A STUDY OF COGNITION IN PRE-ADOLESCENT TWINS

Marieke van Leeuwen

Thesis commitee

Dr. E. Crone

Dr. C. V. Dolan

Dr. S. Durston

Prof.dr. C. Kemner

Prof.dr. N. G. Martin

Dr. T. J. C. Polderman

Prof.dr. J. A. Sergeant

Paranimfen

Drs. H. van Leeuwen

Drs. G. J. D. Teske

This work was supported by grants from the Netherlands Organization for Scientific Research (NWO 051.02.060, 668.772, NWO 575-25-006, NWO 480-04-004, NWO/SPI 56-464-14192) and by the Center for Neurogenomics and Cognition Research (CNCR)

ISBN 978 90 9023637 7

Druk en uitgave mede tot stand gekomen dankzij financiële bijdragen van het CNCR, de J.E. Jurriaanse Stichting en de Vrije Universiteit van Amsterdam

VRIJE UNIVERSITEIT

A STUDY OF COGNITION IN PRE-ADOLESCENT TWINS

ACADEMISCH PROEFSCHRIFT

ter verkrijging van de graad Doctor aan
de Vrije Universiteit Amsterdam,
op gezag van de rector magnificus
prof.dr. L.M. Bouter,
in het openbaar te verdedigen
ten overstaan van de promotiecommissie
van de faculteit der Psychologie en Pedagogiek
op donderdag 18 december 2008 om 13.45 uur
in de aula van de universiteit,
De Boelelaan 1105

door
Marieke van Leeuwen
geboren te Utrecht

promotoren:	prof.dr. D.I. Boomsma
	prof.dr. H.E. Hulshoff Pol
copromotor:	dr. S.M. van den Berg

In herinnering aan oom Jan (Father Gerwin)

TABLE OF CONTENTS

1

Introduction

This thesis reports on the first wave of data collection of a longitudinal twin-sibling study on individual differences before and during adolescence. The longitudinal study specifically focuses on the degree to which individual differences in brain structure, cognition and hormonal levels are the result of genetic and non-genetic differences. This thesis concentrates on cognition and on the relationship between cognition and structural brain parameters. Results for brain structures (voxel based and volumes) and hormones are reported in the thesis of Jiska Peper. This introduction will start with explaining the methodology of twin studies and its assumptions and next proceed with reviewing the literature on cognition, brain development and puberty. An outline of the chapters in the thesis and a description of the sample conclude this introduction.

Twin study methodology

Twin studies are useful for disentangling the etiology of variation in cognition, brain structure and hormonal levels. They can separate variation caused by differences in human DNA sequence and variation caused by differences in environment (Plomin & Kosslyn, 2001). The proportion of genetic variance over the total variance is defined as *heritability*. In twin studies, environmental variance can be decomposed into variance shared by family members (*shared environment*) and variance which is unique for each individual (*unique environment*).

Given a set of assumptions, like random mating and equal environment for monozygotic (MZ) and dizygotic (DZ) twins, heritability estimates are obtained by comparing the resemblance in MZ with the resemblance in DZ twins. When MZ twin correlations are higher than DZ twin correlations, part of the twin resemblance in the phenotype (observed characteristic) is caused by genetic effects. When DZ twin correlations are more than half the size of MZ correlations, the resemblance between twins is at least partly caused by shared environmental effects. The relative importance of unique environmental factors is reflected by differences between MZ twins (1 – rMZ).

In addition to the additive contributions of genes and environment, there may be interactions between genotype and environment. By examining MZ intra-pair average and difference scores (Jinks & Fulker, 1970) one can uncover whether individuals with a certain genotype are more vulnerable to environmental influences. Genetic (and shared environmental) effects add to the similarity of MZ pairs and unique environment to the

differences between MZ pairs. If there is a positive correlation between intra-pair sum and absolute differences in e.g. intelligence, this suggests that individuals with a predisposition for low IQ are more similar than individuals with a predisposition for high IQ, and thus less susceptible to unique environmental influences than either genes or raising conditions (Finkel & Pedersen, 2001). If genotype-environment (GE) interaction is present and not included in the analysis of twin data, it will increase the estimates of environmental variance (whereas G x common environment will increase the genetic variance).

By including parents in the twin design one can study whether there is non-random mating for the trait that is analyzed. Non-random mating may affect the genetic variance in the next generation and thus affect heritability estimates. However, in the classic twin design, the effect of non-random mating is, paradoxically, to decrease heritability estimates. Further, in a twin design which includes parents, genetic and cultural transmission (the parental phenotype influences the environment of their children which influences the measured trait) while taking into account spousal resemblance.

The equal environment assumption is the assumption that the common environment is roughly the same for MZ and DZ twins reared in the same family, i.e. the influence of common environment is independent of zygosity. If this assumption is violated, this biases estimates of genetic influences. This assumption has been tested in several ways and appears reasonable for most traits (Bouchard & Propping, 1993; Kendler, Neale, Kessler, Heath, & Eaves, 1993; Plomin, DeFries, McClearn, & McGuffin, 2001).

The twin design assumes that findings in twins can be extended to the general population, i.e. to non-twins as well as twins. Twins may be treated in a different way than singletons. They are treated as a twosome instead of being approached as a single person with his own personality (Geluk & Hol, 2001). By including additional non-twin siblings in the study it is possible to test several assumptions like equality of means and variances between twins and singletons. The inclusion of siblings also increases the statistical power to detect sources of variance due to additive and non-additive genetic influences, and common environment (Posthuma & Boomsma, 2000). A representation of a twin sibling design is given in Figure 1.1. In this design phenotypic resemblance in identical twins is compared with resemblance in fraternal twins and sibling pairs. In this thesis we will test one possible source of environmental variation that is specific to twins, i.e. the influence of

putting children of a twin pair in separate classes. There hardly is any research comparing the adjustment of twins who are separated versus those kept together at school (Hay, 2004).

The univariate twin-sibling design can easily be generalized to the analysis of multivariate traits, to assess the etiology of covariance between traits and / or across time. The phenotypic association among traits may be caused by overlapping sets of genetic factors and / or environmental factors, or may reflect mechanisms of causality.

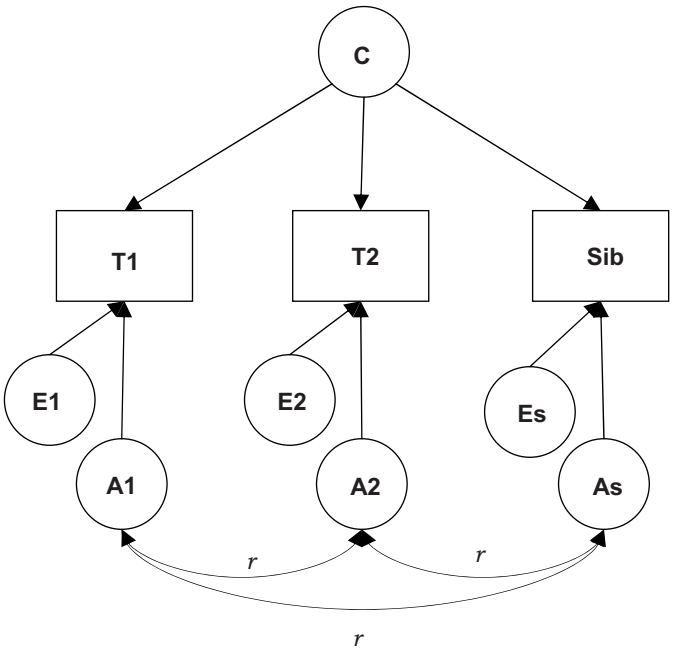


Figure 1.1. T1, T2 and Sib represent measured phenotypes in two twins and their sibling. Phenotypes are influenced by the latent factors: A = additive genetic factors, C = common environmental factors, and E = unique environmental factors. The correlation r between genetic factors is one for identical twins and 0.5 for fraternal twins and sibs

Cognition

We studied several indices of cognition in 9-year old twins and their older siblings. Early adolescence is related to a major change in cognitive thought leading to the development of abstract reasoning (Spear, 2000). Changes in cognition coincide with developmental changes in the brain (Durston & Casey, 2006). Cognition is studied in this thesis in several domains: cognitive control, memory, processing speed, and intelligence.

Cognitive control

Adolescence (the gradual period of transition between childhood and adulthood) is associated with the further development of cognitive control (Davidson, Amso, Anderson, & Diamond, 2006; Durston & Casey, 2006; Lamm, Zelazo, & Lewis, 2006; Levin et al., 1991; Welsh, Pennington, & Groisser, 1991). Cognitive control can be defined as the flexible regulation of thoughts and actions in the presence of competing ones and is essential in many cognitive functions (Durston & Casey, 2006). The key components which are ascribed to cognitive control vary between authors. However, all of these cognitive processes may share one basic neural circuitry: in all tasks measuring cognitive control, people have to ignore interfering information or rules (Casey, Giedd, & Thomas, 2000). The components most often referred to in the context of cognitive control are working memory (the ability to maintain and manipulate information at the same time), inhibition (the ability to inhibit prepotent responses), selective attention (paying attention to relevant stimuli or stimulus properties in the presence of competing ones), and cognitive flexibility, also called task switching (switching between rules; Davidson et al., 2006). Developmental studies of the latter are still scarce, but studies to date have demonstrated that the cost of switching between rules decreases until the age of 15 (Davidson et al., 2006; Huizinga, Dolan, & Van der Molen, 2006).

Other components related to cognitive control include inhibition and selective attention. These constructs refer more or less to the same processes, where subjects have to attend to a relevant event (selective attention) and ignore irrelevant ones (inhibition): selective attention is the flip coin of inhibition (Casey et al., 2000). For inhibition it is not so clear at what age it develops. Depending on the particular task used, some tasks show only a weak developmental curve from early childhood into adulthood and others yield a developmental trend until the age of 12 (Adleman et al., 2002; Huizinga et al., 2006; Lamm et al., 2006; Levin et al., 1991).

Measures for information processing speed try to capture the speed at which an individual completes basic cognitive functions such as item identification or simple discrimination. In the most optimal measure of processing speed cognitive speed is distilled from any motor speed involved in the actual execution of the response (Fry & Hale, 2000). It is assumed that processing speed reflects the speed of cortico-cortical connections in the

brain (Posthuma, De Geus, & Boomsma, 2002). Processing speed increases rapidly during childhood and more slowly during adolescence (Kail, 1991).

Memory

Working memory (WM) can be described as a system with limited capacity, that temporarily maintains and stores information and supports human thought processes by providing an interface between perception, long-term memory and action (Baddeley, 2003). According to (Baddeley, 2003) it consists of four components, namely the visuospatial sketchpad (temporarily stores visual information), the phonological loop (temporarily stores auditory information), the episodic buffer (an interface between the sub-systems of working memory and long-term memory) and the central executive (an attention controller). Working memory gradually develops throughout childhood into late adolescence and adulthood (Huizinga et al., 2006; Luciana, Conklin, Hooper, & Yarger, 2005; Luna, Garver, Urban, Lazar, & Sweeney, 2004; Swanson, 1999).

Short-term memory (STM), the capacity to store material over short periods of time in situations that do not impose other competing cognitive demands (Gathercole, Alloway, Willis, & Adams, 2006), is a central construct in modern theories of memory and cognition (Kail & Hall, 2001). Performance in visuospatial and verbal STM increases linearly from 4 to 14 years and levels of between 14 and 15 years (Alloway, Gathercole, & Pickering, 2006; Gathercole, Pickering, Ambridge, & Wearing, 2004).

Psychometric intelligence

Different cognitive abilities correlate among each other about .30 on average. A general factor (e.g. the first principal component in PCA) accounts for about 40% of the total variance in these tasks (Plomin & Spinath, 2002). This general factor is defined as general intelligence, IQ or *g*, and is supposed to be the driving force of performance in diverse areas of cognition.

A wide range of psychometric tests are available to assess intelligence. In the study described in this thesis the Raven Progressive Matrices (Raven, Raven, & Court, 1998; Raven, 1960) and the Wechsler Intelligence Scales (Wechsler, 1997; Wechsler et al., 2002) were used. The Raven assesses matrix reasoning. Performance on the Raven is considered as a strong predictor of *g*. The Wechsler Intelligence Scales assesses intelligence

by measuring performance in various cognitive domains like verbal STM and WM, arithmetic and vocabulary. Performance on these tasks contribute evenly to a total score which is defined as IQ. Psychometric IQ correlates around .50 with scholastic achievement, and this correlation is fully explained by a common set of genes (Bartels, Rietveld, Van Baal, & Boomsma, 2002). Psychometric IQ is fairly stable, with test-retest reliability scores over three years in children ranging from .80 till .85 for IQ and index scores (Hoekstra, Bartels, & Boomsma, 2007; Livingston, Jennings, Reynolds, & Gray, 2003).

Cognition and intelligence

Cognitive control is significantly correlated with intelligence, with correlations ranging from .38 to .98 (e.g. Ackerman, Beier, & Boyle, 2005; Colom, Rebollo, Palacios, Juan-Espinosa, & Kyllonen, 2004; Conway, Kane, & Engle, 2003; De Ribaupierre & Lecerf, 2006; Fry & Hale, 2000). Correlations between inspection time and IQ are around -.50 (Grudnik & Kranzler, 2001) and are similar for children and adults (Fry & Hale, 2000). For STM and intelligence correlations between .40 and .52 are reported (Colom, Flores-Mendoza, Quiroga, & Privado, 2005; Kane et al., 2004).

Individual differences in cognition

Two studies on the heritability of cognitive control in twelve-year-old children (Polderman et al., 2006; Stins, Van Baal, Polderman, Verhulst, & Boomsma, 2004) and one study in sixteen-year-old adolescents (Hansell et al., 2005) show heritability estimates for tasks measuring cognitive control between 41% and 56%, depending on the task measured and the sample assessed. In two samples of adolescents heritability estimates for inspection time (a measure for processing speed) have been obtained around 40% (Hansell et al., 2005; Luciano et al., 2001). Twin studies on STM in children are scarce. Although no significant heritability was found for verbal STM in children aged 6-13 years (Thapar, Petrill, & Thompson, 1994), this trait was moderately heritable in adolescents (56%; Rijsdijk, Vernon, & Boomsma, 2002).

Psychometric intelligence is a highly heritable trait in adulthood, with 70% genetically explained variance in IQ (Bouchard, Jr., Lykken, McGue, Segal, & Tellegen, 1990; Posthuma, 2002). In children, variance in IQ test performance is for 25 to 50% accounted for by genetic variation between individuals, with independent genetic effects on

verbal and nonverbal abilities (Hoekstra et al., 2007; Jacobs et al., 2001; Plomin, 2003; Turkheimer, Haley, Waldron, D'Onofrio, & Gottesman, 2003). Genetic influences on IQ are not fixed but change over time. In a longitudinal twin study the relative contribution of genetic influences on intelligence increased from age 5 to age 18. The main driving force behind the continuity in general IQ are the genetic influences and to a lesser extent the shared environment (Hoekstra et al., 2007). The stability of IQ and the increased relative contribution of genetic influences on IQ can probably be explained by an amplification of the same genetic influences rather than new genetic influences paying a contribution to the variability in IQ. Thus the genetic influence on intelligence appears to increase with age in children until it reaches maximum control in adulthood.

Structural brain development

Several studies have been completed on the development of brain volumes during childhood and adolescence (e.g. Barnea-Goraly et al., 2005; Casey, Tottenham, Liston, & Durston, 2005; Giedd et al., 1999; Giedd et al., 2006; Gogtay et al., 2004; Lenroot et al., 2007a; Olesen, Nagy, Westerberg, & Klingberg, 2003; Paus et al., 1999; Pruessner, Collins, Pruessner, & Evans, 2001; Sowell, Trauner, Gamst, & Jernigan, 2002; Thompson et al., 2000; Yurgelun-Todd, Killgore, & Young, 2002). These studies show that brain volumes are dynamic and change over time. This is evident from childhood through adolescence and young adulthood. In early adolescence gray matter (i.e. the neuron bodies that generate active electrical signals) volume starts to decrease (Jernigan & Tallal, 1990; Jernigan, Trauner, Hesselink, & Tallal, 1991; Pfefferbaum et al., 1994), whereas up to that period gray and white matter (i.e. myelinated and unmyelinated axons that connect the neuron bodies) volumes have constantly increased in volume (Durston et al., 2001; Giedd et al., 1999; Paus et al., 2001; Paus et al., 1999; Thompson et al., 2000). White matter keeps increasing in volume up into adulthood (Lenroot et al., 2007a).

There is regional and sex specific variability in the progressive and regressive events that occur in brain development (Sowell et al., 2002). In general, males have a steeper increase of white matter volume, and peak 1 to 2 years later in total brain volume and gray matter volume than females. Gray matter starts to decrease at the age of twelve, in boys particularly in the subcortical regions. Later on, but still at the beginning of puberty, cortical gray matter also starts to decrease in frontal and parietal areas (Giedd et al., 1996a;

Giedd et al., 1996b; Giedd et al., 1999; Lenroot et al., 2007a). In contrast to gray matter, white matter keeps increasing in this period particularly in the dorsolateral prefrontal cortex (DLPFC; Reiss, Abrams, Singer, Ross, & Denckla, 1996). Of all brain structures the DLPFC seems to develop last (Giedd, 2004). In addition, in girls, white matter volume increases particularly in the hippocampus, whilst in boys it increases particularly in the amygdala (Giedd et al., 1996b).

Individual differences in brain size

In adults, several studies have quantitatively investigated the contribution of genetic and environmental influences to individual differences in human brain volumes. Genetic factors account for most of the individual differences in whole brain, gray, and white matter volume (82-90%; Baaré et al., 2001). Interestingly, particularly the quantity of gray matter in Broca's and Wernicke's language areas and frontal brain regions (Thompson et al., 2001) is genetically determined in adults. Differences in shape of sulci are also influenced by genetic factors (Le Goualher et al., 2000). The high heritability estimates suggest that differences in brain volumes in adults are to a large extent of genetic origin.

It is poorly understood what factors influence the variation in brain structure in the child's brain. In children, research on genetic and environmental influences on individual differences in brain development measured by structural MRI is just starting. The heritability of brain volumes has been studied in a sample of children aged 5 to 19 years old (Wallace et al., 2006). Heritability estimates for volumes of nearly all brain regions varied between 77 and 89% with the notable exception of the cerebellum and the lateral ventricles whose heritability estimates were respectively 49 and 31%. In the same sample Lenroot et al. (2007b) studied heritability of cortical thickness and observed that heritability estimates depended on age and brain region. Heritability estimates were highest at the age a region showed most developmental changes. Since complex processes like cognitive control and regions associated with them develop latest, these regions are more heritable in adolescents than in children.

Puberty and gonadal hormones

Puberty is often used synonymously with adolescence, although strictly speaking they are not the same. Puberty is the period in which an individual becomes capable of sexual

reproduction, while adolescence refers to the gradual transition between childhood and adulthood. An important biological hallmark of this life stage is the elevated secretion of gonadal steroid hormones. Puberty and adolescence are intricately linked by the brain since this organ is a target for steroid hormones (Sisk & Zehr, 2005). Puberty starts with the reemergence of gonadotropin releasing hormone (GnRH) secretion during the night in the hypothalamus. GnRH signals the synthesis and secretion of the pituitary gonadotropins: luteinizing hormone (LH) and follicle stimulating hormone (FSH). Blood-borne LH and FSH act on target cells in the testes and the ovaries to direct the production of sperm and eggs, as well as the secretion of steroid hormones (Sisk & Foster, 2004). Timing of the first signs of puberty is highly variable across individuals, and the rate of subsequent sexual maturation is also quite varied. It is only recently that the factors which regulate the maturation of hypothalamic-pituitary-gonadal (HPG) axis and modulate the timing of puberty are starting to be understood (Grumbach & Styne, 1998; Sisk & Foster, 2004). Several internal and external signals are needed to permit HPG axis activation (Sisk & Zehr, 2005). In females many of such permissive signals are related to energy balance. Leptin, glucose, insulin, and metabolic fuel ability are important cues indicating that pregnancy can be supported and that sexual maturation can take place (Li, Ji, Wang, & Hu, 2005). The awakening of the HPG axis involves then an increase in excitatory output, a decrease in inhibitory output, and a supporting role of neuropeptides and glial-derived growth factor facilitation (Castellano et al., 2005; Ebling, 2005; Ojeda et al., 2006; Sisk & Zehr, 2005). These mechanisms are under genetic control (Ojeda et al., 2006).

Individual differences in onset of puberty

Based on twin and family studies we know that individual differences in timing of pubertal development are heritable. Heritability estimates range from 50% to 80% (Anderson, Duffy, Martin, & Visscher, 2007; Eaves et al., 2004; Loesch, Hopper, Rogucka, & Huggins, 1995; Meyer, Eaves, Heath, & Martin, 1991; Palmert & Boepple, 2001; Palmert & Hirschhorn, 2003; Van den Berg et al., 2006; Van den Berg & Boomsma, 2007) depending on phenotype definition and method of assessment (Van den Berg et al., 2006). Individual differences in testosterone levels during puberty are also under genetic control. Plasma testosterone, measured in adolescent twin pairs and their parents was found to be influenced by different genetic factors in men and women. In adolescent men,

approximately 60% of the variance in testosterone levels is heritable and different genetic factors may be expressed in adolescence and adulthood. In women (not controlled for time of menstrual cycle), 40% of the variance in testosterone levels is heritable, both in adolescence and in adulthood (Harris, Vernon, & Boomsma, 1998). Timing of puberty and testosterone levels in puberty are genetically related. In a sample of twelve-year-old twins (boys and girls) the correlation between testosterone levels and timing of puberty was .35 and entirely accounted for by a shared genetic etiology (Hoekstra, Bartels, & Boomsma, 2006).

Brain size and IQ

Summarizing, genetic factors seem to affect brain volumes, cognition and gonadal hormone levels. Below I describe what is currently known about how brain structure, cognitive development and hormonal levels are related to each other. The hypothesis that cognitive capacity correlates with brain size has persisted for over a century in evolutionary biology (e.g. Harvey & Krebs, 1990). By now this relationship has been well established and the population correlation in children and in adults between intelligence with total brain volume is estimated at .33 (McDaniel, 2005). Recent findings suggest that particularly the frontal and language-related cortices are linked to intelligence in adults (Haier, Jung, Yeo, Head, & Alkire, 2004; Posthuma et al., 2002; Posthuma et al., 2003; Thompson et al., 2001). To date, a few studies in adults have shown a genetic correlation between brain volumes and cognitive performance (Carmelli, Swan, DeCarli, & Reed, 2002; Posthuma et al., 2002; Posthuma et al., 2003; Thompson et al., 2001). The correlation between gray matter and intelligence and between white matter and intelligence is completely mediated by genes (Posthuma et al., 2002). Regionally, the phenotypic correlations (up to .35) between intelligence and white matter of the superior occipitofrontal, callosal, and left optical radiation and gray matter of the frontal and occipital lobes and the parahippocampal gyrus also can be explained by a common set of genes (Hulshoff Pol et al., 2006).

There are associations between the frontal and language-related cortices and intelligence in children (Frangou, Chitins, & Williams, 2004; Shaw et al., 2006). Children who have thinner gray matter in the left lateral dorsal frontal and parietal areas score better on vocabulary of the Wechsler Intelligence Scale for Children (WISC). Thicker gray matter in the medial occipital region is associated with better performance on block design (Sowell

et al., 2004). Less is known about the relationship between brain volume changes and cognitive development in children. Recently a difference was found in the dynamics of cortical development between children of high and average intelligence (Shaw et al., 2006).

Sex differences in brain volumes

The influence of gonadal hormones on human brain development is the topic of a rapidly expanding field of research. Steroid hormones act both in an activational and an organizational way on the nervous system. Activational effects refer to steroids modifying the activity of target cells to facilitate behavior in specific contexts. Activational effects are transient; they come and go with the presence or absence of hormones and are typically associated with steroid action in adulthood. Organizational effects on the other hand, refer to organizational changes in the nervous system during development that are permanent, persist beyond the period of developmental exposure to hormones, and program activational responses to steroids in adulthood (Sisk & Zehr, 2005). Both vertebrate and invertebrate studies have shown that steroid hormones affect neuronal development and plasticity. They play crucial roles in sexual dimorphism and apoptosis (Kawata, 1995).

Structural changes in adolescence are specific for brain-region and sex (Giedd et al., 1999; Giedd, 2004; Lenroot et al., 2007a; Sowell et al., 2004; Spear, 2000). In adulthood males have larger total brain volume than females, even when controlled for head size (for example: Allen, Damasio, Grabowski, Bruss, & Zhang, 2003; Good et al., 2001; Lemaitre et al., 2005; Nopoulos, Flaum, O'Leary, & Andreasen, 2000). In adults, age related reductions in the frontal and temporal lobes (Cowell et al., 1994) and hippocampus (Pruessner et al., 2001) were greater in men than in women. In children, after correction for overall brain volume, the caudate is relatively larger in girls, and the amygdala is relatively larger in boys (Durston et al., 2001). Moreover, adolescent boys and girls show different developmental trajectories in brain growth (Lenroot et al., 2007a), with girls developing earlier than boys.

Summary and questions

From the above review several conclusions can be drawn. Genetic factors influence variation in intelligence in healthy adolescents and adults. In young children, intelligence is partly genetically determined and partly determined by shared environmental factors. With

age, the heritability of intelligence increases and the influence of common environment decreases. Spouses resemble each other in intelligence (Guttman, 1974; Mascie-Taylor, 1989; Watkins & Meredith, 1981; Watson et al., 2004; Williams, 1975). It is not clear how much spousal resemblance influences heritability estimates for IQ in children. Also, it is not clear to what extent parents provide an environment which enhances the intelligence of their children. In Chapter 5 we analyze data from parents and offspring on psychometric intelligence. We examine the influence of assortative mating, cultural transmission and gene-environment interaction.

In spite of high heritability estimates for intelligence, the actual identification of genes is currently limited to mutations with rather severe neurological effects (De Geus, Wright, Martin, & Boomsma, 2001; Nokelainen & Flint, 2002). Genes that influence normal variation in cognitive ability in children and adolescents have yet to be identified. The search of these genes could be aided using the strategy of endophenotyping - that is, studying confined cognitive components or elements that relate to intelligence variability (De Geus et al., 2001; De Geus & Boomsma, 2001; Deary, 2001; Plomin & Spinath, 2002). However, suitable endophenotypes for intelligence have not been studied in detail in children and adolescents. Previous studies showed that selective attention, working memory and processing speed are suitable endophenotypes in adults (Ando, Ono, & Wright, 2001; Luciano et al., 2002; Posthuma, Mulder, Boomsma, & De Geus, 2002). Therefore, first the reliability of measures to assess these constructs in primary school children and adolescents was investigated. In Chapter 2, we studied the hypothesis that these constructs are suitable endophenotypes for intelligence in children.

In addition to reliability, an endophenotype should be associated with the trait of interest and the association should be due to genetic association (De Geus et al., 2001). The genetic architecture of the covariation among measures in different cognitive domains e.g. between intelligence and endophenotypes remains to be established: are individual differences in each of these domains influenced by overlapping sets of genes or environment? Multivariate genetic studies may solve contrasting findings found in the literature on individual differences in cognitive abilities on the association among cognitive abilities in different domains that have been proposed as endophenotypes for intelligence. This could be a reflection of the *molarity*-versus-*modularity* debate. In the molar view there is one system in which a unitary, general process functions across a wide variety of

cognitive tasks. In the modular view there are numerous distinct cognitive processing units, each responsible for certain non-overlapping cognitive tasks (Petrill, 1997). There is evidence that genetic influences tie together diverse measures of cognitive functioning, whereas environmental effects drive wedges between different dimensions of cognitive processing (Luo, Petrill, & Thompson, 1994; Pedersen, Plomin, & McClearn, 1994). Using multivariate genetic factor analysis, it is possible to establish to what extent the correlation between different cognitive measures is caused by a common set of genes and / or environmental factors (Boomsma & Molenaar, 1986; Martin & Eaves, 1977), and whether the factor structure at the genetic level is consistent with the factor structure at the environmental level. This results in Chapter 3 in a study on the relationship between verbal and visuospatial WM and STM. We tested the hypothesis that seen from a genetic viewpoint WM and STM are in essence part of the same system, and that verbal and visuospatial information are processed using the same memory pathways. Chapter 4 focuses on whether the relationship between reading ability, intelligence, short-term and working memory found in children is mediated by genes.

Genetic factors appear to largely determine variation in brain volumes in adults, particularly gray and white matter of Broca's and Wernicke's language areas and frontal regions. However, in (early) adolescence it is still unclear how strong genetic factors influence variability in brain structure. Also, the influence of puberty and hormonal levels on brain development in humans is not yet studied. Moreover, the genetic relationship between intelligence and brain volumes remains to be resolved in (pre)adolescence. Using a multivariate genetic twin design one can also infer direction of causation (i.e. does the environment influence intelligence which in turn influences brain volume, or the other way around?). If intelligence causally influences brain volume, all genetic and environmental factors that influence intelligence will also, through the causal chain, influence brain volume. Under the causal hypothesis both genetic and environmental correlations should be significant, whereas a significant genetic correlation in the absence of an environmental correlation falsifies the hypothesized causal effect of intelligence (De Moor, Boomsma, Stubbe, Willemsen, & De Geus, in press). Chapter 6 reports on the genetic relation between intelligence and brain volumes in nine-year-old children. The hypothesis that the relation between intelligence and brain volumes was mediated by genes was tested. Moreover, we hypothesized that brain volume causally influences intelligence.

Finally, the last chapter of this thesis concerned the effect of twin separation during primary school on academic achievement and problem behavior. In twin pairs from the young Netherlands Twin Registry (NTR) the assumption was examined that class separation of twin pairs during elementary school years has no long-term effect on academic achievement and problem behavior of these twins.

For this thesis we assessed cognition, brain structure and puberty status in 112 families with nine-year-old twins and their nine-to-fourteen-year-old siblings. Children and their parents were tested for their psychometric IQ using the Raven IQ test. Tasks in the cognitive battery for children were chosen on their ability to measure individual differences between children and to track changes in cognitive development. Using structural magnetic resonance imaging (MRI), a scan of the brain was made. To determine stage of puberty a physical examination was carried out and hormone levels were determined.

Material and Methods

Subjects

The study was approved by the Central Committee on Research involving Human Subjects (CCMO). Twins were recruited from the Netherlands Twin Registry (NTR), established by the Department of Biological Psychology at the Vrije Universiteit (VU) in Amsterdam. Around 40 to 50% of all multiple births in the Netherlands are registered by the NTR (Boomsma, Orlebeke, & Van Baal, 1992; Boomsma et al., 2002; Boomsma et al., 2006). The two birth cohorts (1995-1996) that could be approached consisted of 1754 families and of these 605 twin pairs had brothers and sisters aged between 9 and 14 years and there were 581 twin pairs of whom both twins were eligible for participation. The others met one or more of the exclusion criteria: pacemaker, metal materials in the head -except for dental braces-, chronic use of medication, a major medical history, psychiatric problems as reported by the parents, participation in special education, or physical or sensory disabilities. Based on birthday and zygosity of the twins, 214 families of this group were invited to participate by letter, which included an information brochure for the parents as well as for the children (see appendix). This was sent out one to two months before the ninth birthday of the twins. Two weeks after receiving the letters families were contacted by phone and asked whether they wanted to participate.

