2 error score of 13.9 %. This shows that the ADHD diagnosis was at least partly reflected in the behavioral performance of the participants. Considering the genes, the presence of a *COMT* 158Met allele (i.e. either a genotype of Val/Met or Met/Met) was negatively correlated with the Stroop effect in the Stroop task. Carriers of the Met allele thus exhibited a smaller Stroop effect, appearing to be less dependent on whether the stimulus had conflicting or non-conflicting properties. As for the *DAT1* polymorphism, the presence of the 10-repeat allele (i.e. either a genotype of 9/10-repeat or 10/10-repeat) was negatively correlated with the Stroop effect in the Stroop task. Like carriers of the *COMT* 158Met allele, carriers of the 10-repeat allele seemed to be less affected by the conflict level of the Stroop stimuli and displayed an overall better performance.

Table 6.8. Correlation matrix of the main measures used in this thesis

	ST	PP	NP	BEN	COST	ADHD	D2 CP	D2 E	COMT	DAT1	CHRNA4	5_384	5_169
ST	1.00												
PP	.13°	1.00											
NP	-.01	.37**	1.00										
BEN	.09	.26**	.09	1.00									
COST	.09	.30**	.09	.55**	1.00								
ADHD	.08	.03	.06	.09	.05	1.00							
D2 CP	-.21**	-.12	.04	.11	-.19**	-.09	1.00						
D2 E	.04	.17*	.03	.20**	.12	.16*	-.45**	1.00					
COMT	-.16*	.03	.01	-.01	.02	-.13	.08	-.02	1.00				
DAT1	-.19*	-.07	.04	-.05	.05	.03	-.01	-.08	.05	1.00			
CHRNA4	-.03	-.11	-.08	-.07	-.07	-.13	.07	-.11	-.03	.05	1.00		
5_384	.06	.05	-.01	-.04	-.03	-.14	.01	-.03	.07	-.05	-.04	1.00	
5_169	-.09	-.01	.02	-.01	.02	-.01	.04	-.04	.05	.02	.07	-.58**	1.00

Notes: ° correlation is significant at the 0.10 level (one-tailed); * correlation is significant at the 0.05 level (two-tailed); **correlation is significant at the 0.01 level (two-tailed). Please note that ST = Stroop effect; PP = PP effect in the Negative priming task (the mean RT difference between the attended repetition and control condition); NP = NP effect in the Negative priming task (the mean RT difference between the control condition and the ignored repetition); BENEFITS = RT benefits in the Posner-Cuing task (the mean RT difference between the valid and neutral condition); COSTS = RT costs in the Posner-Cuing task (the mean RT difference between the invalid and neutral condition); ADHD = ADHD diagnosis derived from the ADHD-SR checklist; D2 CP = concentration performance in the D2 test; D2 E = percentage of omission and commission errors in the D2 test in relation to correct responses; COMT = *COMT* Val158Met polymorphism (Val/Val vs. Met+); DAT1 = *DAT1* polymorphism genotypes (9/9 vs. 9/10 vs. 10/10); CHRNA4 = *CHRNA4* rs1044396 genotypes (T/T vs. C/T vs. C/C); 5_384 = *CHRNA5* rs3841324 genotypes (S/S vs. L+); 5_169 = *CHRNA5* rs16969968 genotypes (G/G vs. A+)

Overall summary. It was hypothesized that dopaminergic and cholinergic polymorphisms would modulate selective attention in a differential way – specifically, that the selected dopaminergic polymorphisms would influence the performance in tasks of selective top-down attention, but not in tasks of bottom-up attention. In contrast, it was expected that the selected cholinergic polymorphisms would influence the performance in tasks of selective bottom-up attention, but not in tasks of top-down attention. Carriers of the Met allele of the *COMT* Val158Met polymorphism and carriers of the 10-repeat allele of the *DAT1* polymorphism displayed a better performance in the Stroop task. Neither the *COMT* Val158Met polymorphism nor the *DAT1* polymorphism modulated the performances in the Negative priming task or in the Posner-Cuing task. The *CHRNA4* rs1044396 polymorphism was not found to modulate the attentional performance in any of the three tasks, whereas the *CHRNA5* rs3841324 and *CHRNA5* rs16969968 polymorphisms impacted on the performance in the Stroop task, but did not modulate any other measures of selective attention.

Chapter 7

Discussion of the Attentional Analyses

In this chapter, the attentional analyses will be discussed. First, the hypotheses and results will be summarized (subchapter 7.1). The attentional effects of the dopaminergic polymorphisms will then be discussed in subchapter 7.2 (in the case of the *COMT* Val158Met polymorphism) and in subchapter 7.3 (in the case of the *DAT1* polymorphism). In subchapter 7.4, it will be debated whether these findings align with the dissociation hypothesis of Noudoost and Moore (2011a). Likewise, the effects of the cholinergic polymorphisms will first be discussed individually and then in the wider context of the dissociation hypothesis. Subchapter 7.5 will elaborate on the findings for the *CHRNA4* rs1044396 polymorphism, while subchapter 7.6. will elaborate on the findings for *CHRNA5* polymorphisms. The implications of the cholinergic results for the dissociation hypothesis will be discussed in subchapter 7.7.

7.1. Summary of the attention hypothesis and results

It was the primary objective of this thesis to examine the impact of DA and ACh on selective top-down and bottom-up attention. The thesis was aimed at disentangling the individual contributions of both system on these opposing principles of selective attention. It was hypothesized that the impact of the dopaminergic neurotransmitter system on selective top-down attention is greater than its influence on selective bottom-up attention. Analogously, it was hypothesized that the impact of the cholinergic neurotransmitter system on selective bottom-up attention is greater than its influence on selective top-down attention. This hypothesis – best labelled as the *dissociation hypothesis* – has first been proposed by Noudoost and Moore (2011a). In their review, Noudoost and Moore gave an overview on findings from animal models. This thesis was aimed at examining the possible dissociation of DA and ACh effects in a human sample, via non-invasive methods. Five different polymorphisms had been selected to this end. A double dissociation of the dopaminergic and cholinergic polymorphisms had been expected in regard to selective top-down and bottom-up attention. Specifically, it had been expected that the Met allele carriers of the *COMT* Val158Met polymorphism and the 10-repeat allele carriers of the *DAT1* polymorphism would display a better performance in the Stroop task and Negative priming task. At the same time, it had been expected that these polymorphisms would not (or only to a lesser degree) impact on the performance in the Posner-Cuing task. Likewise, it had been expected that *CHRNA4* rs1044396, *CHRNA5* rs3841324, and *CHRNA5* rs16969968 polymorphisms would impact on the performance in the Posner-Cuing task, but not (or only to a lesser degree) on the performance in the Stroop and Negative priming task. A better performance in this task had been predicted for the T allele carriers of the *CHRNA4* rs1044396 polymorphism, while no prior assumptions about the advantageousness of the *CHRNA5* alleles had been drawn. In line with the dissociation hypothesis, carriers of the Met allele of the *COMT* Val158Met polymorphism and carriers of the 10-repeat allele of the *DAT1* polymorphism displayed a better performance in the Stroop task. Carriers of these alleles

displayed smaller Stroop effects, which suggests that they were less affected by the conflicting properties of the Stroop stimuli. Also as hypothesized, neither the *COMT* Val158Met nor the *DAT1* polymorphism was found to modulate the performance in the Posner-Cuing task. In contrast, the findings in regard to the Negative priming task did not align with the dissociation hypothesis. Neither the *COMT* Val158Met polymorphism nor the *DAT1* polymorphism individually modulated the performance in the Negative priming task. On the cholinergic side, the *CHRNA4* rs1044396 polymorphism was not found to modulate the performance in the Stroop task, Negative priming task or Posner-Cuing task. Likewise, there were no main effects of the *CHRNA5* rs3841324 polymorphism and *CHRNA5* rs16969968 polymorphism on the performance in the three tasks. Significant interactions between the *CHRNA5* polymorphisms and gender were noted in the case of the Stroop task, however. Here, the effects of both polymorphisms were diametrically opposed in men and women. While male carriers of the S/S genotype of the *CHRNA5* rs3841324 polymorphism and of the G/G genotype of the *CHRNA5* rs16969968 polymorphism displayed *smaller* Stroop effects than their male counterparts, the female carriers of the S/S genotype and G/G genotype displayed *larger* Stroop effects than their female counterparts. Thus, the degree to which the participants were affected by the conflicting or non-conflicting properties of the Stroop stimuli seemed partly to be determined by interactions between the selected *CHRNA5* polymorphisms and gender. In sum, the analyses of the cholinergic polymorphisms did not yield the expected results. The polymorphisms were either found to have no influence on the attentional performance in the three tasks (in the case of the *CHRNA4* rs1044396 polymorphism) or impacted on the performance in a task of top-down attention (in the case of the *CHRNA5* rs3841324 and *CHRNA5* rs16969968 polymorphisms).

7.2. The COMT Val158Met polymorphism and selective attention

The *COMT* Val158Met polymorphism modulated the performance in the three tasks largely in the expected direction. In the Stroop task, carriers of the Met allele outperformed carriers of the Val allele. In contrast, there were no associations between the polymorphism and the performance in the Posner-Cuing task. Unexpectedly, the polymorphism also had no effect on the performance in the Negative priming task. Yet, there were indications that the absence of this effect might be due to the characteristics of the specific sample and the characteristics of the present task. In stark contrast to the PP effect, the NP was only significant at a one-tailed level. A large portion of the participants – 43 %, to be precise – did not display the NP effect at all. The possible underlying reasons will be detailed in the General Discussion (chapter 10); at this point it might suffice to say that the ability to touch-type

seemed to have been a driving factor. The participants that were unable to touch-type were slower across all conditions. While the NP effect was highly significant when *only* the touch-typers were considered, the NP effect was not apparent at all when those participants were excluded from the analysis. It can be speculated that the task was too difficult for the participants that were not familiar with the German standard system of keyboard writing. Thus, the NP effect – as a measure of selective top-down attention – did not emerge clearly in the majority of the sample. For this reason, the lacking interaction between the conditions of the Negative priming task and the *COMT* Val158Met genotype is perhaps not surprising and does not devalue the result that has been obtained for the other task of selective top-down attention. In sum, the *COMT* Val158Met polymorphism modulated selective top-down attention (i.e., the performance in the Stroop task), but was not associated with selective bottom-up attention (i.e., the performance in the Posner-Cuing task). The result aligns with previous studies that emphasized the role of the *COMT* Val158Met polymorphism for PFC-dependent subprocesses (cp. chapter 2). In the Stroop task, the task instructions are expected to lead to a prioritizing top-down bias for those stimuli that are deemed goal-relevant. MacDonald and colleagues (2000) found the activity in the DLPFC to be affected during the instruction phase in the Stroop task (cp. chapter 3); they suggested that the DLPFC is involved in the preparation of the response and responsible for the strategic control processes that help to prioritize the less routinized action over a more routinized action. Chen and colleagues (2004) studied the activity of the *COMT* Val158Met polymorphism specifically in DLPFC tissues, as they saw the DLPFC as "a brain region critical to cognitive function and presumably especially impacted by COMT activity" (p. 808). This group found the activity of the Met-containing COMT enzymes to be lower than the activity of Val-containing COMT enzymes. They concluded that these effects are most likely accompanied by higher prefrontal DA levels in Met allele carriers and lower prefrontal DA levels in Val allele carriers. But why is the higher DA level in the DLPFC of Met allele carriers beneficial for the performance in PFC-dependent tasks? According to the *inverted-U curve hypothesis*, the relation between the amount of DA signaling in the PFC and the level of prefrontal function takes the form of an inverted-U curve (Goldman-Rakic et al., 2000; Mattay et al., 2003). According to this hypothesis, both low and high amounts of DA signaling are detrimental for the PFC function. Mattay and colleagues (2003) proposed that Met allele homozygotes are positioned at a more optimal region of the inverted-U curve than Val allele homozygotes; they corroborated this assumption by administering amphetamine to participants, thus increasing the rate of DA signaling. This increase in DA signaling was detrimental for the performance of Met homozygotes in the n-back task and led to a less efficient prefrontal activation response in them. In contrast, the prefrontal activation response became more efficient in Val homozygotes under the administration of amphetamine. In their review article, Dickinson and Elvevåg (2009) concluded that the amount of DA signaling in Met allele carriers is associated with a higher neuronal efficiency. It might seem

paradoxical, but the higher rate of DA signaling in Met allele carriers presumably *decreases* the extent of prefrontal activation overall. Conversely, the lower rate of DA signaling in Val allele carriers is associated with an *increased* extent of prefrontal activation (Mier et al., 2009). This relation between the prefrontal activity and efficiency is termed the *efficiency hypothesis* (Dickinson & Elvevåg, 2009; Cools & D'Esposito, 2011). This hypothesis makes a broad, sweeping assumption about the activity pattern of a wide network of neurons. How might these effects be exerted at a synaptic level? At a synaptic level, the D1 receptors are assumed to play an important role in the modulation of prefrontal efficiency (Mattay et al., 2003; Yang, Seamans, & Gorelova, 1999). In mammalians, D1 receptors are strongly expressed in the forebrain (von Bohlen und Halbach & Dermietzel, 2006). It has been reported that D1-type receptors outnumber D2-type receptors more than 20-fold in this brain area (Goldman-Rakic et al., 2000). D1Rs are located on the dendrites and spines of prefrontal pyramidal cells, in close proximity to putative glutamatergic axon terminals; at the same time, they are also present in GABAergic interneurons (Goldman-Rakic et al., 2000). Goldman-Rakic and colleagues proposed an inverted-U curve model for the relationship between the extent of D1R stimulation and the performance in working memory tasks. This model has later been generalized to describe the assumed relationship between the amount of DA signaling and the PFC function (i.e., the *inverted-U curve hypothesis;* Mattay et al., 2003). Goldman-Rakic and colleagues (2000) assume that low levels of DA transmission do not lead to a D1R-driven enhancement of the glutamatergic inputs to pyramidal cells and interneurons (see figure 7.1). Intermediate levels of DA, however, are assumed to lead to a D1R-driven enhancement of the glutamatergic inputs to pyramidal neurons. High levels of DA transmission are assumed to lead to a D1R-driven enhancement of the glutamatergic inputs to pyramidal neurons *and* interneurons. In short, it is assumed that low levels of DA have no enhancing effect on pyramidal neurons, while intermediate levels of DA result in excitatory effects and high levels of DA result in inhibitory effects.

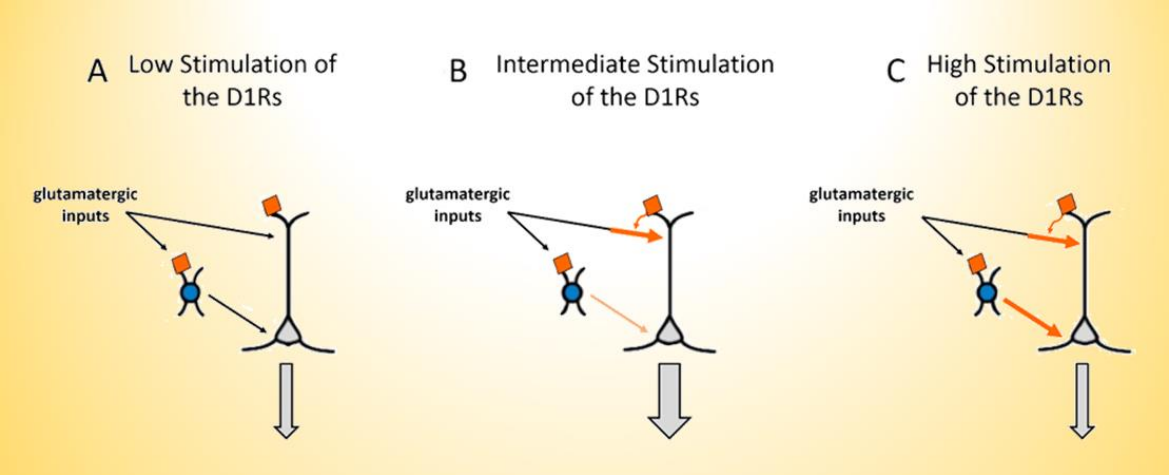


Figure 7.1.: The assumed relation between the D1R stimulation and the firing rate of prefrontal pyramidal neurons (figure taken and adapted from Goldman-Rakic et al., 2000). Gray cells signify prefrontal pyramidal cells, blue cells signify interneurons, and orange rhombi signify D1Rs. In their model, Goldman-Rakic and colleagues assume that amount of prefrontal DA modulates glutamatergic (i.e., excitatory) inputs to pyramidal cells and to interneurons. Low levels of DA should *not* lead to an enhancement of the glutamatergic inputs. Intermediate levels of DA should lead to a higher stimulation of D1Rs and this should enhance the glutamatergic inputs to the pyramidal cells and to the interneurons. Due to differential densities, this enhancement is assumed to be unequal; larger for the glutamatergic inputs to the pyramidal cell than for the glutamatergic inputs to the interneurons. The net effect should be excitatory and increase the firing rate of the pyramidal cell. When the level of DA transmission elevates, Goldman-Rakic and colleagues assume that the effect of the D1Rs on the pyramidal cell reaches a plateau, but increases the effect of the glutamatergic inputs to the GABAergic (i.e., inhibitory) interneurons. This should result in a feed-forward inhibition and decrease the firing rate of the prefrontal pyramidal neurons.

The level of dopaminergic transmission in the PFC directly affects the extent to which the D1Rs are stimulated. These receptors, in turn, can lower the threshold for the neuronal excitability of pyramidal neurons or raise it via inhibitory interneurons, thus ultimately regulating the firing frequency of the pyramidal cells (Goldman-Rakic et al., 2000; Mattay et al., 2003; Yang et al., 1999; Cools & D'Esposito, 2011). Intermediate levels of DA signaling should lead to a more efficient prefrontal activation pattern and thus result in a higher performance level in PFC-dependent tasks. In a manner of speaking, it could be said that more signal is transmitted in comparison to the noise of neuronal firing, or that the signal is "sharpened" (Mattay et al., 2003). This type of modulation might be at the core of the cognitive benefits that Met allele carriers display in PFC-dependent tasks. For Met allele

carriers, Dickinson and Elvevåg (2009) noted a better signal-to-noise ratio in the PFC, a reduced signal variability, and a sharper peak signal – all of which are likely signs of an optimal activation of the D1Rs. Under normal conditions, D1Rs should be more strongly stimulated in the group of Met allele carriers. It has been shown that drugs reduced the prefrontal of efficiency of Met allele carriers by elevating the extent of DA signaling (Cools & D'Esposito, 2011; Mattay et al., 2003). Noudoost and Moore (2011a) highlighted the importance of D1Rs for top-down attention as they elaborated on the transmission of prefrontal attentional signals to the sensory cortices (cp. chapter 3). At this point, the reviewed literature comes full circle. Carriers of the Met allele of the *COMT* Val158Met polymorphism outperformed Val homozygotes in the Stroop task, most likely because this task is PFC-dependent and Met allele carriers exhibit a higher level of prefrontal efficiency.

7.3. The DAT1 polymorphism and selective attention

While the main impact site of the *COMT* Val158Met polymorphism is the PFC, the DAT protein is most strongly expressed in the striatum (Sesack et al., 1998; Lewis et al., 2001). As hypothesized, carriers of the 10-repeat allele of the *DAT1* polymorphism displayed a better performance in the Stroop task. This finding aligns with the report by Rueda and colleagues (2005), who observed that homozygous 10-repeat allele carriers outperformed heterozygous carriers in an Eriksen Flanker task. In this dissertation, the *DAT1* polymorphism did *not* modulate the performance in the Posner-Cuing task. This aligns with the finding of an animal study, where the striatal level of dopaminergic transmission did not influence the performance of rats in an adapted Posner-Cuing task (Ward & Brown, 1996). In sum, the *DAT1* polymorphism modulated selective top-down attention (i.e., the performance in the Stroop task), but did not modulate selective bottom-up attention (i.e., the performance in the Posner-Cuing task). The *DAT1* polymorphism also was not found to modulate the performance in the Negative priming task, but the lack of this effect might be due to specific task and sample characteristics (cp. chapter 10). What is the mechanism of action through which *DAT1* likely affects top-down processing? The DAT binding site density has been reported to be 50 % higher for the 10-repeat allele (VanNess et al., 2005). This points towards a higher amount of DAT activity in carriers of the 10-repeat allele, and thus conversely towards a lower rate of striatal DA transmission. As contoured in chapter 2, *DAT1* might modulate top-down processing either directly through its presence in the PFC or indirectly through its presence in the striatum (Bertolino et al., 2006). Given its low abundance in the PFC, however, it is more likely that the *DAT1* polymorphism modulates cognitive functioning through its primary impact site, the striatum. Cools and D'Esposito (2011) stated:

"While animal work has highlighted the role of basal DA levels specifically in the PFC, human PET work has revealed an important role for basal dopamine in a different brain region, i.e. the striatum. A critical role for the striatum in DA function is not surprising, given that dopaminergic projections are strongest, and receptors most abundant, in the striatum, as well as given existence of strong anatomical connections between the PFC and the striatum […]" (2011, p. 120).

Alterations in the striatal DA transmission in patients of Huntington's disease have been reported to affect the performance in a series of executive tasks, among them the Stroop task (Peinemann et al., 2005). In a PET study, Bäckman and colleagues (2000) found differences in the striatal binding of the D2 receptor to account for a larger extent of age-related cognitive deficits than the actual chronological age. In another PET study, Erixon-Lindroth and colleagues (2005) found the binding of DAT in the striatum to decline during aging; likewise, the performance in a range of tasks (including executive tasks) declined during aging. Interestingly, the degree of DAT-binding mediated the cognitive deficits, leading the authors to conclude that "DAT binding is a powerful mediator of age-related cognitive changes as well as of cognitive functioning in general" (p. 1). These findings indicate that striatal DA exerts effects on cognition that might previously have been subsumed under the domain of prefrontal DA. In human samples, PET offers the most direct form of DA measurement in the CNS; notably, PET scans are not suitable to detect DA in the PFC, but allow the visualization of the striatal DA presence (Cools & D'Esposito, 2011). For this reason, the findings of Bäckman and colleagues (2000) as well as Erixon-Lindroth and colleagues (2005) are especially valuable. It has long been emphasized that the striatum and the cortex are functionally connected. Both areas are presumed to be part of a circuit that also encompasses the thalamus and thus is usually termed the striato-thalamo-cortical circuit (see Newman & Grace, 1999, for an overview on the interconnections within this system). The striatum receives projections from wide areas of the cortex, but in return only transmits to limited frontal regions (Alexander, DeLong, & Strick, 1986). Alexander and colleagues distinguished between a striato-thalamo-cortical circuit that involves the DLPFC (BA 9 and BA 10) and one that involves the lateral orbitofrontal cortex. The DLPFC circuit is depicted in figure 7.2.

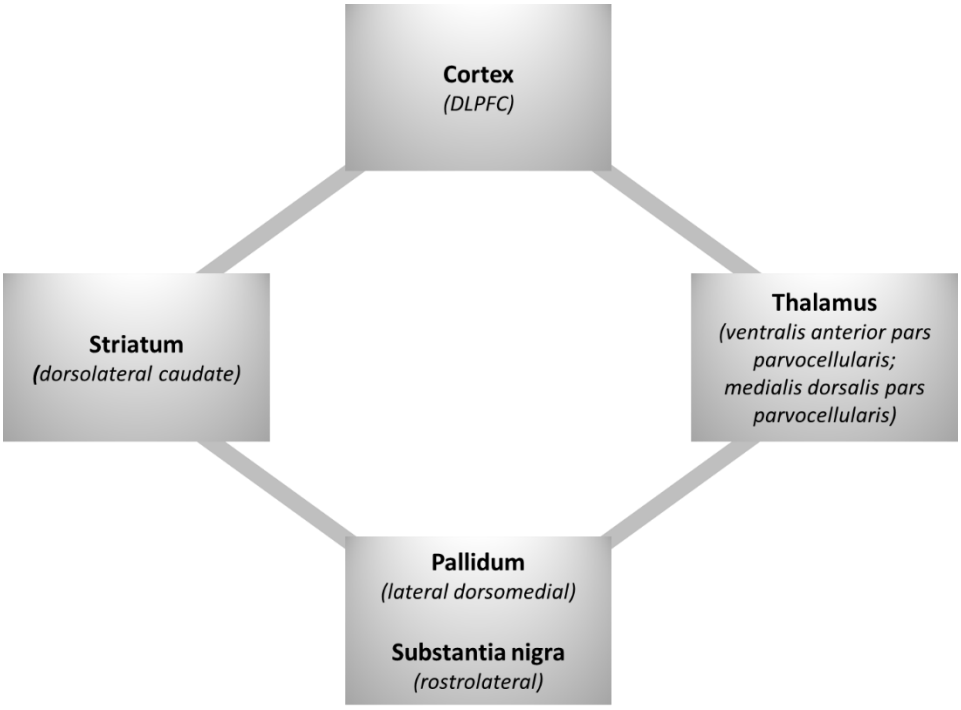


Figure 7.2.: The striato-thalamo-cortical circuit (figure taken and adapted from Alexander et al., 1986). The caudate – an area in which DAT is highly expressed (Shook et al., 2011) – is a key component of the circuit.

Given the deep interconnections between the components of the striato-thalamo-cortical circuit, it is clear why the dopaminergic transmission in the striatum might influence operations at the level of the PFC profoundly. Erixon-Lindroth and colleagues (2005) stated that "alterations in any one component of the […] network may lead to functional and eventually structural changes in other components" (p. 8). Cools and D'Espositio (2011) proposed that the connectivity within the circuit might vary dependent on the baseline levels of DA in the striatum. The striatum might be capable of increasing the single-to-noise ratio in the cortex by facilitating the coordination of cortical activity and suppressing activity that is less relevant for the current task set (Bertolino et al., 2006). A decreased level of striatal DA – which is most likely the case in the presence of the 10-repeat allele – might lead to a more focused response within the circuit (Bertolino et al., 2006). On a cellular level, it is not yet fully understood how the different components of the circuit interact. While D1Rs play a dominant role in the PFC, D2 receptors (D2Rs) are expressed more abundantly in the striatum (Sesack, Aoki, & Pickel, 1994). There are indications that the function level of the striatum and the level of D2R stimulation might also underlie an inverted-U curve relation, with different baseline levels exerting either advantageous or disadvantageous effects on cognitive performance (Cools & D'Esposito, 2011). Since D2Rs are also located on the dopaminergic presynapses, they can function as autoreceptors and

inhibit the DA synthesis and release when stimulated (von Bohlen und Halbach & Dermietzel, 2006). The more DA is released, the more DA diffuses away from the synaptic cleft and reaches the autoreceptors, thus triggering a self-regulatory feedback signal. The sensitivity of the D2R autoreceptors might differ according to the baseline level of the striatal DA transmission; if the level is already close to optimum, for example, the D2Rs might be comparatively more sensitive to ensure the homeostasis of the existing DA levels (Cools & D'Esposito, 2011). In sum, carriers of the 10-repeat allele of the *DAT1* polymorphism outperformed 9-repeat homozygotes in the Stroop task. DAT is the primary catabolic DA agent in the striatum. It can be speculated that a decreased level of striatal DA transmission is more optimal in the context of an inverted-U curve relation between the DA signaling and the striatal function level. Notably, the striatum is a key component of the striato-thalamo-cortico circuit and reciprocally communicates with the DLPFC. This finding further emphasizes the role of the striatum for the modulation of top-down processes.

