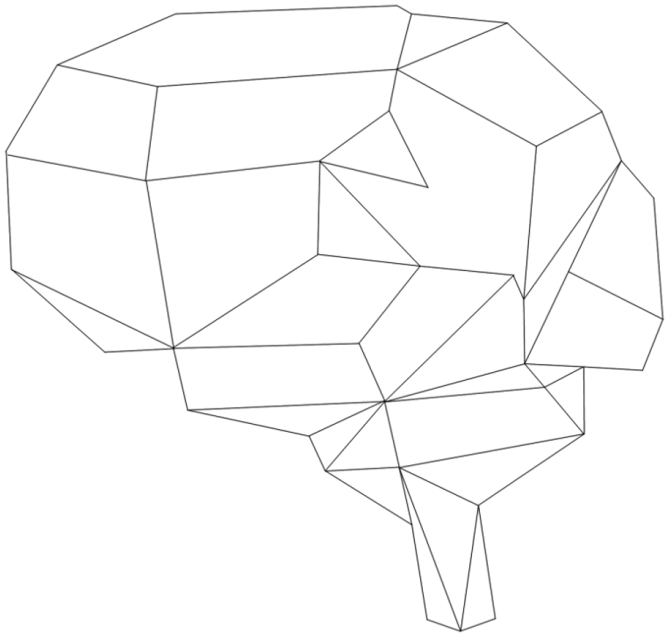
Chapter 1

# GENERAL INTRODUCTION



As I am writing this chapter, the Dutch public broadcasting association KRO-NCRV is airing its second episode in a special series about 'the ADHD-epidemic'. Within one week of announcing the making of this series earlier this year, KRO-NCRV's editorial office had received over two-hundred emails from parents, teachers, and psychiatrists. One week later, members of Parliament were publicly expressing their concerns about (over-) diagnosis and, perhaps even more so, (over-)medicating of youth with ADHD, causing intense debate. Distress about the potential neurotoxic effect of ADHD-medications is bolstered by popular reports comparing them with drugs of abuse such as cocaine and ecstasy. Understandably, people are wary: do we really want to expose our children to ADHD-medications, not knowing whether and how they may affect brain development in the long term? In response to parliamentary questions, the Secretary of the Ministry of Health, Welfare and Sport assured members of parliament that an investigation into the long-term effects of ADHD-medications on the developing brain was underway. In this thesis, I present the first results of that investigation.

In this introductory chapter, I first provide an overview of the scientific state-of-affairs regarding ADHD, stimulant treatment, and the developing brain. Next, I identify questions and challenges that have not satisfactorily been addressed in existing studies. At the end of this first chapter, remaining questions are translated into specific research aims and objectives.

## ATTENTION-DEFICIT/HYPERACTIVITY DISORDER

Attention-deficit/hyperactivity disorder (ADHD) is the most common developmental disorder in childhood, estimated to affect 3-7% of school-age children and 2.5% of adults globally (Polanczyk et al., 2007; Simon et al., 2009). ADHD is characterized by age-inappropriate attention problems and/or impulsivity and hyperactivity (American Psychiatric Association, 2000 and 2013). The clinical presentation of ADHD is heterogeneous and comorbidity with externalizing disorders such as oppositional defiant disorder, internalizing disorders such as mood and anxiety disorders, and other neurodevelopmental disorders such as autism spectrum disorder is frequent (Antshel et al., 2013; Connor et al., 2010; Meinzer et al., 2014). ADHD is regarded as an etiologically multifactorial disorder, with both genetic and environmental factors influencing the clinical presentation and course (Thapar & Cooper, 2016).

Neuropsychological deficits are frequent in ADHD as well. Perhaps the most thoroughly investigated domain is that of executive functioning, a broad concept referring to higher-order processes required for goal-directed behavior and self-regulation. Individuals with ADHD often present with prominent impairment in

executive functioning (Coghill et al., 2014a), which may be reflected in impaired performance on a spectrum of cognitive tasks. Especially poor performance on cognitive control tasks, measuring the ability to inhibit inappropriate responses in favor of more appropriate ones, and on working memory tasks, measuring the capacity to temporarily remember, process, and manipulate information, has consistently been implicated in ADHD (Doyle, 2006; Kasper et al., 2012). Apart from executive functioning impairments, poor performance on tasks measuring sustained attention and vigilance (Huang-Pollock et al., 2012), motivation and reward sensitivity (Luman et al., 2005; Volkow et al., 2011), and timing (Noreika et al., 2013) has also frequently been reported, as well as on other domains. Although individuals with ADHD *as a group* show deficits in each of these domains, individual patients are more likely to present with problems in some but not all of these domains (van Hulst et al., 2015). Approximately 25-30% of children with ADHD appear not to have neuropsychological impairments (Coghill et al., 2014a; Sonuga-Barke et al., 2010).