Measurements

Table 1.1 gives an overview of all data collected in this research project. On one day children were tested at the VU University (VU) in Amsterdam and on the other day at the University Medical Center in Utrecht (UMCU). Children were asked to collect saliva and morning urine at home on two consecutive days prior to testing at the VU. Cheek swabs, for DNA isolation, were collected by the parents as well as the children. Packages containing tubes and written instructions were sent three weeks prior to testing at the VU to the participants by mail. They could return these packages on the day of testing at the VU. Detailed information on the study protocol can be found in the Appendices.

Procedure

At the VU all children and parents underwent cognitive testing at the same time, parents filling in the Raven Advanced Progressive Matrices (Raven et al., 1998). Families arrived at the VU between nine and eleven o'clock in the morning, where they were welcomed and the protocol was explained. After that, children went with a test administrator to separate rooms, where they underwent cognitive assessment. The whole protocol took approximately five hours, including two short breaks and one long lunch break of around 40 minutes. Parents received the results of the subtest of the WISC-III several weeks after testing at the VU.

Most of the families went to the UMCU after they had been to the VU. Average time between testing at the VU and the UMCU was 43 days ranging from 63 days before testing at the VU until 124 days after testing at the VU (SD = 35). The protocol at the UMCU took between 3 to 3.5 hours. At the UMCU a magnetic resonance scan was made of the brain of the children. Prior to the MR scan children were physically examined, weight and height were measured, and they were tested for reading ability and handedness. During scanning of the children, parents were asked to fill out a medical questionnaire about the children.

Table 1.1. *Protocol for testing at VU, UMCU. Overview of hormones and DNA collected*

Testing VU	Hormone and DNA collection (all collected on two consecutive days)
Welcome, short explanation of the research protocol, reception of tubes, offering of drinks and explanation of Raven to the parents	Cortisol in saliva collected at five points (awakening, 15 min, 30 min, 45 min, 12.00 a.m.)
WISC: picture completion, information, coding, similarities, picture arrangement, arithmetic, block design, vocabulary	Testosterone in morning saliva
Delay of gratification	Estrogen in morning urine
Break	LH in morning urine
WISC: object assembly, comprehension, symbol search, digit span	FSH in morning urine
Stroop	Cheek swabs for DNA isolation
Tower of Hanoi	Testing UMCU
Verbal fluency	Three minutes reading test
Break	Handedness inventory
AVLT (a)	Physical examination: Tanner, weight and height
Reading the mind in the eyes	Medical questionnaire
π-inspection task	MRI:
AVLT (b)	SENSE reference scan
Flanker	Scout scan, sagittal T1 weighted
Lunch	Dual Echo - Turbo Spin Echo (DE-TSE) clinical scan, transversal T2 weighted
n-back task	Three Dimensional - Fast Field Echo (3D-FFE) T1 weighted scan
Corsi block tapping test	Diffusion Tensor Imaging (DTI) scan
Raven's Standard Progressive Matrices	Magnetization Transfer Imaging scan

Zygosity

Zygosity was determined at the VU Medical Center based on DNA polymorphisms; eight highly polymorphic di-, tri- and tetranucleotide genetic markers were used. The zygosity testing included a multiplex PCR of markers D2S125, D8S1130, D1S1609, D5S816 and a second multiplex reaction of markers 15 ActC, D21S1437, D7S2846, and D10S1423. These two multiplex PCR reactions were performed essentially by the protocol provided in the website of the Marshfield Institute (http:/ /www.marshmed.org/genetics/). Parents received the results of the zygosity test.

Stage of puberty and hormones

Stage of puberty was physically determined by a trained researcher on the basis of secondary sexual characteristics using the five stages of development devised by Tanner (Marshall & Tanner, 1969; Marshall & Tanner, 1970). Children were asked to collect saliva immediately after waking up. This saliva was later used to measure testosterone levels. Testosterone level in saliva samples was measured employing an immunometric (lumininescention) assay (ILB, Hamburg). The lower limit of detection was 11 pmol/L and inter-assay variation was 25, 15 and 10% for < 20, 50 and 75 pmol/L respectively. FSH, LH, and estrogen levels were determined in morning urine. Total estradiol levels were determined using a competitive (luminiscention) immunoassay (Architect, Abbott Laboratories, Abbott Park., Illinois USA). The intra-assay and inter-assay CVs were 5% and 10% respectively at levels > 150 pmol/L (lower limit of detection) and < 9000 pmol/L (upper limit of detection). FSH and LH levels were measured by means of an immunometric (lumininescention) assay (Architect Abbott Laboratories Diagnostics Division Abbott Park, Illinois USA). Lower limit of detection was for FSH .11 U/L and for LH .1 U/L. Inter-assay was for FSH 6% and 5% for 5 and 18 U/L respectively, and for LH 7 and 6% at 4 and 23 U/L. All assays were carried out by the endocrinological laboratory of clinical chemistry of the VU Medical Center in Amsterdam.

All samples were stored in the refrigerator (4°C) in the family's home until the day of cognitive testing. After taking them to the VU, testosterone samples were stored at -20°C before they went to the laboratory for analysis. Urine samples were immediately brought to the laboratory for analysis. All gonadal hormone levels were assessed by the endocrinological laboratory of clinical chemistry at the VU University Medical Center.

Children were also requested to collect saliva at five later points during the morning using the Salivette sampling device (Starstedt, Rommelsdorf, Germany). These samples were used to measure cortisol levels. The first sample was taken in the morning just before getting up (still lying in bed), and three further samples were taken 15, 30, and 45 minutes after getting up. The last sample was instructed to be taken at noon. The children were instructed to collect saliva on two school days to restrict the awakening time and time of sampling - school starting time is at approximately the same time all over the Netherlands. Each participant was asked to write down the exact sampling time in a time schedule and to note any exceptional events interfering with daily routine. Subjects were

instructed not to brush their teeth before completing saliva sampling to avoid contamination of saliva with blood caused by micro-injuries in the oral cavity. Also, subjects were instructed to thoroughly rinse their mouth with tap water before sampling saliva and not to eat sour food or drink aerated drinks. Subjects were strictly instructed to collect saliva before taking lunch at the last time point. Saliva samples were stored in the refrigerator in the family's home. At the VU, samples were stored at -20°C and consequently sent by courier to the laboratory in Germany (Trier and Düsseldorf) where cortisol in saliva was determined by time-resolved fluorescence immunoassay; inter-assay variability was less than 12%.

Cognitive testing

The cognitive protocol consisted of assessment of intelligence using the Raven Standard Progressive matrices (Raven, 1960) and the WISC– III (Wechsler et al., 2002) with the exception of the subtest *Mazes*. Furthermore, the following aspects of cognitive control were measured: selective attention, planning, verbal fluency and working memory. Two other aspects of memory were tested, namely short and long term memory. In addition, speed of processing was tested and two aspects of social cognition: emotion recognition, and delay of gratification by using candies (Mischel & Metzner, 1962). Additionally, the Raven Advanced Progressive matrices (Raven et al., 1998) was taken by the parents to check for assortment on intelligence. Parents were instructed specifically to take the test on their own, by telling them we were interested in how much they were alike in this test and not in how well they performed.

Stroop

The Stroop test (Stroop, 1935) assesses selective attention; subjects completed 3 cards, each with 10 columns of 10 items and had to name aloud the items on each card, from the top-left corner to the bottom-right corner. Card 1 involved naming the words 'red', 'green', 'yellow' and 'blue' printed in black ink. Card 2 involved naming the colors of squares that are printed in different colors. Card 3 involved naming the ink color that the words 'red', 'green', 'yellow' and 'blue' are printed in. On Card 3 word content and ink color never matched, i.e., all color words were incongruent. Both speed and accuracy were stressed in the instructions. Each card was scored as the time (in seconds, using a stopwatch) to

complete the card and number of mistakes made (if the item was wrongly named or skipped).

Eriksen flanker task

In the Eriksen flanker task (Eriksen & Eriksen, 1974; Eriksen & Schultz, 1979) which measures selective attention, subjects were presented with a horizontal array of five arrows. Subjects were instructed to pay attention to the direction of the center arrow and ignore the four flanking ones. They were told that the cross in the center of the screen between two trials could help them to focus on the middle arrow. Subjects had to press the left key to a left facing central arrow, and the right key to right facing central arrow. The flanking arrows could either all point in the same direction as the target arrow (<<<<< or >>>>>; congruent), or they all pointed in the opposite direction (<<><< or >><>>; incongruent). Children received 40 congruent and 40 incongruent trials in random order after an eight trial practice session. After each ten correct responses a smiley was presented in the center of the screen. On each trial the reaction time (RT) and accuracy were stored.

Tower of Hanoi

To asses planning children were presented a computerized version of the Tower of Hanoi (TOH; Simon, 1975), in which they were asked to solve a set of TOH problems of increasing difficulty level (Klahr & Robinson, 1981; Lehto, 1996). In this version the TOH consisted of 3 pegs with 4 discs in different colors. On top of the screen the children could see a small version of the TOH, which showed the goal position of the discs. Down on the screen was a bigger TOH in the initial position, on which they could move the discs to arrange them into the goal position. Children were told that only one disk was allowed to be moved at a time, that it was forbidden to put a larger disk on a smaller one (this was an impossible move in the computer program, and therefore not counted as a wrong move), and that they could only grab a disc located on top of the peg. It was explained that a disk could be moved from one peg to the other as long as the rules were observed. They were asked to solve the problem within a certain number of moves, as was written on the screen presented before each problem. During the problems the number of moves still to make, was shown in the middle of the screen. They were told that extra moves were not allowed and that if they made a mistake, they again could try to solve the problem from the start.

After a mistake they saw for one second a screen on which was written that they unfortunately made a mistake, before the problem started all over again. They were asked to plan the moves in advance to avoid mistakes. The task started with two practice problems, which could be solved in respectively one and two moves, to make the children familiar with the task and the rules. After that, children were presented eight problems (2, 3, 4, 5, 5, 6, 6 and 7 moves) for each problem the number of mistakes, planning time (time between when a problem was presented and the child starting to make a move) and solution time (time between when a problem was presented and the child solving the problem, excluding the mistake screens) were recorded. Performance was measured by total number of mistakes and total planning and solution time.

Controlled Oral Word Association

The Controlled Oral Word Association task (COWA; a subtest of the Multilingual Aphasia Exam) measures verbal fluency. Per subject category or letter category children were asked to produce as many words as possible in one minute. There were two letter categories, namely ‘r’ and ‘t’ and two subject categories, namely animals and jobs. Total number of correct words per category was recorded.

n-back task

Children had to perform a spatial variant of the *n*-back task to assess visuospatial working memory. The *n*-back used in this protocol was designed after Gevins & Cutillo (1993) and Jansma, Ramsey, Coppola, & Kahn (2000) with increasing levels of difficulty. The children were asked to look at an apple presented on a screen. The apple had four holes in which a caterpillar could appear. Children were told to catch the caterpillar to prevent it from eating the apple, and were instructed to respond to the caterpillar by pushing one of four buttons with the thumb and index finger of both hands. The layout of the four buttons corresponded spatially to the four holes in which the caterpillar could appear. Children had to indicate where the caterpillar was one move back (1-back), two moves back (2-back), or three moves back (3-back). The caterpillar appeared in a hole for one second; after its disappearance there was a warning sound. Children were instructed to respond after this warning sound and could respond until the next caterpillar appeared. Between two caterpillar moves, the apple was empty for one second.

Each level of difficulty was given in sessions of 20 trials. Each condition consisted of a practice session and three sessions in which performance was recorded. Practicing continued until the participants understood the task. The 1-back condition was administered for practice purposes only, performance was recorded on the 2-back and 3-back conditions. Children were motivated during the task by counting the moves of the caterpillar. In the 2-back version the test administrator counted continuously to three and in the 3-back version the administrator counted to four. After each session children received feedback on the number of apples they had saved from the caterpillar (correct responses) and how many had been eaten (incorrect responses). Following the feedback there was a break of 15 seconds. The task requires a continuous response to all stimuli and simultaneous monitoring and update of all movements of the caterpillar. Performance on the task was scored by using the total number of correct responses. Maximum score per condition was 60.

Rey's Auditory Verbal Learning Task

The Rey's Auditory Verbal Learning Task (AVLT; Van den Burg & Kingma, 1999) is a task which measures short term as well as long term verbal memory. A list of 15 unrelated, concrete nouns was presented with a 1 second interval on an audio CD over five learning trials. After each presentation the children had to name as many of the presented words as possible. Next, after a delay interval of some 20–30 minutes (in which the Reading the Mind in the Eyes and the π-inspection task were administered) and with no further presentations of the list, delayed recall was assessed. Performance was measured after each trial by number of correct words, perseverations, intrusions, and double intrusions.

Corsi block tapping task

The Corsi block tapping task (Corsi, 1974) was included to assess short-term spatial memory. Children sat in front of a touch screen monitor on which nine white blocks were displayed unevenly across a gray screen. In succession a number of blocks turned red for one second, after which the screen was blank for three seconds. After reappearance of the blocks, the child had to tap the blocks on the screen in the same sequence in which they had changed color before. When a block was tapped, the block would turn red and stay that way until the end of the run. The computer registered each tap. Each child was given two

practice runs. In these practice runs each person had to memorize two blocks. Immediately after the practice runs the actual test was administered. Actual testing started with a series of two blocks. After every five runs the item length was increased by one block. The test was terminated when the child responded incorrectly to three out of five runs of the same length. The maximum number of blocks that could turn red in succession was nine. Performance was measured by total number of correct runs.

Π- inspection task

The π-inspection task (Brand & Deary, 1982; Luciano et al., 2001) measures speed of processing. The version used in the protocol was designed after (Luciano et al., 2001). For this task subjects had to identify the longer of two lines which were presented by the test administrator as worms that the subjects needed to catch. This task was complicated by the fact that the worms burrowed quickly into the ground (i.e., disappeared quickly from the screen). If subjects caught five worms they had enough worms to catch a fish, which would appear at the lower left-hand side of the screen. It was stressed that it was important to be accurate and that it did not matter how long it took them to catch the worms.

The probability of the longer line appearing on the left or right was equal. The stimulus duration ranged between 14.2 and 2000 ms. A dynamic mask, consisting of two vertical lines shaped as lightning bolts, immediately followed the stimulus to limit further stimulus processing. On each trial, a fixation cross appeared in the Center of the screen for 1 sec (an alerting beep was sounded at the onset of the dot), followed by a blank screen. The π figure was then presented, and the participant's first response (left or right) was noted. Stimulus duration depended on the response of the subject. For every four correct consecutive responses the stimulus duration was decreased, and for every incorrect response the stimulus duration was increased by a step size depending on previous performance (for details, see Luciano et al., 2001). When minimal stimulus duration was achieved, the protocol stopped. If the minimum was not reached within 96 trials the protocol also stopped. This way, for each subject the minimal stimulus duration time could be assessed. On each trial the stimulus duration and whether or not the correct key was pressed, was stored.

Reading the Mind in the Eyes task

The Reading the Mind in the Eyes task assesses emotion recognition (Baron-Cohen, Wheelwright, Hill, Raste, & Plumb, 2001) In this test, children were presented with a series of 36 photographs of the eye-region of the face of 36 actors and actresses. They were asked to choose which of four words best described what the person in the photograph is thinking or feeling (Baron-Cohen et al., 2001; Baron-Cohen, Weelwright, Spong, Scahill, & Lawson, 2005). Before the test started they were told that some of the pictures were easy and other ones were difficult, and that if they did not know the correct answer they had to guess. They were explicitly instructed to ask if they did not know the meaning of the words, because then the experimenter would explain the words. All words were read aloud by the experimenter. Performance was measured by total number of correct descriptions of the emotional state of the actors in the pictures.

Behavioral data

Questionnaires were mailed to parents of twins at ages 0, 2, 3, 5, and 7 years, assessing a wide variety of health and behavior problems in the twins (not in the siblings). In addition, parents were asked about their own height and weight, place of birth, religious background, educational attainment and their socioeconomic status (SES). At registration, data were collected on birth weight and height (Van Baal & Boomsma, 1998) pregnancy and birth complications, medication, smoking and alcohol use during pregnancy, and malformations. When the twins were aged 2, growth data as measured by the Youth Health Services (Boomsma et al., 1992) were collected as well as information about breast feeding, motor development and behavior problems. Questionnaires from age 3 onwards were targeted at the development of psychopathology. At age 3 the CBCL2-3 (Child Behavior Checklist; Achenbach, 1992; Koot, Van den Oord, Verhulst, & Boomsma, 1997) was sent to both parents. Behavioral and emotional syndromes included overactive, oppositional and aggressive behavior, withdrawn/ depressed and anxious behavior, sleep and somatic problems. In addition, the mother of the twins was asked about health problems, and growth, height and weight, and both parents were asked about their religion, profession and education. At age 5 detailed questions were asked about health and behavioral problems, based on a selection of items from the Devereux Child Behavior Rating Scale (Van Beijsterveldt, Verhulst, & Boomsma, 2001). At age 7 years the CBCL4-18 (Achenbach,

1991a; Conners, Sitarenios, Parker, & Epstein, 1998b) was collected. The syndromes overlapped with the CBCL2-3 to a large extent. Teacher data (TRF) were for the first time collected at age 7. Teachers completed the TRF (Achenbach, 1991b) and the Conners' Teaching Rating Scale (Conners, Sitarenios, Parker, & Epstein, 1998a).

At the day of testing at the VU, mothers were asked to fill in a survey concerning the behavior of the sibling of the twins (age appropriate CBCL). In Table 1.2 the number of available surveys in the whole sample is presented. Data on socio-economic status (SES) from the survey mailed out when the twins were 7 years old were analyzed to determine SES. If for some reason SES measured at age 7 of the twins was not available, SES measured at age 3 of the twins was used. SES was based on a full description of the occupation of the parents and classified using a 5 point scale, according to the system used by Statistics Netherlands in which level 1 represents primary school only and level 5 university degree or higher (Fengler, Joung, & Mackenbach, 1997). The highest SES of the two parents determined the SES of the twin pair.

Table 1.2. *Number of available questionnaires per family in the sample*

age	Maternal / Paternal ratings twins	Teacher ratings twins oldest / youngest
0	112 /-	-
2	97 /-	-
3	97 / 84	-
5	110 / 99	-
7	96 / 73	69 / 70
sibs	93 / -	-

Reading ability

One subtest of the 'three minutes reading test' (Cito, 1995; the norms date from 2003) was administered. In one minute subjects had to read out loud as many words as possible from a card containing 120 words.

Handedness

Handedness of each subject was determined on basis of the 10-item version of the Edinburgh Handedness Inventory (Oldfield, 1971).

MRI

Before entering the MR scanner, subjects first practiced in a dummy scanner to get used to the real MR scanner. The imitation scanner is a copy of the real version, except there is no magnetic field. In this way, the child could practice with lying in a tube with a diameter of 60 cm, without moving and hearing loud noises. At their own pace, in the presence of their parents and the investigator, the children could gradually get used to all aspects of MR-acquisition. Only when children were completely comfortable with the procedure, was the actual MR-acquisition carried out. The practice procedure was in accordance with a special protocol for the imitation scanner

Magnetic resonance images were acquired on a Philips Achiva scanner operating at 1.5 Tesla in all subjects. A SENSE head-8 coil was used. Nine image-sequences of the whole head were acquired (see Table 1.1): a short survey scan for immediate verification of head positioning, a clinical scan that was used for neurodiagnostic evaluation and six scientific for quantitative measurements. The total scanning procedure took approximately 33 minutes. In order to make the procedure as comfortable as possible, subjects were able to watch a DVD they brought themselves or listen to their favorite music during scanning. The subjects were always able to communicate with the experimenter or laboratory worker using an intercom. Furthermore, either the experimenter or a parent could be in the scanning room.

Apart from localizer-scans and radiodiagnostic scans, three different scientific scans were carried out: a T1-weighted scan, a Diffusion Tensor Imaging (DTI), and a Magnetization Transfer Ratio (MTR; for a complete overview of the scanning protocol, see Table 1.1). The T1-weighted scan is used for volumetric measurements (total brain, gray and white matter, lateral and 3rd ventricle and cerebellar volumes), and Voxel Based Morphometry (VBM) analysis of regional densities in the brain. The VBM technique is used to detect focal differences in gray and white matter density in one group relative to another group. The DTI scans can be used for fiber tracking measurements. Using this method it is possible to gain information about white matter maturation and axon

orientation in the developing brain. Finally, the MTR scan is used for measuring myelination of fibers. The MTR-scan specifically picks up the signal of fat molecules in brain tissue and therefore offers the opportunity to quantify the amount of myelinated axons or the thickness of the white matter bundles.

Sample Characteristics

Participants

Fifty-two percent (N = 112) of the initially invited families agreed to participate. The most important reasons for parents to refuse to participate were: no time or too much effort (52), children themselves did not want to participate or were scared (13), parents did not want their children to participate in this research (10), and problems with children (9; like hospitalization, psychiatric problems of which we were not aware, and divorce of parents). There was no significant difference between the educational level of mothers who participated and who did not participate in the study (F (1,195) = .68, p = .41). Of these 112 families, 103 twin pairs had full siblings who wanted to participate in the research and did not meet exclusion criteria. Parents signed informed consent statements for the children as well as themselves. Children signed consent forms for themselves (see appendix). Parents were financially compensated for their travel expenses and children received two presents worth €10,-, each one after a testing day.

Mean age of the twins at time of cognitive assessment was 9.1 years, ranging from 8.9 to 9.5 years. There were 23 monozygotic male (MZM), 23 dizygotic male (DZM), 25 monozygotic female (MZF), 21 dizygotic female (DZF) and 20 dizygotic pairs of opposite sex (DOS). For the same sex twin pairs, zygosity determination was based on DNA polymorphisms (90 twin pairs), or on questionnaire items and visual inspection (2 pairs; Rietveld et al., 2000). Mean age of the sibs (N =103) was 11.9 years ranging from 9.9 to 14.9, of whom 59 were female. The mean age of the fathers was 43.9 (SD = 4.2 years), and of the mothers 41.8 (SD = 3.3 years).

Of the 327 subjects 314 (coming from 107 families) came to the UMCU for an MRI scan. Of these 107 families 100 came with siblings, of these siblings 56 were female. Of the total 314 subjects, 11 children did not have a scan-session due to dental braces and 2 because children were too afraid. Therefore, the total number of scanned subjects was 301.

Figure 1.2. Residences of families participating in the research

This group consisted of 22 MZM, 22 DZM, 23 MZF, 21 DZF and 19 DOS twin pairs. Mean age of the twins at time of the MRI scan was 9.2 years (9.0-9.6) and of the sibs was 12.0 (10.0-14.9).

The 112 families participating in the study came from all over the Netherlands as is depicted in Figure 1.2 Data on SES were available for 104 of these families. Median and modal SES was 3, ranging from 2 until 5 (on a scale ranging form 1 to 5, see Figure 1.3). In 2004 in the Netherlands, 9.5% of the Dutch adults belonged to category 1 and 25.4% to category 4 and 5 together. Therefore, SES is somewhat higher for this sample than for the Dutch population (Statistics Netherlands, 2004).

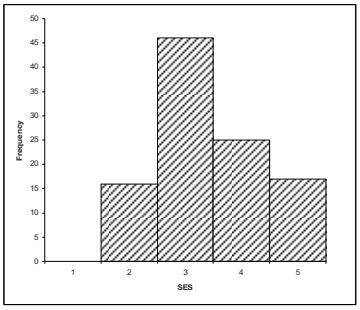


Figure1.3. Distribution of SES in the sample. SES ranges from 1 to 5, in which level 1 represents primary school only and level 5 university degree or higher

Pubertal status

Table 1.3a and b present the Tanner stages of pubertal development of the children in the sample. As can be seen in Table 1.3a most of the twin boys had not reached puberty on the day of testing in the UMCU. Of the 41 brothers of the twins about half had not reached puberty. Also in the group of twin girls most of the girls had not reached puberty as can be seen in Table 1.3b. However, as could be expected, the number of girls that had reached stage 2 was higher than the number of boys that had reached stage 2. Of the 54 sisters of the twins most had reached puberty.

Table 1.3a. *Frequencies of reached Tanner stages in twin boys and older brothers*

		Twins			Siblings		
		Penis Growth	Pubic Hair	Size of Testes	Penis Growth	Pubic Hair	Size of Testes
Tanner stage	1	102	105	105	19	20	22
	2	5	2	2	15	13	15
	3	0	0	0	4	5	4
	4	0	0	0	3	2	0
	5	0	0	0	0	1	0

Table 1.3b. *Frequencies of reached Tanner stages in twin girls and older sisters*

		Twins		Siblings	
		Breast Development	Pubic Hair	Breast Development	Pubic Hair
Tanner stage	1	89	90	0	7
	2	20	16	25	19
	3	0	3	14	10
	4	0	0	12	12
	5	0	0	3	5

Weight, height and BMI

There was no difference in weight, height and BMI between the boys and girls in the sample ($\Delta\chi^2$= 0.314, p = .58; $\Delta\chi^2$= 0.935, p = .33; $\Delta\chi^2$= 2.086, p = .15). When corrected for age, there were no differences in means between twins and siblings. SD was higher in the siblings than in the twins for all three measures.

Handedness

Eighty-nine percent of the children were right handed, 11% were left handed, and 2% were ambidextrous.

Reference List

Achenbach, T.M. (1991a) *Manual for the Child Behavior Checklist/ 4-18 and 1991 Profile.* Burlington, VT: University of Vermont, Department of Psychiatry

Achenbach, T.M. (1991b) *Manual for the Teacher's Report Form and 1991 Profile.* Burlington, VT: University of Vermont, Department of Psychiatry

Achenbach, T.M. (1992) *Manual for the Child Behavior Checklist/ 2-3 and 1992 profile.* Burlington, VT: University of Vermont, Department of Psychiatry

Ackerman, P L, Beier, M E, & Boyle, M O (2005). Working memory and intelligence: the same or different constructs? *Psychological Bulletin, 131*, 30-60.

Adleman, N E, Menon, V, Blasey, C M, White, C D, Warsofsky, I S, Glover, G H, & Reiss, A L (2002). A developmental fMRI study of the Stroop color-word task. *Neuroimage, 16*, 61-75.

Allen, J S, Damasio, H, Grabowski, T J, Bruss, J, & Zhang, W (2003). Sexual dimorphism and asymmetries in the gray-white composition of the human cerebrum. *Neuroimage, 18*, 880-894.

Alloway, T P, Gathercole, S E, & Pickering, S J (2006). Verbal and visuospatial short-term and working memory in children: are they separable? *Child Development, 77*, 1698-1716.

Anderson, C A, Duffy, D L, Martin, N G, & Visscher, P M (2007). Estimation of variance components for age at menarche in twin families. *Behavior Genetics, 37*, 668-677.

Ando, J, Ono, Y, & Wright, M J (2001). Genetic structure of spatial and verbal working memory. *Behavior Genetics, 31*, 615-624.

Baaré, W F, Hulshoff Pol, H E, Boomsma, D I, Posthuma, D, De Geus, E J, Schnack, H G, Van Haren, N E, Van Oel, C J, & Kahn, R S (2001). Quantitative genetic modeling of variation in human brain morphology. *Cerebral Cortex, 11*, 816-824.

Baddeley, A (2003). Working memory: looking back and looking forward. *Nature Reviews Neuroscience, 4*, 829-839.

Barnea-Goraly, N, Menon, V, Eckert, M, Tamm, L, Bammer, R, Karchemskiy, A, Dant, C C, & Reiss, A L (2005). White matter development during childhood and adolescence: a cross-sectional diffusion tensor imaging study. *Cerebral Cortex, 15*, 1848-1854.

Baron-Cohen, S, Wheelwright, S, Hill, J, Raste, Y, & Plumb, I (2001). The "Reading the Mind in the Eyes" Test revised version: a study with normal adults, and adults with Asperger syndrome or high-functioning autism. *Journal of Child Psychology and Psychiatry, 42*, 241-251.

Baron-Cohen, S, Weelwright, S, Spong, A, Scahill, V, & Lawson, J (2005). Studies of theory of mind:Are intuitive physics and intuitive psychology independent? *Journal of Developmental and Learning Disorders, 5*, 47-78.

Bartels, M, Rietveld, M J, Van Baal, G C, & Boomsma, D I (2002). Heritability of educational achievement in 12-year-olds and the overlap with cognitive ability. *Twin Research, 5*, 544-553.

Boomsma, D I, De Geus, E J C, Vink, J M, Stubbe, J H, Distel, M A, Hottenga, J J, Van Beijsterveldt, C E M, Hudziak, J J, Bartels, M, & Willemsen, G (2006). Netherlands Twin Register: from twins to twin families. *Twin Research and Human Genetics, 9*, 849-857.

Boomsma, D I & Molenaar, P C (1986). Using LISREL to analyze genetic and environmental covariance structure. *Behavior Genetics, 16*, 237-250.

Boomsma, D I, Orlebeke, J F, & Van Baal, G C (1992). The Dutch Twin Register: growth data on weight and height. *Behavior Genetics, 22*, 247-251.

Boomsma, D I, Vink, J M, Van Beijsterveldt, T C, De Geus, E J, Beem, A L, Mulder, E J, Derks, E M, Riese, H, Willemsen, G A, Bartels, M, Van den Berg, M, Kupper, N H, Polderman, T J, Posthuma, D, Rietveld, M J, Stubbe, J H, Knol, L I, Stroet, T, & Van Baal, G C (2002). Netherlands Twin Register: A focus on longitudinal research. *Twin Research, 5*, 401-406.

Bouchard, T J, Jr., Lykken, D T, McGue, M, Segal, N L, & Tellegen, A (1990). Sources of human psychological differences: the Minnesota Study of Twins Reared Apart. *Science, 250*, 223-228.

Bouchard T.J., Jr. and Propping P. (Ed.) (1993) *Twins as a tool for behavioral genetics.* Chichester, UK: Wiley

Brand, C.R. & Deary, I.J. (1982). Intelligence and "inspection time.". In: H.J. Eysenck *A model for intelligence.*New York: Springer-Verlag

Carmelli, D, Swan, G E, DeCarli, C, & Reed, T (2002). Quantitative genetic modeling of regional brain volumes and cognitive performance in older male twins. *Biological Psychology, 61*, 139-155.

Casey, B J, Giedd, J N, & Thomas, K M (2000). Structural and functional brain development and its relation to cognitive development. *Biological Psychology, 54*, 241-257.

Casey, B J, Tottenham, N, Liston, C, & Durston, S (2005). Imaging the developing brain: what have we learned about cognitive development? *Trends in Cognitive Sciences, 9*, 104-110.

Castellano, J M, Navarro, V M, Fernandez-Fernandez, R, Nogueiras, R, Tovar, S, Roa, J, Vazquez, M J, Vigo, E, Casanueva, F F, Aguilar, E, Pinilla, L, Dieguez, C, & Tena-Sempere, M (2005). Changes in hypothalamic KiSS-1 system and restoration of pubertal activation of the reproductive axis by kisspeptin in undernutrition. *Endocrinology, 146*, 3917-3925.