7.4. DA and the dissociation hypothesis

Selective top-down mechanism shift attention towards stimuli that are deemed goal-relevant, while selective bottom-up mechanisms shift attention towards highly salient stimuli (Connor et al., 2004). Both types of attention strikingly differ in their degree of controllability and automaticity. Pashler and colleagues (2001) stated that "most human behavior would seem to lie in between [the] two extremes, reflecting the joint impact of high-level goals […] and recent stimuli […]. Teasing apart the principles that govern the interaction of top-down and bottom-up forces is critical to understanding any type of human behavior" (p. 630). In cognitive psychology, these principles are most often disentangled by administering cognitive tasks in human samples, studying various meticulous parameters like RT differences. These studies help to uncover the nature of top-down and bottom-up attention, but do not illuminate their neural foundations in the CNS. Noudoost and Moore (2011a) stressed the importance of disentangling the neural circuits of top-down and bottom-up attention. Specifically, they proposed that "ACh may serve a more unique role in bottom-up attention than it does in top-down attention, whereas the reverse may be true for DA" (p. 589). Their method of choice is the *in vivo* recording of single neurons in dependence on the microiontophoretic application of antagonists or agonists (typically in samples of Rhesus monkeys). While highly illuminating, this type of studies is much too invasive to be realizable in human samples. This is problematic, since the human brain naturally differs from the brain of non-human primates. In fact, the DLPFC has been described as the "hallmark of the human brain" (Weinberger, Berman, & Chase, 1988) and it has even been proposed that "mesocortical prefrontal dopaminergic projections […] could represent an important

element in prefrontal evolution", making "even the comparison to the great apes […] limited" (p. 330). It stands to reason that the psychological lines of research and the neurobiological lines of research need to be intertwined. One way of doing this would be the assessment of naturally occurring genetic variations within the neurotransmitter systems. Being non-invasive, this method is easily applicable in samples of human participants. In this dissertation, the effects of the *COMT* Val158Met polymorphism and *DAT1* polymorphism were studied in regard to the performance in the Stroop task and the Posner-Cuing task, two classical tasks that have been utilized for decades in cognitive psychology and that provide clear-cut measures for selective top-down and bottom-up attention. Carriers of the Met allele and of the 10-repeat allele displayed smaller Stroop effects than Val homozygotes and 9-repeat homozygotes, which indicates that they were less affected by the conflicting stimulus properties in the Stroop task. It is possible that these participants were more capable of representing and maintaining the instructions of the task, and more able to enforce the necessary strategic control processes (MacDonald et al., 2000). At the same time, these polymorphisms did not modulate the performance in the Posner-Cuing task. Thus, this dissertation provides indications that the dopaminergic system indeed appears to contribute more to selective top-down attention than to selective bottom-up attention. This finding complements the evidence that Noudoost and Moore (2011a) accumulated and is the first such dissociative finding in a human sample. In regard to the cellular effects of the DA transmission levels in the brain, it appears it is the dosage that matters. The Swiss scholar Paracelsus said *"no thing is without poison. The dosage makes it either a poison or a remedy; […] it must not be too much nor too little"* (p.210; see Deichmann, Henschler, Holmstedt, & Keil, 1986, for on overview on his legacy). Vouching for a "*middle dose",* as Paracelsus did, seems to be the golden way for the dopaminergic system. It can be assumed that the level of dopaminergic transmission underlies an inverted-U curve relation to the function level in the PFC and in the striatum. Fortunately, the impact sites of both the *COMT* Val158Met polymorphism and the *DAT1* polymorphism are regionally very specific. The *COMT* Val158Met polymorphism most likely contributes to the level of DA signaling in the PFC (affecting D1Rs), while the *DAT1* polymorphism most likely contributes to the level of DA signaling in the striatum (affecting D2Rs). All in all, the results of this dissertation support the assumption that the dopaminergic system is more important for selective top-down attention than for selective bottom-up attention, thus supporting Noudoost's and Moore's dissociation hypothesis.

7.5. The CHRNA4 rs1044396 polymorphism and selective attention

In this dissertation, the *CHRNA4* rs1044396 polymorphism did not modulate the attentional effects in the Stroop task, Negative priming task, or Posner-Cuing task, irrespective of whether the task

in question measured selective top-down or bottom-up attention. The polymorphism also was not associated with the ADHD score – and thus with attentional impairments – or with the performance in the D2 test, which measures general attention and concentration capabilities. These results appear to contradict previous reports, where the relevance of this polymorphism for attentional processes has been highlighted (for an overview, cp. Greenwood et al., 2012). Specifically, Greenwood and colleagues proposed that "the CHRNA4 rs1044396 [polymorphism] is characterized by greater ability of T/T homozygotes compared to the other genotypes to maintain the focus of visuospatial attention with preferential processing of events in the attentional focus compared to events outside the attentional focus" (p. 1332). The tasks that were reviewed by Greenwood and colleagues (2012), however, were based on endogenous shifts of attention. In such tasks, cues are helpful to predict the location of a target. If the *CHRNA4* rs1044396 polymorphism is only relevant for endogenous shifts of spatial attention, the utilized tasks in this dissertation were not best suited to reflect its potential effects. Both in the Stroop and in the Negative priming task, the visuospatial stimulus configuration was no relevant factor. In the Stroop task, the stimuli were presented centrally on the screen; the relevant stimulus property (color) and the irrelevant stimulus property (word content) were spatially not segregated. In the Negative priming task, the target was always the middle letter of a string of three letters, invariant in its position from prime to probe. In the Posner-Cuing task, the visuospatial location was a factor – as covert shifts of attention were required – but since the frequency of valid and invalid trials was equal, the cue did not predict the location target above chance levels. In sum, it can only be speculated why the *CHRNA4* rs1044396 polymorphism did not exert any effects on attention. A possible explanation might be that the polymorphism is only relevant for a specific subset of visuospatial processes (endogenous shifts of attention) and not relevant for selective top-down or bottom-up attention in a much broader sense (for an elaboration on this point, cp. Schneider, Schote, Meyer, Markett, Reuter, & Frings, 2015). It should also be kept in mind that the genotypes of the *CHRNA4* rs1044396 polymorphism were not detectable in the whole sample; the decreased sample size of n = 157 was accompanied by a decrease in the statistical power. Since the effects of individual polymorphisms are quite small, any decreases in power are naturally problematic and make it more difficult to detect any modulations of the behavioral parameters.

7.6. The CHRNA5 polymorphisms and selective attention

The performance in the Stroop task was modulated both by the *CHRNA5* rs3841324 polymorphism and by the *CHRNA5* rs16969968 polymorphism if gender was considered as a covariant. Interestingly, the effects of both polymorphisms were diametrically opposed in men and women. This

result was unexpected, as it had been hypothesized that the *CHRNA5* polymorphisms would impact on the performance in tasks of selective bottom-up attention, but not in tasks of selective top-down attention. Notably, both *CHRNA5* polymorphisms did not only modulate the Stroop performance in a gender-dependent fashion, but also exerted gender-dependent influences on the average response speed in the Stroop task and Negative priming task. Male carriers of the S/S genotype of the *CHRNA5* rs3841324 polymorphism and of the G/G genotype of the *CHRNA5* rs16969968 polymorphism displayed smaller Stroop effects than their male counterparts and were also faster in the Stroop and Negative priming task (cp. chapter 8 and 9). Likewise, female carriers of the S/S genotype and G/G genotype displayed larger Stroop effects than their female counterparts and were also slower in the Stroop and Negative priming tasks. Attention benefits were accompanied by speed benefits; attention disadvantages were accompanied by speed disadvantages. This is an indication that both polymorphisms might modulate attention and response speed (a fundamental factor in information processing) in a parallel fashion. Traditionally, most cholinergic release sites have been assumed to contribute to volume transmission rather than point-to-point transmission (Dani & Bertrand, 2007). In fact, Dani and Bertrand (2007) stated that "although fast, direct nicotinic synaptic transmission drives neuromuscular junctions and autonomic ganglion synaptic transmission, only rare cases of fast nicotinic transmission have been reported in the mammalian brain" (p. 706). In contrast to volume transmission, point-to-point neurotransmission as a mode of intercellular communication is based on a one-to-one ratio of signal sources and signal targets (von Bohlen und Halbach & Dermietzel, 2006). Point-to-point neurotransmission exerts immediate effects, with synapses being meticulously optimized to achieve both speed and precision (Sabatini, & Regehr, 1999). This is a fast and robust way of communication, but also requires a higher metabolic investment (Agnati, Zoli, Strömberg, & Fuxe, 1995). Both point-to-point and volume transmission are depicted in figure 7.3. Sarter and colleagues (2014) challenged the view that the projections of the basal forebrain system exclusively contribute to volume transmission. They distinguished between the *neuromodulatory ACh system,* which results in the stable stimulation of ACh receptors in wide-spread populations of neurons, and the *deterministic ACh system,* which is characterized by its topographically organized, highly structured efferent projections (Sarter et al., 2014).

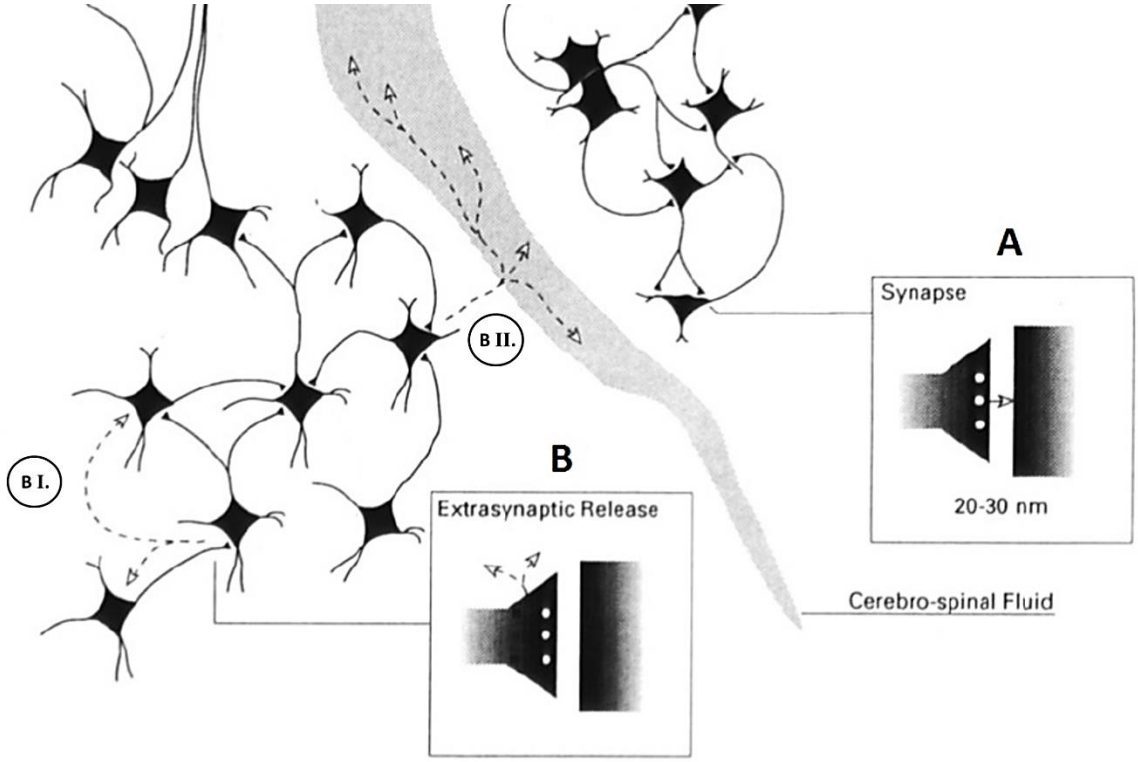


Figure 7.3.: Point-to-point transmission (A) and volume transmission (B) as types of intercellular communication in the CNS (figure taken and adapted from Agnati et al., 1995). Point-to-point transmission is fast, robust, and precise mode of communication between two cells (Agnati et al., 1995). In contrast, volume transmission relies on the extrasynaptical release and diffusion of neurotransmitter molecules and is thus slower, less robust and not as precise. Volume transmission can be further subdivided into short diffusion pathways (BI) and a long diffusion pathways (BII). The short pathway enables the communication between nearby cells (paracrine diffusion) or enables cellular self-stimulation, i.e. autoreceptor feedback signals (autocrine diffusion) (Agnati et al., 1995). The long pathway relies on the transport of neurotransmitter molecules within cerebro-spinal fluid and is a "hormone-like" communication type (Agnati et al., 1995; von Bohlen und Halbach & Dermietzel, 2006).

The deterministic system is capable of generating cholinergic transients – i.e., event-related increases in the release of ACh (Sarter et al., 2014). While volume transmission relies on slow diffusion processes, cholinergic transients can occur in a range of sub-seconds and influence cognitive operations in an immediate and precise manner (Sarter et al., 2014; Sarter, Parikh, & Howe, 2009). It has been proposed that cortical ACh influences the efficacy of information processing, namely the ability "to detect and select stimuli and associations for extended processing and to allocate the appropriate processing resources to these functions" (Sarter & Bruno, 1997, pp. 28). In a cued appetitive response task, changes in the cholinergic activity in the medial prefrontal cortex predicted if cues were detected or missed; while missed cues were not accompanied by cholinergic transients, detected cues were accompanied by these brief increases in ACh activity (Parikh, Kozak, Martinez, & Sarter, 2007).

Similarly, it has been reported that miss-timed cholinergic transients can increase the likelihood of false detections in trials that contain no targets (Sarter et al., 2014). Thus, it seems that the deterministic system influences the target detection. It might also influence the selection of a target amongst distractors or the selection of a task-relevant target property among task-irrelevant stimulus properties. How can the results of the *CHRNA5* polymorphisms be interpreted in the context of the deterministic ACh system? Despite being functional, both polymorphisms have so far not been studied extensively in regard to cognition. It has previously been reported that the *CHRNA5* rs3841324 polymorphism affected the performance in a WCST (Zhang et al., 2010), while the *CHRNA5* rs16969968 polymorphism has been related to memory and IQ parameters (Winterer et al., 2010; Breetvelt et al., 2014). This dissertation provides indications that both polymorphisms might also be relevant in the context of selective top-down attention, which would suggest a deterministic mode of operation. In the Stroop task, the diametrical *CHRNA5* effects canceled each other out and resulted in a non-significant net-effect when gender was not considered as a covariant. While male carriers of the S/S genotype and of the G/G genotype were *less* affected by conflicting stimulus properties in the Stroop task, female carriers of these genotypes were comparatively *more* affected by conflicting stimulus properties. Conversely, the presence of the S/S_G/G diplotype appeared to be beneficial for men, but disadvantageous for women. What mechanism underlies these effects? As the S allele has been linked to higher expression levels of the α5 subunit (Wang et al., 2009) and the G allele has been linked to an elevated nAChR response towards an agonist (Bierut et al., 2008), the S/S_G/G could be the diplotype constellation that results in the comparatively highest level of cholinergic transmission. It can be speculated that the interaction with gender is due to the close link between the α5 subunit and the sex hormone and neurosteroid progesterone. The relationship between the receptor subunit and this neurosteroid will be discussed in detail in chapter 9. At this point it may suffice to say that the progesterone levels in men and women differ greatly; furthermore, there are ample indications that the neurosteroid influences the expression levels of the α5 mRNA. As the α5 subunit is presumably present both in nAChRs in the deterministic and in the neuromodulatory ACh system, interactions of this subunit with progesterone might exert parallel effects in both systems. Naturally, this assumption requires more research.

7.7. ACh and the dissociation hypothesis

The *CHRNA4* rs1044396 polymorphism was not associated with the attentional measures in the Stroop task, Negative priming task, or Posner-Cuing task. In comparison, both *CHRNA5* polymorphisms modulated the performance in the Stroop task in interaction with gender. Noudoost and Moore (2011a) expected ACh to be more important for selective bottom-up attention than for

selective top-down attention. The present study provides no indications that this part of the dissociation hypothesis might hold true. The lack of cholinergic effects on selective bottom-up attention can be interpreted in multiple ways, however (and all of these are necessarily speculative within the framework of this dissertation). For one, Noudoost and Moore (2011a) might be correct and ACh might indeed be more crucial for selective bottom-attention. Second, it is also possible that ACh exerts effects on both selective bottom-up and top-down attention, not being more essential for one type of attention than the other. Third, ACh could influence selective top-down attention more than selective bottom-up attention. In all fairness, Noudoost and Moore (2011a) did not preclude the possibility that the cholinergic system might also play a notable role for top-down attention. In fact, they stated: "as it is known that different neuromodulatory systems interact with one another, including within PFC, the contributions of Ach and DA could be highly complex" (p. 589). One of the main arguments for the dissociation hypothesis – and thus a greater importance of ACh for bottom-up attention – is the deep link between the parabigeminal nucleus (or IPC) and the superior colliculus (Noudoost & Moore, 2011a). The parabigeminal nucleus transmits cholinergic inputs to the superior colliculus (Noudoost & Moore, 2011a). Located on the lateral edge of the midbrain, the nucleus is "so heavily interconnected with the superior colliculus that it has been characterized as a satellite [of the structure]" (Cui & Malpeli, 2003, p. 3128). Studies in birds have shown that this nucleus is sensitive to changes in many different stimulus categories; its neurons increased their firing rate when stimuli mapped to their receptive fields were more salient than competing distractor stimuli (Asadollahi et al., 2010). The parabigeminal nucleus might be responsible for the formation of salience maps (Asadollahi et al., 2010). Noudoost and Moore (2011a) saw this specific link between the nucleus and the superior colliculus as an argument for a potential cholinergic supervision of attentional bottom-up processes. The Guided Search theory (Wolfe, 1994) makes the assumption that the visual field is internally represented through dimension-specific maps (cp. chapter 3). Locations on these maps are differentially activated, both through bottom-up and top-down mechanisms. While bottom-up activations occur due to the inherent salience properties of the stimuli, top-down activations occur due to the preexisting knowledge of goals and strategies. In birds, it has been shown that the IPC reacts not only to stimuli saliences, but is also to top-down projections (Asadollahi et al., 2010). It can be speculated that the parabigeminal nucleus (in mammals) or the IPC (in birds) might be the neuroanatomical structure of Wolfe's dimension-specific activation maps. Given these lines of evidence, it is easy to comprehend why Noudoost and Moore (2011a) argued for a strong involvement of ACh in processes of bottom-up attention. Still, it is perhaps questionable whether this neurotransmitter system is really "more" or "less" involved than the dopaminergic system or even other systems. The sequence of information processing encompasses many brain regions and within these regions, many computational steps; the parabigeminal nucleus is not the first structure to

receive perceptive input, nor is the superior colliculus the last one to process them. The question of quantity might ultimately not be the most expedient one. Noudoost and Moore (2011a) arrived at their dissociation hypothesis by analyzing studies based on single-unit recordings. This is a very different methodological approach than the one that has been realized in this dissertation, where naturally occurring variations within the neurotransmitter systems were assessed. Of course, this approach did not allow the direct manipulation of different neuron populations within brain. It also did not allow to pinpoint the exact neuronal impact sites of the cholinergic polymorphisms. So far, the α5 subunit of the nAChR has been detected in the cortex, thalamus, putamen, and cerebellum, amongst other regions (Paterson & Nordberg, 2000), while the presence of the α4 subunit has also been detected in the cortex, cerebellum, and thalamus (Paterson & Nordberg, 2000; Greenwood et al., 2012). Given the wide-spread distribution of nAChRs, the assessment of genetic variants of nAChR subunits might not have offered a precise enough measure to demonstrate cholinergic effects in the parabigeminal nucleus and the superior colliculus. In contrast, it is perhaps no coincidence that the *COMT* Val158Met polymorphism and the *DAT1* polymorphism modulated the selective top-down attention largely in the expected direction (in comparison, their neural impact sites are very well defined). In sum, the dissociation hypothesis of Noudoost and Moore (2011a) could not be corroborated in regard to ACh and the influence of this neurotransmitter system on selective bottom-up attention. This might have been due to the chosen methodological approach, however. Yet, the present study helps to expand the understanding of the *CHRNA5* polymorphisms, especially in regard to their effect on cognition. Only recently, the *CHRNA5* rs3841324 polymorphism and the *CHRNA5* rs16969968 polymorphism have become the object of an increased number of studies. Both polymorphisms appear to be biologically functional, and as the results of this dissertation indicate, both polymorphisms are likely relevant for selective top-down attention. In future studies, it might be highly illuminating to focus on the effect of these polymorphisms (especially in regard to their interaction with gender).

Chapter 8

The Analyses of Response Speed Effects

This chapter is focused on the second hypothesis of this dissertation, i.e. on the possible effects of the selected cholinergic variants on response speed. As before, the Stroop task, Negative priming task, and Posner-Cuing task were analyzed, while the Dot Probe task was excluded from the analyses. For the *CHRNA4* rs1044396 polymorphism, it was expected that the average RTs in all tasks increased linearly as a function of the C allele dosage. The analyses of this polymorphism will be reported in subchapter 8.1. The *CHRNA5* rs3841324 polymorphism and *CHRNA5* rs16969968 polymorphism were analyzed in an exploratory fashion; no prior assumptions about the effects of the individual alleles were drawn. The analyses of these polymorphisms on response speed will be reported in subchapter 8.2. Please note that the results of the genotyping analyses, the task analyses, and the results of the *CHRNA5* gender and diplotype analyses were already detailed in chapter 6.

8.1. Response speed and the CHRNA4 rs1044396 polymorphism

It was hypothesized that the average RTs in the Stroop task, Negative priming task and Posner-Cuing task would increase linearly with the C allele dosage of the *CHRNA4* rs1044396 polymorphism. Since gender differences in the genotype contribution of the *CHRNA4* rs1044396 polymorphism were noted (cp. chapter 6), gender was included as a covariant in the main analyses. The effect of the *CHRNA4* rs1044396 polymorphism was analyzed via a multivariate analysis of variance (MANOVAs) for the average response speed across all tasks. Polynomial contrasts were built as a second-step analysis to test for linear dosage effects of the *CHRNA4* rs1044396 C allele, both across all tasks and within the individual tasks. Possible speed-accuracy tradeoff effects were also examined.

Overall Task Analyses. The effect of *CHRNA4* rs1044396 on the response speed across the three tasks was tested with a 3 (mean RTs: Stroop task vs. Negative priming task vs. Posner-Cuing task) x 3 (*CHRNA4*: T/T vs. C/T vs. C/C) repeated-measures MANOVA that included gender as a covariant. The analysis yielded a marginally significant main effect of the genotype [F (2, 146) = 2.44, p = .091, η_p^2 = .032]. T allele homozygotes were fastest in each task (M = 487 ms, SD = 60 ms), heterozygote carriers of the C allele displayed intermediate RTs on average (M = 494 ms, SD = 54 ms), and C allele homozygotes were slowest in each task (M = 513 ms, SD = 61 ms). Building polynomial contrasts, a significant linear trend between the *CHRNA4* rs1044396 genotype and the average response speed across all tasks was noted [F (1, 155) = 4.53, p = .035, 95%]. The average response speed was modulated by the C allele dosage of the *CHRNA4* rs1044396 polymorphism (see figure 8.1), linearly increasing with the number of C alleles.

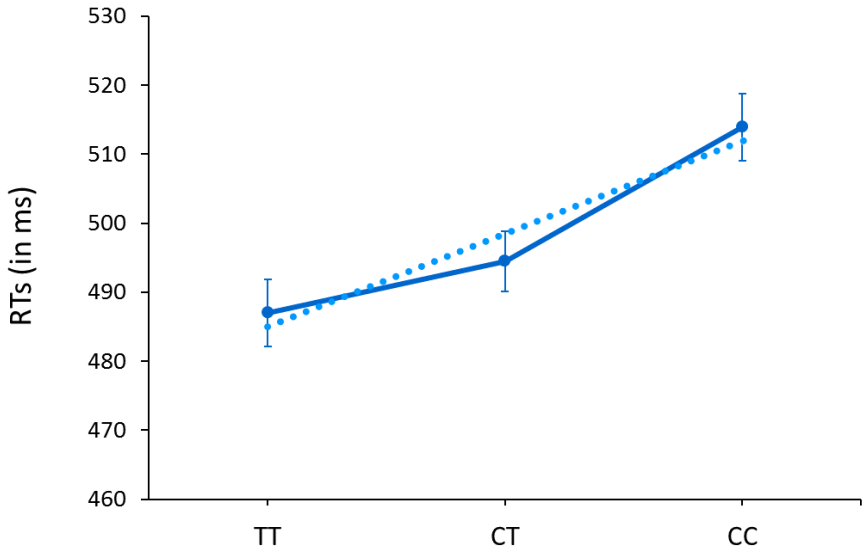


Figure 8.1.: Mean RTs (derived from the Stroop, Negative priming and Posner-Cuing tasks) as a function of the *CHRNA4* rs1044396 genotype. The dashed line depicts the linear trend line. The error bars depict the standard error of the mean.

Single Task Analyses. Next, individual analyses for the Stroop task, Negative priming task and Posner-Cuing task were conducted. Again, polynomial contrasts were built to analyze the effect of the *CHRNA4* rs1044396 polymorphism in the individual tasks.

The mean RTs in the Stroop task were submitted to a MANOVA with the *CHRNA4* rs1044396 genotype (*CHRNA4*: T/T vs. C/T vs. C/C) as factor and gender as a covariant. Building polynomial contrasts, a marginally significant linear trend was noted [F (1, 155) = 3.48, p = .064, 95%]. The trend indicated an increase in the average Stroop RTs with an increase in the number of C alleles (see figure 8.2). While T allele homozygotes displayed an average response speed of M = 411 ms (SD = 72 ms), heterozygote carriers of the C allele displayed an average response speed of M = 419 ms (SD = 77 ms), and C allele homozygotes displayed an average response speed of M = 444 ms (SD = 100 ms). The mean RTs in the Negative task were also submitted to a MANOVA with the *CHRNA4* rs1044396 genotype (*CHRNA4*: T/T vs. C/T vs. C/C) as factor and gender as a covariant. A polynomial contrast analysis showed that the linear trend missed significance [F (1, 156) = 2.27, p = .134]; see figure 8.2. T allele homozygotes displayed an average response speed of M = 667 ms (SD = 93 ms), heterozygote carriers of the C allele displayed an average response speed of M = 679 ms (SD = 94 ms), and C allele homozygotes displayed an average response speed of M = 698 ms (SD = 80 ms). The mean RTs in the Posner-Cuing task were submitted to a MANOVA with the *CHRNA4* rs1044396 genotype (*CHRNA4*: T/T vs. C/T vs. C/C) as factor and gender as a covariant. Building polynomial contrasts, a marginally significant linear trend was noted [F (1, 156) = 3.07, p = .082, 95%]. The trend indicates an increase in the average Posner-Cuing RTs with an increasing number of C alleles (see figure 8.2). While T allele

homozygotes displayed an average response speed of *M* = 383 ms (*SD* = 51 ms), heterozygote carriers of the C allele displayed an average response speed of *M* = 386 ms (*SD* = 42 ms), and C allele homozygotes displayed an average response speed of *M* = 400 ms (*SD* = 42 ms).

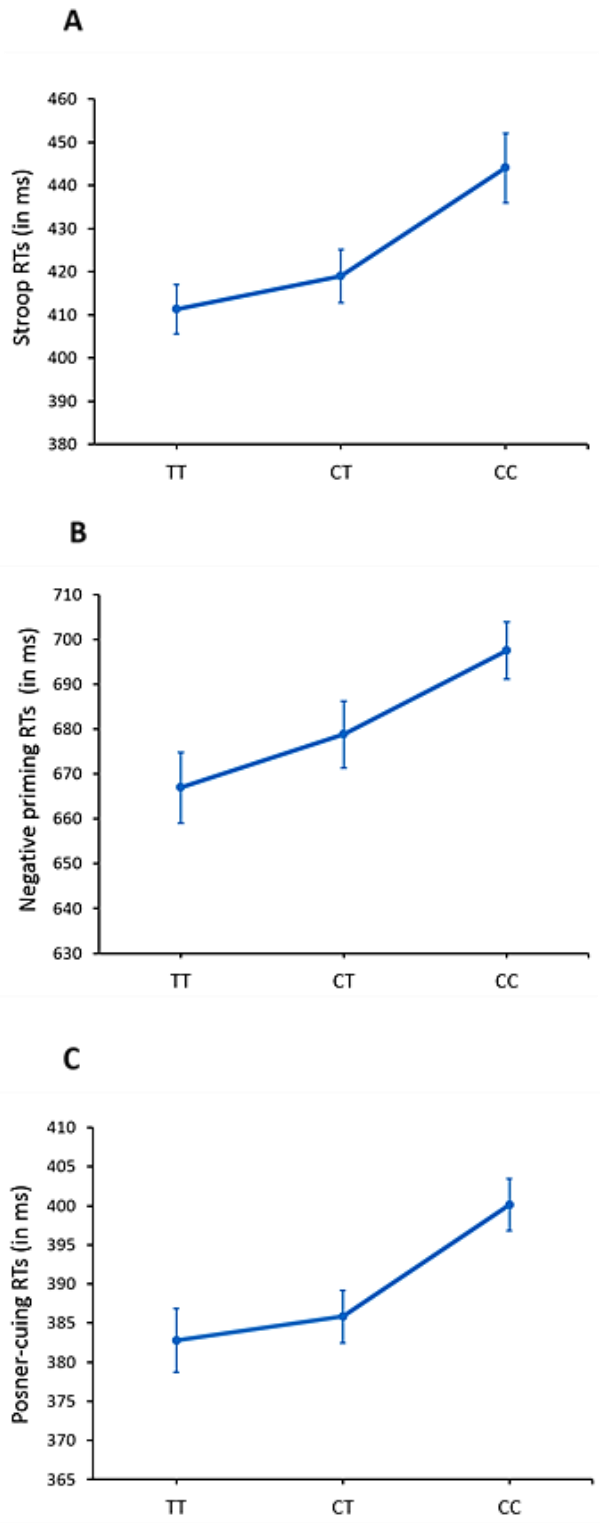


Figure 8.2.: RTs as a function of the *CHRNA4* rs1044396 genotype in the Stroop task (A), Negative priming task (B), and Posner-Cuing task (C). Error bars depict the standard error of the mean.

The average RTs in each task condition are depicted in figure 8.3. in dependence on the *CHRNA4* rs1044396 genotype. Descriptively, the observed speed benefits of the T allele carriers were nearly unequivocally present in each of the individual conditions.