## NEUROBIOLOGY OF ADHD

The dopamine and norepinephrine systems have been implicated in ADHD (Del Campo et al., 2011; Swanson et al., 2007b). Dopamine especially has extensively been studied in this regard. In the human brain, dopamine is abundant in the frontal-striatal pathways, connecting the striatum (caudate nucleus, putamen, and nucleus accumbens) to the orbitofrontal, dorsolateral, and ventrolateral prefrontal cortices, and the pre- and supplementary motor areas (Leh et al., 2007). Neuroimaging studies consistently provide evidence of changes in the frontal-striatal pathways of children and adults with ADHD as compared to their typically developing peers. Such changes entail structural differences, such as local volume reduction in the caudate and putamen (Frodl & Skokauskas, 2012; Nakao et al., 2011), reduced grey matter volume and cortical thickness in prefrontal areas (Depue et al., 2010; Greven et al., 2015; Shaw et al., 2006), and compromised structural integrity of white matter fibers connecting the striatal and frontal regions (Ashtari et al., 2005; Casey et al., 2007; Pavaluri et al. 2009; van Ewijk et al., 2014b). Furthermore, ADHD has been associated with altered activation patterns in frontal-striatal circuits during various cognitive tasks (meta-analysis by Dickstein et al., 2006; review by Cubillo et al., 2012a). Radiotracer techniques such as positron emission tomography (PET) and single photon emission computed tomography (SPECT) have provided more direct evidence of altered dopamine binding patterns in the striatum of patients with ADHD (Zimmer, 2009). Changes in the frontal-striatal system have been linked to severity of ADHD symptoms (Cubillo et al., 2011; Shaw et al., 2011; Yang et al., 2016) and cognitive impairment (Casey et al., 2007; Depue et al., 2010; Valera et al., 2010). Of note, ADHD

has also been associated with more global brain changes (i.e., decrease in total brain volume), as well as with localized brain changes in areas outside the frontal-striatal circuits such as the parietal cortices, thalamus, amygdala, and cerebellum, and altered activation patterns within other networks such as the default-mode network. Such global changes might be associated with altered levels or metabolism of norepinephrine, a neurotransmitter that is widely distributed across the brain, but technological drawbacks have hampered radiotracer studies of norepinephrine metabolism in humans (Zimmer, 2009). For an accessible review on structural and functional brain changes associated with ADHD, see (Cortese, 2012).

## STIMULANT TREATMENT

ADHD treatment typically consists of behavioral interventions (e.g., cognitive behavioral therapy, parent training) and/or pharmacological interventions. Treatment with stimulants such as methylphenidate and d-amphetamine is the pharmacological intervention of first choice in ADHD (NHS NICE guideline, 2008). Robust acute treatment effects, including reduction of hyperactivity symptoms and attention problems, occur in the majority of patients (Chan et al., 2016; Faraone & Buitelaar, 2010; MTA Cooperative Group, 1999). Moreover, stimulant treatment is generally well tolerated and adverse events are rare (e.g. Barbaresi et al., 2006; Storebo et al., 2016; Wilens et al., 2006). Perhaps not surprisingly, treatment with stimulants has become increasingly customary in recent years, resulting in a large and growing number of children, adolescents and adults worldwide being exposed (Dalsgaard et al., 2013; Trip et al., 2009).