Cito. (1995) *Drie Minuten Test.* Arnhem: Cito

Colom, R, Flores-Mendoza, C, Quiroga, M A, & Privado, J (2005). Working memory and general intelligence: The role of short-term storage. *Personality and Individual Differences, 39*, 1005-1014.

Colom, R, Rebollo, I, Palacios, A, Juan-Espinosa, M, & Kyllonen, P C (2004). Working memory is (almost) perfectly predicted by g. *Intelligence, 32*, 277-296.

Conners, C K, Sitarenios, G, Parker, J D, & Epstein, J N (1998a). Revision and restandardization of the Conners Teacher Rating Scale (CTRS-R): factor structure, reliability, and criterion validity. *Journal of Abnormal Child Psychology, 26*, 279-291.

Conners, C K, Sitarenios, G, Parker, J D, & Epstein, J N (1998b). The revised Conners' Parent Rating Scale (CPRS-R): factor structure, reliability, and criterion validity. *Journal of Abnormal Child Psychology, 26*, 257-268.

Conway, A R A, Kane, M J, & Engle, R W (2003). Working memory capacity and its relation to general intelligence. *Trends in Cognitive Sciences, 7*, 547-552.

Cowell, P E, Turetsky, B I, Gur, R C, Grossman, R I, Shtasel, D L, & Gur, R E (1994). Sex differences in aging of the human frontal and temporal lobes. *Journal of Neuroscience, 14*, 4748-4755.

Davidson, M C, Amso, D, Anderson, L C, & Diamond, A (2006). Development of cognitive control and executive functions from 4 to 13 years: Evidence from manipulations of memory, inhibition, and task switching. *Neuropsychologia, 44*, 2037-2078.

De Geus, E J, Wright, M J, Martin, N G, & Boomsma, D I (2001). Genetics of brain function and cognition. *Behavior Genetics, 31*, 489-495.

De Geus, E J & Boomsma, D I (2001). A genetic neuroscience approach to human cognition. *European Psychologist, 6*, 241-253.

De Moor, M H M, Boomsma, D I, Stubbe, J H, Willemsen, G, & De Geus, E J C (in press). Testing causality in the association between regular exercise and symptoms of anxiety and depression. *Archives of General Psychiatry.*

De Ribaupierre, A & Lecerf, T (2006). Relationships between working memory and intelligence from a developmental perspective: Convergent evidence from a neo-Piagetian and a psychometric approach. *European Journal of Cognitive Psychology, 18*, 109-137.

Deary, I J (2001). Human intelligence differences: towards a combined experimental-differential approach. *Trends in Cognitive Sciences, 5*, 164-170.

Durston, S, Hulshoff Pol, H E, Casey, B J, Giedd, J N, Buitelaar, J K, & Van Engeland, H (2001). Anatomical MRI of the developing human brain: what have we learned? *Journal of the American Academy of Child and Adolescent Psychiatry, 40*, 1012-1020.

Durston, S & Casey, B J (2006). What have we learned about cognitive development from neuroimaging? *Neuropsychologia, 44*, 2149-2157.

Eaves, L, Silberg, J, Foley, D, Bulik, C, Maes, H, Erkanli, A, Angold, A, Costello, E J, & Worthman, C (2004). Genetic and environmental influences on the relative timing of pubertal change. *Twin Research, 7*, 471-481.

Ebling, F J (2005). The neuroendocrine timing of puberty. *Reproduction, 129*, 675-683.

Eriksen, B A & Eriksen, C-W (1974). Effects of noise letters upon the identification of a target letter in a nonsearch task. *Perception & Psychophysics, 16*, 143-149.

Eriksen, C W & Schultz, D W (1979). Information processing in visual search: a continuous flow conception and experimental results. *Perception & Psychophysics, 25*, 249-263.

Fengler, M., Joung, I., & Mackenbach, J. (1997). Sociaal-demografische kenmerken en gezondheid: hun relatief belang en onderlinge relaties. In: J.P. Mackenbach & H. Verkleij *Volksgezondheid Toekomst Verkenning 1997, II Gezondheidsverschillen.* (pp. 49-67) Maarssen: Elsevier/De Tijdstroom

Finkel, D. & Pedersen, N.L. (2001). Sources of environmental influence on cognitive abilities in adulthood. In: E.L. Grigorenko & R.J. Sternberg *Family environment and intellectual functioning* (pp. 173-194) Mahwah: Lawrence Erlbaum Associates

Frangou, S, Chitins, X, & Williams, S C R (2004). Mapping IQ and gray matter density in healthy young people. *Neuroimage, 23*, 800-805.

Fry, A F & Hale, S (2000). Relationships among processing speed, working memory, and fluid intelligence in children. *Biological Psychology, 54*, 1-34.

Gathercole, S E, Pickering, S J, Ambridge, B, & Wearing, H (2004). The structure of working memory from 4 to 15 years of age. *Developmental Psychology, 40*, 177-190.

Gathercole, S E, Alloway, T P, Willis, C, & Adams, A M (2006). Working memory in children with reading disabilities. *Journal of Experimental Child Psychology, 93*, 265-281.

Geluk, A & Hol, J (2001). Samen of Apart? Meerlingkinderen naar school, peuterspeelzaal of kinderopvang [Together or Apart? Multiples to school, playground or daycare.].

Gevins, A & Cutillo, B (1993). Spatiotemporal dynamics of component processes in human working memory. *Electroencephalography and Clinical Neurophysiology., 87*, 128-143.

Giedd, J N (2004). Structural magnetic resonance imaging of the adolescent brain. *Annals of the New York Acadamy of Sciences, 1021*, 77-85.

Giedd, J N, Blumenthal, J, Jeffries, N O, Castellanos, F X, Liu, H, Zijdenbos, A, Paus, T, Evans, A C, & Rapoport, J L (1999). Brain development during childhood and adolescence: a longitudinal MRI study. *Nature Neuroscience, 2*, 861-863.

Giedd, J N, Clasen, L S, Lenroot, R, Greenstein, D, Wallace, G L, Ordaz, S, Molloy, E A, Blumenthal, J D, Tossell, J W, & Stayer, C (2006). Puberty-related influences on brain development. *Molecular and Cellular Endocrinology, In Press, Corrected Proof*,

Giedd, J N, Snell, J W, Lange, N, Rajapakse, J C, Casey, B J, Kozuch, P L, Vaituzis, A C, Vauss, Y C, Hamburger, S D, Kaysen, D, & Rapoport, J L (1996a). Quantitative magnetic resonance imaging of human brain development: ages 4-18. *Cerebral Cortex, 6*, 551-560.

Giedd, J N, Vaituzis, A C, Hamburger, S D, Lange, N, Rajapakse, J C, Kaysen, D, Vauss, Y C, & Rapoport, J L (1996b). Quantitative MRI of the temporal lobe, amygdala, and hippocampus in normal human development: ages 4-18 years. *The Journal of Comparative Neurology, 366*, 223-230.

Gogtay, N, Giedd, J N, Lusk, L, Hayashi, K M, Greenstein, D, Vaituzis, A C, Nugent, T F, III, Herman, D H, Clasen, L S, Toga, A W, Rapoport, J L, & Thompson, P M (2004). Dynamic mapping of human cortical development during childhood through early adulthood. *Proceedings of the National Academy of Sciences, 101*, 8174-8179.

Good, C D, Johnsrude, I, Ashburner, J, Henson, R N, Friston, K J, & Frackowiak, R S (2001). Cerebral asymmetry and the effects of sex and handedness on brain structure: a voxel-based morphometric analysis of 465 normal adult human brains. *Neuroimage, 14*, 685-700.

Grudnik, J L & Kranzler, J H (2001). Meta-analysis of the relationship between intelligence and inspection time. *Intelligence, 29*, 523-535.

Grumbach, M.M. & Styne, D. (1998). Puberty: ontogeny, neuroendocrinology, physiology, and disorders. In: P.R. Larsen, H.M. Kronenberg, S. Melmed, & K.S. Polonsky *Williams Textbook of Endocrinology* (pp. 1115-1286) Philadelphia, W.B. Saunders.

Guttman, R (1974). Genetic analysis of analytical spatial ability: Raven's progressive matrices. *Behavior Genetics, 4*, 273-284.

Haier, R J, Jung, R E, Yeo, R A, Head, K, & Alkire, M T (2004). Structural brain variation and general intelligence. *Neuroimage, 23*, 425-433.

Hansell, N K, Wright, M J, Luciano, M, Geffen, G M, Geffen, L B, & Martin, N G (2005). Genetic covariation between event-related potential (ERP) and behavioral non-ERP measures of working-memory, processing speed, and IQ. *Behavior Genetics, 35*, 695-706.

Harris, J A, Vernon, P A, & Boomsma, D I (1998). The heritability of testosterone: a study of Dutch adolescent twins and their parents. *Behavior Genetics, 28*, 165-171.

Harvey, P H & Krebs, J R (1990). Comparing brains. *Science, 249*, 140-146.

Hay, D A (2004). Together or apart? *Twin Research, 7*, iii-iv.

Hoekstra, R A, Bartels, M, & Boomsma, D I (2006). Heritability of testosterone levels in 12-year-old twins and its relation to pubertal development. *Twin Research and Human Genetics, 9*, 558-565.

Hoekstra, R A, Bartels, M, & Boomsma, D I (2007). Longitudinal genetic study of verbal and nonverbal IQ from early childhood to young adulthood. *Learning and Individual Differences, 17*, 97-114.

Huizinga, M, Dolan, C V, & Van der Molen, M W (2006). Age-related change in executive function: Developmental trends and a latent variable analysis. *Neuropsychologia, 44*, 2017-2036.

Hulshoff Pol, H E, Schnack, H G, Posthuma, D, Mandl, R C, Baare, W F, Van Oel, C, Van Haren, N E, Collins, D L, Evans, A C, Amunts, K, Burgel, U, Zilles, K, De Geus, E, Boomsma, D I, & Kahn, R S (2006). Genetic contributions to human brain morphology and intelligence. *Journal of Neuroscience, 26*, 10235-10242.

Jacobs, N, Van Gestel, S, Derom, C, Thiery, E, Vernon, P, Derom, R, & Vlietinck, R (2001). Heritability estimates of intelligence in twins: effect of chorion type. *Behavior Genetics, 31*, 209-217.

Jansma, J M, Ramsey, N F, Coppola, R, & Kahn, R S (2000). Specific versus nonspecific brain activity in a parametric n-back task. *Neuroimage, 12*, 688-697.

Jernigan, T L & Tallal, P (1990). Late childhood changes in brain morphology observable with MRI. *Developmental Medicine and Child Neurology, 32*, 379-385.

Jernigan, T L, Trauner, D A, Hesselink, J R, & Tallal, P A (1991). Maturation of human cerebrum observed in vivo during adolescence. *Brain, 114 (Pt 5)*, 2037-2049.

Jinks, J L & Fulker, D W (1970). Comparison of the biometrical genetical, MAVA, and classical approaches to the analysis of the human behavior. *Psychological Bulletin, 73*, 311-349.

Kail, R (1991). Developmental change in speed of processing during childhood and adolescence. *Psychological Bulletin, 109*, 490-501.

Kail, R & Hall, L K (2001). Distinguishing short-term memory from working memory. *Memory and Cognition, 29*, 1-9.

Kane, M J, Hambrick, D Z, Tuholski, S W, Wilhelm, O, Payne, T W, & Engle, R W (2004). The generality of working memory capacity: a latent-variable approach to verbal and visuospatial memory span and reasoning. *Journal of Experimental Psychology: General, 133*, 189-217.

Kawata, M (1995). Roles of steroid hormones and their receptors in structural organization in the nervous system. *Neuroscience Research, 24*, 1-46.

Kendler, K S, Neale, M C, Kessler, R C, Heath, A C, & Eaves, L J (1993). A test of the equal-environment assumption in twin studies of psychiatric illness. *Behavior Genetics, 23*, 21-27.

Klahr, D & Robinson, M (1981). Formal assessment of problem-solving and planning processes in preschool children. *Cognitive Psychology, 13*, 113-148.

Koot, H M, Van den Oord, E J, Verhulst, F C, & Boomsma, D I (1997). Behavioral and emotional problems in young preschoolers: Cross-cultural testing of the validity of the Child Behavior Checklist/2-3. *Journal of Abnormal Child Psychology, 25*, 183-196.

Lamm, C, Zelazo, P D, & Lewis, M D (2006). Neural correlates of cognitive control in childhood and adolescence: Disentangling the contributions of age and executive function. *Neuropsychologia, 44*, 2139-2148.

Le Goualher, G, Argenti, A M, Duyme, M, Baare, W F, Hulshoff Pol, H E, Boomsma, D I, Zouaoui, A, Barillot, C, & Evans, A C (2000). Statistical sulcal shape comparisons: application to the detection of genetic encoding of the central sulcus shape. *Neuroimage, 11*, 564-574.

Lehto, J (1996). Are executive function tests dependent on working memory capacity? *Quarterly Journal of Experimental Psychology: Human Experimental Psychology, 49A*, 29-50.

Lemaitre, H, Crivello, F, Grassiot, B, Alperovitch, A, Tzourio, C, & Mazoyer, B (2005). Age- and sex-related effects on the neuroanatomy of healthy elderly. *Neuroimage, 26*, 900-911.

Lenroot, R K, Gogtay, N, Greenstein, D K, Wells, E M, Wallace, G L, Clasen, L S, Blumenthal, J D, Lerch, J, Zijdenbos, A P, Evans, A C, Thompson, P M, & Giedd, J N (2007a). Sexual dimorphism of brain developmental trajectories during childhood and adolescence. *Neuroimage, 36*, 1065-1073.

Lenroot, R K, Schmitt, J E, Ordaz, S J, Wallace, G L, Neale, M C, Lerch, J P, Kendler, K S, Evans, A C, & Giedd, J N (2007b). Differences in genetic and environmental influences on the human cerebral cortex associated with development during childhood and adolescence. *Human Brain Mapping,*

Levin, H S, Culhane, K A, Hartmann, J, Evankovich, K, Mattson, A J, Harward, H, Ringholz, G, Ewing-Cobbs, L, & Fletcher, J M (1991). Developmental changes in performance on tests of purported frontal lobe functioning. *Developmental Neuropsychology, 7*, 377-395.

Li, H J, Ji, C Y, Wang, W, & Hu, Y H (2005). A twin study for serum leptin, soluble leptin receptor, and free insulin-like growth factor-I in pubertal females. *Journal of Clininical Endocrinology and Metabolism, 90*, 3659-3664.

Livingston, R B, Jennings, E, Reynolds, C R, & Gray, R M (2003). Multivariate analyses of the profile stability of intelligence tests: high for IQs, low to very low for subtest analyses. *Archives of Clinical Neuropsychology, 18*, 487-507.

Loesch, D Z, Hopper, J L, Rogucka, E, & Huggins, R M (1995). Timing and genetic rapport between growth in skeletal maturity and height around puberty: similarities and differences between girls and boys. *American Journal of Human Genetics, 56*, 753-759.

Luciana, M, Conklin, H M, Hooper, C J, & Yarger, R S (2005). The development of nonverbal working memory and executive control processes in adolescents. *Child Development, 76*, 697-712.

Luciano, M, Smith, G A, Wright, M J, Geffen, G M, Geffen, L B, & Martin, N G (2001). On the heritability of inspection time and its covariance with IQ: A twin study. *Intelligence, 29*, 443-457.

Luciano, M., Wright, M.J., Smith, G.A., Geffen, G.M., Geffen, L.B., & Martin, N.G. (2002). Genetic covariance between processing speed and IQ. In: R. Plomin, J.C. DeFries, I.W. Craig, & P. McGuffin *Behavioral Genetics in the Post Genomic Era* (pp. 163-181) Washington D.C.: American Psychological Association

Luna, B, Garver, K E, Urban, T A, Lazar, N A, & Sweeney, J A (2004). Maturation of cognitive processes from late childhood to adulthood. *Child Development, 75*, 1357-1372.

Luo, D, Petrill, S A, & Thompson, L A (1994). An exploration of genetic g: Hierarchical factor analysis of cognitive data from the western reserve twin project. *Intelligence, 18*, 335-347.

Marshall, W A & Tanner, J M (1969). Variations in pattern of pubertal changes in girls. *Archives of Disease in Childhood, 44*, 291-303.

Marshall, W A & Tanner, J M (1970). Variations in the pattern of pubertal changes in boys. *Archives of Disease in Childhood, 45*, 13-23.

Martin, N G & Eaves, L J (1977). The genetical analysis of covariance structure. *Heredity, 38*, 79-95.

Mascie-Taylor, C G (1989). Spouse similarity for IQ and personality and convergence. *Behavior Genetics, 19*, 223-227.
McDaniel, M A (2005). Big-brained people are smarter: A meta-analysis of the relationship between in vivo brain volume and intelligence. *Intelligence, 33*, 337-346.
Meyer, J M, Eaves, L J, Heath, A C, & Martin, N G (1991). Estimating genetic influences on the age-at-menarche: a survival analysis approach. *American Journal of Human Genetics, 39*, 148-154.
Mischel, W & Metzner, R (1962). Preference for delayed reward as a function of age, intelligence, and length of delay interval. *Journal of Abnormal and Social Psychology, 64*, 425-431.
Nokelainen, P & Flint, J (2002). Genetic effects on human cognition: lessons from the study of mental retardation syndromes. *Journal of Neurology, Neurosurgery, and Psychiatry, 72*, 287-296.
Nopoulos, P, Flaum, M, O'Leary, D, & Andreasen, N C (2000). Sexual dimorphism in the human brain: evaluation of tissue volume, tissue composition and surface anatomy using magnetic resonance imaging. *Psychiatry Research, 98*, 1-13.
Ojeda, S R, Lomniczi, A, Mastronardi, C, Heger, S, Roth, C, Parent, A S, Matagne, V, & Mungenast, A E (2006). Minireview: the neuroendocrine regulation of puberty: is the time ripe for a systems biology approach? *Endocrinology, 147*, 1166-1174.
Oldfield, R C (1971). The assessment and analysis of handedness: The Edinburgh inventory. *Neuropsychologia, 9*, 97-113.
Olesen, P J, Nagy, Z, Westerberg, H, & Klingberg, T (2003). Combined analysis of DTI and fMRI data reveals a joint maturation of white and grey matter in a fronto-parietal network. *Cognitive Brain Research, 18*, 48-57.
Palmert, M R & Boepple, P A (2001). Variation in the timing of puberty: clinical spectrum and genetic investigation. *Journal of Clininical Endocrinology and Metabolism, 86*, 2364-2368.
Palmert, M R & Hirschhorn, J N (2003). Genetic approaches to stature, pubertal timing, and other complex traits. *Molecular Genetics and Metabolism, 80*, 1-10.
Paus, T, Collins, D L, Evans, A C, Leonard, G, Pike, B, & Zijdenbos, A (2001). Maturation of white matter in the human brain: a review of magnetic resonance studies. *Brain Research Bulletin, 54*, 255-266.
Paus, T, Zijdenbos, A, Worsley, K, Collins, D L, Blumenthal, J, Giedd, J N, Rapoport, J L, & Evans, A C (1999). Structural maturation of neural pathways in children and adolescents: in vivo study. *Science, 283*, 1908-1911.
Pedersen, N L, Plomin, R, & McClearn, G E (1994). Is there G beyond g? (Is there genetic influence on specific cognitive abilities independent of genetic influence on general cognitive ability?). *Intelligence, 18*, 133-143.
Petrill, S A (1997). Molarity Versus Modularity of Cognitive Functioning? A Behavioral Genetic Perspective. *Current Directions in Psychological Science, 6*, 96-99.
Pfefferbaum, A, Mathalon, D H, Sullivan, E V, Rawles, J M, Zipursky, R B, & Lim, K O (1994). A quantitative magnetic resonance imaging study of changes in brain morphology from infancy to late adulthood. *Archives of Neurology, 51*, 874-887.
Plomin, R (2003). Genetics, genes, genomics and g. *Molecular Psychiatry, 8*, 1-5.
Plomin, R & Kosslyn, S M (2001). Genes, brain and cognition. *Nature Neuroscience, 4*, 1153-1154.
Plomin, R., DeFries, J.C., McClearn, G.E., & McGuffin, P. (2001) *Behavioral genetics.* New York: Worth Publisher
Plomin, R & Spinath, F M (2002). Genetics and general cognitive ability (g). *Trends in Cognitive Sciences, 6*, 169-176.
Polderman, T J C, Stins, J F, Posthuma, D, Gosso, M F, Verhulst, F C, & Boomsma, D I (2006). The phenotypic and genotypic relation between working memory speed and capacity. *Intelligence, 34*, 549-560.
Posthuma, D, Baare, W F, Hulshoff Pol, H E, Kahn, R S, Boomsma, D I, & De Geus, E J (2003). Genetic correlations between brain volumes and the WAIS-III dimensions of verbal

comprehension, working memory, perceptual organization, and processing speed. *Twin Research, 6*, 131-139.
Posthuma, D (2002). Genetic variation and cognition.1-246.
Posthuma, D & Boomsma, D I (2000). A note on the statistical power in extended twin designs. *Behavior Genetics, 30*, 147-158.
Posthuma, D, De Geus, E J, Baare, W F, Hulshoff Pol, H E, Kahn, R S, & Boomsma, D I (2002). The association between brain volume and intelligence is of genetic origin. *Nature Neuroscience, 5*, 83-84.
Posthuma, D., De Geus, E.J.C., & Boomsma, D.I. (2002). Genetic contributions to anatomical , behavioral, and neurophysiological indices of cognition. In: R. Plomin, J.C. DeFries, I.W. Craig, & P. McGuffin *Behavioral Genetics in the Post Genomic Era* (pp. 141-161) Washington D.C.: American Psychological Association
Posthuma, D, Mulder, E J, Boomsma, D I, & De Geus, E J (2002). Genetic analysis of IQ, processing speed and stimulus-response incongruency effects. *Biological Psychology, 61*, 157-182.
Pruessner, J C, Collins, D L, Pruessner, M, & Evans, A C (2001). Age and gender predict volume decline in the anterior and posterior hippocampus in early adulthood. *Journal of Neuroscience, 21*, 194-200.
Raven, J.C. (1960) *Guide to the Standard Progressive Matrices.* London: H.K. Lewis & Co. Ltd.
Raven, J., Raven, J.C., & Court, J.H. (1998) *Raven manual: Section 4 advanced progressive matrices.* Oxford: Oxford Psychologists Press
Reiss, A L, Abrams, M T, Singer, H S, Ross, J L, & Denckla, M B (1996). Brain development, gender and IQ in children. A volumetric imaging study. *Brain, 119 (Pt 5)*, 1763-1774.
Rietveld, M J, Van der Valk, J C, Bongers, I L, Stroet, T M, Slagboom, P E, & Boomsma, D I (2000). Zygosity diagnosis in young twins by parental report. *Twin Research, 3*, 134-141.
Rijsdijk, F V, Vernon, P A, & Boomsma, D I (2002). Application of hierarchical genetic models to Raven and WAIS subtests: a Dutch twin study. *Behavior Genetics, 32*, 199-210.
Shaw, P, Greenstein, D, Lerch, J, Clasen, L, Lenroot, R, Gogtay, N, Evans, A, Rapoport, J, & Giedd, J (2006). Intellectual ability and cortical development in children and adolescents. *Nature, 440*, 676-679.
Simon, H A (1975). The functional equivalence of problem solving skills. *Cognitive Psychology, 7*, 268-288.
Sisk, C L & Foster, D L (2004). The neural basis of puberty and adolescence. *Nature Neuroscience, 7*, 1040-1047.
Sisk, C L & Zehr, J L (2005). Pubertal hormones organize the adolescent brain and behavior. *Frontiers in Neuroendocrinology, 26*, 163-174.
Sowell, E R, Thompson, P M, Leonard, C M, Welcome, S E, Kan, E, & Toga, A W (2004). Longitudinal mapping of cortical thickness and brain growth in normal children. *Journal of Neuroscience, 24*, 8223-8231.
Sowell, E R, Trauner, D A, Gamst, A, & Jernigan, T L (2002). Development of cortical and subcortical brain structures in childhood and adolescence: a structural MRI study. *Developmental Medicine and Child Neurology, 44*, 4-16.
Spear, L P (2000). The adolescent brain and age-related behavioral manifestations. *Neuroscience & Biobehavioral Reviews, 24*, 417-463.
Statistics Netherlands, C B S (2004). Statline Webselector. http://statline.cbs.nl/Statweb.
Stins, J F, Van Baal, G C, Polderman, T J, Verhulst, F C, & Boomsma, D I (2004). Heritability of Stroop and flanker performance in 12-year old children. *BoiMed Central Neuroscience, 5*, 49-
Stroop, J R (1935). Studies of inference in serial verbal reactions. *Journal of Experimental Psychology, 28*, 643-662.
Swanson, H L (1999). What develops in working memory? A life span perspective. *Developmental Psychology, 35*, 986-1000.
Thapar, A, Petrill, S A, & Thompson, L A (1994). The heritability of memory in the Western Reserve Twin Project. *Behavior Genetics, 24*, 155-160.

Thompson, P M, Cannon, T D, Narr, K L, Van Erp, T, Poutanen, V P, Huttunen, M, Lonnqvist, J, Standertskjold-Nordenstam, C G, Kaprio, J, Khaledy, M, Dail, R, Zoumalan, C I, & Toga, A W (2001). Genetic influences on brain structure. *Nature Neuroscience, 4*, 1253-1258.

Thompson, P M, Giedd, J N, Woods, R P, MacDonald, D, Evans, A C, & Toga, A W (2000). Growth patterns in the developing brain detected by using continuum mechanical tensor maps. *Nature, 404*, 190-193.

Turkheimer, E, Haley, A, Waldron, M, D'Onofrio, B, & Gottesman, I I (2003). Socioeconomic status modifies heritability of IQ in young children. *Psychological Science, 14*, 623-628.

Van Baal, C G & Boomsma, D I (1998). Etiology of individual differences in birth weight of twins as a function of maternal smoking during pregnancy. *Twin Research, 1*, 123-130.

Van Beijsterveldt, C E M, Verhulst, F, & Boomsma, D I (2001). Genetic and environmental influences on problem behaviors in 5-year-old Dutch twins. *Twin Research, 4*, 212-

Van den Berg, S M & Boomsma, D I (2007). The familial clustering of age at menarche in extended twin families. *Behavior Genetics, 37*, 661-667.

Van den Berg, S M, Setiawan, A, Bartels, M, Polderman, T J, Van der Vaart, A W, & Boomsma, D I (2006). Individual differences in puberty onset in girls: Bayesian estimation of heritabilities and genetic correlations. *Behavior Genetics, 36*, 261-270.

Van den Burg, W & Kingma, A (1999). Performance of 225 Dutch school children on Rey's Auditory Verbal Learning Test (AVLT): parallel test-retest reliabilities with an interval of 3 months and normative data. *Archives of Clinical Neuropsychology, 14*, 545-559.

Wallace, G L, Eric, S J, Lenroot, R, Viding, E, Ordaz, S, Rosenthal, M A, Molloy, E A, Clasen, L S, Kendler, K S, Neale, M C, & Giedd, J N (2006). A pediatric twin study of brain morphometry. *Journal of Child Psychology and Psychiatry, 47*, 987-993.

Watkins, M P & Meredith, W (1981). Spouse similarity in newlyweds with respect to specific cognitive abilities, socioeconomic status, and education. *Behavior Genetics, 11*, 1-21.

Watson, D, Klohnen, E C, Casillas, A, Simms, E N, Haig, J, & Berry, D S (2004). Match makers and deal breakers: analyses of assortative mating in newlywed couples. *Journal of Personality, 72*, 1029-1068.

Wechsler, D. (1997) *Wechsler Adult Intelligence Scale-Third edition, Dutch Version.* Amsterdam: The Psychological Corporation Ltd. Harcourt Publishers

Wechsler, D., Kort, W., Compaan, E.L., Bleichrodt, N., Resing, W.C.M., Schittekatte, M., Bosmans, M., Vermeir, G., & Verhaeghe, P. (2002) *Handleiding WISC-III^{NI} [Manual WISC-III^{NI}].* London: The Psychological Corporation Limited, Nederlands Instituut van Psychologen Dienstencentrum

Welsh, M C, Pennington, B F, & Groisser, D B (1991). A normative-developmental study of executive function: a window on prefrontal function in children. *Developmental Neuropsychology, 7*, 131-149.

Williams, T (1975). Family resemblance in abilities: the Wechsler scales. *Behavior Genetics, 5*, 405-409.

Yurgelun-Todd, D A, Killgore, W D, & Young, A D (2002). Sex differences in cerebral tissue volume and cognitive performance during adolescence. *Psychological Reports, 91*, 743-757.

Endophenotypes for intelligence in children and adolescents

The aim of this study was to identify promising endophenotypes for intelligence in children and adolescents for future genetic studies in cognitive development. Based on the available set of endophenotypes for intelligence in adults, cognitive tasks were chosen covering the domains of working memory, processing speed, and selective attention. This set of tasks was assessed in a test-retest design in children and in adolescents. Working memory could be measured reliably using the n-back task and correlated with intelligence in both age groups. For processing speed, assessed with the Π-inspection time task and reaction time on the flanker task, test-retest reliability was good in both age groups, but processing speed only correlated significantly with intelligence in children. Selective attention, i.e., the effect of incongruent flankers on RT and accuracy, showed low reliability and neither correlated with intelligence in adolescents nor in children. Thus, working memory seems a promising endophenotype for intelligence in both children and adolescents. Inspection time and measures of selective attention based on the flanker task do not seem very promising endophenotypes for intelligence in these age groups.

This chapter is published as:

Van Leeuwen, M., Van den Berg, S. M., Hoekstra, R. A., & Boomsma, D. I. (2007). Endophenotypes for intelligence in children and adolescents. *Intelligence, 35,* 369-380

Variance in children's IQ test performance is for 25 to 50% accounted for by genetic variation between individuals (Bartels, Rietveld, Van Baal, & Boomsma, 2002; Rietveld, Dolan, Van Baal, & Boomsma, 2003; Turkheimer, Haley, Waldron, D'Onofrio, & Gottesman, 2003; Jacobs et al., 2001; Plomin, 2003) and in adults for even more than 50% (Posthuma, De Geus, & Boomsma, 2001). In spite of this high heritability actual identification of genes is currently limited to mutations with rather severe neurological effects (De Geus, Wright, Martin, & Boomsma, 2001; Nokelainen & Flint, 2002). Genes that influence normal variation in cognitive ability in children have yet to be identified, although recently, several QTLs (quantitative trait loci, i.e. locations of genes that influence complex traits) have been suggested (Butcher et al., 2005a; Butcher et al., 2005b; Hewitt, 2004; Posthuma et al., 2005). One of the complexities of identifying genes affecting a complex trait such as intelligence is that it is influenced by many genes, and therefore each gene is likely to have a relatively small effect (Plomin, DeFries, McClearn, & McGuffin, 2001). The initial goal of QTL research is not to find *the* gene for intelligence, but rather those genes that contribute to different pathways that explain individual differences in intelligence (Plomin, DeFries, Craig, & McGuffin, 2002).