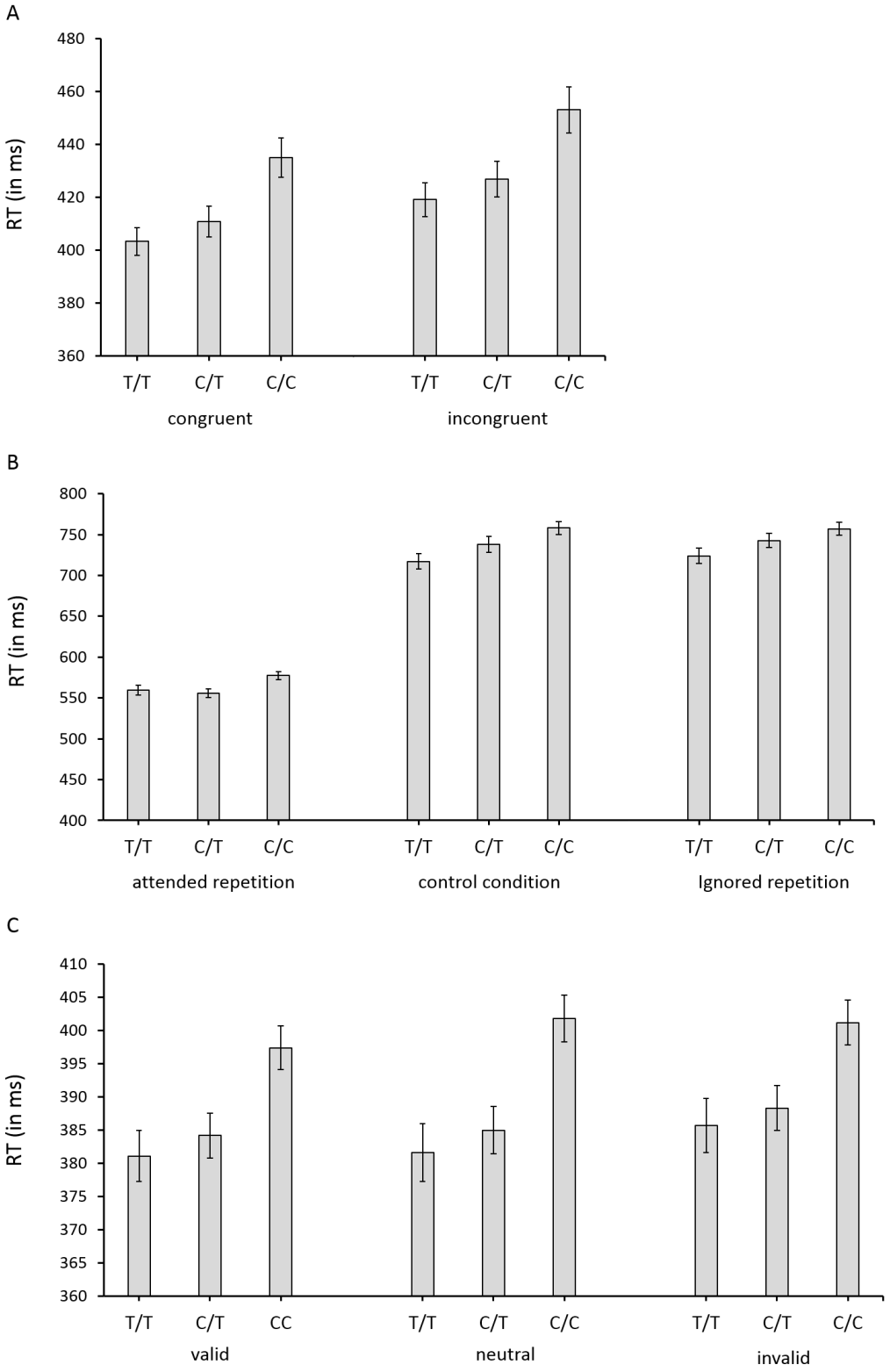


Figure 8.3.: RTs in the conditions of the Stroop task (A), Negative priming task (B), and Posner-Cuing task (C) as afunction of gender and the *CHRNA4* rs1044396 polymorphism. With the exception of the attended repetition of the Negative priming task, T allele homozygotes were fastest in all

conditions, while the C allele homozygotes were slowest. The polymorphism was not associated with any attentional parameters in the three tasks. Error bars depict the standard error of the mean.

Error analyses. When examining response speed, it is especially important to analyze not only the mean RTs but also the error rates. These analyses are necessary to test for the occurrence of speed-accuracy tradeoffs. Speed-accuracy tradeoff takes place when the response speed is improved at the expense of the accuracy (or vice versa). For all tasks, the participants had been instructed to act as fast and as accurate as possible. In the test trials, the participants had gotten a feedback both if they had been too slow or if they had made errors. Nevertheless, there was a possibility that response speed and accuracy might have been differentially favored in the rs1044396 genotype groups. The mean errors in the Stroop task[28] were submitted to a MANOVA with the *CHRNA4* rs1044396 genotype (*CHRNA4*: T/T vs. C/T vs. C/C) as factor and gender as a covariant. There was no significant main effect of the genotype [F (2, 146) < 1, p = .438, η_p^2 = .011] and polynomial contrast analysis showed no linear trend [F (1, 155) < 1, p = .706]. On average, T allele homozygotes made M = 1.0 (SD = 1.0) errors in the Stroop task, while heterozygote carriers of the C allele made M = 0.86 (SD = 0.85) errors, and C allele homozygotes made M = 0.96 (SD = 1.1) errors. The mean errors in the Negative task[29] were also submitted to a MANOVA with the *CHRNA4* rs1044396 genotype (*CHRNA4*: T/T vs. C/T vs. C/C) as factor and gender as a covariant. There was no significant main effect of the genotype [F (2, 147) = 2.07, p = .129, η_p^2 = .027]. A polynomial contrast analysis showed a trend towards a linear relationship, however [F (1, 156) = 3.31, p = .071, 95%]. T allele homozygotes made M = 2.1 (SD = 1.6) errors in the Negative priming task, while heterozygote carriers of the C allele made M = 1.8 (SD = 1.4) errors, and C allele homozygotes made M = 1.5 (SD = 1.2) errors. The errors in the Posner-Cuing task were not further analyzed, as the overall sum of errors was only n = 4 for all participants. In sum, there were no indications for a speed-accuracy tradeoff in the Stroop task, while there was a trend towards a speed-accuracy tradeoff in the Negative priming task. Due to the floor effect for errors in the Posner-Cuing task, no speed-accuracy tradeoff was realizable in this task. Overall, speed-accuracy tradeoffs could be ruled out both for the Stroop task and the Posner-Cuing task. When only these tasks were analyzed, the significant linear trend between the *CHRNA4* rs1044396 genotype and the average response speed still remained apparent, however [F (1, 155) = 4.43, p = .037, 95%].

[28] I.e., the average rate of error across the congruent and incongruent condition.
[29] I.e., the average rate of errors across the attended repetition, control condition, and ignored repetition.

8.2. Response speed and the CHRNA5 rs3841324 and rs16969968 polymorphisms

The *CHRNA5* rs3841324 polymorphism and *CHRNA5* rs16969968 polymorphism were analyzed in an exploratory fashion. As before, the rs3841324 polymorphism was analyzed on basis of comparisons between homozygous S allele carriers and L allele carriers (S/S vs. L+), while the rs16969968 polymorphism was analyzed on basis of comparisons between the homozygous G allele group and the group of A allele carriers (G/G vs. A+). The analyses were first carried out on a level of the individual polymorphisms (both without and with gender as a factor). The diplotypes of both polymorphisms were subsequently analyzed in the context of gender. Main effects of gender were also examined. As a last step, possible speed-accuracy tradeoffs were analyzed.

Effects of the CHRNA5 polymorphisms on response speed. First, possible effects of both CHRNA5 polymorphisms on the average response speed were analyzed without including gender as a covariant. The Stroop-RTs were submitted into a 2 (condition: congruent vs. incongruent) x 2 (*CHRNA5* rs3841324: S/S vs. L+) repeated-measures ANOVA. There was no main effect of the genotype on the average response speed [F (1, 177) < 1, p =.570, η_p^2 = .002]. The Negative priming RTs were also submitted to a 3 (condition: attended repetition vs. control condition vs. ignored repetition) x 2 (*CHRNA5 rs3841324*: S/S vs. L+) repeated-measures ANOVA. Again, no main effect of the genotype on the average response speed was observable [F (1, 178) < 1, p =.912, η_p^2 < .001]. Lastly, the Posner-Cuing-RTs were submitted into a 3 (condition: valid vs. neutral vs. invalid) x 2 (*CHRNA5 rs3841324*: S/S vs. L+) repeated-measures ANOVA. Again, there was no main effect of the genotype [F (1, 178) < 1, p =.419, η_p^2 = .004]. In sum, the *CHRNA5* rs3841324 polymorphism was not found to modulate the average response speed in the Stroop task, Negative priming task or Posner-Cuing task (see figure 8.3). Next, the Stroop-RTs were submitted into a 2 (condition: congruent vs. incongruent) x 2 (*CHRNA5* rs16969968*:* G/G vs. A+) repeated-measures ANOVA. The rs16969968 polymorphism did not modulate the average response speed in this task [F (1, 177) = 1.10, p =.295, η_p^2 = .006]. The Negative priming-RTs were subsequently submitted into a 3 (condition: attended repetition vs. control condition vs. ignored repetition) x 2 (*CHRNA5* rs16969968: G/G vs. A+) repeated-measures ANOVA. Again, no main effect of the genotype was observable [F (1, 178) < 1, p =.592, η_p^2 = .002]. Last, the Posner-Cuing-RTs were also submitted into a 3 (condition: valid vs. neutral vs. invalid) x 2 (*CHRNA5* rs16969968: G/G vs. A+) repeated-measures ANOVA. No effect of the genotype on response speed was notable [F (1, 178) < 1, p = .901, η_p^2 < .001]. Like the *CHRNA5* rs3841324 polymorphism, the *CHRNA5* rs16969968 polymorphism was not found to modulate the average response speed in the three behavioral tasks (see figure 8.4).

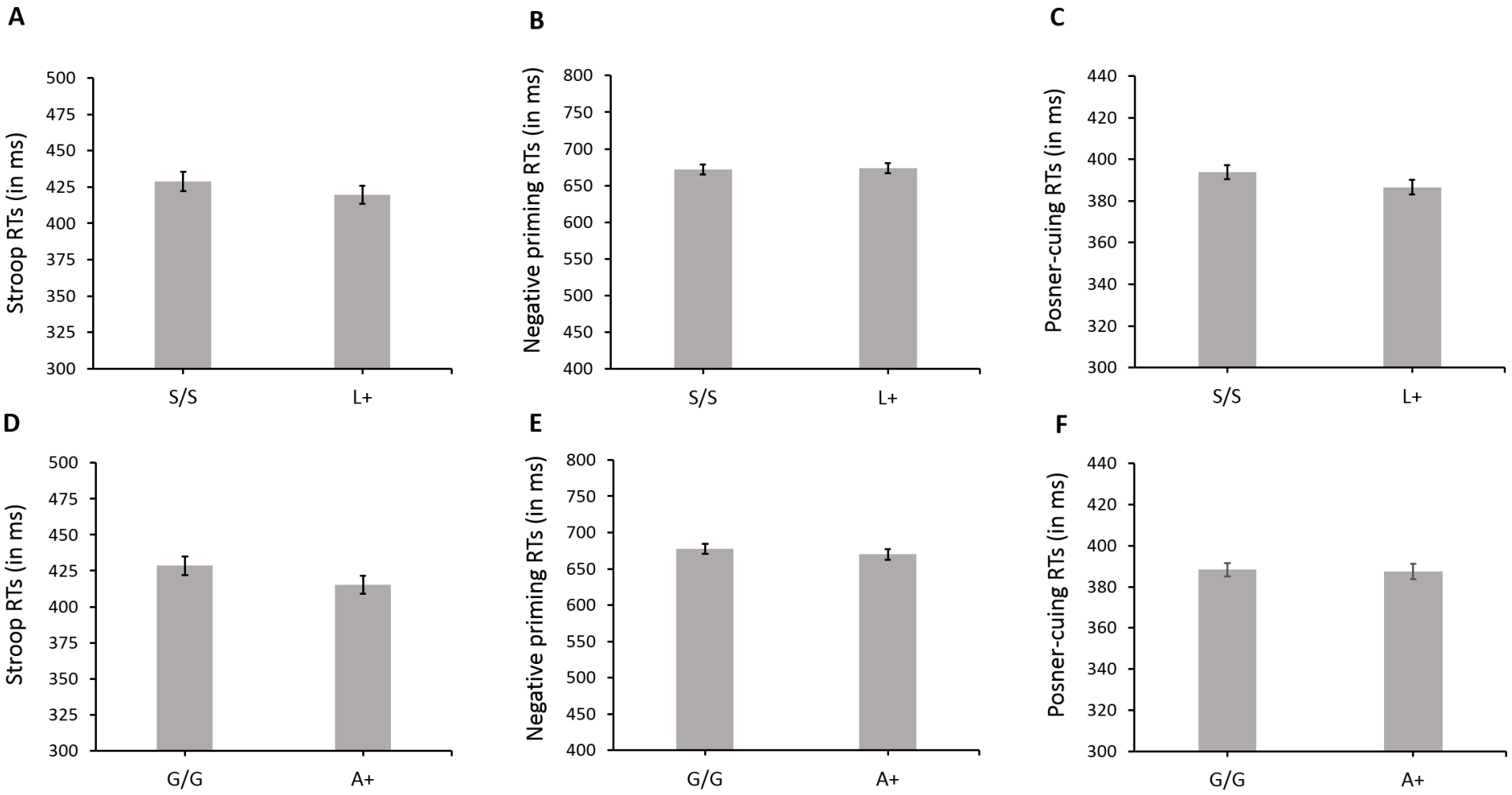


Figure 8.4.: Mean RTs in the Stroop task (A), Negative priming task (B), and Posner-Cuing task (C) as a function of the *CHRNA5* rs3841324 genotype; mean RTs in the Stroop task (D), Negative priming task (E), and Posner-Cuing task (F) as a function of the *CHRNA5* rs16969968 genotype. There were no main effects of the two polymorphisms on the average response speed. The error bars depict the standard error of the mean.

Gender-dependent effects of the CHRNA5 diplotypes on response speed. Next, possible effects of both *CHRNA5* polymorphisms on the average response speed were analyzed by including gender as a covariant in the analyses. The Stroop-RTs were submitted into a 2 (condition: congruent vs. incongruent) x 2 (*CHRNA5* rs3841324: S/S vs. L+) repeated-measures ANOVA with gender as a covariant. There was a significant interaction effect of genotype and gender [F (2, 171) = 13.68, p < .001, η_p^2 = .074]. Both in the congruent and in the incongruent condition, male carriers of the S/S genotype were faster than male carriers of the L allele. In contrast, female carriers of the S/S genotype were slower than female carriers of at least one L allele (see figure 8.5). The Negative priming-RTs were also submitted into a 3 (condition: attended repetition vs. control condition vs. ignored repetition) x 2 (*CHRNA5 rs3841324*: S/S vs. L+) repeated-measures ANOVA with gender as a covariant. The interaction of genotype and gender was also significant in this task [F (1, 172) = 5.36, p = .022, η_p^2 = .030]. In all conditions, male carriers of the S/S genotype were faster than male carriers of at least one L allele. Female carriers of the S/S genotype, on the other hand, were always slower than female carriers of at least one L allele (see figure 8.6). The Posner-Cuing-RTs were submitted into a 3 (condition: valid vs. neutral vs. invalid) x 2 (*CHRNA5 rs3841324*: S/S vs. L+) repeated-measures ANOVA with gender as a covariant. The interaction effect of genotype and gender was not significant in this task [F (1, 172) < 1, p = .334, η_p^2 = .005]. In all conditions, male carriers of the S/S genotype were faster than male carriers of at least one L allele. Female carriers of the S/S genotype, on the other hand, were slower than female carriers of at least one L allele (see figure 8.7). In sum, gender-based analyses of the *CHRNA5* rs3841324 revealed opposite-directed effects on response speed in male and female participants. The presence of the S/S genotype was beneficial in male participants, but disadvantageous in female participants. Male carriers of the S/S genotype were faster in the Stroop task and Negative priming task, while female carriers of the S/S genotype were slower in both tasks. This pattern was also found in the Posner-Cuing task, but the interaction effect between genotype and gender on the average response speed was not significant in this task. This could possibly be due to the task characteristics, as the Posner-Cuing task required only one response key and was on average much faster than the other tasks (it can be speculated that there might have been a floor effect that obstructed the clear emergence of a gender-dependent response speed effect of the *CHRNA5* rs3841324 polymorphism). Next, the Stroop-RTs were submitted into a 2 (condition: congruent vs. incongruent) x 2 (*CHRNA5* rs16969968*:* G/G vs. A+) repeated-measures ANOVA with gender as a covariant. A significant interaction effect between genotype and gender was noted [F (1, 171) = 4.72, p = .031, η_p^2 = .027]. Both in the congruent and in the incongruent condition, male carriers of the G/G genotype were faster than male carriers of the A allele. In both conditions, female carriers of the G/G -genotype were slower than female carriers of at least one A allele (see figure 8.5). The Negative priming-RTs were subsequently submitted into a 3 (condition: attended repetition vs. control condition vs. ignored

repetition) x 2 (*CHRNA5* rs16969968: G/G vs. A+) repeated-measures ANOVA with gender as a covariant. The interaction of genotype and gender on the average response speed was not significant, but showed a trend towards significance [F (1, 172) = 3.03, p = .083, η_p^2 = .017]. In all conditions, male carriers of the G/G genotype were faster than male carriers of at least one A allele, whereas female carriers of the G/G -genotype were slower than female carriers of at least one A allele (see figure 8.6). The Posner-Cuing-RTs were also submitted into a 3 (condition: valid vs. neutral vs. invalid) x 2 (*CHRNA5* rs16969968: G/G vs. A+) repeated-measures ANOVA with gender as a covariant. The interaction effect of genotype and gender on the average response speed in this task was not significant [F (1, 172) < 1, p = .876, η_p^2 < .001]. Male carriers of the G/G genotype were slower than male carriers of at least one A allele in the valid and the neutral condition, but faster in the invalid condition. In all conditions, female carriers of the G/G -genotype were slower than female carriers of at least one A allele (see figure 8.7). In sum, gender-based analyses of the *CHRNA5* rs16969968 revealed opposite-directed effects on response speed in male and female participants. The presence of the G/G genotype was beneficial in male participants, but disadvantageous in female participants. Male carriers of the G/G genotype were faster in the Stroop task and Negative priming task, while female carriers of the G/G genotype were slower in both tasks. In the Posner-Cuing task, this pattern was (descriptively) present for female participants, but not for male participants. There was no significant interaction effect between genotype and gender on the average response speed in this task.

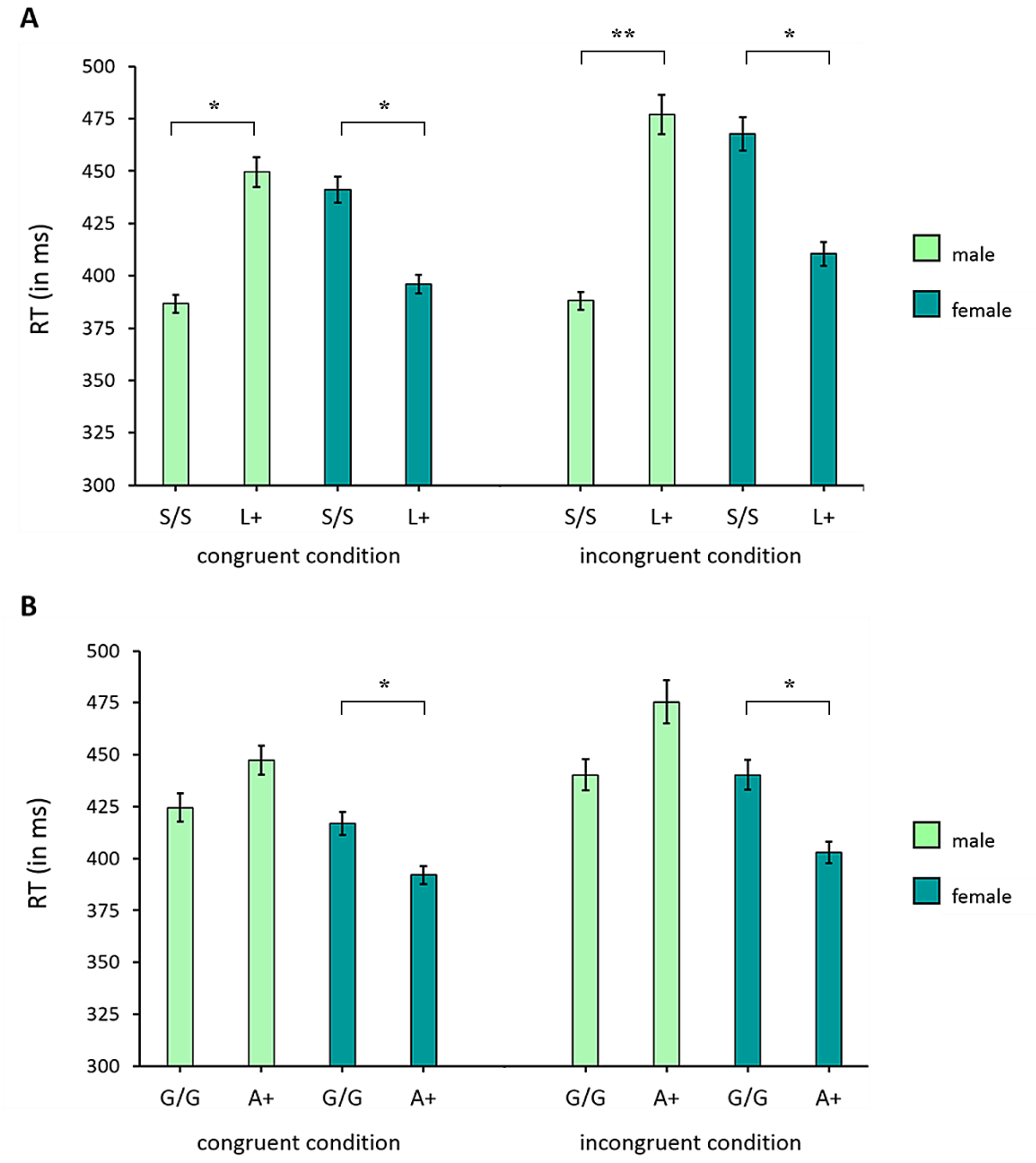


Figure 8.5.: RTs in the Stroop task as a function of gender and of the *CHRNA5* rs3841324 polymorphism (A) and as a function of gender and the *CHRNA5* rs16969968 polymorphism (B). The effects of both polymorphisms were diametrically opposed in men and women. Male carriers of the S/S genotype of the *CHRNA5* rs3841324 polymorphism and of the G/G genotype of the *CHRNA5* rs16969968 polymorphism were faster in the Stroop and Negative priming tasks, while the opposite effects were observed in the female carriers of these genotypes. Error bars depict the standard error of the mean. *Notes:* * $p < 0.05$, ** $p < 0.01$.

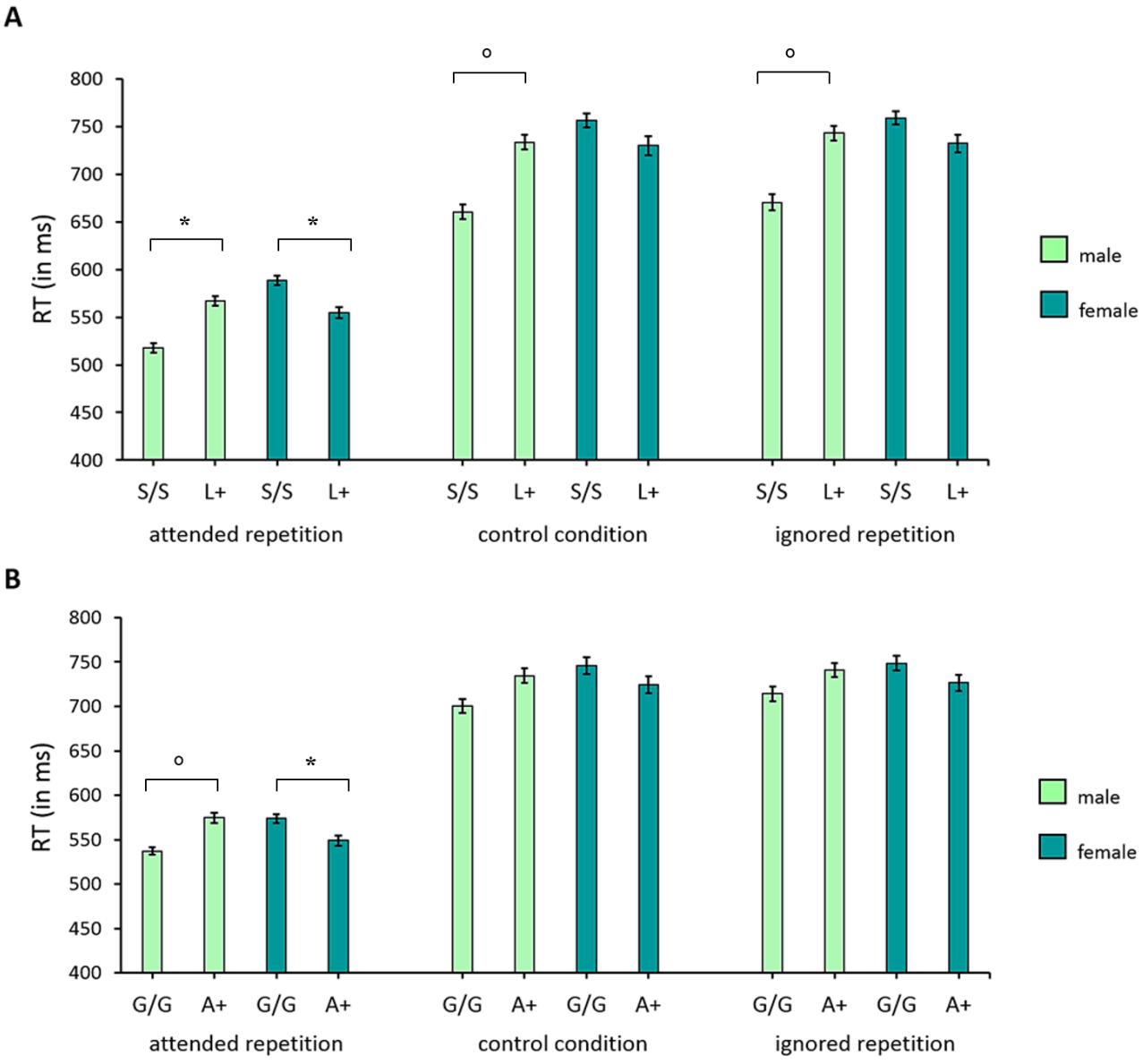


Figure 8.6.: RTs in the Negative priming task as a function of gender and the *CHRNA5* rs3841324 polymorphism (A) and as a function of gender and the *CHRNA5* rs16969968 polymorphism (B). Again, the effects of both polymorphisms were diametrically opposed in men and women. While male carriers of the S/S genotype of the *CHRNA5* rs3841324 polymorphism and of the G/G genotype of the *CHRNA5* rs16969968 polymorphism were faster in the Stroop and Negative priming tasks, female carriers of these genotypes were slower in both tasks. Error bars depict the standard error of the mean. *Notes:* ° $p < 0.10$, * $p < 0.05$.