Stimulant treatment is often initiated shortly after diagnosis, typically between the ages of 6 and 11 (van den Ban et al., 2015). Oftentimes, short-acting, immediate-release formulations are prescribed at first (e.g., generic methylphenidate, Ritalin®, Dexedrine®, or Adderall®), and longer-acting or extended-release formulations are prescribed only at second instance (e.g., Concerta®, Equasym®, or Adderall-XR®). When patients respond well to treatment, stimulant use is often continued throughout childhood and (early) adolescence (van den Ban et al., 2010; Wong et al., 2009). The public and scientific debate about long-term outcomes of stimulant treatment has been intense, especially after the large-scale MTA consortium reported that, in the long term, stimulant treatment was not superior compared to behavioral treatment or even to community care (Jensen et al., 2007; Molina et al., 2009). Individual treatment characteristics such as age of treatment onset, treatment duration, and dose, vary substantially from one patient to the other. There have been reports suggesting that such individual differences with regard to treatment patterns may predict long-term outcomes (Dalsgaard et al., 2014; Mannuzza et al., 2008).

## ACUTE STIMULANT EFFECTS IN THE BRAIN

The immediate effects of stimulants on the brain are relatively well known. Within one hour after a single clinical dose, stimulants block dopamine and norepinephrine reuptake in the presynaptic terminal. As a result, dopamine and norepinephrine levels in the synaptic cleft increase, which in turn enhances stimulation of postsynaptic monoamine receptors (e.g. Arnsten, 2006; Kuczenski & Segal, 1997; Volkow et al., 2001). Magnetic resonance imaging (MRI) techniques offer non-invasive, accessible, and versatile tools to study stimulant effects in the developing human brain. Especially functional MRI studies investigating acute stimulant effects on neuronal activation patterns, measurable as the blood oxygenation level dependent (BOLD-) response that indicates cerebral blood flow, are abundant. In such studies, participants with ADHD typically perform a cognitive task while in the scanner (generally a task of which performance is known to improve upon stimulant intake) on two occasions: once shortly after intake of a single clinical dose of methylphenidate, and once while unmedicated or on placebo. A fairly consistent overall picture emerges across tasks. During tasks tapping cognitive control, methylphenidate appears to normalize brain activation patterns in children with ADHD by enhancing activation in frontostriatal circuits (Epstein et al., 2007; Lee et al., 2010; Rubia et al., 2011a; Vaidya et al., 1998) and parietal brain regions (Rubia et al., 2011b). Methylphenidate has been found to restore default-mode network suppression towards normative levels during cognitive control as well (Liddle et al., 2011; Peterson et al., 2009). Normalization of prefrontal cortex activation has also been observed during emotional processing (Posner et al., 2011). During tasks measuring different aspects of attention, methylphenidate appears to, at least partially, normalize brain activation and functional connectivity patterns (Rubia et al., 2009; Shafritz et al., 2004). Less consistent are the findings of acute methylphenidate effects during working memory tasks. For example, prefrontal cortex activation was found to increase, decrease, or remain unchanged after methylphenidate administration (Kobel et al., 2009; Prehn-Kristensen et al., 2011; Sheridan et al., 2010; Wong & Stevens, 2012). Finally, normalizing effects of a single dose of methylphenidate have also been reported when no tasks were performed, i.e., during so-called resting-state scans (An et al., 2013; Anderson et al., 2002; Teicher et al., 2000). For a review about stimulant-induced changes in brain activation patterns in patients with ADHD, see Rubia et al., 2014.

## LONG-TERM TREATMENT EFFECTS IN THE BRAIN

Considering the substantial acute effects of a single dose of methylphenidate in the brain, it may be expected that repeated exposure to stimulants could cause lasting brain changes as well. Different mechanisms may underlie such lasting changes, two of which I wish to highlight. On a positive account, repeated manifestation of typical, age-appropriate behaviors (i.e., paying attention in class, making non-impulsive decisions) could increase the strength of neural networks underlying such behaviors through a process called activity-induced neuronal plasticity. Similar to how, for instance, juggling practice is known to result in increased grey matter volume in the motor cortex, repeated 'practice' of non-ADHD behavior may lead to attenuation of brain changes associated with untreated ADHD (Kasparek et al., 2015). Repeated positive reinforcement and appraisal of non-ADHD behavior may strengthen such effects.