Genetic influences on cognitive ability are likely to be mediated by a complex network of multiple subcortical and cortical brain structures each influenced in part by its own set of genes (De Geus, et al., 2001). These sets of genes influencing intelligence may be localized and identified using the strategy of endophenotyping that is studying confined cognitive components or elements that relate to intelligence variability. These components can be suggested from neuroscience and may get closer to the actual biological systems involved in intelligence (De Geus, et al., 2001; Deary, 2001; Plomin & Spinath, 2002; De Geus & Boomsma, 2001). It is thought that variation in confined components of intelligence may be influenced only by a subset of all the genes involved in general intelligence. The primary idea behind the endophenotypic approach is that by studying these components it may be easier to isolate and identify the effects of each of these subsets of genes. Although these genes may explain only a small part of general intelligence, they may explain a large part of the variance in the endophenotype itself, thereby improving the statistical power to detect genes for general intelligence (De Geus et al., 2001; De Geus, 2002).

The aim of this study is to identify promising endophenotypes for intelligence in childhood and adolescence that may play crucial roles in future genetic studies in cognitive development. For adults, a small set of endophenotypes for intelligence is already available, as will be outlined below. For children, however, much less is known about the suitability of these cognitive measures as endophenotypes for intelligence. Are children at all able to perform the tasks and are the measures reliable? Are the same constructs involved in children and adolescents as in adults?

A promising endophenotype for intelligence in children should be relatively stable, show reliable within-age individual differences, and should have an association with intelligence that is also theoretically meaningful. The endophenotype should also be heritable and have a strong genetic correlation with intelligence (De Geus et al., 2001). A genetic correlation is the extent to which genetic effects on one trait correlate with genetic effects on another trait independent of the heritability of the two traits (Deary, Spinath, & Bates, 2006). Only when this correlation is strong, it does make sense to look for the genes that explain variability in the endophenotype. With a low genetic correlation, a gene variant found for the endophenotype is probably not involved in the variation of general intelligence. Based on suitable endophenotypes used in adults, promising endophenotypes for intelligence in children and adolescents may cover the followings domains: working memory (particularly working memory capacity), processing speed and selective attention.

Working memory

Working memory in adults is related to intelligence. In a meta-analysis of 86 studies Ackerman, Beier, and Boyle (2005) found a correlation of .48 between working memory and intelligence. Colom, Rebollo, Palacios, Juan-Espinosa, and Kyllonen (2004) showed a large overlap in variance between working memory and general intelligence. In adults the correlation of gray and white matter volume with full scale IQ and the Working Memory dimension is completely mediated by common genetic factors (Posthuma et al., 2002a).

In a Japanese sample of young adults, using a spatial as well as a verbal working memory task, Ando, Ono, and Wright (2001) found that higher-order spatial and verbal cognitive abilities are mediated by a genetic factor they have in common with working memory. This common genetic factor explained 20-22% of variation in working memory

and 64% and 26% of variation in spatial and verbal ability respectively. Thus, in adults working memory is a suitable endophenotype for intelligence.

In children, however, this is less clear. It is known that working memory and intelligence develop in concert in children. In their review on the relationships between processing speed, working memory, and fluid intelligence in children, Fry and Hale (2000) argue that much of the age-related increase in intelligence in children can be attributed to developmental improvements in working memory. The greater the capacity of a child's working memory, the more information the child has available for solving problems.

In children, correlations between intelligence and working memory range from .38 to .67 (Alloway, Gathercole, Willis, & Adams, 2004; Swanson, 2004; Fry & Hale, 1996) and even .82 in children aged 4 to 6 (Swanson & Beebe-Frankenberger, 2004). In a study of De Ribaupierre and Lecerf (2006) working memory accounted for 54% of the variance in the Raven Standard Progressive Matrices (Raven, 1960) in a children and young adult sample.

Furthermore, working memory is associated with scholastic achievement. For instance, links have been found with reading ability (De Jonge & De Jonge, 1996; Cain, Oakhill, & Bryant, 2004; Gathercole, Alloway, Willis, & Adams, 2006) and solving mathematical problems (Adams & Hitch, 1997; Geary, Hoard, Byrd-Craven, & Catherine DeSoto, 2004; Swanson, 2004; Swanson et al., 2004; Lee, Ng, Ng, & Lim, 2004). Moreover, Van der Sluis, Van der Leij, and De Jong (2005) found that when corrected for fluid intelligence most links between working memory and reading and arithmetic-related learning disabilities disappeared, suggesting that most of the relations between working memory and learning disabilities can be explained by IQ.

Using twins at the age of twelve years and their siblings Polderman and colleagues (2006) reported heritability estimates (the proportion of phenotypic differences among individuals that can be attributed to genetic differences in a particular population) for working memory capacity of 54% and 56%. Luciano and colleagues (2001a) found in 16-year-olds that genetic differences explain 48% of the variance in working memory. The genetic correlation between intelligence and working memory is moderate in this age group; a phenotypic correlation (the correlation between observed characteristics) of .26 between intelligence and a delayed response task and a genetic correlation of .34 was found.

Working memory is related to activity in the dorsolateral prefrontal cortex (DL-PFC; Casey et al., 1995), a brain area still developing during childhood (Casey, Giedd, & Thomas, 2000), and to the anterior cingulate, a brain area of which the gray matter density is positively correlated with full-scale IQ in adolescents (Frangou, Chitins, & Williams, 2004). This suggests working memory is still developing in children and is related to intelligence.

Summarizing, in children as well as in adults working memory and intelligence are related. In children variance in working memory is in part explained by genetic factors, and in adolescents and adults a genetic correlation between working memory and intelligence has been found.

Processing speed

Intelligence also co-develops with information processing speed. In children and adults inspection time (a measure for processing speed) and IQ are correlated and the correlations observed in children are similar to the ones found in adults (Fry & Hale, 2000). A meta-analysis conducted by Grudnik and Kranzler (2001) indicated that inspection time and IQ correlate around -.50. De Ribaupierre and colleagues (2006) found in a sample of children and young adults that processing speed (as measured by a task in which subjects had to judge whether two patterns were identical) accounted for 61% of the total variance in the Raven's task. Taken together, processing speed and working memory explained 67% of variance in Raven's performance. Vickers and McDowell (1996) found in a sample with children aged 8 to 10 years a correlation between inspection time and full scale IQ of -.51. Fry and colleagues (1996) found a correlation of -.44 between processing speed and the Raven.

In the literature, a broad range of tasks is used to measure processing speed, ranging from simple inspection time tasks in which subjects have to distinguish the longest of two lines of different length (Brand & Deary, 1982; Luciano et al., 2001b) to complex reaction time tasks in which subject have to memorize 5 digits and have to indicate whether a newly presented digit is one of the memorized digits (Neubauer, Spinath, Riemann, Borkenau, & Angleitner, 2000). This complicates the assessment of processing speed as a possible endophenotype, because the more complex an elementary cognitive task is, the higher the correlation with intelligence (Colom et al., 2004; Neubauer et al., 2000). More

complex tasks are more meaningfully and more strongly related to intelligence, but on the other hand, a suitable endophenotype should not be too complex, since the more complex the task, the more genes are likely to be involved. Therefore a relatively simple task measuring processing speed should be preferred.

In adults and adolescents a high genetic correlation between inspection time and full scale IQ has been found (Posthuma et al., 2001; Luciano et al., 2005). In 13 to 15-year-olds, a genetic correlation of -.63 was reported. A common genetic factor accounted for 36% in the variance of inspection time, the remaining variance in inspection time was accounted for by a unique environmental factor (Luciano et al., 2001b), suggesting that a considerable part of the variance in inspection time could consist of measurement error variance.

Posthuma and colleagues (2001) hypothesized that the genetic factor that influences intelligence as well as speed of processing is a factor that determines axonal myelination in the central nervous system. Fry and Hale (2000) concluded in their review that much of the age related improvement in children in intelligence is due to increases in speed and this seems to be mediated through the effect of speed on working memory, that is, the faster the brain, the more information can be retained in working memory.

Concluding, in both young adolescents and adults there is a relationship between processing speed and intelligence. Moreover, there is a genetic correlation between these two abilities in adolescents and adults. However, it is yet unknown to what extent processing speed is a suitable endophenotype for intelligence in children.

Selective attention

Concepts of selective attention are included in almost all theories of higher cognitive functioning. Dempster (1991) claims that intelligence cannot be understood without reference to inhibitory processes. One of his arguments is that individuals who are more distractible, score generally lower on intelligence tests. Using the flanker task (Eriksen & Eriksen, 1974), Posthuma, Mulder, Boomsma, and De Geus (2002b) showed a significant genetic correlation between IQ and incongruency effects (difference in performance between congruent and incongruent trials) on accuracy, varying between -.37 and -.68 depending on the cohort (old or young) and IQ-scale (verbal or performance IQ). Stins, Van Baal, Polderman, Verhulst, and Boomsma (2004) found little evidence for heritability of

flanker performance in 12-year-old twins. Possible explanations for this discrepant finding can be that the task is not suitable for children and cannot reliably measure inhibitory processes, that twelve-year-olds perform the task in a more prudent way that leads to ceiling effects on accuracy, or that inhibitory processes have not yet fully developed at this age. As far as we know no study has been done up till now relating incongruency effects to intelligence in children. The one study (Censabella & Noël, 2005) we could find reported on the relationship between incongruency effects and learning disabilities. This study could not show that children with learning disabilities exhibit significantly larger incongruency effects than children without learning disabilities.

Slowing down of reaction time and loss of accuracy in flanker task performance as a consequence of incongruencies in the stimuli may reflect an impairment in the top-down inhibitory control of the prefrontal cortex (Posthuma et al., 2002b). During performance on this task the left prefrontal cortex is activated (Fan, Flombaum, McCandliss, Thomas, & Posner, 2003), a brain area shown to be developing between the ages of 7 and 11 years (Sowell, Trauner, Gamst, & Jernigan, 2002) and implied in intelligence (Frangou et al., 2004).

A limitation of all studies relating working memory and processing speed to intelligence, is that none of these studies (Alloway et al., 2004; De Ribaupierre et al., 2006; Fry et al., 1996; Swanson, 2004; Swanson et al., 2004; Vickers et al., 1996) corrected the observed relationships for the biasing effects of measurement error. When one is interested in the relationship between actual traits, rather than relationships between specific measures of traits it is important to make corrections for biases induced in research data by measurement error (Schmidt & Hunter, 1996).

Moreover, the few studies reporting short-term test-retest stability of tasks measuring processing speed, selective attention, and working memory, show that most test-retest reliabilities are rather low in children. Test-retest reliabilities for working memory in children are reported for various tasks, ranging from .52 to .76. However, most of these tasks are verbal in nature. Alloway and colleagues (2004) reported test-retest reliabilities for three working memory tasks in children aged 5 to 8 years. For the *backwards digit recall test* they report a reliability of .53, for the *counting recall test* (children need to count the number of dots in an array, and then recall the tallies of dots in the arrays that were presented) they report a reliability of .74, and for the *sentence completion and recall task*

(the child listens to a series of short sentences with a missing word at the end, produces a word to complete the sentence, and recalls the word she or he produced for each sentence in a sequence) test-retest reliability was .52. In another study test-rest correlations were found of .54 using the *sentence completion and recall* and the *counting span task* (child needs to count yellow dots on a card with blue and yellow dots, after 2 to 5 of these cards, the child has to recall the total number of dots; Kuntsi, Stevenson, Oosterlaan, & Sonuga-Barke, 2001). Archibald and Kerns (1999) found a test-retest reliability of .76 for the *Self-Ordered pointing* in which children had to point to a different drawing in a booklet, whereby every time the location of drawings changed. Vickers and colleagues (1996) found a test-retest reliability of .30 for an inspection time task where children had to discriminate which of two lines was the longest. For the standard Stroop task for interference a test-retest reliability of .81 over three sessions has been reported by Neyens and Aldenkamp (1996).

In the current study, test-retest reliability will be investigated for various tasks measuring working memory, processing speed, and selective attention to identify promising endophenotypes for intelligence in children and adolescents. For working memory the *n*-back task was used (Casey et al., 1995), information processing speed was assessed using the π-inspection time task (Luciano et al., 2001b) and the flanker task (Eriksen et al., 1974). The flanker task was also used to measure selective attention. All tasks were specifically adapted for children. In order to assess the relationship between performance on these tasks and intelligence, all correlations were corrected for test-reliability.

Materials and Methods

Subjects

Three groups of subjects participated in this study. The first group consisted of 108 children who were recruited from the 5th grade of six primary schools located in different social economic areas in the Netherlands. 105 Children returned two to three weeks later for retest, children were 8-11 years of age (M = 8.7, SD = .6). Of these children, 55.4% were female. After completing the test protocol children received a present worth €5,-. The second group consisted of 98 children participating in an ongoing longitudinal study recruited via the Netherlands Twin Registry (NTR). For the current study one twin or sibling was randomly selected from a family (age: M = 9.7, SD = 1.1, 52% female). After

participation they received a present of €10,-. The third group consisted of 30 adolescents in the age range of 14 to 20 (M = 18.4, SD = 1.6), from which 29 returned for retest. In this group 70% was female. Adolescents received a token of €25,-. When children were under 14, their parents signed an informed consent form. If not, participants signed an informed consent themselves.

Testing procedures

Children in the first group were administered the *n*-back task, Eriksen flanker task, and π-inspection time task, as part of a larger neuropsychological test battery. Because of practical reasons not all children were administered the complete battery. Administration of the complete battery required approximately 50 minutes. Children in this group were individually tested during school hours in a quiet room at school. Children in the second group and adolescents were individually tested at the Vrije Universiteit. They were administered the π-inspection time task, Eriksen flanker task and *n*-back task at the end of a larger test battery - including the Wechsler Adult Intelligence Scale-III (WAIS-III) or Wechsler Intelligence Scale for Intelligence-III (WISC-III) - of which administration required approximately 4.5 hours. Retest for children in the first group as well as adolescents took place two to three weeks after initial testing. There was no re-test for the WAIS-III and no retest session for children in the second group.

n-back task

Subjects performed a spatial variant of the *n*-back task, designed after Gevins and Cutillo (1993) and Jansma, Ramsey, Coppola, and Kahn (2000), with increasing levels of difficulty. The task was adapted to make it more attractive for children. Subjects had to look at an apple presented on a screen. The apple had four holes in which a caterpillar could appear. The participants were told to catch the caterpillar to prevent it from eating the apple. They were instructed to respond to the caterpillar by pushing one of four buttons with thumb and index finger of both hands. The layout of the four buttons corresponded spatially to the four holes in which the caterpillar could appear. Subjects had to indicate where the caterpillar was one move back (1-back), two moves back (2-back) or three moves back (3-back). Adolescents received also a session with a delay of 4 moves (4-back). The caterpillar appeared in a hole for 1 second; after its disappearance there was a warning

sound. Subjects had to respond after this warning. Between two caterpillar moves, the apple was empty for one second.

Sessions were given in blocks of 20 trials. After each block participants received feedback on the number of apples they had saved from the caterpillar (correct button presses) and how many had been eaten (incorrect button presses). The 1-back condition consisted of a practice block only. The 2-back, 3-back and 4-back conditions contained one practice block, and three blocks in which performance was measured. Practice blocks were added if the subject did not understand the task. Children were motivated during the task by counting the moves of the caterpillar. In the 2-back version the test administrator counted continuously to three and in the 3-back version the administrator counted to four. The task requires that subjects have to respond to all stimuli and continuously have to monitor and update all movements of the caterpillar. Performance on the task was scored by using the total number of correct responses. Maximum score per condition was 60.

Π-inspection task

The π-inspection task was designed after Luciano et al. (2001b). For this task subjects had to identify the longer of two lines which were presented by the test administrator as worms that the subjects needed to catch. This task was complicated by the fact that the worms burrowed quickly into the ground (i.e., disappeared quickly from the screen). If subjects caught five worms they had enough worms to catch a fish, which would appear at the lower left-hand side of the screen. It was stressed that it was important to be accurate and that it did not matter how long it took them to catch the worms.

The vertical lines measured 22 and 27 mm in length, were 9 mm apart, and joined at the top to a horizontal line 12 mm long. The probability of the longer line appearing on the left or right was equal. The stimulus duration ranged between 14.2 and 2000 ms. A dynamic mask, consisting of two vertical lines (37 mm) shaped as lightning bolts, immediately followed the stimulus and was presented for 300 ms to limit further stimulus processing. On each trial, a fixation cross appeared in the centre of the screen for 1 s (an alerting beep was sounded for 100 ms at the onset of the dot), followed by a blank screen for 100 ms. The π figure was then presented, and the participant's first response (left or right) was noted. The screen was blanked for 750 ms before the next trial was presented. This produced an effective response–stimulus interval of approximately 2 s. Stimulus

duration depended on the response of the subject. The initial duration was 210 ms. For every four correct consecutive responses the stimulus duration was decreased, and for every incorrect response the stimulus duration was increased by a step size depending on previous performance (for details of the actual algorithm, see Luciano et al., 2001b). The algorithm decided when minimal stimulus duration was achieved and stopped the program. If the minimum was not reached within 96 trials the program also stopped. This way, for each subject the minimal stimulus duration time could be assessed. On each trial the stimulus duration and whether or not the correct key was pressed, was stored. Data of the subjects were excluded from the analyses when they responded more often than 10 times before the stimulus had disappeared. The stimulus duration at the last correct trial was used as the measure for inspection time.

Eriksen flanker task

In the Eriksen flanker task (Eriksen & Schultz, 1979) subjects were presented with a horizontal array of five arrows. Before each trial a fixation cross was presented for 1000 ms. The stimulus was then presented for 200 ms. Between two trials there was a random interval of either 2000, 2250, 2500, 2750 or 3000 ms. Subjects were instructed to pay attention to the direction of the centre arrow and ignore the four flanking ones. They were told that the cross in the center of the screen between two trials could help them to focus on the middle arrow. Subjects had to press the left key to a left facing central arrow, and the right key to right facing central arrow. The flanking arrows could either all point in the same direction as the target arrow (<<<<< or >>>>>; congruent), or they all pointed in the opposite direction (<<><< or >><>>; incongruent). Children received 40 congruent and 40 incongruent trials in random order after an eight trial practice session. After each ten correct responses a smiley was presented in the center of the screen. On each trial the reaction time (RT) and whether or not the correct key was pressed, was stored. Maximum score was 40 correct congruent and 40 correct incongruent trials for the children. The adolescents performed a shorter version of the task; this was 20 congruent and 20 incongruent trials. Trials in which reaction time was below 300 ms or exceeded 1500 ms were excluded from analysis. If, as a result of this rule, more than 25% of a subject's trials were excluded, all data from this subject's session were excluded from analysis. Average RT was calculated over the accurate trials. Average RTs on incongruent trials were subtracted from average

RTs on congruent trials and served as a measure for selective attention. The same applies to the error rates that were similarly subtracted as an additional measure for selective attention.

Data analysis

Test-retest reliability and correlations between the endophenotypes and IQ were calculated with Pearson correlation coefficients. Correlations between IQ and the endophenotypes were subsequently corrected for test-retest reliability using the disattenuation formula $\boldsymbol{r}_{xtyt} = \boldsymbol{r}_{xy} / (\boldsymbol{r}_{xx}\boldsymbol{r}_{yy})^{1/2}$, where $\boldsymbol{r}_{xtyt}$ is the correlation between the true scores of the measures x and y, $\boldsymbol{r}_{xy}$ is the observed correlation, and $\boldsymbol{r}_{xx}$ and $\boldsymbol{r}_{yy}$ are the reliabilities of x and y, respectively (Schmidt et al., 1996). For the reliability of the Dutch WISC-III a Cronbach's α of .93 was used (Wechsler et al., 2002) and for the reliability of the Dutch WAIS-III a test-retest correlation was used of .94 (Kessels & Wingbermühle, 2001). Reaction time and inspection time data were log transformed prior to analysis since their distributions were positively skewed.

A partial correlation analysis was conducted to determine the independent contributions of the endophenotypes to IQ. Only variables were included that showed a significant correlation with IQ in one of the two age groups in the previous correlation analyses. To determine how much variance in IQ scores was explained by the endophenotypes, a multiple regression analysis was conducted, in which all variables were entered simultaneously.

Results

Mean IQ score of the children in the second group was 101.6 (*SD* = 14.3). For the adolescents the mean IQ was 108.4 (*SD* = 12.2). Table 2.1 presents a description of the problems encountered during testing. In general the following problems were encountered: During administration of the *n*-back, it was observed that some children were not able to push the button while at the same time paying attention to where to caterpillar went. In the π-inspection task sessions were excluded, because children and adolescents pushed the button before the π-figure disappeared. From the Eriksen flanker task data from some subjects were excluded from analysis, because more than 25% of data were excluded (reaction times were below 300ms or exceeded 1500ms).

Table 2.1. *Problems encountered during testing: (I) children of the first group, (II) children of the second group, (A) adolescents.*

	Test	Retest
2-back	I: 2 subjects not recorded	
3-back	I: 2 subjects not able	I: 3 subjects not able/ did not want to participate
	II: 3 subjects not able	
4-back	A: 1 subject not able	
π-task	I: 8 subjects excluded	I: 2 subjects excluded
	II: 8 subjects excluded	
		A: 1 subject excluded
Flanker	I: 1 subject excluded	I: 2 subjects excluded
	II: 2 subjects excluded	
		A: 1 subject excluded

Note: For the Π task, subjects were excluded when they pressed the keys too early more often than 10 times. For the flanker task, subjects were excluded when more than 25% of their RTs was either < 300 or > 1500 ms.

Results of testing and test-retest reliabilities in children and adolescents are presented in Tables 2.2 and 2.3. Because specific abilities in children are tested, reliabilities of .7 or higher are considered satisfactory, whereas reliabilities of .5 and .6 are considered modest (Kuntsi et al., 2001). As shown in Table 2.2, all test-retest correlations in children exceeded .60, except for accuracy and stimulus congruency effects of the flanker task. The low accuracy test-retest correlation is very likely due to ceiling effects. For the adolescents the same holds true, with an exception of the 2-back and the π-inspection task. The low test-retest correlation on the 2-back can be explained by ceiling effects at the second time of testing.

Table 2.2. *Descriptive statistics and test-retest correlations for n-back, flanker and π-task in children of group I (accuracy is reported in proportion correct, reaction time in ms).*

	N	Mean (*SD*) test	*N* retest	Mean (*SD*) retest	*N*	*r* (95% confidence intervals)
2-back	59	.51 (.24)	59	.66 (.26)	58	.65 (.51-1.0)
3-back	58	.40 (.15)	56	.50 (.19)	56	.70 (.60-1.0)
Flanker RT congruent	76	566 (98)	74	563 (91)	74	.66 (.56-1.0)
Flanker RT incongruent	76	668 (137)	74	638 (106)	74	.62 (.49-.96)
Flanker incongruency effect (RT)	76	102 (86)	74	75 (57)	74	.48 (.29-.76)
Flanker Acc. congruent	76	.95 (.07)	74	.96 (.08)	74	.06 (-.17-.29)
Flanker Acc. incongruent	76	.85 (.19)	74	.90 (.15)	74	.46 (.26-.73)
Flanker incongruency effect (Acc.)	76	.10 (.08)	74	.06 (.06)	74	.29 (.07-.53)
π-task	68	164 (61)	70	128 (50)	63	.65 (.52-1.00)

Table 2.3. *Descriptive statistics and test-retest correlations for n-back, flanker, and π-task in adolescents (accuracy is reported in proportion correct, reaction time in ms).*

	N test	Mean (*SD*) test	*N* retest	Mean (*SD*) retest	*N*	*r* (95% confidence intervals)
2-back	30	.89 (.15)	29	.96 (.09)	29	.16 (-.22-.55)
3-back	30	.72 (.17)	29	.84 (.14)	29	.70 (.48-1.0)
4-back	29	.61 (.15)	29	.69 (.17)	28	.66 (.40-1.0)
Flanker RT congruent	30	434 (72)	28	419 (56)	28	.66 (.40-1.0)
Flanker RT incongruent	30	495 (72)	28	475 (61)	28	.65 (.38-1.0)
Flanker incongruency effect (RT)	30	61 (29)	28	56 (28)	28	.48 (.13-.91)
Flanker Acc. congruent	30	.97 (.05)	28	.96 (.08)	28	.42 (.06-.84)
Flanker Acc. incongruent	30	.96 (.08)	28	.94 (.08)	28	.35 (-.03-.76)
Flanker incongruency effect (Acc.)	30	.01 (.06)	28	.03 (.07)	28	.14 (-.25-.53)
π-task	28	94 (35)	26	70 (17)	25	.58 (.24-1.0)

Table 2.4 presents observed correlations and corrected correlations between IQ and the endophenotypes. In children and adolescents *n*-back performance was significantly related to IQ. Better performance on the *n*-back task was related to higher IQ-scores. No correction for test-retest reliability is reported for the 2-back task in adolescents, since this reliability is influenced by ceiling effects. Reaction time on the congruent and incongruent trials of the flanker was significantly related to IQ for children only; the longer the reaction time, the lower the IQ. Incongruency effects on reaction time, accuracy on the congruent and incongruent trials, as well as incongruency effects on accuracy were not related to IQ in children nor in adolescents. Inspection time was related to IQ in children, the shorter the inspection time the higher the IQ, but was not significantly related to IQ in adolescents.

Table 2.4. *Observed correlations (r_{xy}) and correlations corrected for test-retest (r_{xtyt}) of n-back, flanker, and π-task with IQ in children and adolescents.*

	children			adolescents		
	N	r_{xy}	r_{xtyt}	*N*	r_{xy}	r_{xtyt}
2-back	94	.41**	0.53	30	.66**	-
3-back	95	.44**	0.55	30	.55**	0.68
4-back	-	-		29	.40*	0.51
Flanker RT congruent	96	-.35**	-0.45	30	-.07	-0.09
Flanker RT incongruent	96	-.35**	-0.46	30	-.06	-0.08
Flanker incongruency effect (RT)	96	-.04	-0.06	30	.14	0.21
Flanker Acc. congruent	96	-.10	-0.42	30	.10	0.16
Flanker Acc. incongruent	96	-.06	-0.09	30	.02	0.03
Flanker incongruency effect (Acc.)	96	-.02	-0.04	30	-.06	-0.17
π-task	88	-.28**	-0.36	28	-.33	-0.45

Note. ** = $p < .01$, * = $p < .05$

In Table 2.5 the results of the partial regression analyses are presented. It is important to note that in this table only the subjects are included for whom data are available on all tasks. This leads to a lower correlation between IQ and 3-back performance

and flanker reaction time in children due to a biased sample of relatively smarter subjects: children who performed poorly had a higher probability of not being able to perform all of the cognitive tasks sufficiently well. As can be seen in Table 2.5, none of the tasks contributed completely independently to the variance in intelligence in children. The π-task and the flanker task did not contribute any significant independent part of the variance in intelligence. The 3-back showed significant covariance with intelligence even after the flanker and π–task had been controlled for. Also in adolescents, the 3-back task contributed to the variance in intelligence independently of the other two tasks, while the contribution of performance on the π-task could partly be explained by performance on the 3-back task.

Table 2.5. *Correlations of 3-back, Flanker RT congruent and π-task with IQ controlling for respectively 3-back accuracy, flanker reaction time in congruent trials and π-inspection time.*

r controlled for:	-	3-back	Flanker	π-task	3-back & Flanker	3-back & π-task	Flanker & π-task
Children (N=84)							
3-back	.42**	-	.36**	.35**	-	-	.32**
Flanker RT congruent	-.31**	-.22*	-	-.24*	-	-.19	-
π-task	-.28**	-.15	-.20	-	-.10	-	-
Adolescents (N=27)							
3-back	.57**	-	.57**	.54**	-	-	.54**
Flanker RT congruent	-.03	-.12	-	-.01	-	-.09	-
π-task	-.31	-.23	-.31	-	-.22	-	-

Note. ** = $p < .01$, * = $p < .05$

Regression analyses revealed that 2-back, 3-back, flanker reaction time on the congruent and incongruent trials and inspection time could explain a total of 17% (adjusted R^2) of the variance in IQ in children ($R = .47$, $F(5, 78) = 4.49$, $p < .001$). Since it was clear from the other analyses that flanker reaction time did not contribute to the variance in intelligence, this variable was not included in the regression analysis in adolescents. In this analysis 2-back, 3-back, 4-back and inspection time could explain a total of 45% (adjusted R^2) in the variance of intelligence ($R = .73$, $F(4, 22) = 6.28$, $p < .01$).

Discussion

The goal of this study was to determine whether endophenotypes for intelligence previously used in adults and sometimes in adolescents are promising endophenotypes for intelligence in children and adolescents. A good endophenotype for intelligence must meet the following criteria (De Geus et al., 2001): it must be a reliable trait, it must show evidence of genetic influence, it must be associated with intelligence, the association between endophenotype and intelligence must derive partly from the same genetic source (i.e, there should be a genetic correlation) and the association between endophenotype and intelligence must be theoretically meaningful.

In this paper we examined the reliability and the relation to intelligence of three candidate endophenotypes: working memory, processing speed, and selective attention. The choice for these three endophenotypes was based on prior research, which was mainly conducted in adults.

Working memory

A spatial version of the *n*-back task was used, specifically adapted to measure working memory in children and adolescents. In children 2-back as well as 3-back performance could be measured reliably and in adolescents the *n*-back 3 and 4 could be measured reliably. This result is comparable to the test-retest reliability reported by Hockey and Geffen (2004) who found a test-rest correlation in students on the 3-back of .73. It is also comparable to the results from a spatial working memory task reported by Archibald and colleagues (1999).

In both children and adolescents performance on the *n*-back 2 and 3 was correlated with IQ. In the adolescent group performance on the 4-back task was also correlated with intelligence. The observed correlations in this study are comparable to correlation reported in previous studies (Ackerman et al., 2005; Alloway et al., 2004; Fry et al., 1996; Swanson, 2004).

In this study we found a lower correlation between *n*-back performance and intelligence in children as compared to adolescents. This may be due to the lower performance levels of the children: some children were not able to push a button and meanwhile attend where the caterpillar was going. As a consequence in children the task may measure short term memory, rather than working memory. This interpretation is in line

with that of De Jonge and colleagues (1996) who reported that no distinction could be made between tasks measuring short-term and working memory in children aged 10 to 12 years. This is not surprising since the prefrontal cortex, and particularly the DL-PFC, a brain region involved in working memory, appears to be the last brain region to mature (Casey et al., 2000).

Based on the findings from previous studies as well as from our own, it can be concluded that working memory as measured by the *n*-back task is a suitable endophenotype for intelligence in adolescents and to a somewhat lesser extent in children. Future research should establish whether there is a genetic correlation between performance on this task and intelligence in children and adolescents. Whether in children the task actually measures working memory rather than short-term memory is still a matter of discussion.