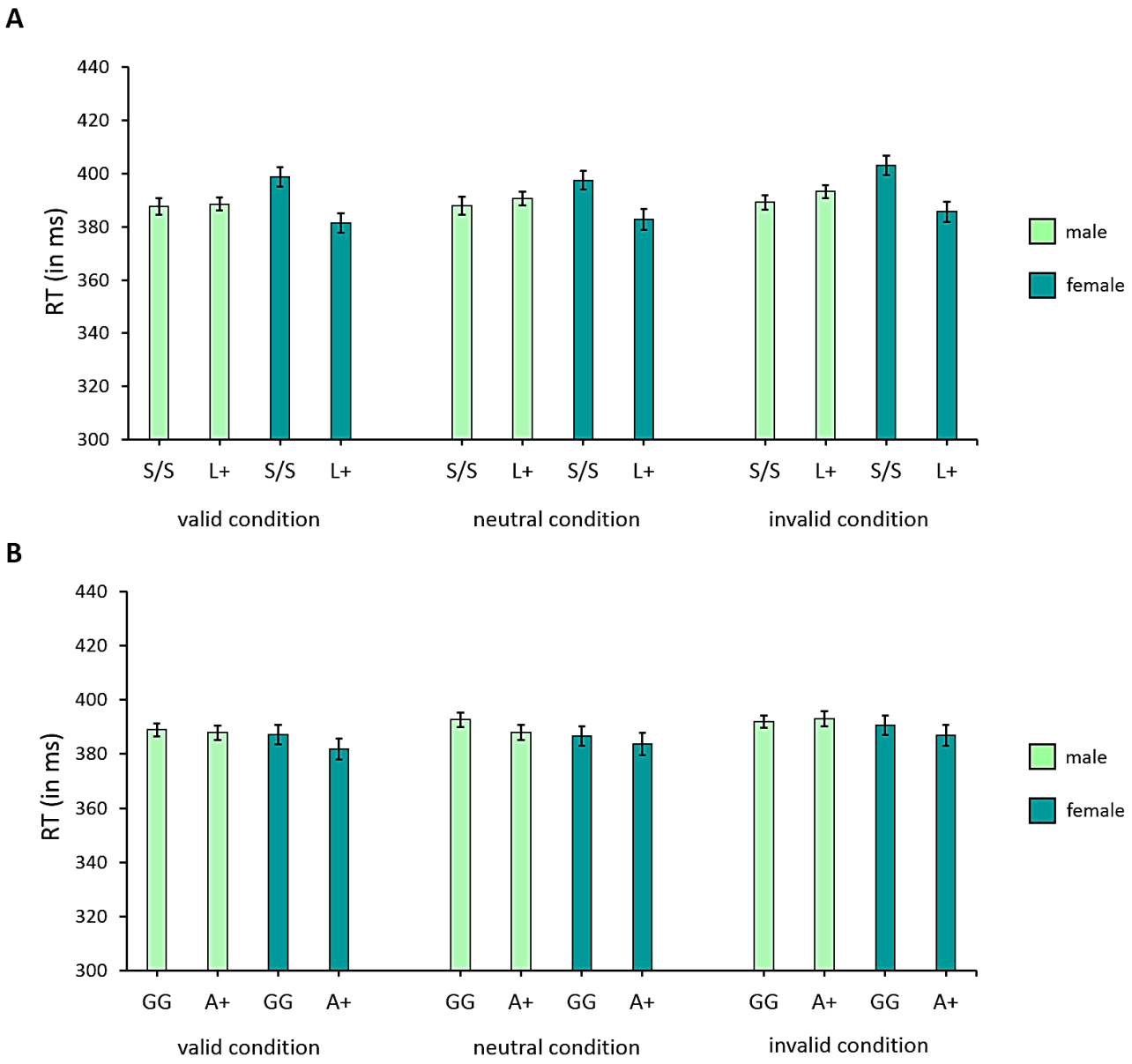


Figure 8.7.: RTs in the Posner-Cuing task as a function of gender and the *CHRNA5* rs3841324 polymorphism (A) and as a function of gender and the *CHRNA5* rs16969968 polymorphism (B). While male carriers of the S/S genotype were faster than male carriers of at least one L allele, and female carriers of the S/S genotype slower than their counterparts, the interaction between the rs3841324 genotype and gender was not significant. Likewise, the interaction between the rs16969968 genotype and gender was also not significant. In fact, male carriers of the G/G genotype were slower than male carriers of the A allele in two of three conditions. Female carriers of the G/G genotype were slower than their counterparts in all conditions. Error bars depict the standard error of the mean.

Diplotype analyses. The mean RTs in the Stroop task were submitted into an ANOVA with the *CHRNA5* diplotypes (*CHRNA5*: S/S_G/G+ vs. S/S_G/G-) as factor and gender as a covariant. A highly significant difference between both groups was noted [F (1, 167) = 13.37, p < .001, η_p^2 = .074]. Male carriers of the S/S_G/G+ diplotype were faster in the Stroop task than their counterparts, while female carriers of the S/S_G/G+ diplotype were slower than their counterparts (see figure 8.8). The mean RTs in the Negative priming task were also submitted into an ANOVA with the *CHRNA5* diplotypes (*CHRNA5*: S/S_G/G+ vs. S/S_G/G-) as factor and gender as a covariant. Again, a significant difference between the two groups was observed [F (1, 168) = 5.13, p = .025, η_p^2 = .030]. Male carriers of the S/S_G/G+ diplotype were faster in the Negative priming task than their counterparts, while female carriers of the S/S_G/G+ diplotype were slower than their counterparts (see figure 8.8). The mean RTs in the Posner-Cuing task were also submitted into an ANOVA with the *CHRNA5* diplotypes (*CHRNA5*: S/S_G/G+ vs. S/S_G/G-) as factor and gender as a covariant. There was no significant effect of the *CHRNA5* diplotypes on the average response speed in this task [F (1, 168) < 1, p = .322, η_p^2 = .006]. The average RTs per task condition, diplotype and gender are listed in table 8.1.

Table 8.1. Mean RTs in ms (standard deviation in brackets) as a function of the CHRNA5 rs3841324 and rs16969968 diplotype groups and gender.

Task	Conditions	male		female	
		S/S_G/G+	S/S_G/G-	S/S_G/G+	S/S_G/G-
		M (SD)	M (SD)	M (SD)	M (SD)
Stroop	congruent	387 (56)	450 (98)	441 (85)	396 (58)
	incongruent	388 (57)	478 (131)	468 (106)	411 (75)
Negative	attended repetition	518 (62)	567 (68)	588 (67)	555 (72)
priming	control	660 (103)	734 (105)	756 (97)	730 (130)
	ignored repetition	670 (112)	742 (105)	759 (92)	732 (123)
Posner-	valid	388 (41)	389 (32)	399 (48)	381 (50)
cuing	neutral	388 (44)	391 (35)	397 (47)	383 (53)
	invalid	389 (35)	394 (32)	403 (48)	386 (50)

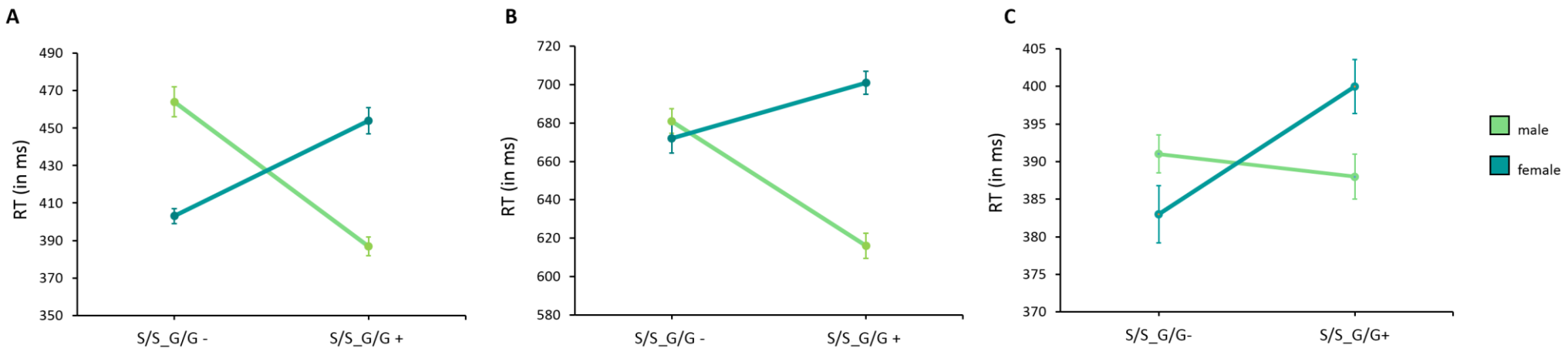


Figure 8.8.: Average RTs in the Stroop task (A), Negative priming task (B), and Posner-Cuing task (C) as a function of the *CHRNA5* diplotypes (S/S_G/G+ vs. S/S_G/G-) and gender. While male carriers of S/S_G/G+ diplotype were faster both in the Stroop task and in the Negative priming task, female carriers of the S/S_G/G+ diplotype were slower both in the Stroop task and in the Negative priming task. The *CHRNA5* diplotypes did not significantly modulate the response speed in the Posner-Cuing task. Error bars depict the standard error of the mean.

Supplementary analysis of gender. To complete the analyses of the *CHRNA5* polymorphisms, it was also examined whether there a main effect of gender on response speed was notable. The mean RTs of each task were entered into a 3 (mean RTs: Stroop vs. Negative priming vs. Posner-Cuing) x 2 (gender: male vs. female) repeated-measures ANOVA. There was no main effect of gender on the average RTs across the three tasks [F (1, 175) = 1.18, p = .279, η_p^2 = .007]. Male participants displayed an average response speed of M = 502 ms (SD = 63 ms), while female participants displayed an average response speed of M = 491 ms (SD = 61 ms).

Error analyses. To test for the occurrence of speed-accuracy tradeoffs, the error rates of the Stroop and Negative priming task were analyzed in relation to the *CHRNA5* polymorphisms and gender. The Stroop errors were submitted into a 2 (condition: congruent vs. incongruent) x 2 (*CHRNA5* rs3841324: S/S vs. L+) repeated-measures ANOVA with gender as a covariant. There was no significant interaction effect of genotype and gender on the average error rate [F (1, 171) < 1, p = .836, η_p^2 < .001]. Next, the Negative priming errors were submitted into a 3 (condition: attended repetition vs. control condition vs. ignored repetition) x 2 (*CHRNA5 rs3841324*: S/S vs. L+) repeated-measures ANOVA with gender as a covariant. The interaction of genotype and gender was also not significant in this task [F (1, 172) < 1, p = .477, η_p^2 = .003]. Next, the Stroop errors were submitted into a 2 (condition: congruent vs. incongruent) x 2 (*CHRNA5* rs16969968*:* G/G vs. A+) repeated-measures ANOVA with gender as a covariant. Again, there was no significant interaction effect between genotype and gender [F (1, 171) < 1, p = .641, η_p^2 = .001]. Subsequently, the Negative priming errors were submitted into a 3 (condition: attended repetition vs. control condition vs. ignored repetition) x 2 (*CHRNA5* rs16969968: G/G vs. A+) repeated-measures ANOVA with gender as a covariant. No significant interaction effect of genotype and gender on the average error rate in this task was observable [F (1, 172) < 1, p = .965, η_p^2 <.001]. In sum, there were no indications that the error rates in the Stroop and Negative priming task were modulated by the *CHRNA5* polymorphisms and gender. Consequentially, speed-accuracy tradeoffs could be ruled out as a possibility.

Summary. Each of the three selected cholinergic variants were found to modulate response speed. T allele homozygotes of the *CHRNA4* rs1044396 polymorphism were fastest in the Stroop task, Negative priming task, and Posner-Cuing task. C allele heterozygotes displayed intermediate RTs, while C allele homozygotes displayed the slowest RTs in each of the three tasks. A significant linear trend of the *CHRNA4* rs1044396 genotype on the average response speed (across all tasks) was noted. Considered as sole factors, neither the *CHRNA5* polymorphisms nor gender modulated the average response speed in the three tasks. Notably, however, interactions between the polymorphisms and gender modulated the response speed in the Stroop and Negative priming tasks. The effects of both

polymorphisms were diametrically opposed in men and women. While male carriers of the S/S genotype of the *CHRNA5* rs3841324 polymorphism and of the G/G genotype of the *CHRNA5* rs16969968 polymorphism were faster in the Stroop and Negative priming tasks, female carriers of these genotypes were slower in both tasks. Likewise, male carriers of the S/S_G/G+ diplotype were faster in the Stroop and Negative priming task than their male counterparts, while female carriers of the S/S_G/G+ diplotype were slower in these tasks than their female counterparts. In regard to response speed, the presence of the S/S_G/G+ diplotype appeared to be beneficial for men, but disadvantageous for women.

Chapter 9

Discussion of the Response Speed Analyses

In this chapter, the analyses of the response speed effects will be discussed. First, the hypotheses and results will be summarized (subchapter 9.1). Subsequently, the impact of the *CHRNA4* rs1044396 polymorphism on response speed be debated (subchapter 9.2). Then, the gender-dependent response speed effects of the *CHRNA5* rs3841324 polymorphism and rs16969968 polymorphism will be discussed in subchapter 9.3.

9.1. Summary of the response speed hypothesis and results

It was the secondary objective of this thesis to examine the effect of the ACh system on response speed. It was expected that each of the three cholinergic polymorphisms would influence the average response speed in the Stroop task, Negative priming task, and Posner-Cuing task – independent of the type of attentional selection that was tapped by the individual tasks. For the *CHRNA4* rs1044396 polymorphism, it was hypothesized that the average RTs would increase linearly with the C allele dosage. For the *CHRNA5* rs3841324 polymorphism and rs16969968 polymorphism, no prior assumptions about the alleles were drawn. These analyses were conducted under an exploratory directive (cp. chapter 4). Each of the three selected cholinergic variants was found to modulate response speed; thus, the general expectation was clearly met. Furthermore, the directed hypothesis for the *CHRNA4* rs1044396 polymorphism could also be corroborated. The T allele homozygotes were fastest in the Stroop task, Negative priming task, and Posner-Cuing task, while the C allele heterozygotes displayed intermediate RTs and the C allele homozygotes were slowest. There was a significant linear trend between the *CHRNA4* rs1044396 genotype and the average response speed across all tasks. While there were no main effects of the *CHRNA5* polymorphisms in regard to response speed, both interacted with gender. Male carriers of the S/S genotype of the *CHRNA5* rs3841324 polymorphism and of the G/G genotype of the *CHRNA5* rs16969968 polymorphism were faster in the Stroop and Negative priming tasks, while female carriers of these genotypes were slower in both tasks. Analogously, male carriers of the S/S_G/G diplotype were faster than their counterparts in the Stroop and Negative priming task, while female carriers of this diplotype were slower than their counterparts in these tasks. The presence of the S/S_G/G diplotype appeared to be beneficial for men, but disadvantageous for women. Thus, both *CHRNA5* polymorphisms were associated with diametrically opposed response speed effects in men and women.

9.2. The CHRNA4 rs1044396 polymorphism and the speed of information processing

The response speed effect of the rs1044396 C allele dosage is no unparalleled finding. In fact, this finding aligns with one reported by Greenwood and colleagues (2005), who had administered a cued visual search task and also found the average response speed to be significantly modulated by this polymorphism. The administered cued visual search task consisted of a conjunction condition (which tapped selective, visuospatial top-down attention) and a pop-out condition (which tapped selective, visuospatial bottom-up attention). They noted both a main effect of the rs1044396 C allele dosage on response speed and an interaction effect with the task condition. In general, the T allele homozygotes were fastest and the C allele homozygotes were slowest in the task, with heterozygotes displaying intermediate RTs (which renders their results strikingly similar to the ones of this dissertation). Furthermore, the disadvantageous effect of the C allele dosage was larger in the more demanding condition (conjunction) than in the less demanding condition (pop-out). The C allele carriers were not only generally slower, but also disproportionately slower than T allele homozygotes in the conjunction condition compared to the pop-out condition. There was no equivalent finding in the present study, as the *CHRNA4* rs1044396 polymorphism did not interact with the conditions of the Stroop task, Negative priming task, or Posner-Cuing task. The C allele homozygotes were always slowest, irrespective of whether the condition was more or less demanding; yet, they were not disproportionately slower in specific conditions. Parasuraman and colleagues (2005) studied the effect of the *CHRNA4* rs1044396 polymorphism on the performance in an endogenous Posner-Cuing task. There were three types of cue validities and two SOA variations, so that the task was comprised of six conditions. Unfortunately, the authors did not report an analysis of the effect of the polymorphism on the average response speed. It was notable, however, that the T allele homozygotes were fastest in all six conditions. In four of the six conditions, the C allele homozygotes were slowest. As this is merely a descriptive observation, it naturally cannot serve to corroborate any hypothesis. Still, this finding provides a tentative further indication of the C allele dosage effect. In this dissertation, the C allele dosage effect was found across a range of different tasks that varied in their degree of complexity and tapped both selective top-down and bottom-up attention. The Posner-Cuing task relied on the detection of the target presence in one of two spatial locations and was dependent on only one response key. The performances in the Stroop task and the Negative priming tasks relied on the identification of a predetermined stimulus property; here, the participants were required to use either two response keys (in the Stroop task) or four response keys (in the Negative priming task). Together with the finding of Greenwood and colleagues (2005), this suggests a generalizability of the response speed effects of the present study. It can be speculated that the *CHRNA4* rs1044396 polymorphism generally impacts on response speed, mostly irrespective of the task type at hand. This leads to several

questions; primarily *where* and *how* the polymorphism influences response speed. More specifically, *where* does the *CHRNA4* rs1044396 polymorphism modulate the stream of information processing? And furthermore, *how* are its effects are implemented on a cellular level? Several distinct processes contribute to response speed: sensory processes (the perception of the stimulus), cognitive processes (the identification of the stimulus and the determination as well as preparation of a suitable response) and motor processes (the execution of the response). The term "information processing" makes the assumption that humans are not merely passive recipients of sensory information, but actively process and alter input. The result of information processing can be implemented in an outwardly observable reaction. Under a simplistic view, the components of response speed can thus be divided into a *cognitive preparation* component (i.e., information processing) and an *execution* component (i.e., the observable action). ACh is present both in the CNS and in the PNS, where the neurotransmitter acts on voluntary muscles (Snyder, 1999). For this reason, genetic variations within the cholinergic system might influence response speed either in the CNS or in the PNS; might impact either on the preparation component or the execution component (or both to varying degrees). The α4 subunit and the α5 subunit only occur in neuronal, CNS-bound nAChRs (Colquhoun et al., 2003). This means that their effects should take place within the CNS, *prior* to the execution of the response. While this assumption narrows the location of the effects of the *CHRNA4* rs1044396 polymorphism down, it does not shed light on which specific process might be modulated. It does not specify whether T allele homozygotes are generally quicker to perceive stimuli, or to identify them, or to determine and prepare the suitable response. Since the *CHRNA4* rs1044396 polymorphism was not associated with the attentional parameters in the Stroop task, Negative priming task, or Posner-Cuing task, it is clear that the speed benefits of the T allele carriers cannot be rooted in the specific attentional processes that are demanded in these tasks. The *CHRNA4* rs1044396 polymorphism did not influence the resolution of the arising interference in the Stroop task and Negative priming task, where attention was voluntarily shifted in accordance with the task instructions; nor did the polymorphism influence the performance in the Posner-Cuing task, where attention was involuntarily shifted due to the salience properties of the stimulus material. The speed advantages of the T allele homozygotes were nearly univocally observable across the different conditions (see figure 8.3). At the moment, there are no studies that pinpoint the possible impact site of the *CHRNA4*-modulated speed effects in the stream of information processing. The tasks that were administered in this dissertation did not allow to disentangle the involved processes. The average RTs that were derived did not reflect when the cognitive preparation ended and was implemented in the response execution, nor did they provide a more fine-grained resolution of the subprocesses within these broader categories. Further studies must be conducted to tackle this issue. In the outlook (chapter 11), one possible approach is contoured. At the moment, the question *where* the *CHRNA4* rs1044396 polymorphism influences the stream of information processing

must go unanswered. It is another important question *how* the polymorphism might modulate response speed. In chapter 7, it has been detailed that ACh can operate in two different modes in the CNS. Specifically, Sarter and colleagues (2014) proposed that there are two largely independent ACh systems; a *deterministic* system that has direct, immediate, event-related effects by impacting on postsynaptic release sites (i.e., contributing to point-to-point neurotransmission) and a *neuromodulatory* system that acts via extrasynaptic volume transmission and results in slow, long-lasting effects. This system requires ACh molecules to diffuse away from their release sites, towards extrasynaptic receptors – which results in large areas being subjected to low concentrations of ACh (Dani & Bertrand, 2007). Diffusion is a comparatively slow process and accordingly, volume transmission has been described as being a "hormone-like" type of transmission (von Bohlen und Halbach & Dermietzel, 2006). Cholinergic effects on the average response speed are much more likely attributable to the neuromodulatory system rather than to the deterministic, fast-acting system[30]. The uniform nature of the response speed effects suggests a stable cholinergic influence across a span of different tasks (cp. figure 8.3), in each case benefitting the T allele carriers. Dani and Bertrand (2007) concluded that cholinergic volume transmission "not only influence[s] the excitability of a neuron but also its set point, which determines the overall electrical and chemical sensitivity and responsiveness" (p. 707-708). They further added that cholinergic volume transmission affects time course and spatial dependence of nAChR signaling. Notably, as the $(\alpha4\beta2)_2$ subtype is the most common nAChR subtype in the brain (Greenwood et al., 2012), genetic variants of the α4 subunit might be quite influential in either system. Since *CHRNA4* rs1044396 is synonymous, it is unclear whether the polymorphism is functional by itself or in a linkage disequilibrium to one or several functional variants in the regulatory or promotor region of the *CHRNA4* gene (as has been suggested by Markett et al., 2011). Winterer and colleagues (2011) reported that the C allele is linked to a higher number of nAChRs in a high-affinity state. Receptors in a high-affinity state require a lower ligand concentration to be occupied and to trigger a physiological response (Greenwood et al., 2012). The finding of Winterer and colleagues (2011) has to be classified as preliminary and requires replication, however. Effects of the *CHRNA4* rs1044396 polymorphism on response speed have not always been found. Kikuno and colleagues (2013) reported no effects on the average response speed in a rapid scene categorization task, while Espeseth and colleagues (2006) found no such main effect in a cued visual discrimination task. In the former study, a sample of $n = 100$ participants was analyzed – a rather small, which might have resulted in too little power to detect the response speed effect. Kikuno and colleagues (2013) analyzed a sample

[30] While the *CHRNA4* rs1044396 polymorphism exerted no effects on attention in the present study (cp. chapter 7), this variant has been repeatedly linked to attentional parameters, especially to endogenous shifts of visuospatial attention (Greenwood et al., 2012). In this regard, the existence of the *CHRNA4* rs1044396 effects on attention – possibly realized via the deterministic ACh system – cannot be precluded. The design of the present study might simply not have been best suited to highlight and illustrate such effects.

of *n* = 230 participants, of which roughly half were 53 to 64 years old and half were 65 to75 years old. In regard to this age setup, the sample is certainly unusual – the majority of *CHRNA4* rs1044396 studies so far have been focused on young adults. The relation between mental speed and age is curvilinear and runs parallel to the development and decline of fluid intelligence (Sheppard & Vernon, 2008). The peak of both response speed and fluid intelligence is located within young adulthood (Sheppard & Vernon, 2008; Horn & Cattell, 1967). The reduction of response speed might possibly drive the decline of fluid intelligence, or could simply be driven by the same underlying factors (Sheppard & Vernon, 2008). In samples of young adults, lifestyle-related impairments of cognition and health problems should be a minimal issue. For these reasons, the effect of genetic variants on response speed should be much more apparent in early adulthood than in the later stages of life[31]. Insofar it is perhaps not surprising that the *CHRNA4* rs1044396 polymorphism was not associated with the average response in a sample of middle-aged and older participants. In sum, it is not yet clear which component of information processing is modulated by the *CHRNA4* rs1044396 polymorphism. The response speed effects are most likely implemented through the neuromodulatory ACh system, however, and could possibly be driven by the affinity state of the α4-containing nAChRs.

9.3. The CHRNA5 polymorphisms and the speed of information processing

The effects of the *CHRNA5* rs3841324 polymorphism and *CHRNA5* rs16969968 polymorphism on response speed were diametrically opposed in men and women. Male carriers of the S/S genotype of the *CHRNA5* rs3841324 polymorphism and of the G/G genotype of the *CHRNA5* rs16969968 polymorphism were faster than their counterparts in the Stroop and Negative priming task. In contrast, female carriers of these genotypes were slower than their counterparts in the Stroop and Negative priming task. This pattern of results was descriptively apparent in the Posner-Cuing task as well, but both polymorphisms failed to modulate the response speed here (irrespective of whether gender was considered as a factor). As expected, the *CHRNA5* polymorphisms were in a strong linkage disequilibrium with each other. All carriers of the S/S genotype of the rs3841324 polymorphism were also carriers of the G/G genotype of the rs3841324 polymorphism[32]. Analyses on a diplotype basis demonstrated that male carriers of the S/S_G/G diplotype outperformed male non-carriers in the Stroop and Negative priming task, while female carriers of this diplotype were disadvantaged and

[31] Behavioral variance can be explained to a larger extent by genetic factors if the environmental factors are more similar. In early adulthood, many of the factors that should later contribute to response speed differences are not yet apparent.

[32] But not vice versa – about 60 % of the carriers of the *CHRNA5* rs16969968 G/G genotype did not simultaneously carry the *CHRNA5* rs3841324 S/S genotype.

displayed slower RTs than female non-carriers. Wei and colleagues (2011) reported that the S/S_G/G diplotype had a protective health effect for women (reducing the risk of developing lung cancer), but no such effect for men. *CHRNA5* is part of a cluster of nAChR genes that appear to be relevant for nicotine consumption and dependency (Bierut et al., 2008), presumably because variants of these genes might change the effectiveness and impact of ACh and cholinergic agonists. In this regard it is notable that accessory α5 subunits provide a greater sensitivity for the nAChR activation (Kuryatov et al., 2008). In a sample of almost three hundred participants, Breetvelt and colleagues (2014) found the A allele of the *CHRNA5* rs16969968 polymorphism to be associated with a better memory function and a higher IQ, particularly in males. This finding is another indication of the relevance of gender for the α5 subunit of the nAChR. In this dissertation, the diametrical *CHRNA5* effects canceled each other out and resulted in a non-significant net-effect when response speed was averaged across the gender categories. Both *CHRNA5* polymorphisms have so far only been sparsely studied in regard to cognitive parameters, but judging from the existing findings it is of the utmost importance to include gender as a covariant in *all* of analyses of these polymorphisms. If gender is omitted, one runs the risk of overlooking quite prominent effects. The most common nAChR in the brain is the $(\alpha4\beta2)_2$ subtype (Greenwood et al., 2012) and the α5 subunit is present in up to 40 % of these receptors (Kuryatov et al., 2008; Mao et al., 2008). Consequently, the α5 subunit is widely distributed in the CNS. Since the α5 subunit is an accessory subunit and as such has a modulating function (Kuryatov et al., 2008), it might fine-tune the actions of a significant portion of the nAChRs in the CNS. Nicotine is the nAChR agonist that has been studied most extensively. Notably, gender effects in in nicotine consumption and addiction are no new discovery. It has been reported that the chronic administration of nicotine led to an up-regulation of the nAChRs in male rats but not in female rats (Koylu, Demirgören, London, & Pögün, 1997) and that nicotine replacement therapy is less effective in women (Wetter et al., 1999). In fact, nicotine underlies a faster metabolism in women (Benowitz, Lessov-Schlaggar, Swan, & Jacob, 2006) and women seem to have more trouble to quit smoking (Benowitz & Hatsukami, 1998). There are also indications that the urge to smoke fluctuates with the menstrual cycle and is highest during the latter luteal phase (Benowitz & Hatsukami, 1998), when the hormone progesterone has reached its peak concentration (Johansson, 1969). The findings of decades of nicotine research suggest that nicotine exerts markedly different effects in men and women. This is certainly no trivial finding, as nicotine as a cholinergic agonist reaches about 80 % of the efficacy of ACh molecules for the nAChR activation (Nelson et al., 2003). Thus, a significant portion of the nicotinic ACh system might underlie gender-specific modulations. It can be argued that these gender differences are partly driven by two closely intertwined factors: the a5 nAChR subunit and the hormone progesterone. Koylu and colleagues (1997) suggested interactions between this sex hormone and the nAChRs to cause the gender-dependent nicotine effects. Wei and colleagues (2011) also proposed that the gender-

dependent effects of the *CHRNA5* rs3841324 polymorphism and *CHRNA5* rs16969968 polymorphism might be caused by interactions between the α5 subunit of the nAChR and progesterone. What are the correlates of the *CHRNA5* diplotypes? The S allele of the rs3841324 polymorphism was linked to higher levels of *CHRNA5* mRNA expression (Wang et al., 2009), while the G allele of the *CHRNA5* rs16969968 polymorphism was linked to an elevated response of the nACh receptor to an agonist (Bierut et al., 2008). Bierut and colleagues (2008) elaborated that α4β2α5 receptors that contained the rs16969968 G allele displayed a maximal response that was more than two times higher than the one displayed by α4β2α5 receptors that did not contain the G allele. In sum, it can be speculated that the S/S_G/G diplotype signifies the allele combination of both polymorphisms that results in the greatest number of nAChRs and the greatest affinity of these receptors. This diplotype, in other words, could be associated with the comparatively highest level of cholinergic transmission. A high rate of cholinergic transmission appears to be beneficial for men, who have much lower levels of endogenous progesterone. In contrast, a high rate of cholinergic transmission appears to be disadvantageous for women, who have much higher – albeit also strongly fluctuating – levels of progesterone. It then stands to reason that male carriers of the S/S_G/G diplotype are more similar to the female *non-carriers* of this diplotype than the female carriers; this would explain why these groups show both response speed advantages and attentional advantages (cp. chapter 6). Progesterone and the α5 subunit of the nAChR seem to be closely intertwined on a cellular level. A putative progesterone responsive element has been located in the promotor of the *CHRNA5* gene, for example (Gangitano, Salas, Perez, & De Biasi, 2009). It has also been shown that progesterone can increase the expression levels of α5 mRNA both *in vivo* and *in vitro* (Gangitano et al., 2009) and can noncompetitively inhibit nAChRs (Valera, Ballivet, & Bertrand, 1992). Traditionally, progesterone has been categorized as a sex hormone and firmly subsumed under the reproductive system. The hormone is produced in the ovaries and in the adrenal glands (Baulieu & Schuhmacher, 2000), and its level are known to fluctuate within the menstrual cycle (Schandry, 2006). Progesterone is present in men as well, albeit at lower levels, and influences the spermatogenesis here (Lue et al., 2013). The role of progesterone is not limited to reproductive functions, however. Baulieu and Schuhmacher (2000) stated that "sex steroids influence neuronal activity within large parts of the nervous system [and] exert a variety of effects that are not necessarily related to reproduction," (p. 605). Progesterone, for example, targets multiple regions in the CNS, among them the hippocampus and cortex (Brinton et al., 2008). Progesterone receptors are widely expressed in the brain (Brinton et al., 2008) and progesterone itself is present both in neurons and glia cells (Baulieu & Schuhmacher, 2000). When progesterone is synthesized de novo in the brain, it is labeled a neurosteroid instead of a hormone (Baulieu & Schuchmacher, 2000). Progesterone molecules are small, hydrophobic and capable of passing the blood-brain barrier (Stein, 2011). Baulieu and Schuhmacher (2000) speculated that the local synthesis of progesterone in the brain may serve to

compensate for drops in progesterone levels due to the menstrual cycle or ageing; in males, the de novo synthesis might be necessary to compensate for the much lower peripheral production levels of progesterone. In many regards, the progesterone system in the brain is yet unchartered territory. A radioligand that is specific for progesterone and can be used in PET studies has only recently been determined (Lee et al., 2010) and utilized in a human sample (Dehdashti et al., 2012). The role of progesterone in the living brain has not yet been investigated via this route, however. In this regard it is relevant that the subjective effects of cocaine in women appear to be partly dependent on the menstrual cycle (Evans, Haney, & Foltin, 2002), specifically on the endogenous progesterone levels (Evans & Foltin, 2005). Cocaine is known to exert many of its reward-related properties by increasing the DA transmission in the nucleus accumbens of the striatum (Ikemoto & Panksepp, 1999). The findings of the Evans group demonstrate that peripherally produced progesterone can exert effects on the processing of psychoactive drugs in a subcortical structure of the CNS. Thus, the stream of information processing might be modulated both by progesterone that has been produced in the brain *and* by progesterone that has been produced outside of the brain. The vastly differing levels of the reproductive progesterone synthesis in women and men might be directly related to gender differences in cognitive functioning. The α5 subunit is most likely one avenue through which progesterone influences the inner workings of the CNS. In future studies of the *CHRNA* rs3841324 polymorphism and *CHRNA5* rs16969968 polymorphism, the role of progesterone should be closely monitored and investigated (cp. chapter 11 for an elaboration on this point). In sum, gender should always be considered as a factor when studying the *CHRNA5* rs3841324 polymorphism and *CHRNA5* rs16969968 polymorphism. Interactions with progesterone might be the driving factor behind the observed gender differences in this dissertation. The diametrical nature of the *CHRNA5* effects means that no gender group was inherently faster in the administered tasks. Naturally, the questions arise *where* and *how* the stream of information processing was influenced by the *CHRNA5* polymorphisms. As for the first question, that is an issue that must be tackled in further studies (cp. chapter 11) and at the moment is similarly uncertain as in the case of the *CHRNA4* rs1044396 polymorphism. In regard to the second question, it is assumed that the neuromodulatory, comparatively more static ACh system is at the core of the *CHRNA5* response speed effects; here influenced via the presumably higher cholinergic transmission in the case of the S/S_G/G diplotype and the interactions of the α5 subunit with progesterone.