Alternatively, on a negative account, lasting brain changes may represent neurochemical damage from repeated exposure to dopaminergic agents, similar to a scar. Dopaminergic nerve terminals may suffer detrimental effects, generally referred to as neurotoxicity, resulting from the accumulation of reactive oxygen species (i.e., oxidative stress) occurring when levels of extracellular dopamine are high and vesicular dopamine storage is disrupted (Berman et al., 2009). Evidence for neurotoxic stimulant effects derives almost exclusively from animal studies in which very high 'binge' (pharmacological) doses of stimulants are administered. Translated to humans, such binge patterns may be more compatible with stimulant abuse (e.g., methamphetamine addiction) or recreational stimulant use (e.g., MDMA/ecstasy) rather than stimulant treatment for ADHD as offered in clinical practice (Lynch et al., 2010). In patients with ADHD, two radiotracer studies have investigated long-term consequences of stimulant treatment for dopamine metabolism in the brain. The results of both studies suggested adverse effects. In the first, patients with ADHD, and especially those with a history of childhood stimulant treatment, presented with down-regulated dopamine metabolism in the striatum, amygdala and midbrain compared to controls (Ludolph et al., 2008). In the second study, individuals with ADHD presented with normative levels of striatal dopamine transporter availability prior to treatment, but showed higher levels after one year of methylphenidate treatment (Wang et al., 2013).

Few MRI studies have evaluated long-term consequences of stimulant treatment. Lasting brain changes after stimulant treatment can be studied using structural MRI, functional MRI, or diffusion MRI, but the latter two techniques have very rarely been employed. Structural MRI studies reporting on grey matter changes have yielded mixed results. Two meta-analyses report indirect evidence that

stimulant treatment is associated with more normative basal ganglia volumes: across different studies, findings of caudate nucleus and putamen volume reduction were more pronounced when predominantly medication-naïve individuals were included, as compared to when more previously stimulant-treated individuals were included (Frodl & Skokauskas, 2012; Nakao et al., 2011). However, large-scale original studies have failed to detect volume differences in the striatum between treated and untreated patients, or associations between treatment duration and striatal volumes (Greven et al., 2015; Onnink et al., 2014; Semrud-Clikeman et al., 2014; Shaw et al., 2014). In other brain regions, including the lateral prefrontal cortex, anterior cingulate cortex, parieto-occipital cortices, thalamus, and cerebellum, treatment effects suggestive of normalization have been reported (Bledsoe et al., 2009; Ivanov et al., 2010; Semrud-Clikeman et al., 2006 and 2012; Shaw et al., 2009). Here, treatment-naive children with ADHD, but not those with a history of stimulant treatment, showed significant volume reductions compared to their typically developing peers. Note that findings of long-term structural brain changes after stimulant treatment are, with one exception (Shaw et al., 2009) based on observational and cross-sectional data, are confined to specific and small portions of the cortex while leaving other abnormalities associated with ADHD unchanged, and have typically not been replicated.

Stimulant treatment may also, in the long-term, affect structural integrity of white matter pathways in the brain, which can be assessed using diffusion MRI. Only two studies, each with few participants, have investigated associations between treatment and white matter integrity. The first compared integrity of the frontal-striatal tracts between children with a relatively short versus a relatively long history of stimulant treatment, and found no differences (De Zeeuw et al., 2012). The second performed a whole-brain hypothesis-free comparison of treatment-naïve children with ADHD, stimulant-treated children with ADHD, and healthy control children (De Luis-García et al., 2015). In this study, stimulant treatment was found to be associated with enhanced structural connectivity in several major white matter bundles, including those of orbitofrontal-striatal pathways.

Functional MRI studies into lasting stimulant treatment effects have been equally rare. Overall, after medication wash-out, a history of stimulant treatment appears not to translate into lasting alterations of brain activation patterns during cognitive control, although some effects restricted to highly specific task conditions have been observed (Konrad et al., 2007; Pliszka et al., 2006). In contrast, during emotion and reward processing, stimulant-naïve adults with ADHD have shown functional brain changes compared to controls, that were not seen in adult patients with a history of childhood stimulant treatment, which may suggest lasting normalization of activation patterns (Schlochtermeier et al., 2011; Stoy et al., 2011).

Thus, in short, there is preliminary evidence for stimulant treatment-induced normalization of specific structural brain changes associated with ADHD, including grey matter volume reductions in the frontal and anterior cingulate cortex. Findings in the basal ganglia have been inconclusive. Studies that evaluate lasting treatment effects on white matter structural connectivity and brain activation patterns are markedly underrepresented and hence do not allow for drawing firm conclusions.