Processing speed

The π-inspection time task was used to measure processing speed. In children as well as adolescents this task showed good test-retest correlations. Test-retest reliability was substantially higher than the one reported in the study of Vickers and colleagues (1996). This discrepancy can possibly be explained by the reward incorporated in our task, which keeps children motivated during the task. The correlation between intelligence and processing speed was lower than what has been found in previous studies (Grudnik et al., 2001). One explanation for this finding is that in studies in which higher correlations with intelligence have been reported, more complex measures for processing speed were used, like for instance Sternberg's memory scanning task. As stated by Neubauer and colleagues (2000) and Colom and colleagues (2004) the more complex an elementary cognitive task is, the higher the correlation with intelligence. At first glance it may seem that inspection time is a suitable endophenotype for children. However, it must be noted that in children the average inspection time was quite long and showed large variation. This suggests that in children whose inspection times are long, it may not be the speed of processing that was measured. Many children seemed somehow unable to deal with this task, showing inspection times of over 500 ms. It is unclear what the task actually measured and therefore the test may be unsuitable as an endophenotype for intelligence in children of this age. An endophenotype must be simple to interpret, since its goal is to facilitate the search for

genes. One of the prerequisites of an endophenotype is that the relationship with the phenotype of interest must be theoretically meaningful.

In adolescents inspection time was not significantly related to IQ. This result may becaused by a lack of statistical power, since the effect size is similar to the one reported by Luciano and colleagues (2001b) and Posthuma et al. (2001). Nevertheless, the amount of variation in intelligence it might explain is limited, particularly after correction for working memory performance (cf. Fry et al., 2000). This finding suggests that inspection time task is of limited added value as an endophenotype in a test battery including a working memory task. We therefore conclude that inspection time as measured by the π-task is not the optimal endophenotype for intelligence, neither in children nor in adolescents.

Selective attention

The flanker task did not measure accuracy and incongruency effects on RT reliably. The task measured reaction time on congruent and incongruent trials reliably, but it can be argued that reaction time is a measure of processing speed rather than selective attention (Fry et al., 1996). In adolescents and children we found no evidence for a relationship between intelligence and incongruency effects. Therefore, it can be concluded that selective attention as measured by flanker incongruency effects is not a suitable endophenotype for intelligence in children and adolescents.

The partial correlation analysis showed that in children working memory as measured by the 3-back task contributed a significant, though not completely independent, part to the variance in intelligence. Processing speed as measured by the flanker task also contributed a small part to the variance of intelligence, though not significantly or independently. In adolescents, working memory as measured by the 3-back contributed a significant part to intelligence that could not by explained by performance on the flanker or π-task. When controlled for working memory and processing speed as measured by the flanker task, no significant contribution of inspection time was left in children or in adolescents.

When exploring the variance contributed by the different tasks to intelligence it becomes clear that the same tasks explain more variance in adolescents than in children. A possible reason for this is that performance on these tasks in children is influenced by different and unknown processes which do not play a role in adolescents. This finding

illustrates that the search for endophenotypes in children may be more complex than in adolescents and adults. Another explanation for the lower contribution of variance by the different cognitive tasks to the variation in intelligence in young children, is that in children variation in intelligence is less influenced by genes than in adolescents and in adults. It is possible that the association of IQ and working memory is mainly due to genetic covariation which will become more pronounced with increasing age.

To conclude, working memory capacity seems a good endophenotype for intelligence in children and adolescents: it can be reliably assessed using our version of the *n*-back and it correlates with intelligence. Processing speed is not an optimal endophenotype for intelligence in children (as measured by reaction time on the flanker task) and adolescents (as measured by the π-task). Once corrected for working memory, it contributes only a very small part to the variance of intelligence. Selective attention, at least when measured as the flanker incongruency effect on RT and accuracy, is not a suitable endophenotype for neither age groups. Future studies will be directed at investigating whether in children and adolescents, working memory is sufficiently heritable and genetically correlated with intelligence to be of use in QTL research.

Acknowledgements

We wish to thank the children and the staff of the following schools for the time and assistance they contributed to this study: Flevoparkschool in Amsterdam, St Michael in De Bilt, Cammingha RK in Bunnik, ABC RK basisschool locatie de Bijenkorf in Gouda, and De Paltrok and De Wissel in Uitgeest. We gratefully acknowledge support from grants by the Netherlands Organisation for Scientific Research, NWO 051-02-060, 668-772 and NWO/SPI 56-464-14192, and the Centre for Neurogenomics and Cognitive Research, Amsterdam.

Reference List

Ackerman, P. L., Beier, M. E., & Boyle, M. O. (2005). Working memory and intelligence: the same or different constructs? *Psychological Bulletin, 131,* 30-60.

Adams, J. W. & Hitch, G. J. (1997). Working memory and children's mental addition. *Journal of Experimental Child Psychology, 67,* 21-38.

Alloway, T. P., Gathercole, S. E., Willis, C., & Adams, A. M. (2004). A structural analysis of working memory and related cognitive skills in young children. *Journal of Experimental Child Psychology, 87,* 85-106.

Ando, J., Ono, Y., & Wright, M. J. (2001). Genetic structure of spatial and verbal working memory. *Behavior Genetics, 31,* 615-624.

Archibald, S. J. & Kerns, K. A. (1999). Identification and description of new tests of executive functioning in children. *Child Neuropsychology, 5,* 115-129.

Bartels, M., Rietveld, M. J. H., Van Baal, G. C. M., & Boomsma, D. I. (2002). Genetic and environmental influences on the development of intelligence. *Behavior Genetics, 32,* 237-249.

Brand, C. R. & Deary, I. J. (1982). Intelligence and "inspection time.". In H.J.Eysenck (Ed.), *A model for intelligence.* (New York: Springer).

Butcher, L. M., Meaburn, E., Dale, P. S., Sham, P., Schalkwyk, L. C., Craig, I. W. et al. (2005a). Association analysis of mild mental impairment using DNA pooling to screen 432 brain-expressed single-nucleotide polymorphisms. *Molecular Psychiatry, 10,* 384-392.

Butcher, L. M., Meaburn, E., Knight, J., Sham, P. C., Schalkwyk, L. C., Craig, I. W. et al. (2005b). SNPs, microarrays and pooled DNA: identification of four loci associated with mild mental impairment in a sample of 6000 children. *Human Molecular Genetics, 14,* 1315-1325.

Cain, K., Oakhill, J., & Bryant, P. (2004). Children's reading comprehension ability: concurrent prediction by working memory, verbal ability, and component skills. *Journal of Educational Psychology, 96,* 31-42.

Casey, B. J., Cohen, J. D., Jezzard, P., Turner, R., Noll, D. C., Trainor, R. J. et al. (1995). Activation of prefrontal cortex in children during a nonspatial working memory task with functional MRI. *NeuroImage, 2,* 221-229.

Casey, B. J., Giedd, J. N., & Thomas, K. M. (2000). Structural and functional brain development and its relation to cognitive development. *Biological Psychology, 54,* 241-257.

Censabella, S. & Noël, M. P. (2005). The inhibition of exogenous distracting information in children with learning disabilities. *Journal of Learning Disabilities, 38,* 400-410.

Colom, R., Rebollo, I., Palacios, A., Juan-Espinosa, M., & Kyllonen, P. C. (2004). Working memory is (almost) perfectly predicted by g. *Intelligence, 32,* 277-296.

De Geus, E. J., Wright, M. J., Martin, N. G., & Boomsma, D. I. (2001). Genetics of brain function and cognition. *Behavior Genetics, 31,* 489-495.

De Geus, E. J. C. (2002). Introducing genetic psychophysiology. *Biological Psychology, 61,* 1-10.

De Geus, E. J. & Boomsma, D. I. (2001). A genetic neuroscience approach to human cognition. *European Psychologist, 6,* 241-253.

De Jonge, P. & De Jonge, P. F. (1996). Working memory, intelligence and reading ability in children. *Personality and Individual Differences, 21,* 1007-1020.

De Ribaupierre, A. & Lecerf, T. (2006). Relationships between working memory and intelligence from a developmental perspective: Convergent evidence from a neo-Piagetian and a psychometric approach. *European Journal of Cognitive Psychology, 18,* 109-137.

Deary, I. J., Spinath, F. M., & Bates, T. C. (2006). Genetics of intelligence. *European Journal of Human Genetics., 14,* 690-700.

Deary, I. J. (2001). Human intelligence differences: towards a combined experimental-differential approach. *Trends in Cognitive Sciences, 5,* 164-170.

Dempster, F. N. (1991). Inhibitory processes: A neglected dimension of intelligence. *Intelligence, 15,* 157-173.

Eriksen, B. A. & Eriksen, C.-W. (1974). Effects of noise letters upon the identification of a target letter in a nonsearch task. *Perception & Psychophysics, 16,* 143-149.

Eriksen, C. W. & Schultz, D. W. (1979). Information processing in visual search: a continuous flow conception and experimental results. *Perception & Psychophysics, 25,* 249-263.

Fan, J., Flombaum, J. I., McCandliss, B. D., Thomas, K. M., & Posner, M. I. (2003). Cognitive and brain consequences of conflict. *NeuroImage, 18,* 42-57.

Frangou, S., Chitins, X., & Williams, S. C. R. (2004). Mapping IQ and gray matter density in healthy young people. *NeuroImage, 23,* 800-805.

Fry, A. F. & Hale, S. (2000). Relationships among processing speed, working memory, and fluid intelligence in children. *Biological Psychology, 54,* 1-34.

Fry, A. F. & Hale, S. (1996). Processing speed, working memory, and fluid intelligence: Evidence for a developmental cascade. *Psychological Science, 7,* 237-241.

Gathercole, S. E., Alloway, T. P., Willis, C., & Adams, A. M. (2006). Working memory in children with reading disabilities. *Journal of Experimental Child Psychology, 93,* 265-281.

Geary, D. C., Hoard, M. K., Byrd-Craven, J., & Catherine DeSoto, M. (2004). Strategy choices in simple and complex addition: Contributions of working memory and counting knowledge for children with mathematical disability. *Journal of Experimental Child Psychology, 88,* 121-151.

Gevins, A. & Cutillo, B. (1993). Spatiotemporal dynamics of component processes in human working memory. *Electroencephalography and Clinical Neurophysiology., 87,* 128-143.

Grudnik, J. L. & Kranzler, J. H. (2001). Meta-analysis of the relationship between intelligence and inspection time. *Intelligence, 29,* 523-535.

Hewitt, J.K. (Ed.). (2004) Variance components methods for mapping quantitative trait loci. (2004). *Behavior Genetics* 34, 125-215.

Hockey, A. & Geffen, G. (2004). The concurrent validity and test-retest reliability of a visuospatial working memory task. *Intelligence, 32,* 591-605.

Jacobs, N., Van Gestel, S., Derom, C., Thiery, E., Vernon, P., Derom, R. et al. (2001). Heritability estimates of intelligence in twins: effect of chorion type. *Behavior Genetics, 31,* 209-217.

Jansma, J. M., Ramsey, N. F., Coppola, R., & Kahn, R. S. (2000). Specific versus nonspecific brain activity in a parametric n-back task. *NeuroImage, 12,* 688-697.

Kessels, R. & Wingbermühle, E. (2001). De WAIS-III^{NI} als neuropsychologisch instrument [The WAIS-III^{NI} as neuropsychological instrument]. *De Psycholoog, 36,* 296-299.

Kuntsi, J., Stevenson, J., Oosterlaan, J., & Sonuga-Barke, E. J. S. (2001). Test-retest reliability of a new delay aversion task and executive function measures. *British Journal of Developmental Psychology, 19,* 339-348.

Lee, K., Ng, S. F., Ng, E. L., & Lim, Z. Y. (2004). Working memory and literacy as predictors of performance on algebraic word problems. *Journal of Experimental Child Psychology, 89,* 140-158.

Luciano, M., Posthuma, D., Wright, M. J., De Geus, E. J., Smith, G. A., Geffen, G. M. et al. (2005). Perceptual speed does not cause intelligence, and intelligence does not cause perceptual speed. *Biological Psychology, 70,* 1-8.

Luciano, M., Wright, M., Smith, G. A., Geffen, G. M., Geffen, L. B., & Martin, N. G. (2001a). Genetic covariance among measures of information processing speed, working memory, and IQ. *Behavior Genetics, 31,* 581-592.

Luciano, M., Smith, G. A., Wright, M. J., Geffen, G. M., Geffen, L. B., & Martin, N. G. (2001b). On the heritability of inspection time and its covariance with IQ: A twin study. *Intelligence, 29,* 443-457.

Neubauer, A. C., Spinath, F. M., Riemann, R., Borkenau, P., & Angleitner, A. (2000). Genetic and environmental influences on two measures of speed of information processing and their relation to psychometric intelligence: Evidence from the german observational study of adult twins. *Intelligence, 28,* 267-289.

Neyens, L. G. J. & Aldenkamp, A. P. (1996). Stability of cognitive measures in children of average ability. *Child Neuropsychology, 3,* 161-170.

Nokelainen, P. & Flint, J. (2002). Genetic effects on human cognition: lessons from the study of mental retardation syndromes. *Journal of Neurology, Neurosurgery, and Psychiatry, 72,* 287-296.

Plomin, R. (2003). Genetics, genes, genomics and g. *Molecular Psychiatry, 8,* 1-5.

Plomin, R., DeFries, J. C., Craig, I. W., & McGuffin, P. (2002). Behavioral genetics. In R.Plomin, J. C. DeFries, I. W. Craig, and P. McGuffin (Eds.), *Behavioral Genetics in the Post Genomic Era* (pp. 3-15). Washington D.C.: American Psychological Association.

Plomin, R., DeFries, J. C., McClearn, G. E., & McGuffin, P. (2001). *Behavioral genetics*. (4 ed.) New York: Worth Publisher.

Plomin, R. & Spinath, F. M. (2002). Genetics and general cognitive ability (g). *Trends in Cognitive Sciences, 6,* 169-176.

Polderman, T. J. C., Stins, J. F., Posthuma, D., Gosso, M. F., Verhulst, F. C., & Boomsma, D. I. The phenotypic and genotypic relation between working memory speed and capacity. *Intelligence,* (in press).

Posthuma, D., De Geus, E. J., Baare, W. F., Hulshoff Pol, H. E., Kahn, R. S., & Boomsma, D. I. (2002a). The association between brain volume and intelligence is of genetic origin. *Nature Neuroscience, 5,* 83-84.

Posthuma, D., De Geus, E. J., & Boomsma, D. I. (2001). Perceptual speed and IQ are associated through common genetic factors. *Behavior Genetics, 31,* 593-602.

Posthuma, D., Luciano, M., Geus, E. J., Wright, M. J., Slagboom, P. E., Montgomery, G. W. et al. (2005). A genomewide scan for intelligence identifies quantitative trait loci on 2q and 6p. *American Journal of Human Genetics, 77,* 318-326.

Posthuma, D., Mulder, E. J., Boomsma, D. I., & De Geus, E. J. (2002b). Genetic analysis of IQ, processing speed and stimulus-response incongruency effects. *Biological Psychology, 61,* 157-182.

Raven, J. C. (1960). *Guide to the Standard Progressive Matrices*. London: H.K. Lewis & Co. Ltd.

Rietveld, M. J. H., Dolan, C. V., Van Baal, G. C. M., & Boomsma, D. I. (2003). A twin study of differentiation of cognitive abilities in childhood . *Behavior Genetics, 33,* 367-381.

Schmidt, F. L. & Hunter, J. E. (1996). Measurement error in psychological research: Lessons from 26 research scenarios. *Psychological Methods, 1,* 199-223.

Sowell, E. R., Trauner, D. A., Gamst, A., & Jernigan, T. L. (2002). Development of cortical and subcortical brain structures in childhood and adolescence: a structural MRI study. *Developmental Medicine & Child Neurology, 44,* 4-16.

Stins, J. F., Van Baal, G. C., Polderman, T. J., Verhulst, F. C., & Boomsma, D. I. (2004). Heritability of Stroop and flanker performance in 12-year old children. *BoiMed Central Neuroscience, 5,* 49.

Swanson, H. L. (2004). Working memory and phonological processing as predictors of children's mathematical problem solving at different ages. *Memory & Cognition, 32,* 648-661.

Swanson, H. L. & Beebe-Frankenberger, M. (2004). The relationship between working memory and mathematical problem solving in children at risk and not at risk for serious math mifficulties. *Journal of Educational Psychology, 96,* 471-491.

Turkheimer, E., Haley, A., Waldron, M., D'Onofrio, B., & Gottesman, I. I. (2003). Socioeconomic status modifies heritability of iq in young children. *Psychological Science, 14,* 623-628.

Van der Sluis, S., Van der Leij, A., & De Jong, P. F. (2005). Working memory in Dutch children with reading- and arithmetic-related LD. *Journal of Learning Disabilities, 38,* 207-221.

Vickers, D. & McDowell, A. (1996). Accuracy in the frequency accrual speed test (FAST), inspection time and psychometric intelligence in a sample of primary school children. *Personality and Individual Differences, 20,* 463-469.

Wechsler, D., Kort, W., Compaan, E. L., Bleichrodt, N., Resing, W. C. M., Schittekatte, M. et al. (2002). *Handleiding WISC-III^{Nl} [Manual WISC-III^{Du}]*. London: The Psychological Corporation Limited, Nederlands Instituut van Psychologen Dienstencentrum.

The genetic and environmental structure of working and short-term memory in young adults and children

The extent to which verbal and visuospatial working memory (WM) and short term memory (STM) tests measure the same or multiple constructs is unclear. Likewise the relationship between WM and STM across development is not known. Here we addressed these questions using genetically informative data, studying two age cohorts (young adults and children) of twins and siblings. Verbal and visuospatial WM and STM were measured using the Corsi block tapping task, n-back task, and the digit span forward and backwards task. Multivariate genetic analyses revealed that two highly correlated common genetic factors, one for verbal and one for visuospatial memory, gave the best description of the covariance structure among the measures. Only in children, specific genetic factors were also present. This led to the following conclusions: At the genetic level two correlated factors are responsible for linking verbal and visuospatial WM and STM in both children and young adults. During the course of development the influence of genetic factors unique to each of these domains disappears. At the environmental level, both in young adults and in children, environmental factors create differences between these domains.

This chapter is submitted as:

Van Leeuwen, M., Van den Berg, S. M., Hoekstra, R. A., & Boomsma, D. I. (in revision). The genetic and environmental structure of working and short-term memory in young adults and children. *Neuropsychology*.

A wealth of studies has focussed on the relation between *short-term memory* (STM) and *working memory* (WM) in adults (e.g. Conway, Cowan, Bunting, Therriault, & Minkoff, 2002; Kane et al., 2004) as well as children (e.g. Alloway, Gathercole, & Pickering, 2006; Bayliss, Jarrold, Baddeley, & Gunn, 2005; Kail & Hall, 2001). However, it is still unclear whether STM and WM are independent or overlapping constructs. The mixed findings could be a result of the tasks used to measure these constructs, different ages of the participants, and different subject populations (e.g. general population vs. undergraduates). We report on a twin study that examined genetic and environmental relation between these constructs in young adults and children from the general population. In both age groups similar tasks were used to measure WM and STM.

According to Cowan (1988, 1995) "STM refers to information [like words or images] in long-term memory that is activated above some threshold. Activated information rapidly returns to an inactive state [leaves the STM] unless it becomes the focus of limited-capacity attentional processes [attention]" (see Kail & Hall, (2001), pg 1). WM can be distinguished from STM by its multi-component character, its combined processing and storage, and functional importance as a system that facilitates cognitive activities (Baddeley, 2003). Working memory is the system that is necessary for the concurrent storage and manipulation of information (Baddeley, 1992). Most studies (e.g. Bayliss, Jarrold, Gunn, & Baddeley, 2003; Kane et al., 2004) examine the relation between STM and WM from the perspective of the WM model of Baddeley (2000) and Baddeley and Hitch (1974). The Baddeley model constitutes of a *central executive* and three storage systems: the *phonological loop*, the *visuospatial sketchpad* and the *episodic buffer*. The central executive is the system responsible for a range of regulatory functions, including attention, the control of action, and problem solving (Baddeley, 1996). The phonological loop comprises a phonological store that can hold memory traces for a few seconds before they fade, and an articulatory rehearsal process. The visuospatial sketchpad is its visuospatial counterpart (Baddeley, 2003). In multiple studies the phonological loop and the visuospatial sketchpad are considered equivalent to STM (e.g. Gathercole, Pickering, Ambridge, & Wearing, 2004). However, to what extent they really are equivalent is still subject to debate (Engle, Tuholski, Laughlin, & Conway, 1999). The episodic buffer provides temporary storage of information held in a multimodal code, which is capable of integrating information from a variety of sources, including long-term memory, into a

unitary episodic representation. The buffer is episodic in the sense that it holds episodes whereby information is integrated across space and potentially extended across time (Baddeley, 2000). The episodic buffer is generally not included in studies on the relation between STM and WM.

The basic methodology to study the relation between STM and WM is assessing these constructs using multiple tasks in the verbal and/ or visuospatial domain (e.g. Colom, Flores-Mendoza, Quiroga, & Privado, 2005; Miyake, Friedman, Rettinger, Shah, & Hegarty, 2001). To measure STM subjects are asked to memorize lists of words, numbers, the location of objects, etcetera, without any intervening tasks (e.g. Alloway et al., 2006). These tasks are generally referred to as (simple) span tasks. WM is tapped by tasks demanding simultaneous storage and processing, which is for instance the case in complex span tasks. In a complex span task (e.g. Swanson, 1992) subjects have to memorize a number of items and meanwhile have to perform another task like counting or answering a question. Another way to combine processing and storage is using tasks that demand subjects to manipulate the information they have to memorize, like in the digit span backwards test (Wechsler, 1997; Wechsler et al., 2002) and the *n*-back task (e.g. Casey, Giedd, & Thomas, 2000; Jansma, Ramsey, Coppola, & Kahn, 2000). In the digit span backwards test subjects have to remember a list of digits in backward order (e.g. Hoshi et al., 2000; Lee, Lu, & Ko, 2007). In an *n*-back task, subjects continuously need to memorize the location of an object, a letter or a picture *n* trials ago in an ongoing stream of stimuli. This makes that subjects constantly need to update information.

Findings in studies on the relation between STM and WM vary from complete independence of STM and WM (Swanson, 1999), up to almost complete overlap between both constructs (Colom et al., 2005). Engle et al. (1999) suggested that age differences could be an explanation for these different findings. He hypothesized that STM and WM share more variance in children than in adults, since children are less skilled in chunking and coding and less routinized in rehearsal. Therefore simple memory tasks are already attention demanding, and thus more comparable to WM tasks.

Also, there is no agreement whether WM and STM are domain specific or domain general constructs. So, it is unclear whether there are domain specific storage and executive function mechanisms for visuospatial and verbal memory tasks. For example, the unitary model (Jones, Beaman, & Macken, 1996) proposes a memory system with one common

factor for verbal and visuospatial short-term memory. In contrast, Haavisto and Lehto (2005) propose not only domain specific storage components for WM, but also domain specific components for the executive function factor of WM. These different views could be a result of studying different populations of participants. For example, Shah and Miyake (1996) and Kane et al. (2004) showed that the dissociation between spatial and verbal measures is less apparent in samples including individuals who are likely to vary widely in general ability (e.g. general population) than in samples including individuals with a restricted range of cognitive abilities (e.g. college students).

In this introduction we discuss the most important studies on the relation between verbal and visuospatial STM and WM. It starts with describing studies which argue for the independence of WM and STM versus studies arguing for an overlap of these constructs. Next, an overview of studies on the domain generality versus the domain-independence of WM and STM is provided.

The independence versus the overlap of WM and STM

In two independent studies on the relation between WM, STM and reading skills in children Swanson (1992) and Swanson and Berninger (1996) found support for the view that WM and STM are independent constructs. In 2001, Kail and Hall (2001) investigated whether STM and WM can be distinguished in children. The data were best described by two factors representing STM and WM, which correlated moderately (r = .3). The authors concluded that STM can already be distinguished from WM during the elementary school years. In a comparable study carried out in undergraduates, Cantor, Engle, and Hamilton (1991) found that two independent factors representing WM and STM gave the best description of the data. Therefore it was concluded that STM and WM tasks reflect two different constructs.

Bayliss et al. (2003) investigated the constraints underlying working memory performance in young adults and children and found support for the Baddeley and Hitch model. Alloway et al. (2006) and Gathercole et al. (2004) tested this model in multiple age cohorts of children. The models they tested consisted of one common factor (WM factor) and two domain specific storage factors. In both studies the WM factor and the domain-specific storage factors shared about half of the variance, showing that processing and storage in WM and STM are not independent. Another important finding of these two

studies was that from six years onwards no significant developmental changes in the relation between STM and WM were observed.

To what extent the relationship between STM and WM can be explained by executive function or short-term storage has been studied extensively (e.g. Bayliss et al., 2005a; Colom et al., 2005; Kane et al., 2004; Shah & Miyake, 1996). Kane et al. (2004), Colom et al. (2005), and Conway et al. (2002) demonstrated in adults that WM and STM are overlapping constructs in both the visuospatial and verbal domain. The three studies do not agree about the nature of this overlap. Colom et al. (2005) and Conway et al. (2002) argue that the high correlation between STM and WM stems from the demand they both place on storage capacity. But according to Kane et al. (2004), who studied subjects from the general population and undergraduates, both STM and WM place a demand on storage and executive functioning. Moreover, Kane et al. (2004) showed that if the sample was limited to undergraduates only, the overlap between WM and STM decreased.

On one side of the continuum multiple studies (Cantor et al., 1991; Kail & Hall, 2001; Swanson, 1992; Swanson & Berninger, 1996) suggest that in young adults as well as in children STM and WM are relatively independent constructs. STM is considered as a memory measure over a short time delay and WM as a executive functioning measure. Others (Alloway et al., 2006; Gathercole et al., 2004) show that WM and STM are not independent, and that the Baddeley and Hitch model is applicable. On the other side of this continuum researchers argue that STM also consists of executive functioning and WM also includes short-term memory (Kane et al., 2004). Summarizing the above studies leads to the conclusion that the extent to which WM and STM tests measure the same or multiple constructs is unclear.

The domain generality versus the domain specificity of WM and STM

Haavisto and Lehto (2005) studied the domain specificity of WM in Air Force Recruits, by examining fluid/spatial and crystallized/verbal intelligence in relation to spatial and verbal WM. The authors found that verbal WM was related to crystallized intelligence and visuospatial WM to fluid intelligence. This shows that complex WM tasks measure separate domain-specific cognitive abilities, which supports the view that the executive functioning component of WM is domain specific. After separating executive functioning from WM in children, Tillman, Nyberg, and Bohlin (in press) showed that visuospatial and verbal

executive functioning both contribute independently to intelligence. This finding argues for a domain specific view of WM in children. Thus, in children as well as in adults, there is evidence for domain specificity of WM.

These findings are in contrast with the Baddeley and Hitch model, which consists of one domain-general factor for executive functioning, and two domain specific storage factors. Alloway et al. (2006), Gathercole et al. (2004) and Kane et al. (2004) demonstrated in their studies on the relation between WM and STM that domain general executive function and domain specific storage already exist at the age of six, stay stable during development and are also present in the adult general population.

Maehara and Saito (2007) examined the domain specificity and domain invariance of WM in a student population, by looking at the effect of *stimulus order* (recall scores are greater in complex span tasks when they start with a task which takes a long time to process and end with a task that requires a short processing time than when these tasks are presented in the reverse order) and *processing time* (processing speed decreases for items that are processed later in time in a complex span task, since at this time subjects have more items to remember). The authors compared the effect of processing tasks and storage in the same and different modalities (verbal and visuospatial). The stimulus order effect was only observed when storage and processing were in the same domain, which demonstrated domain specificity. In contrast, the processing time effect could also be detected when storage and processing were in different domains. This showed that next to domain specific processes, domain general processes also play a role in WM.

By demonstrating that disruption to STM does not depend on the modality of the interfering task Jones, Farrand, Stuart, and Morris (1995) found support for the unitary model in adults. This model suggests one common factor for verbal and visuospatial short-term memory (Jones et al., 1996). Chuah and Maybery (1999) investigated whether this model also applies to children, by studying to what extent age, verbal and spatial searching speed, articulation rate and its visuospatial equivalent, tapping rate, could predict verbal and spatial span. Predictors involving stimuli of the same modality as the span task did not consistently explain more variance than predictors involving stimuli in a different modality than the span task. This finding and a study of Bayliss, Jarrold, Baddeley, Gunn, and Leigh (2005b) gave indirect evidence that the unitary model also applies to children.

Findings in studies on the domain generality and domain specificity of WM and STM are varying. In the strongest version of the domain general view (e.g. Chuah & Maybery, 1999; Jones et al., 1996) it is assumed that sequences of verbal and visuospatial events share a common level of representation, and therefore a memory model with one common factor for verbal and visuospatial short-term memory is proposed. In the domain specific view the assumption is that that there are separate storage mechanisms at work for visuospatial and verbal information. These domain-specific storage mechanisms also drive the domain specificity of WM (e.g. Alloway et al., 2006). In the strongest version of the domain specific view it is assumed that even executive function is domain specific (e.g. Haavisto & Lehto, 2005).

Hence, it is unclear whether STM and WM are independent or overlapping constructs. Moreover, it is unclear whether these constructs are domain general or domain specific in nature. The contrasting findings are a reflection of the *molarity*-versus-*modularity* debate. In the molar view there is one system in which a unitary, general process functions across a wide variety of cognitive tasks. In the modular view there are numerous distinct cognitive processing units, each responsible for certain non-overlapping cognitive tasks (Petrill, 1997). An explanation why cognitive studies find evidence for molarity as well as modularity is that cognition is influenced by genes and environment. There is evidence that genetic influences tie together diverse measures of cognitive functioning, whereas environmental effects drive wedges between different dimensions of cognitive processing (Luo, Petrill, & Thompson, 1994; Pedersen, Plomin, & McClearn, 1994). Genetic evidence points to molarity as evidenced by substantial genetic overlap across different cognitive abilities. In contrast , the different dimensions of cognitive functioning which consistently emerge across many studies seem to be primarily driven by environmental factors (Petrill, 1997). We hypothesize that this also applies to verbal and visuospatial WM and STM. Previous twin studies already showed that differences between individuals in performance on WM and STM tasks can be explained by differences in genotype (Kremen et al., 2007; Kuntsi et al., 2006; Polderman et al., 2006). Using multivariate genetic factor analysis we aim to establish to what extent the correlation between these constructs is caused by a common set of genes and / or environmental factors (Boomsma & Molenaar, 1986; Martin & Eaves, 1977), and whether the factor structure at the genetic level is consistent with the factor structure at the environmental level.

Previously, the genetic structure of verbal and visuospatial WM in young adults has been studied by Ando, Ono, and Wright (2001). Two complex span tasks were administered in which both storage and processing were measured, yielding two scores for each modality. Genetic modeling revealed one common genetic factor influencing all tasks, two modality specific genetic factors, and one storage specific genetic factor. At the environmental level the authors found two specific environmental factors (for verbal storage and for verbal executive functioning) and two common environmental factors (for executive functioning and spatial storage and for executive functioning and verbal storage). At the genetic level, this is an indication for modality specific and modality invariant elements in WM.