Chapter 10

General Discussion

This chapter is focused on a general discussion of the results of the present study. First, the overall hypotheses and results will be summarized (subchapter 10.1). Then, the underlying mechanisms of the gender-dependent attention and speed effects of the *CHRNA5* polymorphisms will be further discussed (subchapter 10.2). Last, the limitations of the study will be detailed and different ways of improvement will be suggested (subchapter 10.3).

10.1. Summary of the overall hypotheses and results

It was the primary objective of this thesis was to examine whether selective top-down and bottom-up attention are differentially modulated by the dopaminergic neurotransmitter system and the cholinergic neurotransmitter system. To this end, two dopaminergic variants (*COMT* Val158Met and the *DAT1*) and three cholinergic variants (*CHRNA4* rs1044396, *CHRNA5* rs3841324, and *CHRNA5* rs16969968) were studied in relation to the participant's performance in a Stroop task, Negative priming task, and Posner-Cuing task. On the basis of the dissociation hypothesis (Noudoost & Moore, 2011a), it was assumed that the *COMT* Val158Met polymorphism and the *DAT1* polymorphism would influence the performance in the top-down driven Stroop and Negative priming tasks, but not (equally) in the bottom-up driven Posner-Cuing task. Likewise, it was assumed that the three cholinergic variants would influence the performance in the bottom-up driven Posner-Cuing task, but not (equally) in the top-down driven Stroop and Negative priming tasks. By and large, the assumptions were confirmed in regard to the dopaminergic system. Both the *COMT* Val158Met polymorphism and the *DAT1* polymorphism did indeed modulate the performance in the Stroop task in the expected fashion, but did not influence the performance in the Posner-Cuing task. Thus, the present study provides indications that the dopaminergic system is especially relevant for processes of selective top-down attention. In regard to the cholinergic system, however, the results provided no indications for the dissociation hypothesis. None of the three cholinergic polymorphisms modulated the performance in the Posner-Cuing task, and thus none of them were found to be especially relevant for processes of selective bottom-up attention. As a second objective of this thesis, it was tested whether variants of the cholinergic system modulated response speed. To this end, the effects of the cholinergic polymorphisms on the average RTs in the three tasks were studied. All three polymorphisms influenced the general response speed. Specifically, a linear relationship between the C allele dosage of the *CHRNA4* rs1044396 polymorphism and the average speed was noted; T allele homozygotes were overall fastest, while C allele homozygotes were overall slowest. The *CHRNA5* polymorphisms interacted with gender on response speed, exerting diametrically opposed effects in men and women. While male carriers of the S/S genotype of the *CHRNA5* rs3841324 polymorphism and of the G/G genotype of the *CHRNA5* rs16969968 polymorphism were faster in the Stroop and Negative priming

tasks, female carriers of these genotypes were slower in both tasks. In sum, there were strong indications that the cholinergic system impacts on response speed (cp. chapter 9). At the moment, it is still up to debate whether the observed effects are attributable to the neuromodulatory or the deterministic ACh system. This point will be further discussed in the next subchapter.

10.2. One system or two systems?

The *CHRNA5* rs3841324 polymorphism and by the *CHRNA5* rs16969968 polymorphisms modulated both selective top-down attention and response speed in interaction with gender, causing diametrically opposed effects in men and women. Male carriers of the S/S_G/G diplotypes were both faster in the Stroop task, Negative priming task, and Posner-Cuing task, and displayed smaller Stroop effects. In contrast, female carriers of the S/S_G/G diplotypes were both slower in the Stroop task, Negative priming task, and Posner-Cuing task, and displayed larger Stroop effects. The S/S_G/G diplotype appears to be allele combination of both polymorphisms that leads to the comparatively highest level of cholinergic transmission. It can be speculated that a neurosteroid like progesterone – greatly varying in its levels in men and women – might cause the gender-dependent effects (cp. chapter 9). In this dissertation, it has been proposed that the effects of the *CHRNA5* polymorphisms on attention are likely exerted through the fast-acting, deterministic ACh system, while their effects on response speed are exerted through the slower, neuromodulatory ACh system. Likewise, it has been proposed that the effects of the *CHRNA4* rs1044396 polymorphism on response speed are exerted through the neuromodulatory system. The response speed effects occurred independent of task interactions, with the three cholinergic polymorphisms exerting almost uniform effects across the individual tasks and task conditions. Dani and Bertrand (2007) stated that the neuromodulatory ACh system influences the "set point, which determines the overall electrical and chemical sensitivity and responsiveness [of neurons]" (p. 708). Likewise, Agnati and colleagues (1995) stated that this type of intercellular communications "[allows] a global mode of operation for the CNS. In fact, [volume transmission] allows the setting of 'entire provinces of the CNS' […] to the appropriate functional state so that they work as a unit" (p. 712). In short, volume transmission as a comparatively slow-acting and stable type of intercellular communication is ideally suited to pre-set or configure large populations of neurons. This type of influence might be called an *offline* influence. It can be speculated that the neuromodulatory system modulates the framework in which information processing takes place. The framework might determine if information processing is more or less efficient and thus, more or less fast. Yet, the neuromodulatory system is too slow to actually exert immediate effects; to not only build the frame, but also interact with the subprocesses that take place within its edges. In contrast, the effect of the *CHRNA5* polymorphism on selective top-down attention are assumed to rely on the

deterministic system. ACh can modulate the detection and possibly also selection of targets; this type of influence relies on brief bursts of cholinergic activity that take place in a millisecond range (Sarter et al., 2014). The underlying neuronal mechanism is point-to-point neurotransmission, which must operate *online,* directly modulating the way the information is processed and the task is carried out. Both *CHRNA5* polymorphisms interacted with the conditions of the Stroop task. Male carriers of the S/S_G/G diplotype were less affected by conflicting properties of the Stroop stimuli than their male counterparts, while female carriers of this diplotype were more affected than their female counterparts. Yet, the notion of two systems being responsible can be challenged. Is it strictly necessary to assume that the observed effects are caused by the two ACh systems in the CNS? Is it not also possible that both the speed of information processing and the attentional processing are favorably modulated by beneficial or detrimental *offline* settings? Naturally, this question cannot be fully answered in this dissertation. However, a tentative indication for this line of argumentation seems to be that the *CHRNA5* effects on attention and speed effects were quite uniform, almost mirroring each other[33]. In contrast, the C*HNRA4* rs1044396 polymorphism exerted clear effects on response speed, but no effects on attention. This finding contraindicates that the notion that the neuromodulatory system might be responsible for modulating both the response speed and the attention effects. If this system were responsible for pre-setting the framework both for the general speed of information processing and for the speed of the interference resolution (the latter being required in the Stroop task), the *CHRNA4* rs1044396 polymorphism should also have modulated the attentional performance in the Stroop task. This was not the case; the polymorphism did not influence the Stroop effect. Thus, it is more plausible that *two* ACh systems are at the core of the observed effects. Most likely, nAChRs that contain the α4 subunit and α5 subunit are present in both systems and the *CHRNA5* polymorphisms interact with gender in the same fashion in both cases. While largely autonomic, the neuromodulatory and deterministic ACh system can also interact. Sarter and colleagues (2014) reported that elevated levels of ACh neuromodulation increased the likelihood of cholinergic transients. Interestingly, both system can also be affected by global regulatory signals that fine-tune the efficacy of entire networks (Agnati et al., 1995). More research is certainly required to understand the connections between both systems and to disentangle their effects, but at the moment it is indeed expected that the *two* systems caused the observed effects of the cholinergic variants on attention and response speed.

[33] Both the *CHRNA5* rs3841324 polymorphism and the *CHRNA5* rs16969968 modulated the Stroop effect, but not the NP effect or the Posner effect. The Stroop effect was the largest effect out of these three; naturally this should have facilitated the emergence of a significant cholinergic modulation.

10.3. Limitations of the study

The Dot Probe task. Regarding the tasks, the study has several limitations. First and foremost, the Dot Probe task did not yield the expected Dot Probe effect. The participants were faster in invalid trials (where the target appeared at the location of a preceding neutral stimulus) compared to valid trials. This led to the removal of the Dot Probe task from all subsequent analyses. The typical Dot Probe effect consists in faster responses to neutral targets that appear at a location where an emotional stimulus has previously been presented (Wenzel, 2012). However, this "typical" effect is especially noticeable if *disorder-specific* cues are used in a clinical sample of participants with that disorder (Wenzel, 2012). In fact, sometimes – as in this study – the opposite results are reported in samples of healthy participants (Wenzel, 2012). The inconsistencies of the Dot Probe effect have been systematically examined by Schmukle (2005). Schmukle tested the retest reliability and internal consistency of word-based and picture-based versions of the Dot Probe task over a time period of one week. The retest reliability was estimated by correlating the same key indices over a one-week interval. The internal consistencies of the Dot Probe versions were estimated via the split half method and via Cronbach's alpha. Schmukle reported that the utilized Dot Probe tasks were internally inconsistent and unstable. It made no difference whether the Dot Probe task was word-based or picture-based or whether the utilized threat stimuli were of a socially threatening or physically threatening nature. Schmukle summarized that the Dot Probe tasks neither measured a trait-like variable (which would require both stableness and internal consistency), nor a state-like variable (which would require internal consistency). Thus, it is not surprising that the reported Dot Probe effects in the literature are so inconsistent. As Schmukle stated, the Dot Probe tasks utilized by him measured only error variance and therefore precluded the possibility of an effect replication. He concluded that the Dot Probe task is "completely unreliable" and not suited "to measure the attentional allocation in non-clinical samples" (p. 595). In the present study, the sample consisted primarily of students and thus was non-clinical. Therefore, sizeable influences of existing disorders on the performance in the Dot Probe task were unlikely. In the future, it would be advisable to exclude the Dot Probe task from all studies that are aimed at the examination of cognitive functions in healthy participants. The removal of the Dot Probe task from this study equaled the loss of the second task of selective bottom-up attention. Therefore, the construct of selective bottom-up attention was only tapped by the Posner-Cuing task. Ultimately, this prevented the evaluation of the convergent validity of this approach. Concerning the measurement of traits, Campbell and Fiske (1959) stated that each employed test is a *trait-method unit,* where "the systematic variance among test scores can be due to responses to the measurement features as well as responses to the trait features" (p. 81). They reasoned that "more than one trait as well as more than one method must be employed in the validation process […] in order to estimate

the relative contributions of trait and method variance" (p. 81). The present study was aimed at disentangling the contributions of DA and ACh on selective top-down and bottom-up attention. The employed design allowed the representation of the dopaminergic and cholinergic system by several variants and, at the same time, allowed the representation of the selective top-down and bottom-up system by several tasks. Thus, the design was aimed at the evaluation of convergent validity (by testing whether all dopaminergic variants affected top-down attention, while all cholinergic variants affected bottom-up attention) and the evaluation of discriminant validity (by testing whether the dopaminergic variants did *not* affect bottom-up attention, while the cholinergic variants did *not* affect top-down attention). The removal of the Dot Probe task from the analyses was unfortunate insofar as it reduced the generalizability of the Posner-Cuing results. All of these results would have been strengthened in their interpretability if the Dot Probe task would have served as a reliable comparison task. In future studies, it might be worthwhile to try to replicate the effects of the selected dopaminergic polymorphisms in other tasks, for example in a visual search task (where it would be expected that the polymorphisms influence the performance in the conjunction condition, but not in the pop-out condition).

The Negative priming task. In regard to the Negative priming task, several aspects are worth a critical discussion. While both the PP effect and the NP effect were observed, the NP effect was significant only in a one-tailed test and was of a small effect size. In fact, a sizeable fraction of the participants did not display a NP effect at all. Since the NP effect is not as robust as the Stroop effect (Frings et al., 2014; Siegrist, 1997), it is not surprising that the effect was not present in some participants. A rate of about 40 % non-responders – as in this study – is excessive, however. This high rate might have been due to specific task and sample characteristics. A control variable was found to contribute to the task performance, explaining some of the error variance. Particularly, the ability to *touch-type* explained about 10 % of the variance that was observed in this task. About a quarter of the participants (n = 46) indicated that they were able to touch-type according to the German standard system of touch-typing. In this system, the left middle finger is placed on the key "D", the left index finger is placed on the key "F", the right index finger is placed on the key "J", and the right middle finger is placed on the key "K". This is exactly the finger placement that was required in the Negative priming task, where exactly these four letters had to be identified. The differences between the touch-typing participants and the rest of the sample were quite pronounced. The touch-typing participants were overall faster in the NP task; in addition, an overall larger percentage of them displayed the NP effect (74 % as compared to 51 % of the other participants). In particular, the touch-typing participants displayed larger NP effects and smaller PP effects than their counterparts. In the subsample of touch-typing participants, the comparison of the control condition and ignored repetition led to a significant

NP effect in a two-tailed test. If the touch-typing participants were entirely excluded from the analysis of the whole sample, no NP effect was apparent anymore. Thus, the observed NP effect in this study appeared to have been partly driven by the person-specific factor of the ability to touch-type. As can be seen in table 10.1., the participants that were unable to touch-type benefitted from the attended repetition (where the target was repeated) to a disproportionately larger degree than the touch-typing participants. If the prime target was not repeated as the probe distractor, the responses of these participants were slower and not discriminative of the condition (hence the lacking NP effect in this subset of the sample).

Table 10.1. Negative priming RTs in ms (standard deviations in the brackets) as a function of the ability to touch-type

	attended repetition	control condition	ignored repetition
	M (SD)	*M (SD)*	*M (SD)*
Touch-typing	525 (61)	651 (93)	665 (99)
No touch-typing	571 (70)	755 (116)	756 (111)

It can be assumed that the participants who were unable to touch-type were more strongly affected by a response change. The smaller NP effects in this group might have be due to participants trying to retrieve the information about the key position and thus being obstructed in focusing solely on the execution of the response. This aligns with one of the most prominent theoretical accounts of the NP effect. It is mostly consensus that not only inhibitory mechanisms, but also retrieval processes contribute to the NP effect (Tipper, 2001; Frings et al., 2014). The Episodic Retrieval account assumes that retrieval processes are at the core of the NP effect. Specifically, it is assumed that the probe target in the ignored repetition serves as a retrieval cue for the prime episode (Neill & Valdes, 1992; Neill, Valdes, Terry, & Gorfein, 1992; for an overview, see Mayr & Buchner, 2007, and Fox, 1995). The retrieved information contains the responses that were required for the stimuli, among them the *do-not-respond* information that is linked to the prime distractor. It is assumed that the incompatibility of the probe instruction *(respond to the stimulus)* is at conflict with the prime instruction *(respond to the stimulus)* and results in a time-consuming solution processes that is ultimately reflected in the NP effect (Mayr & Buchner, 2007). Mayr and Buchner stated that “the successful retrieval of the prime episode is a necessary precondition for the [NP] effect to occur”, and further added that the NP effect

should increase dependent on the probability of the prime episode retrieval. Conway and colleagues (1999) examined the influence of the memory load on the performance in a Negative priming task. The experiment consisted of five-trial groups. After each trial, a word (experiment 1) or polygon (experiment 2) was presented to the participants. After the fifth trial, the participants had to indicate whether the presented word or polygon matched one of the previously presented stimuli. Thus, the memory load increased linearly within the trial groups. Conway and colleagues reported that NP effects were only observable at a memory load of zero, i.e. the first trial. The experiments demonstrate that the NP effect can be eliminated by the memory load of a secondary task (in dependence of the individual level of performance). Conway and colleagues (1999) concluded that the processes that underlie NP effect are resource-dependent. In the present study, it is likely that the participants who were not able to touch-type were not as confident in the placement of their fingers. As a consequence, they might have had to retrieve the respective information frequently. For example, if the middle letter in a probe display was "K", the appropriate response *(press the key "K" with the right middle finger)* was probably not as readily accessible for them as for the touch-typing participants. In this way, the ability to touch-type could have influenced the performance in the Negative priming task in the present study. As several theories have branched off from the Episodic Retrieval account, different explanations for the impact of the touch-typing ability are possible. For example, the theory of *Transfer-(In)appropriate Processing* (short, the TIP/TAP theory) also implies that a particular stimulus restores the processing operations that have previously been applied to it (Leboe, Whittlesea, & Milliken, 2005; for an overview, see Frings et al., 2014). In the attended repetition, this processing transfer would be *appropriate* (TAP) and lead to RT benefits. In the ignored repetition, this processing transfer would be *inappropriate* (TIP) and lead to interference and a reduction of response speed. The TIP/TAP theory focuses on the similarity between the encoding and retrieval of phases and is thus more specific than the Episodic Retrieval account, which relies on the retrieval of the relatively vague *respond* tags and *do-no-respond* tags (Frings et al., 2014). Many of the assumptions of the Episodic Retrieval account and the TIP/TAP theory overlap and the TIP/TAP theory would also postulate that a high memory load would interfere with the inappropriate processing transfer in the ignored repetition. Whatever theoretical approach might be best suited for explaining the impact of touch-typing, it is clear that the utilized NP task was too difficult for a part of the sample. The lack of familiarity with the finger placement seemed to obstruct the occurrence of the NP effect in some participants. There are several ways this could have been circumvented. For one, the block of training trials could have been prolonged to familiarize all participants with the key positions. However, this probably would not been an optimal solution, since it is unlikely that the performance level of the non-touch-typing participants could have been elevated to the performance level of the touch-typing participants, where the skill is probably hugely automatized. Additionally, and given that it was primarily a student sample, the non-

touch-typing participants probably also had an established system of finger positioning and movement that they were familiar with[34]. This system might also have been automatized to a large degree, so that their individual system and the finger placement system that was required in the study probably interfered. Shiffrin and Schneider (1977) examined the effect of automaticity by letting participants judge whether a stimulus in a display-set matched previously learned stimuli of a memory-set. After over 2000 learning trials, the task was reversed. The former distractors became the targets and the former targets became the distractors. While the participants had reached a hit rate of about 90 % after 900 trails of the original task, they required about 2100 trials to achieve this hit rate after the task had been reversed. This finding demonstrates the cost of unlearning established stimulus-response mappings. For this reason, the extension of practice trials would not have been an economic countermeasure. Instead, it would have been more appropriate to utilize a neutral response system, like a separate response keypad. In future studies, a larger training block should be implemented to ensure that the stimulus-response mappings are suitably practiced and internalized. Additionally, the individual performance level in regard to the stimulus-response mappings could be assessed. In the present study, the participants completed 24 training trials of the Negative priming task. They received feedback if their response had been wrong or too slow. However, the training phase had a fixed length and was not dependent on the individual performance. It might have been beneficial to implement a threshold of speed and accuracy that the participants would have had to reach to proceed to the test trials. These are points that should be addressed appropriately in future Negative priming tasks. Notably, the selected tasks in this study enabled the simultaneous examination of selective attention and response speed and differed in the degree of difficulty. The Posner-Cuing task was a comparatively simple task, as it required only the detection of the target and not – as in the Stroop task or Negative priming task – the identification of the target in regard to differing stimulus dimensions. In the Posner-cuing task, the participants had to react when the target appeared, regardless of what color it had or which letter it signified. While one response key was utilized in the Posner-Cuing task, two response keys were utilized in the Stroop task and four response keys were utilized in the Negative priming task (thus, the tasks ranged from one implemented stimulus-response mapping to four stimulus-response mappings). The selected tasks offered a scope of different dependent measures and degrees of difficulty, which increased the generalizability and interpretability of the response effects that were

[34] It should be noted that it was assessed whether the participants used the German standard system of finger placement and positioning. This system is sometimes called “Zehnfingerschreiben” (translation: *writing with ten fingers*), “Blinschreiben” (translation: *writing blindly*), or “Tastschreiben” (translation: *touch-writing*). The direct English translation is “touch-typing”. Being able to touch-type is the goal of learning the German standard system. However, it was not directly *tested* whether the touch-typing participants indeed were able to touch-type in the true sense of the word. Yet, the internalization of the standard rules requires practice, and the findings support the notion that the touch-typing participants indeed were likely able to touch-type. As for the other participants, it should be assumed that they used individual typing systems and differed in the degree to which these systems had been automatized.

obtained for the cholinergic polymorphisms. However, it is a limitation of this study that the tasks did not allow to infer *how* the sequence of information processing might have been influenced by the cholinergic polymorphisms. In the next chapter, an alternative approach will be considered.

Polymorphisms. The functional effects of the polymorphisms could only be inferred; this is a limitation of this study. In the next chapter, additional ways of evaluating neurotransmitter variations will be discussed. In future studies, it would be beneficial to utilize several of these ways to maximize the convergent validity.

Sample. In future studies, it would be advisable to collect ethnic data. While the risk of population stratification was low (Steffens et al., 2006), it would be beneficial to preclude its possibility altogether. In addition, it might be useful to collect more data on the medical histories of the participants, particularly specific information about the current medication or the possible prevalence of psychiatric disorders. This is relevant, since disorders are associated with the dysregulation of neurotransmitter systems. In schizophrenia, for example, the dopaminergic system is assumed to be overactive (due to an elevation of the high-affinity D2 receptors; Seeman, 2011). Disorders can influence the performance in attention tasks; participants with schizophrenia generally display impairments in Negative priming tasks, for example (Frings et al., 2014). Given the frequencies provided by Bailer and colleagues (2008), it is of course not likely that a significant fraction of the participants of the present study suffered from schizophrenia. While about a fifth of the sample may have been affected by psychiatric disorders, those disorders must have varied and their effects should have been diverse, thus not unanimously driving the performance in the tasks. Nonetheless – and with the goal to reduce error variance in mind – it should be considered to collect data on the medical histories in future studies.

Chapter 11

Outlook

In sum, this dissertation provides further evidence for the impact of the dopaminergic system on selective top-down attention and the impact of the cholinergic system on response speed. This chapter will be focused on ways in which future studies might build on these findings. A different method of assessing response speed will be shortly discussed (subchapter 11.1). Furthermore, additional ways of approaching and manipulating the DA and ACh neurotransmitter systems will be detailed in subchapter 11.2 and 11.3, respectively.

11.1. Disentangling response speed components

It is a limitation of the present study that the Stroop task, Negative priming task, and Posner-Cuing did not allow to deduce what component of the sequence of information processing might have been influenced by the cholinergic polymorphisms. In this regard, it is advisable to investigate the impact of the cholinergic polymorphisms through an electroencephalographic (EEG) approach. Due to their high temporal resolution, EEG measures allow to dissociate processes prior and after the stimulus onset. Perri and colleagues (2014) administered a go/no-go task to healthy participants and compared accuracy-matched subsamples who had displayed either a high speed or who had displayed a low speed. They reported that the fast group showed an elevated activity of the supplementary motor area even before the stimulus onset (as measured via the Bereitschaftspotential); this indicates different pre-stimulus baselines between the fast and the slow group. A higher response speed after stimulus onset was associated with the visual, event-related N1 and N2 potentials. In the fast group, both the N1 and the N2 components were characterized by larger amplitudes. Perri and colleagues described the neural path of a "speed system", which encompasses the supplementary motor area, the late extrastriate area and the posterior parietal cortex. At the moment, it has not been determined what part of information processing is influenced by the selected cholinergic polymorphism and neither has it been determined whether the *CHRNA4* rs1044396 polymorphism impacts on the same component as the *CHRNA5* polymorphisms. Given the different distributions of the receptor subunits within the CNS, it cannot be precluded that the polymorphisms might act on different stations of the neural speed path that has been outlined by Perri and colleagues. An EEG study would greatly help to illuminate these two issues.

11.2. Alternative ways of assessing and manipulating the DA System

The study of polymorphisms is a common way of examining naturally occurring variations within neurotransmitter systems, especially since many options that are viable in animal studies are not employable in human samples due to their invasiveness. In the present study, the functional effects of the polymorphisms could only be inferred. In case of the *COMT* Val158Met polymorphism, for instance, it was assumed that the presence of Met alleles is accompanied by higher prefrontal levels of DA (cp. Chen et al., 2004). In future studies, it would be worthwhile to assess neurotransmitter variations in multiple ways to further strengthen the validity of the results. Several of these methods will be shortly discussed.