## CHALLENGES AHEAD

The available literature provides important directions for the work in thesis. Here, I discuss several questions and issues that remain unanswered in the existing literature, which have guided our research aims and objectives described in the next section. First, more research attention is warranted regarding long-term effects of stimulant treatment on non-volumetric brain outcomes, such as activation patterns and structural and functional connectivity, but also advanced cortical surface measures such as cortical thickness and shape, deserve more research attention. Especially functional neuroimaging can be informative, as it has the potential to bridge the gap between brain structure on the one hand, and clinical outcomes of ADHD and treatment on the other. After all, intuitively, the suggested normalization of structural brain changes after long-term stimulant treatment (Frodl & Skokauskas, 2012; Nakao et al., 2011) appears at odds with the rapid return of ADHD symptoms when stimulant treatment is temporarily ceased.

Second, a developmental perspective will be critical. The brain undergoes rapid developmental changes throughout childhood, adolescence, and young adulthood. Moreover, different brain regions and neural networks show distinct developmental trajectories (Raznahan et al., 2011; Wierenga et al., 2014). As a result, stimulant treatment effects are likely to be both developmentally sensitive (i.e., dependent on age of treatment, as has previously been shown in animal studies [van der Marel et al., 2014 and 2015]) and regionally specific. Moreover, treatment effects may become apparent only after adolescence or young adulthood, when neurodevelopment enters a more stable phase, which emphasizes the importance of long-term follow-up and developmentally matched reference groups, including typically developing adolescents.

Third, differences between patients that may predispose lasting treatment effects, such as genetic variation, remain largely unexplored. Similar to clinical or behavioral response to treatment, the brain's response to stimulants is likely to differ between individuals. As one example directly linked to the biological substrates of ADHD, an acute dose of methylphenidate was found to attenuate neural response to reward in adults with ADHD carrying the 9R risk allele of the dopamine transporter

gene, but not in those without the risk allele (Aarts et al., 2015). Lasting brain changes may be predisposed by genetic makeup in a similar fashion. Investigating these and other sources of interindividual differences (such as age, gender, and symptom severity) will not only advance our understanding of the neurobiological substrates of stimulant treatment, but may also potentially be of prognostic value to patients in the future.

Finally, fourth, the distinction between previously-treated and stimulant-naive patients with ADHD is a practical and often used yet overly simplistic representation of complex stimulant treatment trajectories. In practice, patients with ADHD initiate and discontinue treatment at different ages, require different dosages, do or do not receive other psychoactive medications, etc. More specifically, animal studies have shown that lasting treatment effects may be more pronounced when treatment occurred at younger age (van der Marel et al., 2014). Moreover, acute beneficial effects of stimulants were found to be reversed by concurrent antipsychotic exposure in rats (Cheng & Li, 2013). Studying long-term treatment effects thus requires consideration of individual differences in treatment trajectories of stimulant and non-stimulant medications. Taken one step further, recreational stimulant use patterns may be informative as well, as these are typically characterized by infrequent, irregular, high-dose exposure, which is very different from the everyday low-dose stimulant exposure in ADHD treatment. Here, too, different trajectories of stimulant exposure and their associated neural and behavioral outcomes are likely to provide insight into the working mechanisms of stimulant treatment. Furthermore, such information could in the future contribute to optimized clinical decision-making.

## AIMS, OBJECTIVES, AND CHAPTER OUTLINE

The first and foremost overall aim of this thesis is to describe long-term effects, or the absence thereof, of stimulant treatment on the developing brain in children, adolescents and young adults with ADHD. The second aim of this thesis is to advance our knowledge about mechanisms underlying stimulant effects in the developing brain of young people affected with ADHD. To these aims, we formulated the following objectives: 1) to investigate structural brain correlates of long-term stimulant treatment, with a special focus on grey and white matter aspects of the frontal-striatal circuits; 2) to explore functional brain correlates of long-term stimulant treatment; 3) to investigate how individual differences in relevant patient characteristics (e.g., gender, age, genetic makeup) may predict long-term treatment outcomes; 4) to explore which treatment characteristics (e.g., dose, age of treatment onset, concurrent non-stimulant treatment) are predictive of long-term treatment

outcomes; and 5) to evaluate long-term clinical and cognitive effects of stimulant treatment beyond the level of acute changes that are well-known.