In two age cohorts we address the question whether STM and WM are independent or overlapping constructs and whether they are domain general or domain specific in nature. The oldest, the young adult, cohort consists of 18-year-old twins coming from 186 families and the youngest, the child, cohort are 9-year-old twins and their siblings coming from 112 families. Verbal and visuospatial WM and STM are operationalized by administering one task in every domain. The genetic and environmental structure underlying the relation between verbal and visuospatial STM and WM is examined with multivariate genetic analyses (Boomsma & Molenaar, 1986; Martin & Eaves, 1977). By investigating their relationship in a genetically informative design it is possible to elucidate the previous mixed findings, which, we hypothesize, are a result of genetic molarity and environmental modularity. Based on the existing literature three models for the structure of verbal and visuospatial STM and WM are compared separately for the genetic and environmental factor structure:

1. WM and STM are independent, but modality invariant constructs. This will be reflected in a common factor for WM and a common factor for STM. Since studies arguing for the independence of WM and STM do find moderate correlations between the constructs, the two factors are allowed to correlate.
2. STM and WM are overlapping, but modality specific constructs. This will be reflected in a common factor for verbal memory and a common factor for visuospatial memory. The verbal memory and visuospatial memory factors are permitted to correlate.

3. STM and WM are overlapping, modality invariant constructs, one common factor will describe the relation between visuospatial and verbal STM and WM.

By comparing model fit in the young adult and child cohort we can asses whether there are any developmental changes in the underlying environmental and genetic structure of verbal and visuospatial WM and STM. We hypothesize that STM is more demanding for children than for young adults and therefore (see Engle et al., 1999) that STM and WM will share more variance in children than in young adults. This hypothesis ties in with the differentiation hypothesis, whose origins can be traced to the 'Law of Diminishing Returns' of Spearman (1927), and which states that cognitive abilities become increasingly more differentiated during development (Garret, 1946; Reinert, 1970). We hypothesize that the differentiation of cognitive abilities with increasing age will be reflected only at the genetic level. We expect that genetic factors will be more task specific in young adults than in children.

Material and Methods

Participants

Young adult cohort

Twin families were recruited via the Netherlands Twin Register (Boomsma et al., 2002; Boomsma et al., 2006). This cohort consisted of 186 families of eighteen-year-old twin pairs (M = 18.2, SD = .21) and one of their siblings (N = 93, M = 18.5, SD = 5.74) who take part in a longitudinal study of cognition and behavioral problems (Bartels, Rietveld, Van Baal, & Boomsma, 2002; Hoekstra, Bartels, & Boomsma, 2007). The group comprised 33 monozygotic male twin pairs (MZM), 34 dizygotic male twin pairs (DZM), 44 monozygotic female twin pairs (MZF), 38 dizygotic female twin pairs (DZF), and 37 dizygotic twin pairs of opposite sex (DOS). The zygosity of the same sex twin pairs was determined by DNA analyses (139 pairs), blood group polymorphisms (9 pairs) or longitudinally collected questionnaire items (Rietveld et al., 2000; 1 pair). There were 46 male and 47 female additional siblings in this cohort. The study was approved by the Central Committee on Research involving Human Subjects (CCMO). When children were

under 18, their parents signed an informed consent form. If they were aged 18 years or older, participants signed an informed consent themselves.

Data from one sibling were excluded from analyses since this boy had severe learning difficulties. Data of the Corsi block tapping task of 33 participants (7% of the sample) were excluded since these participants had a score of 10 or lower. A score of 10 or lower means that they made mistakes when memorizing two or three blocks in a row, suggesting they probably did not understand the task properly.

Child cohort

The group of participants in this cohort consisted of 112 nine-year-old twin pairs (M = 9.1, SD = .10) and one of their siblings aged nine to fourteen (N = 100, M = 11.8, SD = 1.16). Children were recruited from the NTR. This group is taking part in an ongoing study on the development of cognition and brain structure (Van Leeuwen, Van den Berg, & Boomsma, 2008), and includes 23 MZM pairs, 23 DZM pairs, 25 MZF, 21 DZF pairs, and 20 DOS pairs. For the same sex twin pairs, zygosity determination was based on DNA polymorphisms (90 twin pairs), or on questionnaire items (2 pairs; Rietveld et al., 2000). There were 44 male and 56 female siblings. The study was approved by the CCMO, and parents signed an informed consent form for their children.

Three families did not complete the Corsi block tapping task, one sibling did not complete the 2-back, and two siblings did not take the WISC. Ten children were not able to complete the n-back task and eight children could not complete the Corsi.

Testing Procedures

In both cohorts all participants were individually tested at the VU University in separate rooms by experienced test administrators, so each participant was tested by a different administrator. For the young adult cohort a testing day consisted of two parts; in the morning participants completed a medical test protocol and after lunch they completed a psychological test protocol. The psychological test protocol including a break took about three and a half hours to complete and included the Corsi block tapping task, the *n*-back task and an intelligence test. Twins and siblings of 16 years of age and above completed the Wechsler Adult Intelligence Scale for Adults-III (WAIS-III; Wechsler, 1997), children under 16 were administered the Wechsler Intelligence Scale for Children-III (WISC-III;

Wechsler et al., 2002). For the child cohort a testing day consisted of a psychological test protocol only. Testing lasted for about five hours (including three breaks). Children completed as part of a larger test battery the Corsi block tapping task, the *n*-back task and the WISC-III.

Corsi Block Tapping Task

The Corsi block tapping task (Corsi, 1974) was included to assess short-term spatial memory. Participants sat in front of a touch screen monitor on which nine white blocks were displayed unevenly across a grey screen. In succession a number of blocks turned red for one second, after which the screen was blank for three seconds. After reappearance of the blocks, the participant had to tap the blocks on the screen in the same sequence in which they had changed color before. When a block was tapped, the block would turn red and stay that way until the end of the run. The computer registered each tap. Each participant was given two practice runs. In these practice runs each person had to memorize two blocks. Immediately after the practice runs the actual test was administered. Actual testing started with a series of two blocks. After every five runs the item length was increased by one block. The test was terminated when the participant responded incorrectly to three out of five runs of the same length. The maximum number of blocks that could turn red in succession was nine. Performance was measured by total number of correct runs.

N-back Task

Participants had to perform a spatial variant of the *n*-back task (Van Leeuwen, Van den Berg, Hoekstra, & Boomsma, 2007) to assess visuospatial working memory. The *n*-back used in this protocol was designed after Gevins and Cutillo (1993) and Jansma et al. (2000) with increasing levels of difficulty. The participants were asked to look at an apple presented on a screen. The apple had four holes in which a caterpillar could appear. Participants were told to catch the caterpillar to prevent it from eating the apple, and were instructed to respond to the caterpillar by pushing one of four buttons with the thumb and index finger of both hands. The layout of the four buttons corresponded spatially to the four holes in which the caterpillar could appear. Participants had to indicate where the caterpillar was one move back (1-back), two moves back (2-back), three moves back (3-back), or four moves back (4-back). The caterpillar appeared in a hole for one second; after its

disappearance there was a warning sound. Participants were instructed to respond after this warning sound and could respond until the next caterpillar appeared. Between two caterpillar moves, the apple was empty for one second.

Sessions were given in sessions of 20 trials. Each condition consisted of a practice session and three sessions in which performance was recorded. Practicing continued until the participants understood the task. After each session participants received feedback on the number of apples they had saved from the caterpillar (correct responses) and how many had been eaten (incorrect responses). Following the feedback there was a break of 15 seconds. The task requires a continuous response to all stimuli and simultaneous monitoring and update of all movements of the caterpillar. Performance on the task was scored by using the total number of correct responses. Maximum score per condition was 60.

In the young adult cohort the 1-back and 2-back conditions were administered for practice purposes only, performance was recorded on the 3-back and 4-back conditions. For this cohort the sum score on the 3-back condition was used. Test-retest correlation of the 3-back condition in young adults is .70 (Van Leeuwen et al., 2007). In the child cohort the 4-back condition was not administered and only the 1-back condition was used solitary for practice. Children were motivated during the task by counting the moves of the caterpillar. In the 2-back version the test administrator counted continuously to three and in the 3-back version the administrator counted to four. For this cohort we used performance on the 2-back condition. For children the test-retest on 2-back is .65 (Van Leeuwen et al., 2007).

Digit Span

Digit span forwards (DSF) of the WAIS-III or WISC-III was used to measure verbal short-term memory. In this task participants had to recall lists of numbers. The test started with a trial of two numbers. If participants recalled one out of two trials correctly, the list increased with one digit. Increments proceeded, until participants had both of two trials wrong. Performance was scored as the total number of correct trials. To measure verbal working memory the digit span backwards task (DSB) was used. This time the participants had to recall lists of numbers in reverse order. Test-restest correlation for digit span (forward and backward together) of the WAIS-III is .74 (Kooij, Rolfhus, Wilkins, Yang, & Zhu, 2004). The split half coefficient for the internal consistency of digit span of the WISC-III is .67 (Wechsler et al., 2002).

Data Analysis

All data analyses were performed using the software package Mx (Neale, Boker, Xie, & Maes, 2006). First, general covariance matrices, means, and the effect of sex and age on the means were estimated in a saturated model. Means were estimated separately for MZ twins, DZ twins and siblings. The 12 x 12 covariance matrices (i.e. 4 variables x 3 family members) were estimated separately for MZ and DZ twin families. In the saturated model separate covariances were estimated for MZ twin pairs, DZ twin pairs and twin-sibling pairs. The phenotypic, MZ, DZ and twin-sibling correlations were derived by standardizing the corresponding covariances. Since a large number of parameters were estimated, this model yields a good description of the data.

First, several assumptions such as equality of means and variances in twins and siblings were tested by fitting a series of nested models in which the means and variances for MZ and DZ twins and for twins and siblings were equated. The assumption that the 4 variables covaried in the same way within twins and siblings was tested by constraining the phenotypic covariances among measures to be the same in twins and siblings. Next, the assumption was tested that the resemblance in DZ twins is similar to the resemblance in non-twin siblings. We continued equating parameters until the most parsimonious model with still acceptable fit was established. The choice for the best fitting model was based on likelihood-ratio tests. The difference between minus twice the log likelihoods (-2 LL) of two nested models asymptotically follows a χ^2 distribution. The degrees of freedom are given by the difference in the number of parameters estimated in the two nested models. A high increase in χ^2 against a low gain of degrees of freedom denotes a worse fit of the submodel compared to the full model. The means and the covariance structure between family members and between traits was tested for equality across the age cohorts. All data were analyzed, including data from families with incomplete twin pairs or without an additional sibling, using the raw data option in Mx.

Genetic Modeling

Univariate analysis

To get a first impression of the relative influence of genes and environment on individual differences in memory performance, MZ, DZ, and sibling correlations were inspected. If

MZ twin correlations are higher than DZ twin and twin-sibling correlations, part of the individual differences are caused by genetic effects, comprising of *additive genetic effects* (A) and *non-additive genetic effects* (D). If DZ twin and twin-sibling correlations are more than half the size of MZ correlations, the resemblance between twins is at least partly caused by *shared environmental effects* (C; environmental effects shared among offspring brought up in the same family). If MZ twin correlations are more than twice as high as DZ twin and twin-sibling correlations, D is likely to contribute to individual differences in memory performance. Differences within MZ twin pairs reflect the importance of *unique environment* (E). To have sufficient power to detect D or C large samples are required (Boomsma, Busjahn, & Peltonen, 2002; Plomin, DeFries, McClearn, & McGuffin, 2001). Based on the limited sample size and on inspection of the MZ, DZ and twin-sibling correlations we decided to fit a genetic model in which the relative contributions of A end E were estimated.

Formally, a trait or *phenotype* (P; i.e. observed characteristic of an individual that results from the combined effect of genes and environment) can be represented at the individual level as:

$$P_{ij} = a*A_{ij} + e*E_{ij},$$

where i = 1,2, … 112 (families) and j = 1, 2, or 3 (family members) and A and E are factors (latent variables, that are not observed directly). A and E are standardized to have unit variance. Figure 3.1 represents the phenotypes in one twin pair and one additional sibling in a genetic path model. $P_{Twin\ 1}$, $P_{Twin\ 2}$ and $P_{sibling}$ represent the phenotypes measured in these participants. The variance in P due to A and E is given by the square of a and e, respectively, so that Var (P) = $a^2 + e^2$, which means that the observed variance in a population is attributed to variance caused by genes and variance caused by environment. Note that e^2 also contains variance due to measurement error.

MZ twins are practically identical at the DNA sequence level and therefore genetic effects are nearly perfectly correlated in MZ twins. DZ twins and siblings share on average half of their segregating genes so that the expected genetic correlation between their additive genetic effects (A) is ½ (see also Figure 3.1). By definition the correlation among the unique environmental effects (E) in twins and siblings is zero. Therefore the covariance within MZ twin pairs can be modeled as:

$$Cov_{MZ}(P_{Twin\ 1}, P_{Twin\ 2}) = a^2,$$

and within DZ twin pairs and twin-sibling pairs as:

$$Cov_{DZ}(P_{Twin\ 1}, P_{Twin\ 2}) = Cov(P_{Twin\ 1}, P_{Sibling}) = \tfrac{1}{2}\, a^2.$$

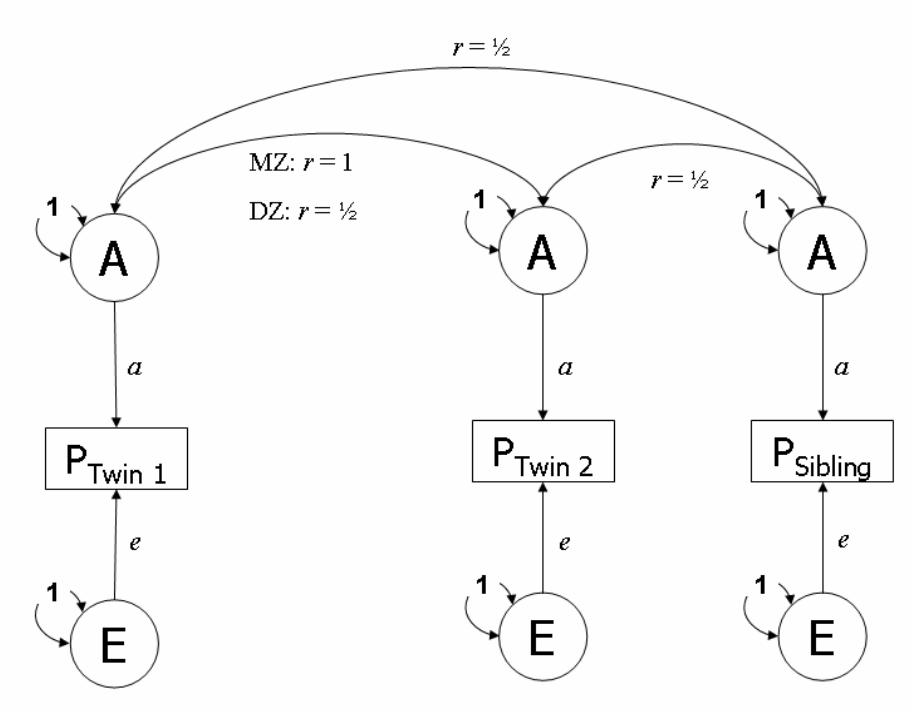


Figure 3.1. Univariate AE path model, with one twin pair and one additional sibling. A stands for the additive genetic factor, E for the unique environmental factor, and P for phenotype. As follows from the figure the variance in the observed measure equals *a*2 + *e*2, the covariation within MZ twin pairs equals *a*2 and within DZ twin pairs and between twins and siblings ½ *a*2 .

Multivariate analysis

To determine to what extent the covariation among the four measures is due to correlated genetic and environmental effects, multivariate genetic factor analysis was applied. In a multivariate analysis the *cross twin - cross trait correlations* between MZ and DZ twins and between twins and siblings contain information on the etiology of the association between traits. An example of a cross twin – cross trait correlation is the correlation between visuospatial WM in twin 1 and verbal WM in twin 2. These cross-correlations are estimated in the saturated model. Larger MZ cross-correlations compared to the DZ and twin-siblings cross-correlations indicate that part of the covariation between the two traits is determined by correlated genetic factors.

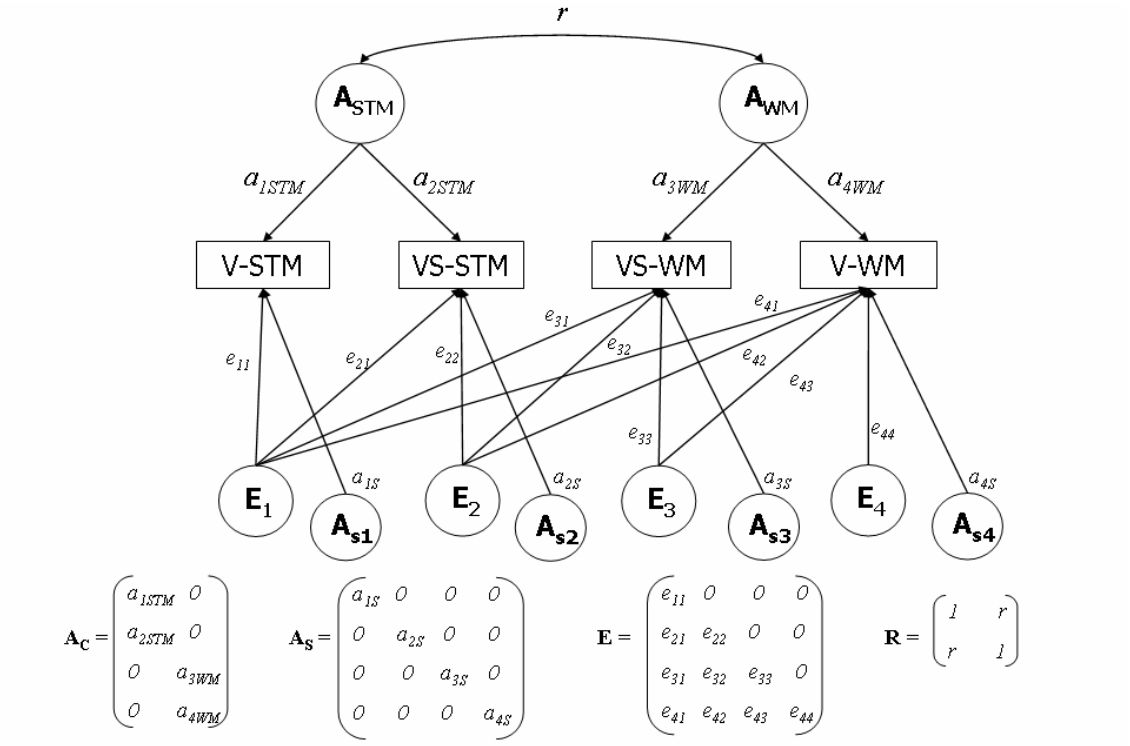


Figure 3.2. Model 1, the working memory – short-term memory model (WM-STM model) fitted on the underlying genetic (A) structure, with specific A for each variable and a saturated environmental (E) structure, and the corresponding matrices with factor loadings. In case of model 2, the visuospatial-verbal (VS-V) model, the order of the common A factors would be A_{VS} and A_V, and the variable order would be VS-WM, VS-STM, V-STM, and V-WM.

Additive genetic and unique environmental effects were modeled using a saturated four factor structure with all *factor loadings* (the loadings of the observed variables on the A or E factors) represented in two 4×4 lower triangular matrices (one for A and one for E, see matrix **E** in Figure 3.2). In a saturated factor structure all possible contributions are parameterized; therefore it yields the best possible fit to the data. First, it was tested whether genes contributed significantly to the variation in and the covariation among the four measures. This was accomplished by assessing the deterioration of model fit of the saturated four factor model after the A factor was dropped from the model.

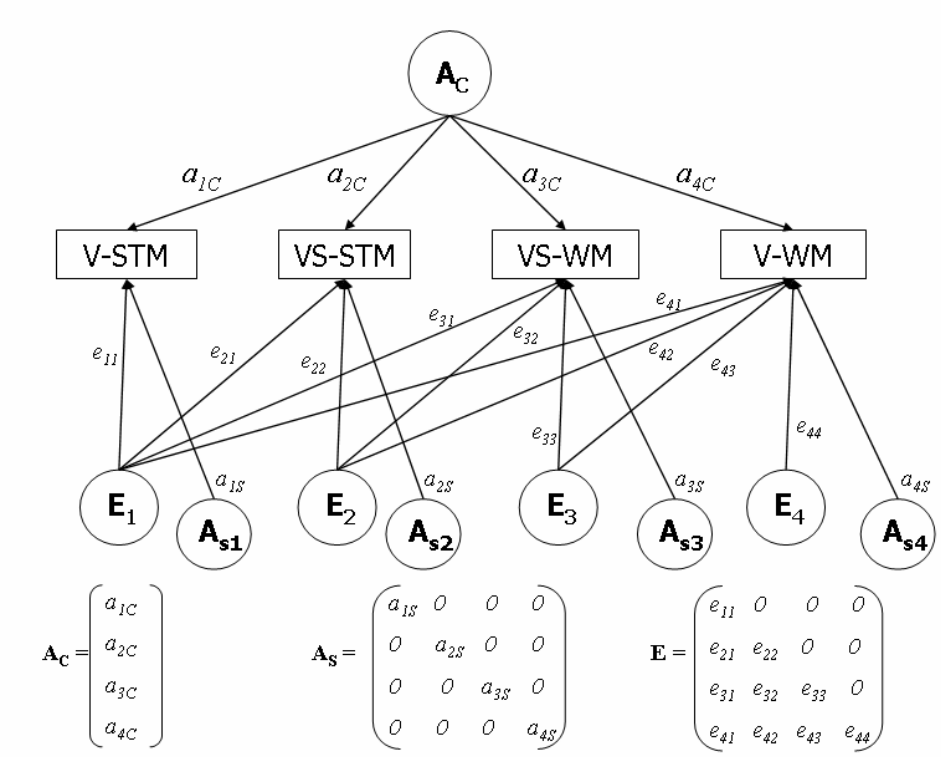


Figure 3.3. One common genetic factor (A_C), with specific genetic factors (A_S) for each variable and saturated enviromental (E) structure, and the corresponding matrices with factor loadings. V= verbal, VS=visuospatial, WM=working memory, STM= short term memory

After establishing the significance of A, based on the existing literature, six models were evaluated, to asses the underlying A structure and E structure. Model fitting started with three models to asses the underlying A structure:

1. Two common genetic factors, one for WM and one for STM (WM-STM model; see Figure 3.2, also for matrix specifications). The assumption of this model is that there is a group of common genes influencing performance on the WM tasks and a group of common genes for the STM tasks. These common genetic factors were permitted to correlate.
2. Two common genetic factors, one for verbal memory (VM) and one for visuospatial memory (VSM; VSM-VM model; see Figure 3.2). The two genetic factors were allowed to correlate with each other.

3. One genetic factor model (see Figure 3.3, also for matrix specifications). The assumption in this model is that all tasks are influenced by one set of genes.

In all models the four variables could be influenced by genetic effects specific to that task. E was modeled as a lower triangular matrix. In this way every possible contribution of E is modeled, and therefore all variance caused by E is captured. In models 1 and 2 the correlation between the two common A factors was bound between zero and one. The path diagram depicting these models is shown in Figure 3.2. This figure also contains the specification of the matrices of factor loadings. $\mathbf{A}_C$ is 4 × 2 matrix with the common genetic factor loadings, $\mathbf{A}_S$ is a 4 × 4 diagonal matrix containing the specific genetic factor loadings, and **E** is a 4 × 4 lower triangular matrix containing the unique environmental factor loadings. Within an individual, the variance in P (where P now is a four-variate phenotype) due to the two common A factors, the specific A factor and the saturated E structure is then given by:

$$\mathbf{V}_P = \mathbf{A}_C \times \mathbf{R} \times \mathbf{A}_C\text{'} + \mathbf{A}_S \times \mathbf{A}_S\text{'} + \mathbf{E} \times \mathbf{E}\text{'}$$

where ' indicates a transposed matrix, $\mathbf{V}_P$ is a 4×4 symmetrical variance/covariance matrix containing the variances of the four variables and the covariances between these variables, and **R** is a 2 × 2 standardized symmetrical matrix, with on the off-diagonal the correlation between the two A factors. Model 3 is represented in Figure 3.3, which shows one common genetic factor, which influences all four phenotypes.

Since the two correlated common genetic factor models are not nested, the three models were compared against the four variate AE model. From the three models the best fitting model was selected and subsequently model fit was improved by dropping parameters which did not significantly contribute to model fit. Consecutively, the same procedure was repeated for the factor structure of E: fitting the same three models for E with a saturated A structure. In the final model, the best fitting model for A was joined with the best fitting model for E.

Results

Means, standard deviations, and age and sex effects are reported in Table 3.1. Means were equal for MZ and DZ twins and siblings (young adult cohort: $\Delta\chi^2 = 7.850$, $\Delta df = 8$, $p = .45$; child cohort: $\Delta\chi^2 = 9.349$, $\Delta df = 8$, $p = .31$).

Table 3.1. *Maximum likelihood estimates of means, SD and age regression of the variables*.

Young adult cohort						
Variable		*N*	Mean	*SD*		Age regression
V-STM	DSF	454	8.95	1.74		-
VS-STM	Corsi	421	19.34	3.51		.14
VS-WM	3-back	442	36.25	10.95		.20
V-WM	DSB	454	6.90	1.93		-
Child Cohort						
Variable		*N*	Mean	*SD* twins	*SD* sibs	Age regression
V-STM	DSF	322	7.23	1.53	2.15	.47
VS-STM	Corsi	310	12.80	3.91	4.78	1.24
VS-WM	2-back	313	29.69	10.40	15.77	3.38
V-WM	DSB	323	4.72	1.37	2.09	.45

Note. V = verbal; VS = visuospatial; STM = short-term memory; WM = working memory

There were no significant effects of sex on the means of the four tasks in the young adult as well as the child cohort. A significant effect of age on the means of the Corsi and *n*-back (performance increased with age) was found in the young adult cohort. In the child cohort there was a significant age effect on the means of all variables. All subsequent models were corrected for these effects. Constraining the means, the within person variance covariance matrices and the between person variance covariance matrices across both cohorts resulted in a significant deteriorations of fit (means: $\Delta\chi^2 = 298.750$, $\Delta df = 12$, $p = .00$; within person: $\Delta\chi^2 = 487.291$, $\Delta df = 20$, $p = .00$; between person: $\Delta\chi^2 = 56.892$, $\Delta df = 30$, $p = .00$). Therefore in all subsequent analyses data of the young adult and the child group could not be analyzed simultaneously.

Table 3.2. *Phenotypic, MZ and DZ/twin sibling correlations*

	Variable		DSF	Corsi	*n*-back	DSB
Young adult cohort						
Phenotypic correlations	V-STM	DSF	1.00			
	VS-STM	Corsi	0.27	1.00		
	VS-WM	3-back	0.22	0.48	1.00	
	V-WM	DSB	0.44	0.36	0.33	1.00
MZ and DZ/twin sibling correlations	V-STM	DSF	.49 / .17	0.03	0.03	0.09
	VS-STM	Corsi	0.26	.38 / .14	0.10	0.04
	VS-WM	3-back	0.18	0.40	.31 / .17	0.06
	V-WM	DSB	0.41	0.39	0.25	.39 / .08
Child Cohort						
Phenotypic correlations	V-STM	DSF	1.00			
	VS-STM	Corsi	0.21	1.00		
	VS-WM	2-back	0.17	0.31	1.00	
	V-WM	DSB	0.26	0.31	0.22	1.00
MZ and DZ/twin sibling correlations	V-STM	DSF	0.57 / .20	.09	.10	.27
	VS-STM	Corsi	0.13	0.58 / .16	.10	.14
	VS-WM	2-back	0.09	0.24	0.57 / .16	.12
	V-WM	DSB	0.30	0.36	0.24	0.40 / .12

Note. Maximum likelihood estimates of phenotypic (upper parts) and MZ and DZ/twin-sibling correlations (lower parts) between the variables corrected for age and sex. On the diagonal on the left side the MZ correlations and on the right the DZ/twin-sibling correlations, below the diagonal MZ cross correlations and above the diagonal DZ/twin-sibling cross correlations. V = verbal; VS = visuospatial; STM = short-term memory; WM = working memory

In the young adult cohort the variances and covariances among the four measures were equal for twins and siblings ($\Delta\chi^2$ = 13.652, Δdf = 10, p = .19). DZ covariances and twin-sibling covariances could be equated ($\Delta\chi^2$ = 8.494, Δdf = 10, p = .58). Therefore, in all subsequent genetic models DZ and twin-sibling covariances were equated. This way also twin-sibling covariance contributed to the estimation of A and E, which amplified the

power of the study (Posthuma & Boomsma, 2000). In the child cohort variances in twins and siblings could not be equated ($\Delta\chi^2 = 22.687$, $\Delta df = 10$, $p = .01$). In the siblings there was more variation in the DSF, 2-back and DSB. We corrected for this variance inequality by multiplying the variances of the DSF, 2-back and DSB in the siblings by a factor which equated these three variances between twins and siblings. DZ covariation could be equated to twin-sibling covariation ($\Delta\chi^2 = 9.121$, $\Delta df = 10$, $p = .52$).

In both cohorts the phenotypic correlations amongst variables were modest to moderate (see Table 3.2). In the lower parts of Table 3.2 the MZ and DZ/twin-sibling correlations are displayed on the diagonal. MZ correlations were higher than DZ/twin-sibling correlations in both cohorts, indicating genetic influence on the variance of the four variables. Below the diagonal MZ cross correlations and above the diagonal DZ/twin-sibling cross correlations are presented. In both cohorts most MZ cross correlations were higher than DZ/twin-sibling cross correlations, suggesting that genes play a role in the covariation amongst the four variables.

Model fitting results of the young adult cohort are presented in the top of Table 3.3. As was indicated by the higher MZ (cross) correlations than DZ/twin-sibling (cross) correlations, A could not be dropped from the four variate AE model without a significant deterioration of fit (see Table 3.3). Therefore, it can be concluded that genes play a significant role in variation in, and the covariation amongst the four measures.

Next, the three four-variate factor models as described above were fitted for the A and E structure separately. Comparing the three models (WM-STM model, VSM-VM model, and one common factor) for the underlying genetic structure revealed that the VSM-VM model was the best fitting model. In this model two genetic factor explained the genetic covariance amongst the four measures. All four specific genetic factors could be dropped from the model without a significant reduction of fit. Thus, none of the four measures was influenced by genes specific to that measure.