Methods of assessment. The most direct way of estimating DA levels in the CNS is through PET, though the downside of this approach lies no doubt in its costliness and invasiveness (Volkow, Fowler, Wang, Baler, & Telang, 2009; Jongkees, Hommel, & Colzato, 2014). A more indirect method has recently been introduced. DA that is released in the hypothalamus acts as an inhibiting factor for prolactin (von Bohlen und Halbach & Dermietzel, 2006). Reuter and colleagues (2007) utilized this effect by determining the blood plasma levels of prolactin in relation to the *COMT* Val158Met polymorphism and the *ANKK1/DRD2* Taq1A polymorphism. They found the polymorphisms to interact on the mean prolactin levels. This study demonstrates that peripheral measures like the plasma concentrations of hormones can help to illuminate the effect of polymorphisms in the CNS. In comparison, taking a blood sample is still a relatively invasive and costly approach, however. The ability to discriminate colors can also be used as a dopaminergic marker. This approach is still new, but promising. In their overview, Jongkees and colleagues (2014) summarized that disorders that are associated with altered DA levels have been linked to impairments in color vision, especially of the blue-yellow type; this finding is all the more noteworthy as DA is highly concentrated in the amacrine and interplexiform cells of the retina (Witkovsky, 2004; Jongkees et al., 2014). Cocaine-dependent, recently deprived patients displayed reduced blue cone b-wave amplitudes and also had a significantly lower concentration homovanillic acid in their cerebrospinal fluid (Roy, Roy, Berman, & Gonzalez, 2003; Jongkees et al., 2014). Since homovanillic acid is a DA metabolite, this suggests that reduced levels of DA are accessible by measuring the function of the blue-yellow color spectrum. It has been proposed that impairments in this color discrimination capability might be indicative of a hypodopaminergic state in the CNS (Roy et al., 2003; Jongkees et al., 2014). A study of Colzato and colleagues (2014a) provides further evidence for this assumption. In their study, the participants were tested in regard to color discrimination and also took part in a Simon task (Simon & Small, 1969). In the task, two acoustic stimuli were presented to the participants. One stimulus required a key-press

on the left side of the keyboard, while the other stimulus required a key-press on the right side of the keyboard. Via earphones, the stimuli were presented either to the left or to the right ear. The probability was equal for both locations; the location was wholly irrelevant for the task at hand. Nevertheless, RTs were faster and more accurate if the stimulus was presented at the same side as the required response (i.e., when a left-presented stimulus required a key-press on the left side of the keyboard). This consequence of stimulus-response compatibility is termed the *Simon effect.* The effect is presumed to be driven by the automatic activation of the response location by the stimulus location (Fischer, Plessow, Kunde, & Kiesel; 2010; for an overview, see Lu & Proctor, 1995). Colzato and colleagues (2014a) reported that participants with an impairment along the blue-yellow color spectrum displayed larger Simon effects, thus exhibiting less control in a response conflict task than participants with unimpaired blue-yellow color discrimination. In the study of this dissertation, Met allele carriers of the *COMT* Val158Met polymorphism and 10-repeat allele carriers of the *DAT1* polymorphism displayed a better performance in the Stroop task, which was reflected in smaller Stroop effects. It would be worthwhile to examine the link between impairments of blue-yellow color discrimination and the effects of both polymorphisms. It can be speculated that Val allele carriers not only display a worse performance in tasks of selective top-down attention, such as the Stroop task, but also display comparatively larger impairments in blue-yellow color discrimination. At this point, the link between color discrimination and the dopaminergic system is not yet well understood, however. In addition, Colzato and colleagues (2014a) reported merely a correlative finding; it is unclear whether the color discrimination capability truly reflected endogenous levels of DA (instead of the general efficiency of information processing, for example). Even if a link between the color discrimination capability and the dopaminergic system can be established, further questions remain unanswered. For example, retinal DA is released both in reaction to light and circadian rhythmicity (Witkovsky, 2004). It is unclear whether impairments in color discrimination are attributable to *stable* or *transient* hypodopaminergic states. Both could be the case. Comparatively lower levels of DA could be due to stable interindividual factors (like the presence of the *COMT* Val158 allele) or to more transient factors. Drugs that lead to an increased DA transmission (like cocaine) can cause a reduction in the endogenous DA release, for example (Weiss, Markou, Lorang, & Koob, 1992; Martinez et al., 2009). Since the levels of DA vary in the different brain structures and are not uniform, it is unclear what an impairment of color discrimination reflects. It would be interesting to include a color discrimination test in future studies of the *COMT* Val158Met polymorphism and *DAT1* polymorphism. The test is non-invasive and *might* provide an additional way of assessing the DA system. At this point it cannot be recommended that the test replaces any other method of assessment, however. First, the correlates and possible mechanisms of the blue-yellow color impairment should be understood more clearly.

Methods of manipulation. Neurotransmitter systems can also be studied through external manipulations. Amphetamine, for example, has been administered to study the dopaminergic system in regard to working memory (Mattay et al., 2003), while nicotine, atropine or scopolamine have been administered to study the cholinergic system in regard to attention (Witte et al., 1997; Davidson, Cutrell, & Marrocco, 1999). The disadvantages of pharmacological challenges lie in their profound invasiveness and the need for a close medical supervision of the experiment. A less invasive method of manipulation has been established by Colzato and colleagues (2013), who tested the effect of tyrosine on the working memory performance of participants. Tyrosine is the precursor for the biosynthesis of catecholamines and can be supplied by food (Kuhar et al., 1999). The tyrosine hydroxylase enzyme catalyzes the conversion of tyrosine to L-DOPA, which in turn acts as the precursor of DA. This conversion is the rate-limiting step of the catecholamine biosynthesis (Kuhar et al., 1999). Employing a within-subjects design, Colzato and colleagues administered a tyrosine solution and a placebo solution to participants who had fasted overnight. The participants were tested in an n-back task with two conditions. In the less demanding condition (*1-back*), the participants had to indicate whether a currently presented letter matched the letter that had been presented in the previous trial. In the more demanding condition *(2-back)*, the participants had to indicate whether the previously presented letter matched the letter that had been presented two trials ago. The performance in the 1-back condition was comparable after tyrosine and placebo consumption. In the 2-back condition, however, the consumption of tyrosine increased the accuracy and the number of correct rejections while reducing the rate of false alarms. According to the authors, DA could be one of the cognitive resources that were depleted by the execution of the demanding task condition, so that the supplementation of tyrosine replenished the resource pool and increased the performance level. The results are similar to the one reported by Mattay and colleagues (2003), who administered amphetamine to participants and tested them in an n-back task. In this study, the effect of amphetamine was also dependent on the difficulty of the task. While Val homozygotes of the *COMT* Val158Met polymorphism benefitted from the drug at all levels of memory load, Met homozygotes were affected (i.e., impaired) only at the highest memory load condition. The effect of tyrosine consumption was also examined in regard to the performance in a go/no-go task[35] (Colzato, Jongkees, Sellaro, van den Wildenberg, & Hommel, 2014b). In this task, the participants were required to indicate the direction of green arrows (which could point either to the left or right side), while withholding a response to arrows that underwent a color change from green to red after a delay interval. The average RTs in the *go* trials (where exclusively green arrows were presented) reflected the speed of the

[35] A specific variation of the go/no-go task was administered: the stop-signal task. In a typical go/no-go task, stimuli either require a response or do not require a response. In a stop-signal task, the response-requiring go signal can change into a stop signal which indicates that no response should be made. Thus, the stop-signal task is a more demanding variant of the go/no-go task.

response execution. The average RTs in the *no-go* trials (where eventually red arrows were presented) reflected the efficiency of response inhibition. A response-dependent algorithm ensured that the participants inhibited the response in only about half of the no-go trials. As before, a within-subject design was employed. The consumption of tyrosine had no effect on the average response speed in the go trials. In contrast, tyrosine led to faster RTs in the no-go trials, therefore indicating a facilitation of response inhibition under increased DA transmission. Again, the authors speculated that additional tyrosine might have counteract the depletion of cognitive control resources in more demanding tasks conditions. They did, however, not yet take into consideration that there are four prominent DA pathways in the CNS (cp. chapter 2) or that these pathways – which target different brain regions – are intricately connected in different circuits (cp. chapter 7). In this regard, it is unclear how the elevation of the overall DA transmission affected the individual pathways. In future studies, the effect of tyrosine on cognitive parameters should best be pinpointed in regard to the affected brain areas. One way of achieving this could be the assessment of dopaminergic variants with high regional specificities. Thus, as a next step, the effect of tyrosine should be investigated in relation to the effect of dopaminergic variants, such as the *COMT* Val158Met polymorphism or *DAT1* polymorphism. It has been proposed that a medium range of DA activity is optimal for the PFC function, while high and low levels of DA activity are detrimental for the PFC function (Goldman-Rakic et al., 2000; Mattay et al., 2003). There are indications that Val allele carriers are positioned at a less optimal section of this inverted-U curve (Mier et al., 2009), but can be shifted to a more optimal section through pharmacological manipulations that increase the rate of DA signaling, like the administration of amphetamine (Mattay et al., 2003). The consumption of tyrosine should have an analogous effect to pharmacological agents. Since tyrosine should increase the baseline level of DA transmission, it can be assumed that its consumption would be beneficial for carriers of the Val allele, but detrimental for the carriers of the Met allele. This conclusion was also drawn by the Colzato group ("*Val carriers stand to benefit from [tyrosine] supplementation];"* Jongkees et al., 2014, p. 2). To test the hypothesis, the performance of Met and Val allele carriers should be compared after the consumption of a placebo and after the consumption of tyrosine. A range of PFC-dependent tasks are suitable to study possible interactions between the *COMT* Val158Met polymorphism and tyrosine. In this dissertation, carriers of the Met allele outperformed Val homozygotes in the Stroop task by being disproportionately faster in the incongruent condition. If the Stroop task were to be replicated under the condition of either placebo or tyrosine consumption, it can be assumed that the performance level of Met allele carriers would decrease while the performance level of Val allele carriers would increase, as both groups should be shifted on their respective positions on the inverted-U curve (see figure 11.1). This finding would strengthen the evidence for the inverted-U curve hypothesis and demonstrate that the natural food component tyrosine can exert effects analogous to those of pharmacological manipulations (Mattay

et al., 2003; Tunbridge et al., 2004). It would also expand the findings of the Colzato group and show that effect of tyrosine is dependent on the interaction with stable interindividual factors. In addition, it would serve to generalize findings that have been obtained from tasks of working memory (Colzato et al., 2013; Mattay et al., 2003) and inhibitory control (Colzato et al., 2014b) to the domain of selective top-down attention.

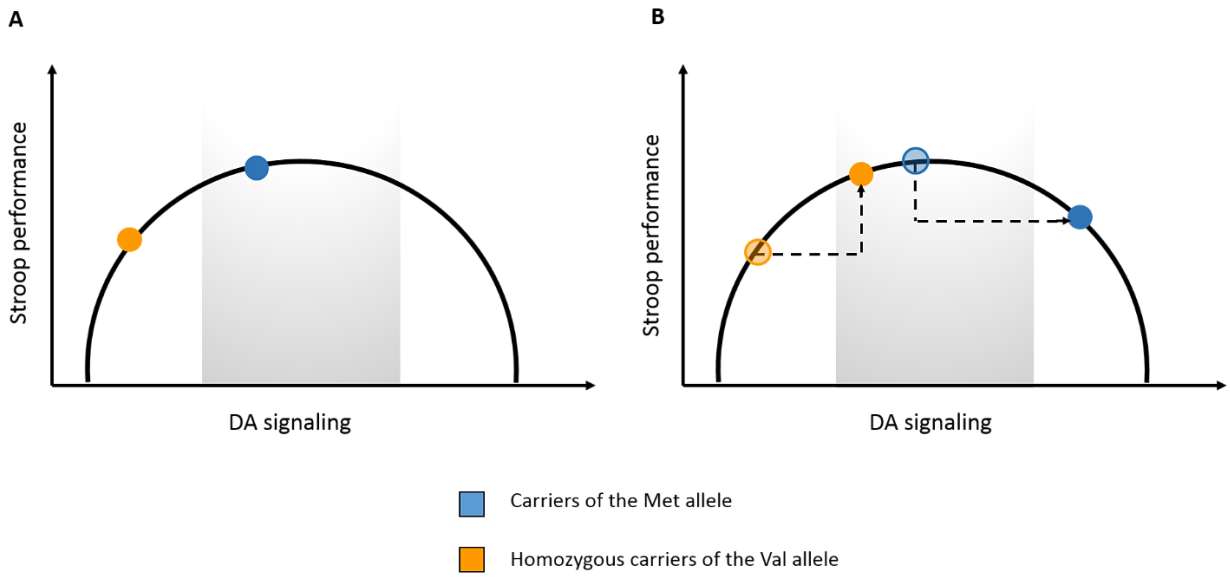


Figure 11.1: The assumed relation between the level of DA signaling and the performance in the Stroop task (note that higher performance levels indicate smaller Stroop effects). The optimal range of DA signaling is depicted in grey. The graphic depicts the assumed position of Val allele homozygotes and Met allele carriers on the inverted-U curve between DA signaling and PFC function under the condition of either placebo consumption (A) or tyrosine consumption (B). Under the consumption of a placebo, the level of prefrontal DA should be significantly higher in Met allele carriers (Chen et al., 2004) and result in a higher performance level in the Stroop task; it stands to reason that the Met allele carriers are positioned at more optimal position of the curve (Mattay et al., 2003). In contrast, tyrosine should elevate the rate of DA transmission and thus have a disadvantageous influence on the performance of Met allele carriers and a beneficial influence on the performance of Val homozygotes. Tyrosine should lead to a shift of their respective positions on the curve so that the Met allele carriers are shifted *outside* of the optimal range and the Val homozygotes are *shifted* inside of the optimal range (also cp. Mattay et al., 2003). The graphic was created in reference to Mattay and colleagues (2003).

The processing of tyrosine is dependent on the tyrosine hydroxylase enzyme, which is already close to saturation under normal conditions (Lü et al., 2009). Thus, the advantage of tyrosine administration lies in the direct manipulation of the dopaminergic system without the possibility of causing undesirable side effects (i.e., “overdosing” the participants). It future studies, it would be advisable to use multiple ways of assessing (and manipulating) the DA system, which would considerably increase the convergent validity and would further strengthen the interpretability of the obtained results.

11.3. Alternative ways of assessing and manipulating the ACh System

Methods of assessment. In the present study, the attention and response speed effects of the selected *CHRNA5* polymorphisms were dependent on gender; both polymorphisms exerted diametrically opposed effects in men and women. It has been speculated that these effects were due to interactions between the α5 subunit of the nAChR and the sex hormone and neurosteroid progesterone (cp. chapter 9). Consequently, the role of progesterone should be closely monitored and investigated In future studies of the two polymorphisms. This entails collecting data on the menstrual cycles of the female participants. The functioning of the nicotinic ACh system also appears to be significantly affected by oral contraceptives. It has been reported that the metabolism of nicotine is generally faster in women than in men, but also faster in women that took oral contraceptives than in women that did not take contraceptives (Benowitz et al., 2006). In future studies, it should be considered to match the female participants in regard to the phase of their menstrual cycle and in regard to the kind of oral contraceptive that is taken (if any). Furthermore, would be worthwhile to take blood samples from *all* participants to measure the plasma levels of progesterone. Likewise, it would be informative to devise an experiment in which progesterone is externally administered, so that the effects of this hormone on the selected *CHRNA5* polymorphisms and response speed as well as attention could be directly investigated. The different venues of examining the link between the α5 subunit und progesterone are depicted in figure 11.2.

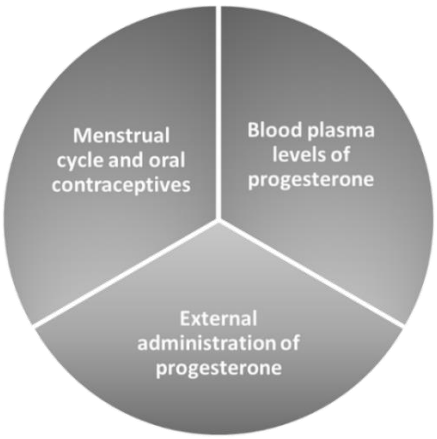


Figure 11.2.: Recommended venues of studying progesterone effects on the α5 subunit of the nAChR and response speed. In future studies, the blood plasma levels of progesterone should be measured in all participants. It is also important to control for the effect of the menstrual cycle and the intake of oral contraceptives in female participants. These two routes would serve to specify the already existing, circulating levels of progesterone in the participants. The most illuminating route of studying progesterone effects would be the external administration of progesterone, as it would presumable shift the performance levels of the participants. For example, the response speed advantages of the male S/S_G/G diplotype carriers should either be attenuated

or entirely eliminated under the administration of progesterone (as their progesterone levels should become more similar to the levels of the female participants). It is assumed that a high rate of cholinergic transmission is beneficial at low levels of progesterone, while a low rate of cholinergic transmission is beneficial at high levels of progesterone.

Methods of manipulation. Analogous to the investigation of the dopaminergic system through tyrosine, the cholinergic system can possibly be studied through the consumption of choline. ACh is synthesized by the enzyme choline acetyltransferase in a single step, through mediating the transfer of an acetyl group from acetyl coenzyme A to choline (Oda, 1999). Acetyl coenzyme A is produced by the glucose metabolism in the mitochondrion (Oda, 1999; von Bohlen und Halbach & Dermietzel, 2006), while choline can be provided either from food – more specifically, the phosphatidylcholine group of phospholipids – or through the hydrolysis of ACh molecules (von Bohlen und Halbach & Dermietzel, 2006; Snyder, 1999). Early studies suggested that the food supplementation by choline can counteract some of the cognitive deficits that are experienced by patients of Alzheimer's Disease (Cacabelos et al., 1992; 1996). In these studies, a daily dose of one gram choline was administered to the patients for a minimum of one month. The administration of choline has been studied mainly in regard to clinical samples or in the context of an age-dependent cognitive decline. There are indications that the consumption of choline improves the memory function after traumatic brain injuries (Wortzel & Arciniegas, 2012) and generally in elderly participants (Spiers, Myers, Hochanadel, Lieberman, & Wurtman, 1996; Alvarez et al., 1997; Sánchez et al., 2006; for an overview, see Secades, 2011), as well as improving the response speed of elderly participants (Bettini & Gorini, 2002; Secades, 2011). Fortunately, the administration of choline is quite unlikely to result in side effects. Secades described the tolerability of choline as "excellent" and the side effects as "rare" and "never severe" (2011, p. 47). All of the selected polymorphisms of this dissertation impacted on response speed. Open questions about their biological functionality still remain, though – for example in regard to their specific contributions to the neuromodulatory ACh system and the deterministic ACh system. To illuminate this question, the response speed of participants should investigated in a double-blind study with a within-subject, carry-over design. It would be advisable to test the performance in a series of tasks; most suitably the Stroop task, Negative priming task, and Posner-Cuing task, to achieve a maximal comparability and allow the replication of the results of this dissertation. After the consumption of a placebo, homozygous carriers of the *CHRNA4* rs1044396 polymorphism should display the highest response speed. Likewise, it is expected that male carriers of the S/S genotype of the *CHRNA5* rs3841324 polymorphism and of the G/G genotype of the *CHRNA5* rs16969968 polymorphism would display a higher response speed than their counterparts, while female carriers of these genotypes would be slower than their counterparts. The consumption of choline could impact on response speed

in a number of different ways. For example, it can be hypothesized that the response speed of all participants increases uniformly, conserving the relative positions of the homozygous rs1044396 T allele carriers compared to the heterozygous carriers and to the homozygous C allele carriers. The findings of Witte and colleagues (1997) provide a tentative indication for this assumption, since these authors reported that an elevated rate of cholinergic transmission overall led to reduced RTs. Alternatively, it can be hypothesized that the relative advantage of the rs1044396 T allele carriers increases even more when the level of cholinergic transmission is elevated. However, it is also possible that their advantage would collapse after the consumption of choline, because too high or too low levels of ACh signaling might exert detrimental effects. In fact, the relation between response speed and the level of ACh signaling might take the form of an inverted-U shaped curve, as has been reported for the PFC function and the level of DA signaling (Mattay et al., 2003). This assumption appears to be the most probable one and should be tested by administering different doses of choline. While low increases in the cholinergic transmission might improve the performance level of T allele homozygotes, higher increases might alleviate or reverse these effects. Inverted-U relations shaped relations between the dose of a substance and its direct effects are highly common. In fact, Calabrese and Baldwin (2001) concluded that "the most fundamental shape of the dose response is neither linear nor threshold but, in fact, U-shaped" (p. 285). Braida and colleagues (1996) reported that memory deficits in rats could be reversed by the administration of inhibitors cholinesterase (the enzyme which is responsible for the degradation of ACh); they specifically noted an inverted-U shaped response curve, with higher doses of the inhibitor leading to detrimental instead of beneficial effects. Similar diverse effects of choline consumption are also imaginable in the case of the *CHRNA5* polymorphisms. While the outcome of such a choline study underlies speculation, it would most likely serve to illuminate the workings of the ACh systems and conducting it would therefore be highly advisable.

References

Agnati, L. F., Zoli, M., Strömberg, I., & Fuxe, K. (1995). Intercellular communication in the brain: wiring versus volume transmission. *Neuroscience*, *69*, 711-726.

Alexander, G. E., DeLong, M. R., & Strick, P. L. (1986). Parallel organization of functionally segregated circuits linking basal ganglia and cortex. *Annual Review of Neuroscience, 9,* 357–381.

Allport, A. (1987). Selection for action: some behavioral and neurophysiological considerations of attention and action. In H. Heuer & A. F. Sanders (Eds.), *Perspectives on Perception and Action* (pp. 395-419). Hillsdale, NJ: Lawrence Erlbaum Associates.

Allport, A. (1989). Visual attention. In M. I. Posner (Ed.), Foundations of cognitive science (pp. 631–682). Cambridge, MA: MIT Press.

Allport, A. (1993). Attention and control: Have we been asking the wrong questions? A critical review of twenty-five years. In Meyer, D. E., & Kornblum, S., (Eds.), *Attention and performance XIV* (pp. 183-218). Cambridge, MA: MIT Press.

Alvarez, X. A., Laredo, M., Corzo, D., Fernandez-Novoa, L., Mouzo, R., Perea, J. E., ... & Cacabelos, R. (1997). Citicoline improves memory performance in elderly subjects. *Methods and findings in experimental and clinical pharmacology*, *19*, 201-210.

Anderson, J. R. (2007). Kognitive Psychologie. *Eine Einführung.* Spektrum-Verlag, Heidelberg.

Ansorge, U., & Leder, H. (2011). *Wahrnehmung und Aufmerksamkeit*. Wiesbaden: VS Verlag für Sozialwissenschaften.

Asadollahi, A., Mysore, S. P., & Knudsen, E. I. (2010). Stimulus-driven competition in a cholinergic midbrain nucleus. *Nature Neuroscience*, *13*, 889-895.

Axelrod, J. & Tomchick, R. (1958). Enzymatic O-methylation of epinephrine and other catechols. *Journal of Biological Chemistry, 233,* 702–705.

Bäckman, L., Ginovart, N., Dixon, R.A., Robins-Wahlin, T.-B., Wahlin, 2., Halldin, C., Farde, L., 2000. Age-related cognitive deficits mediated by changes in the striatal dopamine system. *American Journal of Psychiatry, 157,* 635– 637.

Bailer, J., Schwarz, D., Witthöft, M., Stübinger, C., & Rist, F. (2008). Prävalenz psychischer Syndrome bei Studierenden einer deutschen Universität. *PPmP-Psychotherapie· Psychosomatik· Medizinische Psychologie*, *58*, 423-429.

Baulieu, E. E., & Schumacher, M. (2000). Progesterone as a neuroactive neurosteroid, with special reference to the effect of progesterone on myelination. *Steroids*, *65*, 605-612.

Behrmann, M., Zemel, R. S., & Mozer, M. C. (1998). Object-based attention and occlusion: evidence from normal participants and a computational model. *Journal of Experimental Psychology: Human Perception and Performance*, *24*, 1011.

Benowitz, N. L., & Hatsukami, D. (1998). Gender differences in the pharmacology of nicotine addiction. *Addiction biology*, *3*, 383-404.

Benowitz, N. L., Lessov-Schlaggar, C. N., Swan, G. E., & Jacob, P. (2006). Female sex and oral contraceptive use accelerate nicotine metabolism*. *Clinical Pharmacology & Therapeutics*, *79*, 480-488.

Berger, A., Henik, A., & Rafal, R. (2005). Competition between endogenous and exogenous orienting of visual attention. *Journal of Experimental Psychology: General*, *134*, 207.

Berrettini, W. H., & Doyle, G. A. (2012). The CHRNA5–A3–B4 gene cluster in nicotine addiction. *Molecular psychiatry*, *17*, 856-866.

Bertocci, B., Miggiano, V., Da Prada, M., Dembic, Z., Lahm, H. W., & Malherbe, P. (1991). Human catechol-O-methyltransferase: cloning and expression of the membrane-associated form. *Proceedings of the National Academy of Sciences*, *88*, 1416-1420.

Bertolino, A., Blasi, G., Latorre, V., Rubino, V., Rampino, A., Sinibaldi, L., ... & Dallapiccola, B. (2006). Additive effects of genetic variation in dopamine regulating genes on working memory cortical activity in human brain. *The Journal of neuroscience*, *26*, 3918-3922.

Bettini, R., & Gorini, M. (2002). I tempi di reazione in corso di trattamento con citicolina. *La Clinica terapeutica*, *153*, 50.

Bierut, L., Stitzel, J., Wang, J., Hinrichs, A., Grucza, R., Xuei, X., ... & Goate, A. (2008). Variants in nicotinic receptors and risk for nicotine dependence. *American Journal of Psychiatry*, *165*, 1163-1171.

Blondel, A., Sanger, D. J., & Moser, P. C. (2000). Characterisation of the effects of nicotine in the five-choice serial reaction time task in rats: antagonist studies. *Psychopharmacology, 149,* 293–305.

Braida, D., Paladini, E., Griffini, P., Lamperti, M., Maggi, A., & Sala, M. (1996). An inverted U-shaped curve for heptylphysostigmine on radial maze performance in rats: comparison with other cholinesterase inhibitors. *European Journal of Pharmacology*, *302*, 13-20.

Breetvelt, E., Aukes, M., Boks, M., Cerrone, K., Grobbee, R., & Kahn, R. (2014). *Association of cognitive function with α5 nicotinic acetylcholine receptor genotype.* Twenty-second World Congress of Psychiatric Genetics, Copenhagen, Denmark.

Briand, K. A. (1998). Feature integration and spatial attention: More evidence of a dissociation between endogenous and exogenous orienting. *Journal of Experimental Psychology: Human Perception and Performance*, *24*, 1243.

Briand, K. A., & Klein, R. M. (1987). Is Posner's" beam" the same as Treisman's" glue"?: On the relation between visual orienting and feature integration theory. *Journal of Experimental Psychology: Human Perception and Performance*, *13*, 228.

Brickenkamp, R., Schmidt-Atzert, D. & Liepmann, D. (2010). *Test d2 - Revision*. Göttingen: Hogrefe.

Brinton, R. D., Thompson, R. F., Foy, M. R., Baudry, M., Wang, J., Finch, C. E., ... & Nilsen, J. (2008). Progesterone receptors: form and function in brain. *Frontiers in Neuroendocrinology*, *29*, 313-339.

Broadbent, D. (1958). *Perception and Communication*. London: Pergamon Press.

Broadbent, D. E., Cooper, P. F., FitzGerald, P., & Parkes, K. R. (1982). The cognitive failures questionnaire (CFQ) and its correlates. *British Journal of Clinical Psychology*, *21*, 1-16.

Brosch, T., Pourtois, G., Sander, D., & Vuilleumier, P. (2011). Additive effects of emotional, endogenous, and exogenous attention: Behavioral and electrophysiological evidence. *Neuropsychologia*, *49*, 1779-1787.

Brown, A. B., Biederman, J., Valera, E. M., Doyle, A. E., Bush, G., Spencer, T., ... & Seidman, L. J. (2010). Effect of dopamine transporter gene (SLC6A3) variation on dorsal anterior cingulate function in attention-deficit/hyperactivity disorder. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics*, *153*, 365-375.

Brown, A. B., Biederman, J., Valera, E., Makris, N., Doyle, A., Whitfield-Gabrieli, S., ... & Seidman, L. (2011). Relationship of *DAT1* and adult ADHD to task-positive and task-negative working memory networks. *Psychiatry Research: Neuroimaging*, *193*, 7-16.

Brozowski, T. S., Brown R. M., Rosvold, H. E., & Goldman, P. S. (1979). Cognitive deficits caused by regional depletion of dopamine in prefrontal cortex of Rhesus monkey. *Science,* 205, 929–932.

Buschman, T. J., & Miller, E. K. (2007). Top-down versus bottom-up control of attention in the prefrontal and posterior parietal cortices. *Science*, *315*, 1860-1862.

Cacabelos, R., Alvarez, X. A., Franco, A., Fernández-Novoa, L., Caamaño, J., & Valle-Inclán, F. (1992). Therapeutic effects of CDP-choline in Alzheimer's disease and multi-infarct dementia: psychometric assessment and immune function. *Annals of General Psychiatry*, *3*, 233-245.

Cacabelos, R., Caamano, J., Gomez, M. J., Fernández-Novoa, L., Franco-Maside, A., & Alvarez, X. A. (1996). Therapeutic Effects of CDP-Choline in Alzheimer's Disease-Cognition, Brain Mapping, Cerebrovascular Hemodynamics, and Immune Factorsa. *Annals of the New York Academy of Sciences*, *777*, 399-403.

Calabrese, E. J., & Baldwin, L. A. (2001). Hormesis: U-shaped dose responses and their centrality in toxicology. *Trends in Pharmacological Sciences*, *22*, 285-291.

Campbell, D. T., & Fiske, D. W. (1959). Convergent and discriminant validation by the multitrait-multimethod matrix. *Psychological Bulletin*, *56*, 81-105.

Cave, K. R., & Wolfe, J. M. (1990). Modeling the role of parallel processing in visual search. *Cognitive Psychology*, *22*, 225-271.

Chen, J., Lipska, B. K., Halim, N., Ma, Q. D., Matsumoto, M., Melhem, S., ... & Weinberger, D. R. (2004). Functional Analysis of Genetic Variation in Catechol-O-Methyltransferase (*COMT*): Effects on mRNA, Protein, and Enzyme Activity in Postmortem Human Brain. *The American Journal of Human Genetics*, *75*, 807-821.

Cherry, E. C. (1953). Some experiments on the recognition of speech, with one and with two ears. *The Journal of the Acoustical Society of America*, *25*, 975-979.

Clark, K. L., & Noudoost, B. (2014). The role of prefrontal catecholamines in attention and working memory. *Frontiers in Neural Circuits*, *8*.