These objectives are addressed in eight chapters. **CHAPTER 2** provides an extensive overview of the available literature regarding acute and long-term stimulant treatment effects on the brain in children and adolescents with ADHD. Chapter two is followed by seven empirical chapters, all except one (chapter eight) based on data from the NeuroIMAGE study (**BOX 1**). First, we investigate associations between long-term stimulant treatment and brain structure and function, while taking into account individual differences regarding patient characteristics and treatment patterns. In **CHAPTER 3**, we compare grey matter cortical thickness across the cortical mantle between individuals with and without ADHD, followed by a detailed evaluation of the effects of cumulative stimulant dose, age of stimulant treatment onset, and other treatment parameters. In **CHAPTER 4** we describe changes in structural connectivity of the frontal-striatal white matter pathways in patients, and how these pathways may be affected by long-term stimulant treatment. Next, in **CHAPTER 5**, we investigate whether reward processes in the brain are influenced by long-term stimulant treatment. To this end, we evaluate whether brain activation patterns during anticipation and/or reception of a monetary reward are associated with history of stimulant treatment even while off-medication.

In chapters six through eight, more focus is placed on patient- and treatment-related features that may mediate or moderate treatment effects in the brain. In **CHAPTER 6**, we evaluate how patient characteristics, i.e., age and genetic variability in two dopaminergic genes, may interact to affect outcome of long-term stimulant treatment at the level of brain structure. In **CHAPTER 7**, we investigate whether stimulant treatment effects in the basal ganglia may be altered by augmentation with low-dose atypical antipsychotics such as risperidone, a dopaminergic antagonist with neurochemical properties opposing those of stimulants. In **CHAPTER 8** we take a sidestep, and look at the effects of non-medical, recreational use of stimulants, which may include prescription medications but mainly entailed cocaine and ecstasy (3,4-methylenedioxymethamphetamine, MDMA). Recreational stimulant exposure patterns are very different from typical ADHD treatment patterns. In the Youth At Risk (YAR) sample, based in the University of California, San Diego (**BOX 2**), we evaluate the effect of incidental, high-dose stimulant exposure on the development of hippocampus structure and memory performance.

Finally, after having explored the neural correlates of stimulant exposure in multiple ways, we turn to the analysis of clinical and cognitive outcomes of stimulant treatment in **CHAPTER 9**. Here, we make use of the valuable longitudinal aspect of the

NeuroIMAGE sample, and relate changes in clinical and cognitive outcomes over six years to stimulant intake in that exact same timeframe.

**BOX 1.** The NeuroIMAGE study

The NeuroIMAGE study is a follow-up phase (2009-2012) of the Dutch part of the International Multicenter ADHD Genetics project (IMAGE; 2003-2006). It is a multi-site family-based cohort study designed to investigate the course of ADHD and its cognitive and neurobiological underpinnings. More than 1,000 children, adolescents, and young adults from almost 500 families participated in NeuroIMAGE (70.3% ADHD, 56% male, average age = 17.0 years old). Data collection included diagnostic interviews, behavioral questionnaires, cognitive assessment, structural and functional neuroimaging, collection of lifetime pharmacy transcripts, and genotyping. More information about the NeuroIMAGE study can be found online (www.neuroimage.nl) and in von Rhein et al., 2015a. Chapters three through seven and chapter nine are based in the NeuroIMAGE cohort.

**BOX 2.** The YAR study

The Youth At Risk (YAR) study is a prospective longitudinal neuroimaging study of adolescents at elevated risk for substance use problems. The study started in 2002 and is based in San Diego, California. At baseline, a total of 295 healthy boys and girls between twelve and fourteen years of age, with no or minimal exposure to alcohol or substances, were recruited. The sample was enriched with children who had a family history of substance use disorders, which put them at high risk for developing substance use problems themselves. Initial assessment included substance use interviews, behavioral questionnaires, cognitive testing, and structural and functional neuroimaging. After enrollment, participants were administered substance use interviews every six months, and were invited for complete assessment including an MRI scan every year. The YAR-study is now in its 12$^{th}$ year of funding, and participants are currently in their mid-twenties. Chapter eight is based in the YAR cohort.