Table 3.3. *Model fitting results.*

		Young adult cohort						
model		*df*	*-2LL*	cpm	$\Delta\chi^2$	*Δdf*	*p*	*AIC*
1.	Four variate AE model	1745	12965.267					9475.267
2.	Four variate E model	1755	13022.693	1	57.426	10	.00	9512.693
3.	2 fac A (WM-STM), spec A, sat E	1746	12974.632	1	9.365	1	.00	9482.632
4.	2 fac A (VSM-VM) , spec A, sat E	1746	12967.279	1	2.012	1	.16	9475.279
5.	Common fac A, spec A, sat E	1747	12974.632	1	9.365	2	.01	9480.632
6.	2 fac A (VSM-VM), sat E	1750	12971.277	4	3.998	3	.41	9471.277
7.	2 factor E (WM-STM) , spec E, sat A	1746	12966.064	1	0.797	1	.63	9474.064
8.	2 factor E (VSM-VM) , spec E, sat A	1746	12965.221	1	-0.046	1	inc.	9473.221
9.	Common fac E, spec E, sat A	1747	12966.064	1	0.797	2	.67	9472.064
10.	Common fac for VS, spec E, sat A	1750	12970.547	8	5.326	4	.26	9470.547
11.	**2 fac A (VSM-VM), one fac for E VS, spec E**	**1755**	**12981.129**	**1**	**15.862**	**10**	**.10**	**9471.129**
		Child cohort						
1.	Four variate AE model	1237	8777.668					6303.668
2.	2 fac A (VSM-VM), one fac for E VS, spec E	1247	8800.753	1	23.085	10	.01	6306.753
3.	**2 fac A (VSM-VM), one fac for E VS, spec A, spec E**	**1243**	**8782.614**	**1**	4.946	**6**	**.55**	**6296.614**

Note. Best fitting model bold faced. -2LL = -2 log likelihood; df = degrees of freedom; cpm = compared to model; AIC = Akaike's Information Criterion; A = additive genetic factor; E = environmental factor; VM = verbal memory; VS(M) = visuospatial (memory); STM = short-term memory; WM = working memory; spec = specific, sat = saturated, fac = factor

Fitting the three models on the underlying E structure revealed that the unique environmental influences were also best captured by the VSM-VM model. The verbal factor could be dropped without a significant deterioration of fit. Hence, only visuospatial WM and STM are influenced by the same environmental factor; this factor explains part of the covariance between these measures.

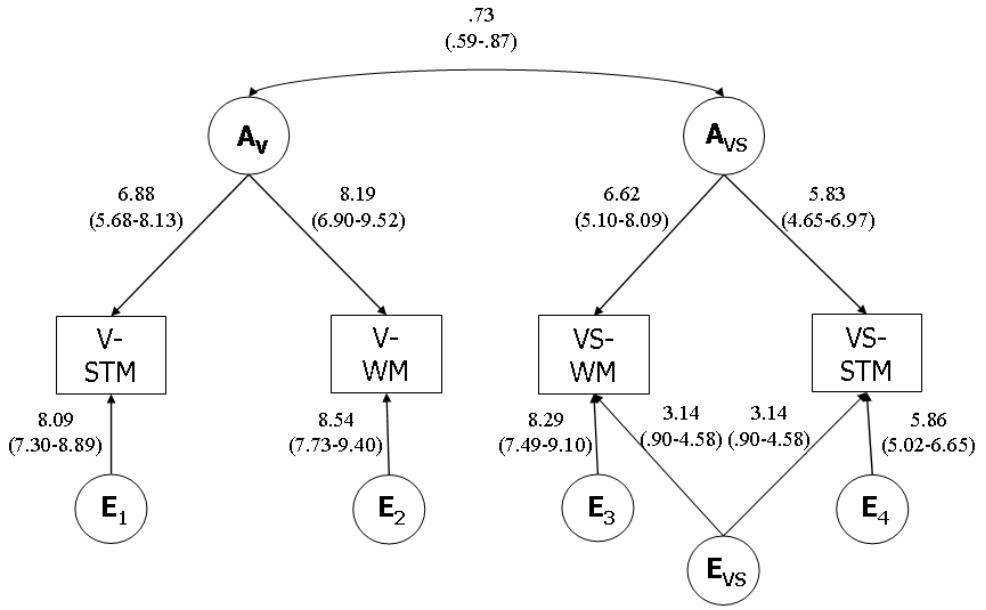


Figure 3.4. Best fitting genetic factor model in the young adult group with (unstandardized) factor loadings and their confidence intervals between brackets. A= genetic factor; E = environmental factor; V = verbal; VS = visuospatial; STM = short-term memory; WM = working memory

Thus, the final AE model in the young adult cohort consisted of two correlated factors for verbal and visuospatial memory explaining all genetic variance, one common environmental factor for the visuospatial memory tasks, and one specific E factor for each variable (see Figure 3.4 and Table 3.4). The factor loadings and their confidence intervals are given in Figure 3.4. The correlation between the genetic factor for verbal memory and visuospatial memory was .73. The environmental factor between the VSM tasks explained 20% of the observed correlation between the visuospatial tasks. All other phenotypic correlations and the remaining covariance between the VSM tasks were explained by the two common genetic factors. Approximately 40% of the individual variation in all tasks could be explained by genetic variation. The remaining variation was explained by variation in unique environmental factors.

In the child cohort dropping A from the four variate AE model also led to a significant deterioration of fit ($\Delta\chi^2 = 373.661$, $\Delta df = 10$, $p < .01$). We first tested whether the genetic factor model obtained in young adults also gave a good description for the relation amongst visuospatial and verbal WM and STM tasks in children. Therefore we

started model fitting with the final model of the young adult cohort (model 11, Table 3.3 and Figure 3.4).

Table 3.4. *Parameter estimates of the variance due to the additive genetic (A) and environmental factors (E) in the young adult cohort.*

	Unstandardised solution		Standardized solution	
Variable	A	E	A (heritability)	E
V-STM	47.32	65.48	.42	.58
VS-STM	33.97	44.15	.43	.57
VS-WM	43.79	78.59	.36	.64
V-WM	67.14	72.95	.48	.52

Note. Estimates are based on the best-fitting model. On the left of the table the unstandardised solutions and on the right standardized solutions. In the standardized solution A and E add up to 1.00. A = additive genetic factor; E = environmental factor; V = verbal; VS = visuospatial; STM = short-term memory; WM = working memory

Table 3.5. *Parameter estimates of the variance due to the additive genetic (A) and environmental factors (E) in the child cohort.*

	Unstandardised solution				Standardized solution			
Variable	A total	A common	A specific	E	A total	A common	A specific	E
V-STM	39.76	18.95	20.81	45.08	.47	.22	.25	.53
VS-STM	26.98	17.26	9.72	30.44	.47	.30	.17	.53
VS-WM	22.67	7.7	14.97	26.08	.47	.16	.31	.53
V-WM	24.50	24.50	0	44.77	.35	.35	.00	.65

Note. Estimates are based on the best-fitting model. On the left of the table the unstandardised solutions and on the right standardized solutions. In the standardized solution A total and E add up to 1.00. A = additive genetic factor; E = environmental factor; V = verbal; VS = visuospatial; STM = short-term memory; WM = working memory

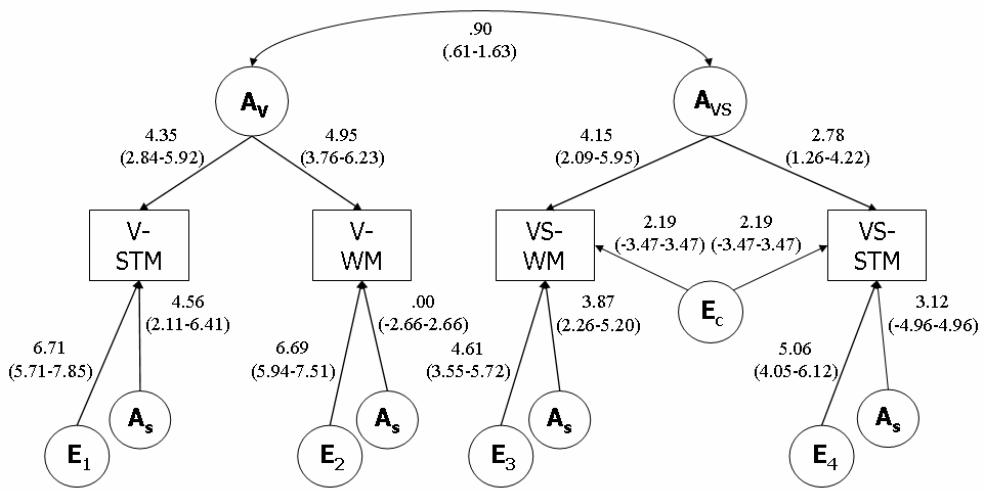


Figure 3.5. Best fitting genetic factor model in the child group with (unstandardized) factor loadings and their confidence intervals between brackets. A = genetic factor; E = environmental factor; V = verbal; VS = visuospatial; STM = short-term memory; WM = working memory

Model fitting results are presented in Table 3.3. The model which fitted best in the young adults led to a significant deterioration of fit compared to the saturated AE model. Adding specific genetic factors for each variable improved fitting results significantly. Consequently the child model consisted of 1) two correlated genetic factors for verbal and visuospatial memory, which explained all genetic covariation among the four measures; 2) one specific genetic factor for each variable, which explained additional genetic variance in each of the variables; 3) a common environmental factor for the visuospatial tasks, which explained the environmental covariance in the memory tasks in the visuospatial domain; 4) a specific environmental factor for each variable (see Figure 3.5 and Table 3.5). Hence, all phenotypic correlation between WM and STM could be explained by two common genetic factors, except for the phenotypic correlation between the visuospatial tasks which was for 29% explained by the common environmental factor. The two common genetic factors explained half of this genetic variance, except for verbal WM, where the two common genetic factors explained all genetic variance. Furthermore, about half of the variation in all tasks could be explained by genetic variance. The remaining variation in memory in children could be explained by variation in environmental factors unique to each variable. The genetic correlation between the two common genetic factors was .90, and not significantly different from 1. The common environmental factor and the specific genetic

factors for visuospatial STM and verbal WM also did not reach significance. So, in the child cohort the model is not as crystallized as in the young adult cohort.

Discussion

In this study we addressed the following questions: Are STM and WM independent or overlapping constructs and are they domain general or domain specific in nature? These questions were investigated by looking at their relationship in a genetically informative design. Thirdly, are there any developmental changes in the genetic and environmental factor structure of visuospatial and verbal WM and STM? This question was addressed by studying the genetic and environmental structure of these constructs in young adults and children.

Both in the young adult and child cohort low correlations were observed between the measures of verbal and visuospatial WM and STM at the phenotypic level. These correlations were comparable to the correlations observed in the studies of Alloway et al. (2006) and Gathercole et al. (2004). Since findings at the phenotypic level of this study are comparable to previous studies and there were no mean differences between twins and siblings, we think it is safe to assume that the findings in our twin sibling population are representative for the general population.

In the young adults three different factor models were compared: a WM-STM model, a VSM-VM model and a one common factor model. These models were fitted to the genetic and the environmental structure of visuospatial and verbal WM and STM. At the non-observed (latent) level two highly correlated common genetic factors were found, one for verbal and one for visuospatial memory, which explained most of the phenotypic correlations between visuospatial and verbal WM and STM. A common environmental factor for visuospatial memory explained the remaining phenotypic covariance. All other environmental factors were uncorrelated. The specific environmental factors could reflect measurement errors unique to each task, or unique experiences which make people perform better on one task but not on the other. The common environmental factor can possibly be explained by the fact that both tasks were administered right after each other at the end of the testing day, and might therefore reflect weariness at the end of the day of testing in some participants. On the other hand this common environmental factor can also be a true finding, since multiple studies report a higher correlation between visuospatial STM and

WM, than between verbal STM and WM (Bayliss et al., 2005a; Miyake et al., 2001; Shah & Miyake, 1996).

As in the adult cohort in the child cohort most of the phenotypic correlations were explained by a genetic factor for verbal and a genetic factor for visuospatial memory. The remaining phenotypic covariation between the visuospatial tasks was explained by an additional common environmental factor. However, the results in the children also indicated significant differences in the genetic structure of cognition in children as opposed to young adults: STM and WM were also influenced by specific genetic factors.

In the young adult and child cohort two genetic factors, one for visuospatial and one for verbal memory explained most of the phenotypic correlations between the visuospatial and verbal memory tasks, indicating that these abilities are influenced by two related sets of genes. This suggests that at the genetic level visuospatial and verbal memory are two different, but highly correlated systems. WM and STM on the other hand can not be distinguished, suggesting that at the genetic level these two systems are part of one unitary system. At the environmental level variation was mainly explained by specific environmental factors, except for one common environmental factor for the visuospatial memory tasks. This means that variability in verbal and visuospatial STM and WM is caused by environmental influences which are specific to each of the variables. Environmental events (e.g. experience) make that visuospatial and verbal WM and STM are distinct cognitive processing units. This is in concordance with the neuroconstructivist view which states that cognitive modules are a consequence of the developmental process of modularization and specific environmental interactions (Karmiloff-Smith, 2006; Karmiloff-Smith, 1998).

Based on the fact that two highly correlated common genetic factors described the data best, it can be concluded that seen from a genetic perspective WM and STM are domain specific constructs which can not be distinguished from each other. At the environmental level a different pattern is seen; visuospatial and verbal WM and STM hardly share any common environmental variance, and seem to be different abilities. So it seems that from a genetic perspective STM consists of executive functioning and WM also includes short-term memory, which supports the theory of Kane et al. (2004). Further, it seems that at the genetic level there are separate storage and executive function mechanisms at work for visuospatial and verbal information. (Haavisto & Lehto, 2005;

Tillman et al., in press). The mixed findings reported in studies on WM and STM could be a consequence of genetic molarity and environmental modularity. These findings are in concordance with the view of Price et al. (2000) and Petrill (1997) who suggest that genetic influences are responsible for linking diverse areas of cognitive functioning (genetic molarity), whereas environmental effects create differences between different domains of cognitive functioning (environmental modularity; e.g. Luo, et al. (1994) and Pedersen, et al. (1994)).

In children, it was shown that, apart from two common genetic factors, specific genetic factors also explain part of the variability in the four abilities: each ability is also influenced by a genetic factor which is specific to that ability. Thus in children, verbal and visuospatial WM and STM are only partly overlapping abilities at this age. So, in contrast to our expectations the correlation between genetic factors that represent different domains of cognition increases with age. This means that with age different cognitive abilities start to develop into one general system. A similar finding had been reported by Casto, DeFries, and Fulker (1995), Hoekstra et al. (2007), Price et al. (2000) and Rietveld, Dolan, Van Baal, and Boomsma (2003). They concluded that genetic effects on cognitive abilities may be more modular in early development and become increasingly molar later in life.

One limitation of this study is that only one measure for each construct was used. Using multiple indicators would have made our claims stronger. However, in a longitudinal study design it is not feasible to let children return multiple times to finish one test battery without a significant loss of participants on future test occasions.

Based on this study we can speculate what the common genetic factors for visuospatial and verbal memory represent. From twin studies it is known that brain structure is highly heritable (Baaré et al., 2001; Hulshoff Pol et al., 2006): differences in brain structure between people are caused by genetic variability between people. The study of Posthuma et al. (2003) showed that WM performance and brain volumes are genetically related. Since there is a genetic relation between memory performance and brain structure, it is possible that the genetic correlation between verbal and visuospatial WM and STM represents processing of STM and WM by the same brain structures. The two different factors for verbal and visuospatial memory probably reflect processing by respectively the visual and auditory cortex. Whether this truly is the case, should be addressed by future studies, combining measures of brain structure and memory performance. For future studies

it would also be of interest to learn more about what kind of processes are captured by the two common genetic factors. Do they capture storage, executive functioning, general intelligence, or a combination? Another direction of research would be to take a closer look at the specific genetic factors found in children: what do they represent and when do they disappear? Also it is important to see if these findings can be replicated using different STM and WM tasks. Finally, by following the child group longitudinally we can establish if the effect of increasing genetic correlations between memory measures with age can be replicated.

To conclude, two major findings were obtained in this study. First, two genetic factors are responsible for explaining the association between verbal and visuospatial WM and STM, whereas environmental factors create differences between these domains. This means that performance on visuospatial and verbal WM and STM is influenced by two highly correlated sets of genes. Therefore, from a genetic viewpoint one could say that WM and STM are in essence part of the same system, and verbal and visuospatial information are processed using two partly overlapping memory pathways. Second, during the course of development the specific genetic factors, which create differences between the four abilities, disappear. This suggests that with aging these cognitive abilities start to become part of two genetic systems, one for verbal memory and one for visuospatial memory.

Reference List

Alloway, T P, Gathercole, S E, & Pickering, S J (2006). Verbal and visuospatial short-term and working memory in children: are they separable? *Child Development, 77*, 1698-1716.

Ando, J, Ono, Y, & Wright, M J (2001). Genetic structure of spatial and verbal working memory. *Behavior Genetics, 31*, 615-624.

Baaré, W F, Hulshoff Pol, H E, Boomsma, D I, Posthuma, D, De Geus, E J, Schnack, H G, Van Haren, N E, Van Oel, C J, & Kahn, R S (2001). Quantitative genetic modeling of variation in human brain morphology. *Cerebral Cortex, 11*, 816-824.

Baddeley, A (1992). Working memory. *Science, 255*, 556-559.

Baddeley, A (2000). The episodic buffer: a new component of working memory? *Trends in Cognitive Sciences, 4*, 417-423.

Baddeley, A (2003). Working memory: looking back and looking forward. *Nature Reviews Neuroscience, 4*, 829-839.

Baddeley, A.D. & Hitch, G. (1974). Working-memory. In: G.H. Bower *The psychology of learning and motivation: Advances in research and theory* (pp. 47-90) New York: Academic Press

Baddeley, A (1996). Exploring the Central Executive. *The Quarterly Journal of Experimental Psychology Section A, 49*, 5-28.

Bartels, M, Rietveld, M J H, Van Baal, G C M, & Boomsma, D I (2002). Genetic and environmental influences on the development of intelligence. *Behavior Genetics, 32*, 237-249.

Bayliss, D M, Jarrold, C, Baddeley, A D, & Gunn, D M (2005a). The relationship between short-term memory and working memory: complex span made simple? *Memory, 13*, 414-421.

Bayliss, D M, Jarrold, C, Baddeley, A D, Gunn, D M, & Leigh, E (2005b). Mapping the developmental constraints on working memory span performance. *Developmental Psychology, 41*, 579-597.

Bayliss, D M, Jarrold, C, Gunn, D M, & Baddeley, A D (2003). The complexities of complex span: Explaining individual differences in working memory in children and adults. *Journal of Experimental Psychology: General, 132*, 71-92.

Boomsma, D, Busjahn, A, & Peltonen, L (2002). Classical twin studies and beyond. *Nature Reviews Genetics, 3*, 872-882.

Boomsma, D I, De Geus, E J C, Vink, J M, Stubbe, J H, Distel, M A, Hottenga, J J, Van Beijsterveldt, C E M, Hudziak, J J, Bartels, M, & Willemsen, G (2006). Netherlands Twin Register: from twins to twin families. *Twin Research and Human Genetics, 9*, 849-857.

Boomsma, D I & Molenaar, P C (1986). Using LISREL to analyze genetic and environmental covariance structure. *Behavior Genetics, 16*, 237-250.

Boomsma, D I, Vink, J M, Van Beijsterveldt, T C, De Geus, E J, Beem, A L, Mulder, E J, Derks, E M, Riese, H, Willemsen, G A, Bartels, M, Van den Berg, M, Kupper, N H, Polderman, T J, Posthuma, D, Rietveld, M J, Stubbe, J H, Knol, L I, Stroet, T, & Van Baal, G C (2002). Netherlands Twin Register: A focus on longitudinal research. *Twin Research, 5*, 401-406.

Cantor, J, Engle, R W, & Hamilton, G (1991). Short-term memory, working memory, and verbal abilities: How do they relate? *Intelligence, 15*, 229-246.

Casey, B J, Giedd, J N, & Thomas, K M (2000). Structural and functional brain development and its relation to cognitive development. *Biological Psychology, 54*, 241-257.

Casto, S D, DeFries, J C, & Fulker, D W (1995). Multivariate genetic analysis of Wechsler Intelligence Scale for Children--Revised (WISC-R) factors. *Behavior Genetics, 25*, 25-32.

Chuah, Y M & Maybery, M T (1999). Verbal and spatial short-term memory: common sources of developmental change? *Journal of Experimental Child Psychology, 73*, 7-44.

Colom, R, Flores-Mendoza, C, Quiroga, M A, & Privado, J (2005). Working memory and general intelligence: The role of short-term storage. *Personality and Individual Differences, 39*, 1005-1014.

Conway, A R A, Cowan, N, Bunting, M F, Therriault, D J, & Minkoff, S R B (2002). A latent variable analysis of working memory capacity, short-term memory capacity, processing speed, and general fluid intelligence. *Intelligence, 30*, 163-183.

Corsi, P M (1974). Human memory and the medial temporal region of the brain. *Dissertation Abstracts International, 34*, 819B (University Microfilms No. AAI05-77717)-

Cowan, N. (1995) *Attention and memory: An integrated framework.* Oxford: Oxford University Press

Cowan, N (1988). Evolving conceptions of memory storage, selective attention, and their mutual constraints within the human information-processing system. *Psychological Bulletin, 104*, 163-191.

Engle, R W, Tuholski, S W, Laughlin, J E, & Conway, A R A (1999). Working memory, short-term memory, and general fluid intelligence: A latent-variable approach. *Journal of Experimental Psychology: General, 128*, 309-331.

Garret, H E (1946). A developmental theory of intelligence. *American Psychologist, 1*, 372-378.

Gathercole, S E, Pickering, S J, Ambridge, B, & Wearing, H (2004). The structure of working memory from 4 to 15 years of age. *Developmental Psychology, 40*, 177-190.

Gevins, A & Cutillo, B (1993). Spatiotemporal dynamics of component processes in human working memory. *Electroencephalography and Clinical Neurophysiology., 87*, 128-143.

Haavisto, M L & Lehto, J E (2005). Fluid/spatial and crystallized intelligence in relation to domain-specific working memory: A latent-variable approach. *Learning and Individual Differences, 15*, 1-21.

Hoekstra, R A, Bartels, M, & Boomsma, D I (2007). Longitudinal genetic study of verbal and nonverbal IQ from early childhood to young adulthood. *Learning and Individual Differences, 17*, 97-114.

Hoshi, Y, Oda, I, Wada, Y, Ito, Y, Yutaka, Y, Oda, M, Ohta, K, Yamada, Y, & Mamoru, T (2000). Visuospatial imagery is a fruitful strategy for the digit span backward task: a study with near-infrared optical tomography. *Cognitive Brain Research, 9*, 339-342.

Hulshoff Pol, H E, Schnack, H G, Posthuma, D, Mandl, R C, Baare, W F, Van Oel, C, Van Haren, N E, Collins, D L, Evans, A C, Amunts, K, Burgel, U, Zilles, K, De Geus, E, Boomsma, D I, & Kahn, R S (2006). Genetic contributions to human brain morphology and intelligence. *Journal of Neuroscience, 26*, 10235-10242.

Jansma, J M, Ramsey, N F, Coppola, R, & Kahn, R S (2000). Specific versus nonspecific brain activity in a parametric n-back task. *Neuroimage, 12*, 688-697.

Jones, D.M., Beaman, P., & Macken, W.J. (1996). The object-oriented episodic record model. In: S.E. Gathercole *Models of short-term memory* (pp. 209-237) Hove, England: Erlbaum

Jones, D, Farrand, P, Stuart, G, & Morris, N (1995). Functional equivalence of verbal and spatial information in serial short-term memory. *Journal of Experimental Psychology: Learning, 21*, 1008-1018.

Kail, R & Hall, L K (2001). Distinguishing short-term memory from working memory. *Memory and Cognition, 29*, 1-9.

Kane, M J, Hambrick, D Z, Tuholski, S W, Wilhelm, O, Payne, T W, & Engle, R W (2004). The generality of working memory capacity: a latent-variable approach to verbal and visuospatial memory span and reasoning. *Journal of Experimental Psychology: General, 133*, 189-217.

Karmiloff-Smith, A (2006). The tortuous route from genes to behavior: A neuroconstructivist approach. *Cognitive, Affective & Behavioral Neuroscience, 6*, 9-17.

Karmiloff-Smith, A (1998). Development itself is the key to understanding developmental disorders. *Trends in Cognitive Sciences, 2*, 389-398.

Kooij, A.P., Rolfhus, E., Wilkins, C., Yang, Z., & Zhu, J. (2004) *WAIS-III Nederlandstalige bewerking. Technisch rapport hernormering. [WAIS-III Dutch Version. Technical report restandardization].* Amsterdam: Harcourt Test Publishers

Kremen, W S, Jacobsen, K C, Xian, H, Eisen, S A, Eaves, L J, Tsuang, M T, & Lyons, M J (2007). Genetics of verbal working memory processes: A twin study of middle-aged men. *Neuropsychology, 21*, 569-580.

Kuntsi, J, Rogers, H, Swinard, G, Borger, N, Van der Meere, J, Rijsdijk, F, & Asherson, P (2006). Reaction time, inhibition, working memory and 'delay aversion' performance: genetic influences and their interpretation. *Psychological Medicine, 36*, 1613-1624.

Lee, Y s, Lu, M j, & Ko, H p (2007). Effects of skill training on working memory capacity. *Learning and Instruction, 17*, 336-344.

Luo, D, Petrill, S A, & Thompson, L A (1994). An exploration of genetic g: Hierarchical factor analysis of cognitive data from the western reserve twin project. *Intelligence, 18*, 335-347.

Maehara, Y & Saito, S (2007). The relationship between processing and storage in working memory span: Not two sides of the same coin. *Journal of Memory and Language, 56*, 212-228.

Marquardt, N D & Bayless, J D (1999). Verbal recall and learning vs. working memory in children and adolescents. *Archives of Clinical Neuropsychology, 14*, 13-13.

Martin, N G & Eaves, L J (1977). The genetical analysis of covariance structure. *Heredity, 38*, 79-95.

Miyake, A, Friedman, N P, Rettinger, D A, Shah, P, & Hegarty, M (2001). How are visuospatial working memory, executive functioning, and spatial abilities related? A latent-variable analysis. *Journal of Experimental Psychology: General, 130*, 621-640.

Mulder, J.L., Dekker, R., & Dekker, P.H. (1996) *Verbale Leer en Geheugen Test Handleiding [Verbal Learning and Memory Test Manual].* Amsterdam, The Netherlands: Harcourt Test Publishers

Neale, M., Boker, S.M., Xie, G., & Maes, H.H. (2006) *Mx: statistical modelling.* Richmond, VA: Department of Psychiatry, Medical College of Virginia

Pedersen, N L, Plomin, R, & McClearn, G E (1994). Is there G beyond g? (Is there genetic influence on specific cognitive abilities independent of genetic influence on general cognitive ability?). *Intelligence, 18*, 133-143.

Petrill, S A (1997). Molarity Versus Modularity of Cognitive Functioning? A Behavioral Genetic Perspective. *Current Directions in Psychological Science, 6*, 96-99.

Plomin, R., DeFries, J.C., McClearn, G.E., & McGuffin, P. (2001) *Behavioral genetics.* New York: Worth Publisher

Polderman, T J C, Stins, J F, Posthuma, D, Gosso, M F, Verhulst, F C, & Boomsma, D I (2006). The phenotypic and genotypic relation between working memory speed and capacity. *Intelligence, 34*, 549-560.

Posthuma, D, Baare, W F, Hulshoff Pol, H E, Kahn, R S, Boomsma, D I, & De Geus, E J (2003). Genetic correlations between brain volumes and the WAIS-III dimensions of verbal comprehension, working memory, perceptual organization, and processing speed. *Twin Research, 6*, 131-139.

Posthuma, D & Boomsma, D I (2000). A note on the statistical power in extended twin designs. *Behavior Genetics, 30*, 147-158.

Price, T S, Eley, T C, Dale, P S, Stevenson, J, Saudino, K, & Plomin, R (2000). Genetic and environmental covariation between verbal and nonverbal cognitive development in infancy. *Child Development, 71*, 948-959.

Reinert, G. (1970). Comparative factor analytic studies of intelligence troughout the human life-span. In: L.R. Goulet & P.B. Baltes *Life-span Developmental Psychology* (pp. 467-484) New York: Academic Press

Rietveld, M J, Van der Valk, J C, Bongers, I L, Stroet, T M, Slagboom, P E, & Boomsma, D I (2000). Zygosity diagnosis in young twins by parental report. *Twin Research, 3*, 134-141.

Rietveld, M J H, Dolan, C V, Van Baal, G C M, & Boomsma, D I (2003). A twin study of differentiation of cognitive abilities in childhood . *Behavior Genetics, 33*, 367-381.

Shah, P & Miyake, A (1996). The separability of working memory resources for spatial thinking and language processing: An individual differences approach. *Journal of Experimental Psychology: General, 125*, 4-27.

Spearman, C. (1927) *The abilities of man: Their nature and measurement.* New York: MacMillan

Swanson, H L (1992). Generality and modifiability of working memory among skilled and less skilled readers. *Journal of Educational Psychology, 84*, 473-488.

Swanson, H L (1999). What develops in working memory? A life span perspective. *Developmental Psychology, 35*, 986-1000.

Swanson, H L & Berninger, V W (1996). Individual differences in children's working memory and writing skill. *Journal of Experimental Child Psychology, 63*, 358-385.

Tillman, C M, Nyberg, L, & Bohlin, G (in press). Working memory components and intelligence in children. *Intelligence.*

Van den Burg, W & Kingma, A (1999). Performance of 225 Dutch school children on Rey's Auditory Verbal Learning Test (AVLT): parallel test-retest reliabilities with an interval of 3 months and normative data. *Archives of Clinical Neuropsychology, 14*, 545-559.

Van Leeuwen, M, Van den Berg, S M, Hoekstra, R A, & Boomsma, D I (2007). Endophenotypes for intelligence in children and adolescents. *Intelligence, 35*, 369-380.

Van Leeuwen, M, Van den Berg, S M, & Boomsma, D (2008). A twin-family study of general IQ. *Learning and Individual Differences, 18*, 76-88.

Wechsler, D. (1997) *Wechsler Adult Intelligence Scale-Third edition, Dutch Version.* Amsterdam: The Psychological Corporation Ltd. Harcourt Publishers

Wechsler, D., Kort, W., Compaan, E.L., Bleichrodt, N., Resing, W.C.M., Schittekatte, M., Bosmans, M., Vermeir, G., & Verhaeghe, P. (2002) *Handleiding WISC-III^{Nl} [Manual WISC-III^{Nl}].* London: The Psychological Corporation Limited, Nederlands Instituut van Psychologen Dienstencentrum

The genetic relationship between reading ability, intelligence and memory in children.

This study investigated the genetic relationship between reading ability, IQ, working memory (WM) and short-term memory (STM) in 111 nine-year-old twin pairs and their siblings (9-14 years old). Reading ability was assessed using the one minute reading test, IQ using the WISC, verbal STM and WM using digit span forward and backward, and visuospatial STM and WM using the Corsi block tapping task and the n-back task. The relationship between reading ability and the other measures was completely explained by two common genetic factors: a common factor for reading ability, IQ, WM and STM and a common factor for reading ability and verbal memory. Genetic variation in reading ability was for about 53% explained by genetic variation in IQ and memory performance. Thus, based upon the genetic factors involved in reading ability, children with reading disabilities can be divided in three groups: 1) children who are low in IQ and therefore have problems with reading; 2) children who have normal IQ and a deficit in STM and therefore experience problems with reading; 3) children with low IQ and deficits in STM, this group experiences more reading problems than the other two.