Clarke, P. B., Schwartz, R. D., Paul, S. M., Pert, C. B., & Pert, A. (1985). Nicotinic binding in rat brain: autoradiographic comparison of [3H] acetylcholine, [3H] nicotine, and [125I]-alpha-bungarotoxin. *The Journal of Neuroscience*, *5*, 1307-1315.

Colquhoun, D., Shelley, C., Hatton, C., Unwin, N., & Sivilotti, L. (2003). Nicotinic acetylcholine receptors. *Burger's Medicinal Chemistry and Drug Discovery*, 2, 357–406.

Colzato, L. S., Jongkees, B. J., Sellaro, R., & Hommel, B. (2013). Working memory reloaded: tyrosine repletes updating in the N-back task. *Frontiers in Behavioral Neuroscience*, *7*. doi: 10.3389/fnbeh.2013.00200

Colzato, L. S., Jongkees, B. J., Sellaro, R., van den Wildenberg, W. P., & Hommel, B. (2014b). Eating to stop: Tyrosine supplementation enhances inhibitory control but not response execution. *Neuropsychologia, 62,* 398–402

Colzato, L. S., Sellaro, R., Hulka, L. M., Quednow, B. B., & Hommel, B. (2014a). Cognitive control predicted by color vision, and vice versa. *Neuropsychologia*, *62*, 55-59.

Connor, C. E., Egeth, H. E., & Yantis, S. (2004). Visual attention: bottom-up versus top-down. *Current Biology*, *14*, R850-R852.

Cools, R., & D'Esposito, M. (2011). Inverted-U–Shaped Dopamine actions on human working memory and cognitive control. *Biological Psychiatry*, *69*, 113-125.

Corbetta, M. (1998). Frontoparietal cortical networks for directing attention and the eye to visual locations: identical, independent, or overlapping neural systems? *Proceedings of the National Academy of Sciences*, *95*, 831-838.

Corteen, R. S., & Dunn, D. (1974). Shock-associated words in a nonattended message: A test for momentary awareness. *Journal of Experimental Psychology*, *102*, 1143-1144.

Corteen, R. S., & Wood, B. (1972). Autonomic responses to shock-associated words in an unattended channel. *Journal of Experimental Psychology*, *94*, 308-313.

Cowan, N. (1995). *Attention and memory: An integrated framework.* Oxford Psychology Series, No. 26. New York: Oxford University Press.

Craddock, N., Owen, M. J., & O'Donovan, M. C. (2006). The catechol-O-methyl transferase (COMT) gene as a candidate for psychiatric phenotypes: evidence and lessons. *Molecular psychiatry*, *11*, 446-458.

Cui, H., & Malpeli, J. G. (2003). Activity in the parabigeminal nucleus during eye movements directed at moving and stationary targets. *Journal of Neurophysiology*, *89*, 3128-3142.

Dani, J. A., & Bertrand, D. (2007). Nicotinic acetylcholine receptors and nicotinic cholinergic mechanisms of the central nervous system. *Annual Review of Pharmacology and Toxicology, 47*, 699-729.

Davidson, M. C., Cutrell, E. B., & Marrocco, R. T. (1999). Scopolamine slows the orienting of attention in primates to cued visual targets. *Psychopharmacology*, *142*, 1-8.

Dehdashti, F., Laforest, R., Gao, F., Aft, R. L., Dence, C. S., Zhou, D., ... & Welch, M. J. (2012). Assessment of progesterone receptors in breast carcinoma by PET with 21-18F-fluoro-16α, 17α-[(R)-(1'-α-furylmethylidene) dioxy]-19-norpregn-4-ene-3, 20-dione. *Journal of Nuclear Medicine*, *53*, 363-370.

Deichmann, W. B., Henschler, D., Holmstedt, B., & Keil, G. (1986). What is there that is not poison? A study of the Third Defense by Paracelsus. *Archives of Toxicology*, *58*, 207-213.

Deumens, R., Blokland, A., & Prickaerts, J. (2002). Modeling Parkinson's disease in rats: an evaluation of 6-OHDA lesions of the nigrostriatal pathway. *Experimental Neurology*, *175*, 303-317.

Deutsch, J. A., & Deutsch, D. (1963). Attention: some theoretical considerations. *Psychological Review*, *70*, 80-90.

Dickinson, D., & Elvevåg, B. (2009). Genes, cognition and brain through a COMT lens. *Neuroscience*, *164*, 72-87.

Duga, S., Soldà, G., Asselta, R., Bonati, M. T., Dalprà, L., Malcovati, M., & Tenchini, M. L. (2001). Characterization of the genomic structure of the human neuronal nicotinic acetylcholine receptor CHRNA5/A3/B4 gene cluster and identification of novel intragenic polymorphisms. *Journal of Human Genetics*, *46*, 640-648.

Duncan, J. (1984). Selective attention and the organization of visual information. *Journal of Experimental Psychology: General*, *113*(4), 501.

Durston, S., Fossella, J. A., Casey, B. J., Pol, H. H., Galvan, A., Schnack, H. G., ... & Van Engeland, H. (2005). Differential effects of DRD4 and DAT1 genotype on fronto-striatal gray matter volumes in a sample of subjects with attention deficit hyperactivity disorder, their unaffected siblings, and controls. *Molecular Psychiatry*, *10*, 678-685.

Dutta, A., & Gutfreund, Y. (2014). Saliency mapping in the optic tectum and its relationship to habituation. *Frontiers in Integrative Neuroscience*, *8*, doi: 10.3389/fnint.2014.00001.

Egan, M. F., Goldberg, T. E., Kolachana, B. S., Callicott, J. H., Mazzanti, C. M., Straub, R. E., ... & Weinberger, D. R. (2001). Effect of COMT Val108/158 Met genotype on frontal lobe function and risk for schizophrenia. *Proceedings of the National Academy of Sciences*, *98*, 6917-6922.

Egly, R., Driver, J., & Rafal, R. D. (1994). Shifting visual attention between objects and locations: evidence from normal and parietal lesion subjects. *Journal of Experimental Psychology: General*, *123*, 161-177.

Eriksen, C. W., & James, J. D. S. (1986). Visual attention within and around the field of focal attention: A zoom lens model. *Perception & Psychophysics*, *40*, 225-240.

Erixon-Lindroth, N., Farde, L.,Wahlin, T.-B. R., Sovago, J., Halldin, C., & Bäckman, L. (2005). The role of the striatal DA transporter in cognitive aging. Psychiatry Research, 138, 1–12.

Espeseth, T., Endestad, T., Rootwelt, H., & Reinvang, I. (2007). Nicotine receptor gene CHRNA4 modulates early event-related potentials in auditory and visual oddball target detection tasks. *Neuroscience*, *147*, 974-985.

Espeseth, T., Greenwood, P. M., Reinvang, I., Fjell, A. M., Walhovd, K. B., Westlye, L. T., ... & Parasuraman, R. (2006). Interactive effects of APOE and CHRNA4 on attention and white matter volume in healthy middle-aged and older adults. *Cognitive, Affective, & Behavioral Neuroscience*, *6*, 31-43.

Espeseth, T., Sneve, M. H., Rootwelt, H., & Laeng, B. (2010). Nicotinic receptor gene CHRNA4 interacts with processing load in attention. *PloS one*, *5*, e14407.

Evans, S. M., & Foltin, R. W. (2005). Exogenous progesterone attenuates the subjective effects of smoked cocaine in women, but not in men. *Neuropsychopharmacology*, *31*, 659-674.

Evans, S. M., Haney, M., & Foltin, R. W. (2002). The effects of smoked cocaine during the follicular and luteal phases of the menstrual cycle in women. *Psychopharmacology*, *159*, 397-406.

Eysenck, M., & Keane, M. T. (2010). *Cognitive Psychology – A Student's Handbook.* Hove: Psychology Press.

Feng, Y., Niu, T., Xing, H., Xu, X., Chen, C., Peng, S., ... & Xu, X. (2004). A common haplotype of the nicotine acetylcholine receptor α4 subunit gene is associated with vulnerability to nicotine addiction in men. *The American Journal of Human Genetics*, *75*, 112-121.

Fischer, R., Plessow, F., Kunde, W., & Kiesel, A. (2010). Trial-to-trial modulations of the Simon effect in conditions of attentional limitations: Evidence from dual tasks. *Journal of Experimental Psychology: Human Perception and Performance*, *36*, 1576-1594.

Floderus, Y., Ross, S. B., & Wetterberg, L. (1981). Erythrocyte catechol-o-methyltransferaseactivity in a Swedish population. *Clinical Genetics, 19,* 389 –392.

Floresco, S. B., & Magyar, O. (2006). Mesocortical dopamine modulation of executive functions: beyond working memory. *Psychopharmacology*, *188*, 567-585.

Flowers, J. H., Warner, J. L., & Polansky, M. L. (1979). Response and encoding factors in "ignoring" irrelevant information. *Memory & Cognition*, *7*, 86-94.

Folk, C. L., Remington, R. W., & Johnston, J. C. (1992). Involuntary covert orienting is contingent on attentional control settings. *Journal of Experimental Psychology: Human perception and performance*, *18*, 1030.

Folk, C. L., Remington, R. W., & Wright, J. H. (1994). The structure of attentional control: contingent attentional capture by apparent motion, abrupt onset, and color. *Journal of Experimental Psychology: Human perception and performance*, *20*, 317.

Fossella, J., Sommer, T., Fan, J., Wu, Y., Swanson, J. M., Pfaff, D. W., & Posner, M. I. (2002). Assessing the molecular genetics of attention networks. *BMC Neuroscience, 3,* 1-11.

Fox, E. (1995). Negative priming from ignored distractors in visual selection: A review. *Psychonomic Bulletin & Review*, *2*, 145-173.

Fox, E., Russo, R., Bowles, R., & Dutton, K. (2001). Do threatening stimuli draw or hold visual attention in subclinical anxiety? *Journal of Experimental Psychology: General, 130,* 681–700.

Franke, B., Hoogman, M., Arias Vasquez, A., Heister, J. G. A. M., Savelkoul, P. J., Naber, M., ... & Buitelaar, J. K. (2008). Association of the dopamine transporter (SLC6A3/DAT1) gene 9–6 haplotype with adult ADHD. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics*, *147*, 1576-1579.

Frings, C., Schneider, K. K., & Fox, E. (2014). *The Negative Priming Paradigm: An Update.* Manuscript submitted for publication.

Gangitano, D., Salas, R., Teng, Y., Perez, E., & De Biasi, M. (2009). Progesterone modulation of α5 nAChR subunits influences anxiety-related behavior during estrus cycle. *Genes, Brain and Behavior*, *8*, 398-406.

Giros, B., Jaber, M., Jones, S. R., Wightman, R. M., & Caron, M. G. (1996). Hyperlocomotion and indifference to cocaine and amphetamine in mice lacking the dopamine transporter. *Nature*, *379*, 606-612.

Glickstein, S. B., DeSteno, D. A., Hof, P. R., & Schmauss, C. (2005). Mice lacking dopamine D2 and D3 receptors exhibit differential activation of prefrontal cortical neurons during tasks requiring attention. *Cerebral Cortex*, *15*, 1016-1024.

Gogos, J. A., Morgan, M., Luine, V., Santha, M., Ogawa, S., Pfaff, D., & Karayiorgou, M. (1998). Catechol-O-methyltransferase-deficient mice exhibit sexually dimorphic changes in catecholamine levels and behavior. *Proceedings of the National Academy of Sciences*, *95*, 9991-9996.

Goldman-Rakic, P. S., Castner, S. A., Svensson, T. H., Siever, L. J., & Williams, G. V. (2004). Targeting the dopamine D1 receptor in schizophrenia: insights for cognitive dysfunction. *Psychopharmacology*, *174*, 3-16.

Goldman-Rakic, P. S., Muly III, E. C., & Williams, G. V. (2000). D1 receptors in prefrontal cells and circuits. *Brain Research Reviews*, *31*, 295-301.

Gray, J. A., & Wedderburn, A. A. I. (1960). Shorter articles and notes grouping strategies with simultaneous stimuli. *The Quarterly Journal of Experimental Psychology*, *12*, 180-184.

Green, A. E., Munafò, M. R., Deyoung, C. G., Fossella, J. A., Fan, J., & Gray, J. R. (2008). Using genetic data in cognitive neuroscience: From growing pains to genuine insights. *Nature Neuroscience Review, 9,* 710–720.

Greenwood, P. M., Fossella, J. A., & Parasuraman, R. (2005). Specificity of the effect of a nicotinic receptor polymorphism on individual differences in visuospatial attention. *Journal of Cognitive Neuroscience*, *17*, 1611-1620.

Greenwood, P. M., Parasuraman, R., & Espeseth, T. (2012). A cognitive phenotype for a polymorphism in the nicotinic receptor gene CHRNA4. *Neuroscience & Biobehavioral Reviews*, *36*, 1331-1341.

Grossman, M. H., Emanuel, B. S., & Budarf, M. L. (1992). Chromosomal mapping of the human catechol-O-methyltransferase gene to 22q11. 1→ q11. 2. *Genomics*, *12*, 822-825.

Gudelsky, G. A. (1981). Tuberoinfundibular dopamine neurons and the regulation of prolactin secretion. *Psychoneuroendocrinology*, *6*, 3-16.

Hahn, B., Ross, T. J., & Stein, E. A. (2006). Neuroanatomical dissociation between bottom–up and top–down processes of visuospatial selective attention. *Neuroimage*, *32*, 842-853.

Hahn, B., Shoiab, M., & Stolerman, I. (2002). Nicotine-induced enhancement of attention in the five-choice serial reaction time task: the influence of task demands. *Psychopharmacology, 162,* 129–137.

Herrero, J. L., Roberts, M. J., Delicato, L. S., Gieselmann, M. A., Dayan, P., & Thiele, A. (2008). Acetylcholine contributes through muscarinic receptors to attentional modulation in V1. *Nature*, *454*, 1110-1114.

Hindmarch, I., Kerr, J. S., & Sherwood, N. (1990). Effects of nicotine gum on psychomotor performance in smokers and non-smokers. *Psychopharmacology, 100,* 535-541.

Hommel, B. (2010). Grounding attention in action control: The intentional control of selection. In B. J. Bruya (Ed.), *Effortless attention: A new perspective in the cognitive science of attention and action* (pp. 121−140). Cambridge, MA: MIT Press.

Hopfinger, J. B., & West, V. M. (2006). Interactions between endogenous and exogenous attention on cortical visual processing. *NeuroImage*, *31*, 774-789.

Horn, J. L., & Cattell, R. B. (1967). Age differences in fluid and crystallized intelligence. *Acta Psychologica*, *26*, 107-129.

Houghton, G., & Tipper, S. P. (1994). A model of inhibitory mechanisms in selective attention. In Dagenbach, D. and Carr, T.H., (Eds), *Inhibitory Processes in Attention, Memory, and Language (*pp. 53–112). Academic Press.

Houghton, G., & Tipper, S. P. (1996). Inhibitory mechanisms of neural and cognitive control: Applications to selective attention and sequential action. *Brain and Cognition*, *30*, 20-43.

Ikemoto, S., & Panksepp, J. (1999). The role of nucleus accumbens dopamine in motivated behavior: a unifying interpretation with special reference to reward-seeking. *Brain Research Reviews*, *31*, 6-41.

Illing, R. B. (1996). The mosaic architecture of the superior colliculus. *Progress in brain research*, *112*, 17-34.

James, W. (1890). The principles of psychology. *Harvard UP, Cambridge, MA*.

Janes, A. C., Smoller, J. W., & David, S. P. (2012). Association between CHRNA5 genetic variation at rs16969968 and brain reactivity to smoking images in nicotine dependent women. *Drug and Alcohol Dependence*, *120*, 7-13.

Johansson, E. D. (1969). Progesterone levels in peripheral plasma during the luteal phase of the normal human menstrual cycle measured by a rapid competitive protein binding technique. *Acta Endocrinologica*, *61*, 592-606.

Johnston, W. A., & Heinz, S. P. (1978). Flexibility and capacity demands of attention. *Journal of Experimental Psychology: General*, *107*, 420-435.

Jongkees, B. J., Hommel, B., & Colzato, L. S. (2014). People are different: tyrosine's modulating effect on cognitive control in healthy humans may depend on individual differences related to dopamine function. *Frontiers in Psychology*, *5*. doi: 10.3389/fpsyg.2014.01101

Jonides, J. (1981). Voluntary versus automatic control over the mind's eye. In J. Long & A. Baddeley (Eds.), *Attention and performance IX* (pp. 187-203). Hillsdale, NJ: Erlbaum.

Kahneman, D., & Chajczyk, D. (1983). Tests of the automaticity of reading: dilution of Stroop effects by color-irrelevant stimuli. *Journal of Experimental Psychology: Human Perception and Performance*, *9*, 497.

Kang, A. M., Palmatier, M. A., & Kidd, K. K. (1999). Global variation of a 40-bp VNTR in the 3'-untranslated region of the dopamine transporter gene (SLC6A3). *Biological Psychiatry*, *46*, 151-160.

Kikuno, Y., Matsunaga, T., & Saiki, J. (2013). Polymorphism in the CHRNA4 gene is associated with rapid scene categorization performance. *Attention, Perception, & Psychophysics*, *75*, 1427-1437.

Klein, R. M. (1994). Perceptual-motor expectancies interact with covert visual orienting under conditions of endogenous but not exogenous control. *Canadian Journal of Experimental Psychology/Revue canadienne de psychologie expérimentale*, *48*, 167-181.

Klein, R. M. (2000). Inhibition of return. *Trends in Cognitive Sciences*, *4*, 138-147.

Klein, R. M., & Lawrence, M. A. (2012). On the Modes and Domains of Attention. In M. I. Posner (ed.), *Cognitive Neuroscience of Attention* (p. 11-29). New York: Guilford Press.

Kosslyn, S. M., & Rosenberg, R. S. (2001). *Psychology: The brain, the person, the world*. Boston: Allyn & Bacon.

Koster, E. H., Crombez, G., Verschuere, B., & De Houwer, J. (2004). Selective attention to threat in the dot probe paradigm: Differentiating vigilance and difficulty to disengage. *Behaviour Research and Therapy*, *42*, 1183-1192.

Koylu, E., Demirgören, S., London, E. D., & Pöğün, S. (1997). Sex difference in up-regulation of nicotinic acetylcholine receptors in rat brain. *Life sciences*, *61*, PL185-PL190.

Krauzlis, R. J., Lovejoy, L. P., & Zénon, A. (2013). Superior colliculus and visual spatial attention. *Annual Review of Neuroscience*, *36*.

Kuhar, M. J., Couceyro, P., & Lambert, P. D. (1999). Biosynthesis of Catecholamines. In Siegel, G. J., Agranoff, B. W., Albers, R. W., Fisher, S. K., & Uhler, M. D. (Eds.), *Basic Biochemistry – Molecular, Cellular and Medical Aspects* (pp. 243-262). Philadelphia: Lippincott Williams & Wilkins.

Kuhl, J. (1990). *Fragebogen zur Erfassung der Handlungskontrolle: HAKEMP-90.* Non-published questionnaire, Universität Osnabrück.

Kuryatov, A., Onksen, J., & Lindstrom, J. (2008). Roles of accessory subunits in α4β2* nicotinic receptors. *Molecular pharmacology*, *74*, 132-143.

LaBerge, D. (1983). Spatial extent of attention to letters and words. *Journal of Experimental Psychology: Human Perception and Performance*, *9*, 371-379.

Lachman, H. M., Papolos, D. F., Saito, T., Yu, Y. M., Szumlanski, C. L., & Weinshilboum, R. M. (1996). Human catechol-O-methyltransferase pharmacogenetics: description of a functional polymorphism and its potential application to neuropsychiatric disorders. *Pharmacogenetics and Genomics*, *6*, 243-250.

Lang, P.J., Bradley, M.M., & Cuthbert, B.N. (2008). International affective picture System (IAPS): Affective ratings of pictures and instruction manual. Technical Report A-8. University of Florida, Gainesville, FL.

Lavie, N., & Driver, J. (1996). On the spatial extent of attention in object-based visual selection. *Perception & Psychophysics*, *58*, 1238-1251.

Leboe, J. P., Whittlesea, B. W. A., & Milliken, B. (2005). Selective and nonselective transfer: Positive and negative priming in a multiple-task environment. *Journal of Experimental Psychology: Learning, Memory, and Cognition, 31,* 1001-1029.

Lee, J. H., Zhou, H. B., Dence, C. S., Carlson, K. E., Welch, M. J., & Katzenellenbogen, J. A. (2010). Development of [F-18] fluorine-substituted Tanaproget as a progesterone receptor imaging agent for positron emission tomography. *Bioconjugate Chemistry*, *21*, 1096-1104.

Lewis, D.A., Melchitzky, D.S., Sesack, S.R., Whitehead, R.E., Auh, S. & Sampson, A. (2001). Dopamine transporter immunoreactivity in monkey cerebral cortex - Regional, laminar, and ultrastructural localization. *Journal of Comparative Neurology, 432,* 119–136.

Loewi, O. (1921). Über humorale Übertragbarkeit der Herznervenwirkung. *Pflügers Archiv, 189,* 239-242.

Logan, G. D. (1985). Skill and automaticity: Relations, implications, and future directions. *Canadian Journal of Psychology, 39,* 367–386.

Lu, C. H., & Proctor, R. W. (1995). The influence of irrelevant location information on performance: A review of the Simon and spatial Stroop effects. *Psychonomic Bulletin & Review*, *2*, 174-207.

Lü, J. M., Wang, X., Marin-Muller, C., Wang, H., Lin, P. H., Yao, Q., & Chen, C. (2009). Current advances in research and clinical applications of PLGA-based nanotechnology. *Expert Review of Molecular Diagnostics, 9,* 325-341.

Lue, Y., Wang, C., Lydon, J. P., Leung, A., Li, J., & Swerdloff, R. S. (2013). Functional role of progestin and the progesterone receptor in the suppression of spermatogenesis in rodents. *Andrology*, *1*, 308-317.

Lukas, R. J., Changeux, J. P., le Novère, N., Albuquerque, E. X., Balfour, D. J., Berg, D. K., ... & Wonnacott, S. (1999). International Union of Pharmacology. XX. Current status of the nomenclature for nicotinic acetylcholine receptors and their subunits. *Pharmacological Reviews*, *51*, 397-401.

Lumb, P. L. K. (1995). Cognitive failures and performance differences: Validation studies of a German version of the cognitive failures questionnaire. *Ergonomics*, *38*, 1456-1467.

Lundström K, Salminen M, Jalanko A, Savolainen R, Ulmanen I (1991): (1991). Cloning and Characterization of Human Placental Catechol--Methyltransferase cDNA. *DNA and cell biology*, *10*, 181-189.

Lundström, K., Tenhunen, J., Tilgmann, C., Karhunen, T., Panula, P., & Ulmanen, I. (1995). Cloning, expression and structure of catechol-O-methyltransferase. *Biochimica et Biophysica Acta (BBA)-Protein Structure and Molecular Enzymology*, *1251*, 1-10.

MacDonald III, A. W., Cohen, J. D., Stenger, V. A., & Carter, C. S. (2000). Dissociating the role of the dorsolateral prefrontal and anterior cingulate cortex in cognitive control. *Science*, *288*, 1835-1838.

MacLeod, C. M. (1991). Half a century of research on the Stroop effect: an integrative review. *Psychological Bulletin*, *109*, 163.

MacLeod, C. M., & Dunbar, K. (1988). Training and Stroop-like interference: evidence for a continuum of automaticity. *Journal of Experimental Psychology: Learning, Memory, and Cognition*, *14*, 126.

MacLeod, C., Mathews, A., & Tata, P. (1986). Attentional bias in emotional disorders. *Journal of Abnormal Psychology*, *95*, 15.

Maczko, K. A., Knudsen, P. F., & Knudsen, E. I. (2006). Auditory and visual space maps in the cholinergic nucleus isthmi pars parvocellularis of the barn owl. *The Journal of Neuroscience*, *26*, 12799-12806.

Mao, D., Perry, D. C., Yasuda, R. P., Wolfe, B. B., & Kellar, K. J. (2008). The α4β2α5 nicotinic cholinergic receptor in rat brain is resistant to up-regulation by nicotine in vivo. *Journal of Neurochemistry*, *104*, 446-456.

Markett, S., Montag, C., & Reuter, M. (2011). The nicotinic ACh receptor gene CHRNA4 is associated with negative emotionality. *Emotion, 11,* 450-455.

Martinez, D., Greene, K., Broft, A., Kumar, D., Liu, F., Narendran, R., ... & Kleber, H. (2009). Lower level of endogenous dopamine in patients with cocaine dependence: findings from PET imaging of D2/D3 receptors following acute dopamine depletion. *American Journal of Psychiatry*, *166*, 1170-1177.

Mattay, V. S., Goldberg, T. E., Fera, F., Hariri, A. R., Tessitore, A., Egan, M. F., ... & Weinberger, D. R. (2003). Catechol O-methyltransferase val158-met genotype and individual variation in the brain response to amphetamine. *Proceedings of the National Academy of Sciences*, *100*, 6186-6191.

Matsumoto, M., Weickert, C. S., Akil, M., Lipska, B. K., Hyde, T. M., Herman, M. M., ... & Weinberger, D. R. (2003). Catechol-O-methyltransferase mRNA expression in human and rat brain: evidence for a role in cortical neuronal function. *Neuroscience*, *116*, 127-137.

May, C. P., Kane, M. J., & Hasher, L. (1995). Determinants of negative priming. *Psychological Bulletin*, *118*, 35.

Mayr, S., & Buchner, A. (2007). Negative priming as a memory phenomenon: A review of 20 years of negative priming research. *Zeitschrift für Psychologie/Journal of Psychology*, *215*, 35.

Meiser, J., Weindl, D., & Hiller, K. (2013). Complexity of dopamine metabolism. *Cell Communication and Signaling*, *11*. doi:10.1186/1478-811X-11-34

Mesulam, M. M. (1985). Attention, confusional states, and neglect. *Principles of Behavioral Neurology*, *3*, 125-68.

Mier, D., Kirsch, P., & Meyer-Lindenberg, A. (2009). Neural substrates of pleiotropic action of genetic variation in COMT: a meta-analysis. *Molecular Psychiatry*, *15*, 918-927.

Mignone, F., Gissi, C., Liuni, S., & Pesole, G. (2002). Untranslated regions of mRNAs. *Genome Biology*, *3*, 1-10.

Miller, E. K., & Cohen, J. D. (2001). An integrative theory of prefrontal cortex function. *Annual Review of Neuroscience*, *24*, 167-202.

Miyake, A., Friedman, N. P., Emerson, M. J., Witzki, A. H., Howerter, A., & Wager, T. D. (2000). The unity and diversity of executive functions and their contributions to complex "frontal lobe" tasks: A latent variable analysis. *Cognitive Psychology*, *41*, 49-100.

Mogg, K., & Bradley, B. P. (1999). Some methodological issues in assessing attentional biases for threatening faces in anxiety: A replication study using a modified version of the probe detection task. *Behaviour Research and Therapy*, *37*, 595-604.

Moors, A., & De Houwer, J. (2006). Automaticity: a theoretical and conceptual analysis. *Psychological Bulletin*, *132*, 297.

Moray, N. (1959). Attention in dichotic listening: Affective cues and the influence of instructions. *The Quarterly Journal of Experimental Psychology*, *11*, 56-60.

Müller, H. J., & O'Grady, R. B. (2000). Dimension-based visual attention modulates dual-judgment accuracy in Duncan's (1984) one-versus two-object report paradigm. *Journal of Experimental Psychology: Human Perception and Performance*, *26*, 1332.

Müller, H. J. & Rabbitt, P. M. (1989). Reflexive and voluntary orienting of visual attention: Time course of activation and resistance to interruption. *Journal of Experimental Psychology: Human Perception and Performance, 15*, 315–330.

Müsseler, J., & Prinz, W. (2002). *Allgemeine Psychologie*. Heidelberg: Spektrum, Akademischer Verlag.

Müsseler, J. (2008). *Allgemeine Psychologie* (2nd edition). Heidelberg: Spektrum, Akademischer Verlag.

National Library of Medicine (US). Genetics Home Reference [http://ghr.nlm.nih.gov/]. Bethesda (MD): The Library; 2014 Sep 29. CHRNA5; [cited 2014 Sep 19]. Available from: http://ghr.nlm.nih.gov/gene/CHRNA5.

Neill, W. T. (1997). Episodic retrieval in negative priming and repetition priming. *Journal of Experimental Psychology: Learning, Memory, and Cognition*, *23*, 1291.

Neill, W. T., & Valdes, L. A. (1992). Persistence of negative priming: Steady state or decay? *Journal of Experimental Psychology: Learning, Memory, and Cognition*, *18*, 565-576.

Neill, W. T., Valdes, L. A., Terry, K. M., & Gorfein, D. S. (1992). Persistence of negative priming: II. Evidence for episodic trace retrieval. *Journal of Experimental Psychology: Learning, Memory, and Cognition*, *18*, 993.

Neisser, U. (1967). *Cognitive Psychology.* New York: Appleton-Century-Crofts.

Nelson, M. E., Kuryatov, A., Choi, C. H., Zhou, Y., & Lindstrom, J. (2003). Alternate stoichiometries of α4β2 nicotinic acetylcholine receptors. *Molecular Pharmacology*, *63*, 332-341.

Newman, J., & Grace, A. A. (1999). Binding across time: the selective gating of frontal and hippocampal systems modulating working memory and attentional states. *Consciousness and Cognition*, *8*, 196-212.

Nirenberg, M. J., Vaughan, R. A., Uhl, G. R., Kuhar, M. J., & PickelI, V. M. (1996). The dopamine transporter is localized to dendritic and axonal plasma membranes of nigrostriatal dopaminergic neurons. *The Journal of Neuroscience*, *76*, 436-447.

Noudoost, B., & Moore, T. (2011a). The role of neuromodulators in selective attention. *Trends in Cognitive Sciences*, *15*, 585–591.

Noudoost, B., & Moore, T. (2011b). Control of visual cortical signals by prefrontal dopamine. *Nature*, *474*, 372-375.

Oda, Y. (1999). Choline acetyltransferase: the structure, distribution and pathologic changes in the central nervous system. *Pathology International*, *49*, 921-937.

Online Mendelian Inheritance in Man, OMIM®. Johns Hopkins University, Baltimore, MD. MIM Number: *118505, 04/25/2012. World Wide Web URL: http://omim.org/entry/ 118505?search= rs16969968&highlight=rs16969968.

Palmatier, M. A., Pakstis, A. J., Speed, W., Paschou, P., Goldman, D., Odunsi, A., ... & Kidd, K. K. (2004). COMT haplotypes suggest P2 promoter region relevance for schizophrenia. *Molecular Psychiatry*, *9*, 859-870.

Parasuraman, R., Greenwood, P. M., Kumar, R., & Fossella, J. (2005). Beyond Heritability Neurotransmitter Genes Differentially Modulate Visuospatial Attention and Working Memory. *Psychological Science*, *16*, 200-207.

Parikh, V., Kozak, R., Martinez, V., & Sarter, M. (2007). Prefrontal acetylcholine release controls cue detection on multiple timescales. *Neuron, 56,* 141–154.

Pashler, H., Johnston, J. C., & Ruthruff, E. (2001). Attention and performance. *Annual Review of Psychology*, *52*, 629-651.

Paterson, D., & Nordberg, A. (2000). Neuronal nicotinic receptors in the human brain. *Progress in Neurobiology*, *61*, 75-111.

Peinemann, A., Schuller, S., Pohl, C., Jahn, T., Weindl, A., & Kassubek, J. (2005). Executive dysfunction in early stages of Huntington's disease is associated with striatal and insular atrophy: a neuropsychological and voxel-based morphometric study. *Journal of the Neurological Sciences*, *239*, 11-19.

Perri, R. L., Berchicci, M., Spinelli, D., & Di Russo, F. (2014). Individual differences in response speed and accuracy are associated to specific brain activities of two interacting systems. *Frontiers in Behavioral Neuroscience*, *8*. doi: 10.3389/fnbeh.2014.00251

Pierce, R. C., & Kumaresan, V. (2006). The mesolimbic dopamine system: the final common pathway for the reinforcing effect of drugs of abuse? *Neuroscience & Biobehavioral Reviews*, *30*, 215-238.

Posner, M. I. (1980). Orienting of attention. *The Quarterly Journal of Experimental Psychology*, *32*, 3-25.

Posner, M. I., & Cohen, Y. (1984). Components of visual orienting. In H. Bouma & D. Bonwhuis (Eds.), *Attention and performance X: Control of language processes* (pp. 551–556). Hillsdale, NJ: Erlbaum.

Posner, M. I., Nissen, M. J., & Ogden, W. C. (1978). Attended and unattended processing modes: The role of set for spatial location. In Pick Jr., H. L. & Saltzman, E. (Eds.), *Modes of perceiving and processing information* (pp. 137–157).

Posner, M. I., Snyder, C. R., & Davidson, B. J. (1980). Attention and the detection of signals. *Journal of Experimental Psychology: General*, *109*, 160-174.

Purves, D., Augustine, G. J., Fitzpatrick, D., Katz, L. C., LaMatnia, A.-S., … & Williams, S. M. (2001). Neuroscience. Sunderland: Sinhauer Associates. Available from: http://www.ncbi.nlm.nih.gov/books/NBK10991/

Remington, R. W., Folk, C. L., & McLean, J. P. (2001). Contingent attentional capture or delayed allocation of attention? *Perception & Psychophysics*, *63*, 298-307.

Reuter, M., Schmitz, A., Corr, P., & Hennig, J. (2007). Molecular genetics support Gray's personality theory: the interaction of COMT and DRD2 polymorphisms predicts the behavioural approach system. *The International Journal of Neuropsychopharmacology*, *10*, 1-12.

Riederer, P., Eckert, A., Thome, J., & Müller, W. E. (2010). Neurotransmission und Signaltransduktion. In Riederer, P., & Laux, G. (2010). *Grundlagen der Neuro-Psychopharmakologie: ein Therapiehandbuch*. Springer.

Riederer, P. F. & Laux, G. (2010). *Grundlagen der Neuro-Psychopharmakologie.* Wien: Springer.

Rivett, A. J., Francis, A., & Roth, J. A. (1983). Localization of Membrane-Bound Catechol-O-Methyltransferase. *Journal of Neurochemistry*, *40*, 1494-1496.

Rösler, M., Retz, W., Retz-Junginger, P., Thome, J., Supprian, T., Nissen, T., Stieglitz, R.-D., Blocher, D., Hengesch, G., & Trott, G. E. (2004). Instrumente zur Diagnostik der Aufmerksamkeitsdefizit-/Hyperaktivitätsstörung (ADHS) im Erwachsenenalter. *Nervenarzt, 9,* 888–895.

Rothbart, M. K., Ahadi, S. A., Hershey, K. L., & Fisher, P. (2001). Investigations of temperament at three to seven years: The Children's Behavior Questionnaire. *Child development*, *72*, 1394-1408.

Roy, A., Roy, M., Berman, J., & Gonzalez, B. (2003). Blue cone electroretinogram amplitudes are related to dopamine function in cocaine-dependent patients. *Psychiatry Research*, *117*, 191-195.

Rueda, M. R., Rothbart, M. K., McCandliss, B. D., Saccomanno, L., & Posner, M. I. (2005). Training, maturation, and genetic influences on the development of executive attention. *Proceedings of the National Academy of Sciences, 102,* 14931-14936.

Sabatini, B. L., & Regehr, W. G. (1999). Timing of synaptic transmission. *Annual Review of Physiology*, *61*, 521-542.

Saccone, S. F., Hinrichs, A. L., Saccone, N. L., Chase, G. A., Konvicka, K., Madden, P. A., ... & Bierut, L. J. (2007). Cholinergic nicotinic receptor genes implicated in a nicotine dependence association study targeting 348 candidate genes with 3713 SNPs. *Human Molecular Genetics*, *16*, 36-49.

Salmi, J., Rinne, T., Koistinen, S., Salonen, O., & Alho, K. (2009). Brain networks of bottom-up triggered and top-down controlled shifting of auditory attention. *Brain Research*, *1286*, 155-164.

Sánchez, S., García, M. E., Carrizalez, Y., Cháves, L., Rodríguez, U., Cárdenas, J., ... & González, Y. M. (2006). Efectividad y tolerabilidad de la citicolina (somazina®): en el tratameinto de pacientes con deterioro cognitivo tipo demencia. *Archivos venezolanos de farmacología y terapéutica*, *25*, 101-103.

Sarter, M., & Bruno, J. P. (1997). Cognitive functions of cortical acetylcholine: toward a unifying hypothesis. *Brain Research Reviews*, *23*, 28-46.

Sarter, M., Givens, B., & Bruno, J. P. (2001). The cognitive neuroscience of sustained attention: where top-down meets bottom-up. *Brain Research Reviews*, *35*, 146-160.

Sarter, M., Lustig, C., Howe, W. M., Gritton, H., & Berry, A. S. (2014). Deterministic functions of cortical acetylcholine. *European Journal of Neuroscience*. Advance online publication. doi: 10.1111/ejn.12515

Sarter, M., Parikh, V., & Howe, W. M. (2009). Phasic acetylcholine release and the volume transmission hypothesis: time to move on. *Nature Reviews Neuroscience*, *10*, 383-390.

Schandry, R. (2006). *Biologische Psychologie*. Weinheim: Beltz Verlag.

Schmukle, S. C. (2005). Unreliability of the dot probe task. *European Journal of Personality*, *19*, 595-605.

Schneider, K. K., Schote, A. B., Meyer, J., & Frings, C. (2014). Genes of the Dopaminergic System Selectively Modulate Top-down but not Bottom-up Attention. *Cognitive, Affective, & Behavioral Neuroscience.* Advance online publication. doi: 10.3758/s13415-014-0320-9

Schneider, K. K., Schote, A. B., Meyer, J., & Frings, C. (in press). Sex matters! Interaction of gender and polymorphisms of a Cholinergic Receptor Gene modulates response speed. *NeuroReport.*

Schneider, K. K., Schote, A. B., Meyer, J., Markett, S., Reuter, M. & Frings, C. (2015). Individual response speed is modulated by variants of the gene encoding the alpha 4 sub-unit of the nicotinic acetylcholine receptor (CHRNA4). *Behavioural Brain Research.* Advance online publication. doi: 10.1016/j.bbr.2015.01.041

Secades, J. J. (2011). Citicoline: pharmacological and clinical review, 2010 update. *Revista de neurología, 52,* S1-62.

Seeman, P. (2011). All roads to schizophrenia lead to dopamine supersensitivity and elevated dopamine D2High receptors. *CNS Neuroscience & Therapeutics*, *17*, 118-132.

Servan-Schreiber, D., Printz, H. & Cohen, J. D. (1990) A Network Model of Catecholamiine Effects: Gain, Signal-to-Noise Ratio, and Behavior. *Science, 249,* 892–895.

Sesack, S. R., Aoki, C., & Pickel, V. M. (1994). Ultrastructural localization of D2 receptor-like immunoreactivity in midbrain dopamine neurons and their striatal targets. *The Journal of neuroscience*, *14*, 88-106.

Sesack, S.R., Hawrylak, V.A., Matus, C.V., Guido, M.A. & Levey, A.I. (1998). Dopamine axon varicosities in the prelimbic division of the rat prefrontal cortex exhibit sparse immunore activity for the dopamine transporter. *Journal of Neuroscience, 18,* 2697–2708.

Sheppard, L. D., & Vernon, P. A. (2008). Intelligence and speed of information-processing: a review of 50 years of research*. Personality and Individual Differences, 44, 535–551.*

Sherva, R., Wilhelmsen, K., Pomerleau, C. S., Chasse, S. A., Rice, J. P., Snedecor, S. M., ... & Pomerleau, O. F. (2008). Association of a single nucleotide polymorphism in neuronal acetylcholine receptor subunit alpha 5 (CHRNA5) with smoking status and with 'pleasurable buzz'during early experimentation with smoking. *Addiction*, *103*, 1544-1552.

Shiffrin, R. M., Dumais, S. T., & Schneider,W. (1981). Characteristics of automatism. In Long, J. & Baddeley, A. (Eds.), *Attention and performance IX* (pp. 223_238). Hillsdale, NJ: Lawrence Erlbaum Associates, Inc.

Shiffrin, R. M., & Schneider, W. (1977). Controlled and automatic human information processing: II. Perceptual learning, automatic attending and a general theory. *Psychological Review*, *84*, 127-190.

Shook, D., Brady, C., Lee, P. S., Kenealy, L., Murphy, E. R., Gaillard, W. D., ... & Vaidya, C. J. (2011). Effect of dopamine transporter genotype on caudate volume in childhood ADHD and controls. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics*, *156*, 28-35.

Siegrist, M. (1997). Test-retest reliability of different versions of the Stroop test. *The Journal of Psychology*, *131*, 299-306.

Simon, J. R., & Small Jr, A. M. (1969). Processing auditory information: interference from an irrelevant cue. *Journal of Applied Psychology*, *53*, 433.

Snyder, S. H. (1999). *Drugs and the Brain* (2. Aufl.). New York: W. H. Freeman and Company.

Spiers, P. A., Myers, D., Hochanadel, G. S., Lieberman, H. R., & Wurtman, R. J. (1996). Citicoline improves verbal memory in aging. *Archives of Neurology*, *53*, 441-448.

Squire, R. F., Noudoost, B., Schafer, R. J., & Moore, T. (2013). Prefrontal contributions to visual selective attention. *Annual Review of Neuroscience*, *36*, 451-466.

Steffens, M., Lamina, C., Illig, T., Bettecken, T., Vogler, R., Entz, P., *et al.* (2006). SNP-based analysis of genetic substructure in the German population. *Human Heredity, 62,* 20–29.

Stein, D. G. (2011). Is progesterone a worthy candidate as a novel therapy for traumatic brain injury?. *Dialogues in Clinical Neuroscience*, *13*, 352.

Steinlein, O., Sander, T., Stoodt, J., Kretz, R., Janz, D., & Propping, P. (1997). Possible association of a silent polymorphism in the neuronal nicotinic acetylcholine receptor subunit α4 with common idiopathic generalized epilepsies. *American Journal of Medical Genetics*, *74*, 445-449.

Steinlein, O., Smigrodzki, R., Lindstrom, J., Anand, R., Köhler, M., Tocharoentanaphol, C., & Vogel, F. (1994). Refinement of the localization of the gene for neuronal nicotinic acetylcholine receptor α4 subunit (CHRNA4) to human chromosome 20q13. 2-q13. 3. *Genomics*, *22*, 493-495.

Steinlein, O., Weiland, S., Stoodt, J., & Propping, P. (1996). Exon–intron structure of the human neuronal nicotinic acetylcholine receptor α4 subunit (CHRNA4). *Genomics*, *32*, 289-294.

Sternberg, R. (2008). *Cognitive psychology*. New York: Wadsworth.

Stieglitz, R.-D. (2000). *Diagnostik und Klassifikation psychischer Störungen.* Göttingen: Hogrefe.

Stroop, J. R. (1935). Studies of interference in serial verbal reactions. *Journal of Experimental Psychology*, *18*, 643.

Tenhunen, J., Salminen, M., Lundström, K., Kiviluoto, T., Savolainen, R., & Ulmanen, I. (1994). Genomic organization of the human catechol-o-methyltransferase gene and its expression from two distinct promoters. *European Journal of Biochemistry, 223*, 1049 –1059.

Theeuwes, J. (2010). Top–down and bottom–up control of visual selection. *Acta Psychologica*, *135*, 77-99.

Tipper, S. P. (1992). Selection for action: The role of inhibitory mechanisms. *Current Directions in Psychological Science, 1*, 105–109.

Tipper, S. P. (2001). Does negative priming reflect inhibitory mechanisms? A review and integration of conflicting views. *The Quarterly Journal of Experimental Psychology, 54*, 321-343.

Treisman, A. M. (1960). Contextual cues in selective listening. *The Quarterly Journal of Experimental Psychology*, *12*, 242-248.

Treisman, A. M. (1964a). Monitoring and storage of irrelevant messages in selective attention. *Journal of Verbal Learning and Verbal Behavior*, *3*, 449-459.

Treisman, A. M. (1964b). The Effect of Irrelevant Material on the Efficiency of Selective Listening. *The American Journal of Psychology, 77,* 533–546.

Treisman, A. M. (1985). Preattentive processing in vision. *Computer Vision, Graphics, and Image Processing*, *31*, 156-177.

Treisman, A. M., & Gelade, G. (1980). A feature-integration theory of attention. *Cognitive Psychology*, *12*, 97-136.

Trepel, M. (2008). *Neuroanatomie – Struktur und Funktion*. München: Urban & Fischer.

Tukey, J. W. (1977). *Exploratory data analysis.* Reading, MA: Addison-Wesley.

Tunbridge, E. M., Bannerman, D. M., Sharp, T., & Harrison, P. J. (2004). Catechol-o-methyltransferase inhibition improves set-shifting performance and elevates stimulated dopamine release in the rat prefrontal cortex. *The Journal of Neuroscience*, *24*, 5331-5335.

Tunbridge, E. M., Harrison, P. J., & Weinberger, D. R. (2006). Catechol-o-Methyltransferase, Cognition, and Psychosis: Val- 158- Met and Beyond. *Biological Psychiatry*, *60*, 141-151.

Valera, S., Ballivet, M., & Bertrand, D. (1992). Progesterone modulates a neuronal nicotinic acetylcholine receptor. *Proceedings of the National Academy of Sciences*, *89*, 9949-9953.

Vandenbergh, D. J., Persico, A. M., Hawkins, A. L., Griffin, C. A., Li, X., Jabs, E. W., & Uhl, G. R. (1992). Human dopamine transporter gene (DAT1) maps to chromosome 5p15. 3 and displays a VNTR. *Genomics*, *14*, 1104-1106.

VanNess, S. H., Owens, M. J., & Kilts, C. D. (2005). The variable number of tandem repeats element in DAT1 regulates in vitro dopamine transporter density. *BMC Genetics*, *6*. doi: 10.1186/1471-2156-6-55

Volkow, N. D., Fowler, J. S., Wang, G. J., Baler, R., & Telang, F. (2009). Imaging dopamine's role in drug abuse and addiction. *Neuropharmacology*, *56*, 3-8.

Volkow, N. D., Wang, G. J., Fowler, J. S., Gatley, S. J., Logan, J., Ding, Y. S., ... & Pappas, N. (1998). Dopamine transporter occupancies in the human brain induced by therapeutic doses of oral methylphenidate. *American Journal of Psychiatry*, *155*, 1325-1331.

Volkow, N. D., Wang, G. J., Newcorn, J., Fowler, J. S., Telang, F., Solanto, M. V., ... & Pradhan, K. (2007). Brain dopamine transporter levels in treatment and drug naive adults with ADHD. *Neuroimage*, *34*, 1182-1190.

von Bohlen und Halbach, O. V. B., & Dermietzel, R. (2006). *Neurotransmitters and neuromodulators: handbook of receptors and biological effects*. John Wiley & Sons.

Wang, J. C., Grucza, R., Cruchaga, C., Hinrichs, A. L., Bertelsen, S., Budde, J. P., ... & Goate, A. M. (2009). Genetic variation in the CHRNA5 gene affects mRNA levels and is associated with risk for alcohol dependence. *Molecular Psychiatry*, *14*, 501-510.

Ward, N. M., & Brown, V. J. (1996). Covert orienting of attention in the rat and the role of striatal dopamine. *The Journal of Neuroscience*, *16*, 3082-3088.

Wei, C., Han, Y., Spitz, M. R., Wu, X., Chancoco, H., Akiva, P., ... & Amos, C. I. (2011). A Case–Control Study of a Sex-Specific Association between a 15q25 Variant and Lung Cancer Risk. *Cancer Epidemiology Biomarkers & Prevention*, *20*, 2603-2609.

Weinberger, D. R., Berman, K. F., & Chase, T. N. (1988). Mesocortical dopaminergic function and human cognition. *Annals of the New York Academy of Sciences*, *537*, 330-338.

Weinshilboum, R. M., Otterness, D. M., & Szumlanski, C. L. (1999). Methylation pharmacogenetics: catechol O-methyltransferase, thiopurine methyltransferase, and histamine N-methyltransferase. *Annual Review of Pharmacology and Toxicology*, *39*, 19-52.

Weiss, F., Markou, A., Lorang, M. T., & Koob, G. F. (1992). Basal extracellular dopamine levels in the nucleus accumbens are decreased during cocaine withdrawal after unlimited-access self-administration. *Brain Research*, *593*, 314-318.

Wentura, D., & Frings, C. (2012). *Kognitive Psychologie.* Wiesbaden: VS Verlag für Sozialwissenschaften.

Wenzel, S.F. (2012). Dot-Probe. In C. Bermeitinger (Hrsg.), *Paradigmen der Kognitiven Psychologie: Affektive Reize I* (S. 73-108). Berlin: Uni-Edition.

Wesnes, K., & Warburton, D. M., (1984). Effects of scopolamine and nicotine on human rapid information processing performance. *Psychopharmacology, 82,* 147-150.

Wetter, D. W., Fiore, M. C., Young, T. B., McClure, J. B., de Moor, C. A., & Baker, T. B. (1999). Gender differences in response to nicotine replacement therapy: Objective and subjective indexes of tobacco withdrawal. *Experimental and Clinical Psychopharmacology*, *7*, 135-144.

Winterer, G., Musso, F., Konrad, A., Vucurevic, G., Stoeter, P., Sander, T., et al., (2007). Association of attentional network function with exon 5 variations of the CHRNA4 gene. *Human Molecular Genetics, 16,* 2165–2174.

Winterer, G., Rujescu, D., Maier, W., Steinlein, O.K., Bertrand, D., (2011). CHRNA4 Exon 5 genotype affects nicotinic receptor sensitivity and is associated with clinically high-functioning

schizophrenia rapid drug treatment-response and superior prefrontal function. In: Paper Presented at the Nicotinic Acetylcholine Receptors, Wellcome Trust Conference.

Witkovsky, P. (2004). Dopamine and retinal function. *Documenta Ophthalmologica*, *108*, 17-39.

Witte, E., Davidson, M. C., & Marrocco, R. (1997). Effects of altering brain cholinergic activity on covert orienting of attention: comparison of monkey and human performance. *Psychopharmacology, 134*, 324-334.

Wolfe, J. M. (1994). Guided search 2.0 a revised model of visual search. *Psychonomic Bulletin & Review*, *1*, 202-238.

Wolfe, J. M. (2007). Guided search 4.0: Current progress with a model of visual search. In W. Gray (Ed.), *Integrated models of cognitive systems* (pp. 99–119). New York: Oxford.

Wolfe, J. M., & Gancarz, G. (1997). Guided Search 3.0. In *Basic and clinical applications of vision science* (pp. 189-192). Springer Netherlands.

Wortzel, H. S., & Arciniegas, D. B. (2012). Treatment of post-traumatic cognitive impairments. *Current Treatment Options in Neurology*, *14*, 493-508.

Yang, C. R., Seamans, J. K., & Gorelova, N. (1999). Developing a neuronal model for the pathophysiology of schizophrenia based on the nature of electrophysiological actions of dopamine in the prefrontal cortex. *Neuropsychopharmacology*, *21*, 161-194.

Youdim, M. B., Edmondson, D., & Tipton, K. F. (2006). The therapeutic potential of monoamine oxidase inhibitors. *Nature Reviews Neuroscience*, *7*, 295-309.

Xie, T., Ho, S. L., & Ramsden, D.B. (1999). Characterization and implications of estrogenic down-regulation of human catechol-o-methyltransferase gene transcription. *Molecular Pharmacology, 56,* 31–38.

Xu, H., Jeong, H. Y., Tremblay, R., & Rudy, B. (2013). Neocortical somatostatin-expressing GABAergic interneurons disinhibit the thalamorecipient layer 4. *Neuron*, *77*, 155-167.

Zaehle, T., Sandmann, P., Thorne, J. D., Jäncke, L., & Herrmann, C. S. (2011). Transcranial direct current stimulation of the prefrontal cortex modulates working memory performance: combined behavioural and electrophysiological evidence. *BMC Neuroscience*, *12*. doi: 10.1186/1471-2202-12-2.

Zhang, H., Kranzler, H. R., Poling, J., & Gelernter, J. (2010). Variation in the nicotinic acetylcholine receptor gene cluster CHRNA5–CHRNA3–CHRNB4 and its interaction with recent tobacco use influence cognitive flexibility. *Neuropsychopharmacology*, *35*, 2211-2224.

Appendix

A1: the ADHD SR

ADHS-Selbstbeurteilungsskala (ADHS-SB) (Rösler et al. 2004)

Nachfolgend finden Sie einige Fragen über Konzentrationsvermögen, Bewegungsbedürfnis und Nervosität. Gemeint ist damit Ihre Situation, wie sie sich gewöhnlich darstellt.

Wenn die Formulierungen auf Sie nicht zutreffen, kreuzen Sie bitte »nicht zutreffend« an. Wenn Sie der Meinung sind, dass die Aussagen richtig sind, geben Sie bitte an, welche Ausprägung – leicht/mittel/schwer – Ihre Situation am besten beschreibt.

0	trifft nicht zu
1	leicht ausgeprägt (kommt gelegentlich vor)
2	mittel ausgeprägt (kommt oft vor)
3	schwer ausgeprägt (kommt nahezu immer vor)

Bitte kreuzen Sie die entsprechende Antwortalternative an. Lassen Sie bitte keinen Punkt aus.

Beispiel:

Ich bin unaufmerksam gegenüber Details oder mache Sorgfaltsfehler bei der Arbeit.	☐ 0	☐ 1	☐ 2	☒ 3

In diesem Fall ist die 3 (»schwer ausgeprägt«) angekreuzt: Das würde bedeuten, dass Sie stark ausgeprägt und nahezu immer Aufmerksamkeitsprobleme haben.

1. Ich bin unaufmerksam gegenüber Details oder mache Sorgfaltsfehler bei der Arbeit.	☐ 0	☐ 1	☐ 2	☐ 3
2. Bei der Arbeit oder sonstigen Aktivitäten (z. B. Lesen, Fernsehen, Spiel) fällt es mir schwer, konzentriert durchzuhalten.	☐ 0	☐ 1	☐ 2	☐ 3
3. Ich höre nicht richtig zu, wenn jemand etwas zu mir sagt.	☐ 0	☐ 1	☐ 2	☐ 3
4. Es fällt mir schwer, Aufgaben am Arbeitsplatz, wie sie mir erklärt wurden, zu erfüllen.	☐ 0	☐ 1	☐ 2	☐ 3
5. Es fällt mir schwer, Projekte, Vorhaben oder Aktivitäten zu organisieren.	☐ 0	☐ 1	☐ 2	☐ 3

6. Ich gehe Aufgaben, die geistige Anstrengung erforderlich machen, am liebsten aus dem Weg. Ich mag solche Arbeiten nicht oder sträube mich innerlich dagegen.	☐ 0	☐ 1	☐ 2	☐ 3
7. Ich verlege wichtige Gegenstände (z. B. Schlüssel, Portemonnaie, Werkzeuge).	☐ 0	☐ 1	☐ 2	☐ 3
8. Ich lasse mich bei Tätigkeiten leicht ablenken.	☐ 0	☐ 1	☐ 2	☐ 3
9. Ich vergesse Verabredungen, Termine oder telefonische Rückrufe.	☐ 0	☐ 1	☐ 2	☐ 3
10. Ich bin zappelig.	☐ 0	☐ 1	☐ 2	☐ 3
11. Es fällt mir schwer, längere Zeit sitzen zu bleiben (z. B. im Kino, Theater).	☐ 0	☐ 1	☐ 2	☐ 3
12. Ich fühle mich unruhig.	☐ 0	☐ 1	☐ 2	☐ 3
13. Ich kann mich schlecht leise beschäftigen. Wenn ich etwas mache, geht es laut zu.	☐ 0	☐ 1	☐ 2	☐ 3
14. Ich bin ständig auf Achse und fühle mich wie von einem Motor angetrieben.	☐ 0	☐ 1	☐ 2	☐ 3
15. Mir fällt es schwer abzuwarten, bis andere ausgesprochen haben. Ich falle anderen ins Wort.	☐ 0	☐ 1	☐ 2	☐ 3
16. Ich bin ungeduldig und kann nicht warten, bis ich an der Reihe bin (z. B. beim Einkaufen).	☐ 0	☐ 1	☐ 2	☐ 3
17. Ich unterbreche und störe andere, wenn sie etwas tun.	☐ 0	☐ 1	☐ 2	☐ 3
18. Ich rede viel, auch wenn mir keiner zuhören will.	☐ 0	☐ 1	☐ 2	☐ 3
19. Diese Schwierigkeiten hatte ich schon im Schulalter.	☐ 0	☐ 1	☐ 2	☐ 3
20. Diese Schwierigkeiten habe ich immer wieder, nicht nur bei der Arbeit, sondern auch in anderen Lebenssituationen, z. B. Familie, Freunde und Freizeit.	☐ 0	☐ 1	☐ 2	☐ 3
21. Ich leide unter diesen Schwierigkeiten.	☐ 0	☐ 1	☐ 2	☐ 3
22. Ich habe wegen dieser Schwierigkeiten schon Probleme im Beruf und auch im Kontakt mit anderen Menschen gehabt.	☐ 0	☐ 1	☐ 2	☐ 3

Bitte prüfen Sie, ob Sie alle Fragen beantwortet haben.

Table A2. Gender frequencies of the *COMT* Val158Met polymorphism

	COMT Val158Met	
	Val/Val	*Met+*
male	13	36
female	33	93

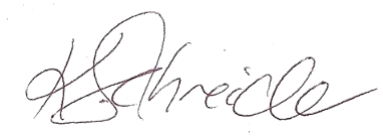
Katja Kerstin Schneider

Zum Sarkbrunnen 5

54296 Trier

Eidesstattliche Erklärung

Hiermit erkläre ich an Eides statt, dass ich die vorgelegte Dissertation selbst angefertigt und alle von mir benutzten Quellen und Hilfsmittel in der Arbeit angegeben habe.



_____

(Katja Kerstin Schneider, Trier, 4. Februar 2015)