This chapter is based on:

Van Leeuwen, M., Van den Berg, S. M., Peper, J. S., Hulshoff Pol, H.E., & Boomsma, D. I. (submitted). The genetic relationship between reading ability, intelligence and memory in children. *Behavior Genetics*

Dyslexia, or specific reading disability, is fairly widespread, with a prevalence estimated between 5% and 17.5% (Démonet, Taylor, & Chaix, 2004). "[It] is defined as an unexpected, specific, and persistent failure to acquire efficient reading skills despite conventional instruction, adequate intelligence, and sociocultural opportunity" (see Démonet et al. (2004), pg 1451). Several authors argue against the use of discrepancy scores (based on differences between IQ and reading achievement scores) to identify children with reading disability (Stanovich, 1993; Sternberg & Grigorenko, 2002).

The relationship between reading ability and IQ has been well established in affected groups and in non-affected groups with reading disability (Tiu, Jr., Thompson, & Lewis, 2003). Several studies in children have investigated the heritability of reading ability in relation to intelligence (e.g. Cardon, Dilalla, Plomin, DeFries, & Fulker, 1990; Thompson, Detterman, & Plomin, 1991; Tiu, Jr., Wadsworth, Olson, & DeFries, 2004). Most of these studies are based on the Colorado Twin Study of Reading Disability (Alarcón & DeFries, 1997; Brooks, Fulker, & DeFries, 1990; Gayán & Olson, 2003; Hawke, Wadsworth, & DeFries, 2006; Pennington, Gilger, Olson, & DeFries, 1992; Tiu, Jr. et al., 2004). That sample consists of 640 twins with reading disabilities and 436 control twins, with an overall mean age of 12 years (Pennington et al., 1992). These studies generally report heritability estimates for reading ability of around 50% and a moderate phenotypic correlation between reading ability and intelligence completely mediated by genes. This suggests that reading ability and intelligence have a common genetic origin.

Alarcón & DeFries (1997) investigated whether the genetic background of reading ability and intelligence was the same for twin pairs which were selected for reading disabilities as for control twin pairs. There were no fundamental differences between the groups in heritability of general cognitive ability. However, the genetic and phenotypic variances and covariances amongst the reading measures were larger for the affected than for the control group, with higher heritabilities for reading performance in the affected. This is in concordance with the hypothesis that DNA polymorphisms for reading disability are more prevalent in this group. The phenotypic correlations between reading performance and cognitive ability were larger for the control group than the affected group (respectively $r = .76$ and $r = .41$), as well as the genetic correlations (respectively $r = .81$ and $r = .52$).

In the literature on individual differences in reading ability the relationship between reading ability, working memory (WM) and short-term memory (STM), is a subject of debate (Cohen-Mimran & Sapir, 2007; Gathercole, Alloway, Willis, & Adams, 2006; Kercher & Sandoval, 1991; Swanson & Jerman, 2007). STM is the capacity to store material over short periods of time in situations that do not impose other competing cognitive demands (Gathercole et al., 2006). WM is the system that is necessary for the concurrent storage and manipulation of information (Baddeley, 1992). WM constitutes of the central executive and three storage systems: the phonological loop, the visuospatial sketchpad and the episodic buffer. The central executive is the system responsible for a range of regulatory functions, including attention, the control of action, and problem solving (Baddeley, 1996). The phonological loop comprises a phonological store that can hold memory traces for a few seconds before they fade, and an articulatory rehearsal process. The visuospatial sketchpad is its visuospatial counterpart (Baddeley, 2003). In multiple studies the phonological loop and the visuospatial sketchpad are considered equivalent to STM (e.g. Gathercole, Pickering, Ambridge, & Wearing, 2004). The episodic buffer provides temporary storage of information held in a multimodal code, which is capable of integrating information from a variety of sources, including long-term memory, into a unitary episodic representation (Baddeley, 2000).

The findings on the relationship between reading disability and STM deficits are rather mixed. For instance Kercher and Sandoval (1991) and Swanson and Ashbaker (2000) did find that children with reading disability performed poorly on STM tasks while Gathercole et al. (2006) and Swanson and Jerman (2007) did not find a relation between reading disability and STM. A possible explanation for a relation between STM and reading disability is that in children with reading disability the ability to code information phonemically or verbally is affected, which is an important aspect of STM (Kercher & Sandoval, 1991). This theory is in concordance with the difficulties in phonemic coding strategies observed in dyslexic children (Snowling, 1980).

Most studies agree that children with reading disabilities score poorly on WM tasks (Gathercole et al., 2006; Swanson, 2003; Swanson & Ashbaker, 2000; Swanson & Berninger, 1995), although Van der Sluis, Van der Leij, and De Jong (2005) did not find any WM deficits in children with reading disability. Further, Swanson and Berninger (1995) and Swanson and Ashbaker (2000) found that WM contributed independently from

STM to the reading deficits found in less skilled readers. A possible explanation for the relation between WM and reading disabilities is that impairments of WM result in reading disability because the system serves as a bottleneck for learning: children with low WM skills will have difficulties in meeting the routine WM demands of structured learning activities that are necessary for learning to read (Gathercole et al., 2006). Since reading ability, STM and WM all are correlated with intelligence (e.g. Alarcón & DeFries, 1997; Colom, Abad, Rebollo, & Chun Shih, 2005; Kane et al., 2004; Polderman et al., 2006), another explanation for the relation between WM, STM and reading ability is that all three are correlated by means of their correlation with intelligence.

The genetic relation between reading ability and memory, and especially WM is less well established. In children, only one study investigated the relation between verbal STM and reading ability in a genetically informative design (Wadsworth, DeFries, Fulker, & Olson, 1995). A moderate correlation between reading ability and verbal STM was observed, which was for 80% accounted for by genetic factors.

Here, we will explore whether there is a phenotypic correlation between WM, STM and reading ability independent of intelligence. Moreover, we want to investigate, whether STM and WM independently contribute to reading disability. By relating intelligence, WM, STM and reading ability in a genetically informative design we will be able to test whether the phenotypic correlations between WM, STM, and intelligence and reading ability stem from common genes. We hypothesize that memory and intelligence contribute independently to the variance in reading ability. In addition, we hypothesize that the phenotypic correlation between the three constructs and reading ability can be explained by common sets of genes. Recurring findings show that a disorder is more severe when underlying dissociated deficits co-occur. So, if we find that the genetic association between memory and reading ability is independent from the association between IQ and reading ability, this suggests that a combination of deficits in theses three areas is a sign of the severity of the reading disability rather than a symptom of reading disability per se (Bishop, 2006).

We will test these hypotheses in a sample of nine-year-old twins and their siblings aged nine to fourteen. Children were tested for reading ability, memory performance and IQ.

Material and Methods

Participants

The group of participants in this study consisted of 112 nine-year-old twin pairs (M = 9.1, SD = .1) and one of their siblings aged nine to fourteen (N = 100, M = 11.8, SD = 1.2). Children were recruited from the Netherlands Twin Registry (NTR; (Boomsma et al., 2002). This group takes part in a study on the development of cognition and brain structure (Peper et al., in press; Van Leeuwen, Van den Berg, & Boomsma, 2008), and included 23 MZM, 23 DZM, 25 MZF, 21 DZF, and 20 DOS 56 female siblings. The study was approved by the Central Committee on Research involving Human Subjects (CCMO), and parents signed an informed consent form for their children.

Of these participants 21 MZM, 22 DZM (including 1 incomplete), 23 MZF, 21 DZF (including 1 incomplete pair), and 19 DOS (including 3 incomplete pairs) pairs and 82 siblings (44 female) were tested for reading disability.

Protocol

All children were individually tested at VU University in separate rooms by experienced test administrators. A testing day consisted of a psychological test protocol only. Testing lasted for about five hours (including three breaks). Children completed as part of a larger test battery the Corsi block tapping task, the n-back task and the WISC-III. Most of the families went, after they had been to the VU University, to the University Medical Centre of Utrecht (UMCU) for a magnetic resonance (MR) scan. Children were tested for reading ability prior to the MR scan. Average time between testing at the VU and the UMCU was 43 days (VU before UMCU) ranging from 63 days before testing at the VU until 124 days after testing at the VU (SD = 35).

Measures

Reading: One minute reading test

One subtest of the ‘one minute reading test’ (OMRT; Cito, 1995; the norms date from 2003) was administered as a measure of technical reading ability, or oral reading fluency. Children were instructed to read out loud as many words as possible in one minute without making errors from a card containing 120 unrelated words. The OMRT is a standardized

test frequently used in Dutch education as a measure of early reading ability ?(Van der Sluis et al., 2005) and corresponds well with other instruments (Moelands, Kamphuis, & Verhoeven, 2008). Nine year old children are suspected to be dyslexic when they score below 28 words a minute. Test-retest reliability in the nine year olds is .92 (Moelands et al., 2008).

Short-term spatial memory: Corsi block tapping task

The Corsi block tapping task (Corsi, 1974) was included to assess short-term spatial memory. Children sat in front of a touch screen monitor on which nine white blocks were displayed unevenly across a gray screen. In succession a number of blocks turned red for one second, after which the screen was blank for three seconds. After reappearance of the blocks, the child had to tap the blocks on the screen in the same sequence in which they had changed color before. When a block was tapped, the block would turn red and stay that way until the end of the run. The computer registered each tap. Each child was given two practice runs. In these runs each person had to memorize two blocks. Immediately after the practice runs the actual test was administered, starting with a series of two blocks. After every five runs the item length was increased by one block. The test ended when the child responded incorrectly to three out of five runs of the same length. The maximum number of blocks that could turn red in succession was nine. Performance was measured by total number of correct runs.

Visuospatial working memory: *n*-back task

Children had to perform a spatial variant of the *n*-back task to assess visuospatial working memory. The *n*-back used in this protocol was designed after Gevins and Cutillo (1993) and Jansma, Ramsey, Coppola, and Kahn (2000) with increasing levels of difficulty. The children were asked to look at an apple presented on a screen. The apple had four holes in which a caterpillar could appear. Children were told to catch the caterpillar to prevent it from eating the apple, and were instructed to respond to the caterpillar by pushing one of four buttons with the thumb and index finger of both hands. The layout of the four buttons corresponded spatially to the four holes in which the caterpillar could appear. Children had to indicate where the caterpillar was one move back (1-back), two moves back (2-back), or three moves back (3-back). The caterpillar appeared in a hole for one second; after its

disappearance there was a warning sound. Children were instructed to respond after this warning sound and could respond until the next caterpillar appeared. Between two caterpillar moves, the apple was empty for one second.

Sessions were given in sessions of 20 trials. Each condition consisted of a practice session and three sessions in which performance was recorded. Practicing continued until the participants understood the task. The 1-back condition was administered for practice purposes only, performance was recorded on the 2-back and 3-back conditions. Children were motivated during the task by counting the moves of the caterpillar. In the 2-back version the test administrator counted continuously to three and in the 3-back version the administrator counted to four. After each session children received feedback on the number of apples they had saved from the caterpillar (correct responses) and how many had been eaten (incorrect responses). Following the feedback there was a break of 15 seconds. The task requires a continuous response to all stimuli and simultaneous monitoring and update of all movements of the caterpillar. Performance on the task was scored by using the total number of correct responses. Maximum score per condition was 60. For this study we used performance on the 2-back condition. For children the test-retest on 2-back is .65 (Van Leeuwen, Van den Berg, Hoekstra, & Boomsma, 2007).

Verbal working and short-term memory: digit span forwards and backwards

Digit span forwards (DSF) of the WISC-III (Wechsler et al., 2002) was used to measure verbal short-term memory. In this task participants had to recall lists of numbers. The test started with a trial of two numbers. If participants recalled one out of two trials correctly, the list increased with one digit. Increments proceeded, until participants had both of two trials wrong. Performance was scored as the total number of correct trials. To measure verbal working memory the digit span backwards task (DSB) was used. This time the participants had to recall lists of numbers in reverse order. The test-retest coefficient over three years of digit span (forward and backward together) of the WISC- Revised is .53 (Livingston, Jennings, Reynolds, & Gray, 2003), the split half coefficient for the internal consistency of digit span of the WISC-III is .67 (Wechsler et al., 2002).

IQ: WISC

Psychometric IQ was measured with the Dutch adaptation of the WISC-III (Wechsler et al., 2002). IQ was based on 10 subtests (information, similarities, arithmetic, vocabulary, comprehension, block design, picture completion, picture arrangement, object assembly and digit-symbol substitution). The two digit span subtests were not included in the total IQ score. Cronbach's α for total IQ is .93 (Wechsler et al., 2002).

Statistical Analyses

All data analyses were performed with the Mx software package (Neale, Boker, Xie, & Maes, 2006). First, general covariance matrices, means and sex regressions on the means were estimated in a saturated model. By fitting nested models in which the means and variances between twins and siblings were equated, several assumptions were tested such as equality of means between twins and siblings. After testing equality of means, sex and age effects on the means were tested. We continued equating parameters until the most parsimonious model with still acceptable fit was established. The choice for the best fitting model was based on likelihood-ratio tests. The difference between minus twice the log likelihoods (*-2LL*) of two nested models, asymptotically follows a χ^2 distribution. The degrees of freedom are given by the difference in the number of parameters estimated in the two nested models. A high increase in χ^2 against a low gain of degrees of freedom denotes a worse fit of the sub model compared to the full model. All data were analyzed, including data from incomplete twin pairs using the raw data option in Mx.

Genetic Modeling

In an extended twin design and given a number of assumptions, MZ, DZ and sibling correlations contain information on the relative influence of genetic and environmental factors on the variability in traits. When MZ twin correlations are higher than DZ twin and twin-sibling correlations, part of the twin resemblance in the phenotype is caused by genetic factors (comprising of additive effects of alleles at one or more loci (A) and non-additive effects of alleles (D)). When DZ twin correlations are more than half the size of MZ correlations, the resemblance between twins is at least partly caused by shared environmental factors (C; common environmental factors shared between siblings brought

up in the same family). Differences between MZ twins reflect the importance of unique environment (E). Large sample sizes are required to have sufficient power to detect D or C (Boomsma, Busjahn, & Peltonen, 2002; Plomin, DeFries, McClearn, & McGuffin, 2001). Based on the limited sample size and on inspection of the MZ and DZ correlations acquired in the saturated models we decided to fit a genetic model in which the relative contributions of A end E were estimated. The phenotype for an individual can be represented as:

$$P_{ij} = a^{*}\text{Aij} + e^{*}E_{ij},$$

where i = 1,2, …or 112 (families) and j = 1, 2 and 3 (twin 1, twin 2 and sibling) and A and E are latent variables (factors) standardized to have unit variance. The variance in P due to A and E is given by the square of a and e, respectively, so that Var (P) = $a^2 + e^2$. Note that e^2 also contains variance due to measurement error. MZ twins have the same DNA sequence and therefore genetic factors are perfectly correlated in MZ twins. DZ twins and siblings share on average half of their segregating genes, so that the expected correlation between their additive genetic factors (A) is ½. By definition the correlation between unique environmental factors (E) is zero. Therefore the covariance within MZ twin pairs is: Cov (MZ) = a^2, and within DZ twin pairs and siblings: Cov (DZ) = ½ a^2.

To determine to what extent the covariation between the reading ability, memory and IQ was due to genetic and environmental effects, multivariate genetic modeling was applied. In a six-variate saturated AE model the factor loadings of the A and E factors are modeled in lower triangular matrices of dimensions 6 × 6 (IQ, four memory measures, and reading ability), where matrix **A** contains the genetic factor loadings, and matrix **E** the environmental factor loadings. The model is then represented as follows:

$$\mathbf{p}_{ij} = \mathbf{A} \times \mathbf{a}_{ij} + \mathbf{E} \times \mathbf{e}_{ij}$$

where i = 1,2, …or 112 (families) and j = 1, 2 and 3 (twin 1, twin 2, and sibling), vector **p** denotes the 6 phenotypes and has the dimension 6 × 1. Vectors $\mathbf{a}_{ij}$ and $\mathbf{e}_{ij}$ have the dimensions 6 × 1 and contain the genetic and environmental factors. The random factors are standardized to have unit variance. The variance in **p** due to **a** and **e** is than given by:

$$\mathbf{V_P} = \mathbf{A} \times \mathbf{A}' + \mathbf{E} \times \mathbf{E}'$$

where matrix $\mathbf{V_P}$ is a symmetric matrix of 6 × 6, **A** and **E** are triangular matrices of 6 × 6, and ’ indicates transposition. To test whether variation in genes contributed significantly to the variability in IQ, memory and reading ability, deterioration of model fit of the saturated six factor model was assessed after the A factor was dropped from the model.

Consecutively non-significant parameters were dropped from the model until the most parsimonious model with still acceptable fit was established.

Results

Data of one MZM family were excluded since the mother did not speak Dutch and the children's Dutch language skills seemed to be delayed. Three families did not complete the Corsi block tapping task, one sibling did not make the 2-back, and in two siblings the Wechsler Intelligence Scale for Children-III (WISC-III; Wechsler et al., 2002) was not assessed. Ten children were not able to complete the n-back task and eight children could not complete the Corsi.

Table 4.1. *Maximum likelihood estimates of means, SD twins, SD sibs and age regression of the variables in the child cohort.*

Variable		*N*	Mean twins	*SD* twins	Mean sibs	*SD* sibs	Age regression
IQ	WISC	323	100.87	13.35	103.34	15.69	-
VS-STM	Corsi	319	12.82	3.98	12.82	4.73	1.25
V-STM	DSF	321	7.31	1.54	7.31	2.09	0.46
VS-WM	2-back	312	30.04	10.46	30.04	15.52	3.42
V-WM	DSB	322	4.63	1.37	5.15	1.87	0.47
Reading	OMRT	291	58.83	18.96	78.74	18.06	6.83

Note. V = verbal; VS = visuospatial; STM = short-term memory; WM = working memory; DSF = digit span forward; DSB = digit span backward; OMRT = one minute reading test

Means, standard deviations, and age and sex effects are reported in Table 4.1. Means were not equal for MZ and DZ twins and siblings ($\Delta\chi^2 = 29.882$, $\Delta df = 12$, $p < .01$). Means were higher for siblings on IQ, reading ability and DSB. Twins could read on average 58 words in one minute, ranging from seven to 100 words. According to Cito (1995) children of this age are suspected to be dyslexic when they score below 28 words a minute. Six percent of the twins indeed had a score lower than 28. The siblings scored between 36 and 120 words a minute, with an average of 79 words. Number of children with possible dyslexia in the group siblings was 7 (9%). There were no significant effects of sex

on the means of the six measures. A significant effect of age on the means of all variables, except IQ (this test is already standardized for age), was observed. All subsequent models were corrected for these effects.

Table 4.2. *Phenotypic and MZ and DZ/twin sibling correlations*

Variable			WISC	Corsi	DSF	2-Back	DSB	OMRT
Phenotypic correlations	IQ	WISC						
	VS-STM	Corsi	.28					
	V-STM	DSF	.25	.21				
	VS-WM	2-back	.35	.32	.17			
	V-WM	DSB	.34	.30	.26	.21		
	Reading	OMRT	.42	.24	.44	.24	.37	
MZ and DZ/twin sibling correlations	IQ	WISC	.74 / .54	.17	.20	.22	.25	.31
	VS-STM	Corsi	.29	.56 / .17	.09	.10	.14	.10
	V-STM	DSF	.22	.13	.54 / .20	.09	.27	.18
	VS-WM	2-back	.33	.27	.09	.55 / .15	.12	.12
	V-WM	DSB	.28	.27	.26	.22	.41 / .12	.30
	Reading	OMRT	.44	.23	.46	.23	.35	.84 / .40

Note. Upper part of the table maximum likelihood estimates of phenotypic correlations between the variables corrected for age and sex. Lower part of the table MZ and DZ/twin-sibling correlations, on the diagonal on the left side the MZ correlations and on the right the DZ/twin-sibling correlations, below the diagonal MZ cross correlations and above the diagonal DZ/twin-sibling cross correlations. V = verbal; VS = visuospatial; STM = short-term memory; WM = working memory; DSF = digit span forward; DSB = digit span backward; OMRT = one minute reading test

Variances in twins and siblings could not be equated ($\Delta\chi^2 = 58.67$, $\Delta df = 21$, $p < .01$). In the siblings there was more variation in all tasks except for the Corsi. We corrected for this variance inequality by multiplying the variances of these tasks in the siblings by a factor which equated these three variances between twins and siblings. DZ covariation could be equated to twin-sibling covariation ($\Delta\chi^2 = 19.54$, $\Delta df = 21$, $p = .55$).

Phenotypic correlations among variables were moderate (see Table 4.2). In the lower parts of Table 2 the MZ and DZ/twin-sibling correlations are displayed on the

diagonal. MZ correlations were higher than DZ/twin-sibling correlations in both cohorts, suggesting genetic influences on the six variables. Below the diagonal MZ cross correlations and above the diagonal DZ/twin-sibling cross correlations are presented. MZ cross correlations were higher than DZ/twin-sibling cross correlations, suggesting that genes play a role in the covariation amongst the six measures.

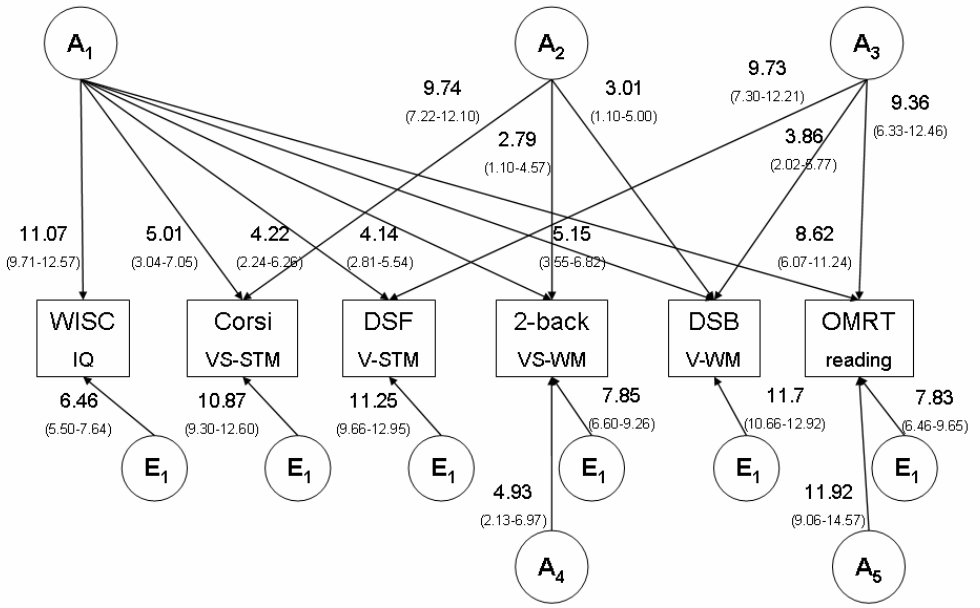


Figure 4.1. Best fitting AE model with parameter estimates (95% confidence intervals). 95% confidence intervals are given in *brackets*. Only twin one is shown in the figure. V = verbal; VS = visuospatial; STM = short-term memory; WM = working memory; DSF = digit span forward; DSB = digit span backward; OMRT = one minute reading test

Indeed, additive genetic effects could not be dropped from the AE model without a significant deterioration of fit ($\Delta\chi^2 = 219.67$, $\Delta df = 21$, $p < .01$). Therefore, it can be concluded that genes play a significant role in variation in, and the covariation amongst the six measures. Further model fitting showed that all environmental covariation could be dropped from the model, without a significant deterioration of fit ($\Delta\chi^2 = 7.579$, $\Delta df = 15$, $p = .94$). Thereafter, non-significant parameters were dropped from the model. The most parsimonious model is represented in Figure 4.1 with specific environmental factors only and five genetic factors: 1) a genetic factor common to all variables; 2) a genetic factor common to visuospatial STM and verbal and visuospatial WM 3) a genetic factor common

to verbal memory and reading ability; 4) a specific genetic factor for visuospatial WM; 5) a specific genetic factor for reading ability (compared to the model without environmental covariation: $\Delta\chi^2 = 5.988$, $\Delta df = 7$, $p = .54$). Heritability estimates are presented in Table 4.3. Heritability ranged between 27% and 83%. The highest heritabilities were seen for IQ (75%) and reading ability (83%). Genetic variation in the reading measure is largely independent of IQ and memory related traits: of the total genetic variance, roughly half (47%) is not shared with the other phenotypes in the model ($(8.62^2 + 9.36^2) / (8.62^2 + 9.36^2 + 11.92^2)$).

Table 4.3. *Heritability estimates*

Variable		h^2
IQ	WISC	75%
VS-STM	Corsi	50%
V-STM	DSF	47%
VS-WM	2-back	44%
V-WM	DSB	27%
Reading	OMRT	83%

Note. V = verbal; VS = visuospatial; STM = short-term memory; WM = working memory; DSF = digit span forward; DSB = digit span backward; OMRT = one minute reading test

Visuospatial WM and STM do not contribute independently from intelligence to the genetic variability in reading performance. The verbal memory tasks contribute independently to the variability in reading performance. Genetic covariance shared between reading ability and verbal WM is for 45% independent of IQ: (9.36*3.86) / (9.36*3.86 + 8.62*5.15). For the shared genetic correlation between reading ability and verbal STM this is 71%: (9.36*9.73) / (9.36*9.73 + 8.62*4.22). Verbal STM and WM do no contribute independently from each other to the genetic covariance in reading ability. Fifty-three percent of the genetic variation in reading ability is explained by variation shared with intelligence and memory performance.

Discussion

In this study we investigated in a genetically informative design whether the phenotypic correlation between IQ, WM, STM and reading ability stems in part from overlapping sets of genes and whether IQ, WM and STM independently contribute to the genetic and environmental variance in reading ability. Results showed that the phenotypic correlation between IQ, WM, STM and reading ability is explained by common sets of genes (genetic pleiotropy). This shows that genetic influences are responsible for linking IQ, WM, STM and reading ability, whereas environmental effects create differences between these constructs. This finding is in concordance with the view of Price et al. (2000) and Petrill (1997): Genetic studies tend to show substantial genetic overlap which explains the association found between different cognitive abilities. In contrast, environmental factors primarily drive the different dimensions of cognitive functioning which also consistently emerge across many studies.

The genetic analyses also revealed a common genetic factor for IQ, WM, STM and reading ability. This common genetic factor probably represents general intelligence, or *g*. Thus, children low in *g* are less skilled in reading, STM and WM. This suggests that there is no etiological separation between low IQ, deficits in WM, STM and reading.

Variation in reading ability is further explained by genes specific to reading ability and a set of genes common to verbal memory and reading ability. The specific factor for reading ability explains half of the genetic variation in reading ability. The verbal memory factor is as important for explaining variation in reading ability as the *g* factor. Verbal STM and WM contributed respectively 71% and 45% of their variance independently of IQ to variation in reading ability. This factor could represent the problems children with reading disability have in the ability to code information phonemically or verbally, which is an important aspect of verbal STM (Kercher & Sandoval, 1991), but also of verbal WM. So, there is an etiological separation between low intelligence and lower reading ability and deficits in verbal memory and reading.

Three genetic factors influence variability in reading ability, a genetic factor which represents *g*, a genetic factor representing verbal coding and a genetic factor specific to reading ability. Children who have a genetic predisposition for low *g*, still can have a genetic predisposition for average verbal coding and vice versa, but also a combination of a

genetic predisposition for low *g* and verbal coding deficits is possible. This suggests that at different groups of children with reading disability can be differentiated: children who are low in general intelligence and therefore are less skilled in reading; children who have normal IQ and a deficit in coding information phonemically or verbally and therefore experience problems with reading; and children with low IQ and deficits in phonemic and verbal coding, this group experiences more reading problems than the other two. This hypothesis is supported by the findings of Alarcón and DeFries (1997) which revealed that phenotypic and genetic correlations between intelligence and reading ability were larger for a control group than for the group affected with reading disabilities. This study also showed that genetic and phenotypic variances and covariances amongst the reading measures were larger for the affected group, suggesting that reading disability is caused by one or more genes with major effects.

Our finding argues against the use of discrepancy scores to identify reading disability, because using this strategy would miss the children with most severe reading disability. Further research in children with reading disabilities should aim at distinguishing these groups of children, so adequate reading methods for each of these three groups can be developed.

To conclude, this study shows that reading ability is related to intelligence, WM and STM. This relation is completely mediated by two common genetic factors: a factor which has reading ability in common with IQ, WM and STM and a common factor for reading ability and verbal memory.

Reference List

Alarcón, M & DeFries, J C (1997). Reading performance and general cognitive ability in twins with reading difficulties and control pairs. *Personality and Individual Differences, 22*, 793-803.

Baddeley, A (1992). Working memory. *Science, 255*, 556-559.

Baddeley, A (2000). The episodic buffer: a new component of working memory? *Trends in Cognitive Sciences, 4*, 417-423.

Baddeley, A (2003). Working memory: looking back and looking forward. *Nature Reviews Neuroscience, 4*, 829-839.

Baddeley, A (1996). Exploring the Central Executive. *The Quarterly Journal of Experimental Psychology Section A, 49*, 5-28.

Bishop, D V M (2006). Developmental cognitive genetics: How psychology can inform genetics and vice versa. *The Quarterly Journal of Experimental Psychology, 59*, 1153-1168.

Boomsma, D, Busjahn, A, & Peltonen, L (2002). Classical twin studies and beyond. *Nature Reviews Genetics, 3*, 872-882.

Boomsma, D I, Vink, J M, Van Beijsterveldt, T C, De Geus, E J, Beem, A L, Mulder, E J, Derks, E M, Riese, H, Willemsen, G A, Bartels, M, Van den Berg, M, Kupper, N H, Polderman, T J, Posthuma, D, Rietveld, M J, Stubbe, J H, Knol, L I, Stroet, T, & Van Baal, G C (2002). Netherlands Twin Register: A focus on longitudinal research. *Twin Research, 5*, 401-406.

Brooks, A, Fulker, D W, & DeFries, J C (1990). Reading performance and general cognitive ability: A multivariate genetic analysis of twin data. *Personality and Individual Differences, 11*, 141-146.

Cardon, L R, Dilalla, L F, Plomin, R, DeFries, J C, & Fulker, D W (1990). Genetic correlations between reading performance and IQ in the Colorado Adoption Project. *Intelligence, 14*, 245-257.

Cito. (1995) *Drie Minuten Test.* Arnhem: Cito

Cohen-Mimran, R & Sapir, S (2007). Deficits in working memory in young adults with reading disabilities. *Journal of Communication Disorders, 40*, 168-183.

Colom, R, Abad, F J, Rebollo, I, & Chun Shih, P (2005). Memory span and general intelligence: A latent-variable approach. *Intelligence, 33*, 623-642.

Démonet, J F, Taylor, M J, & Chaix, Y (2004). Developmental dyslexia. *Lancet, 363*, 1451-1460.

Gathercole, S E, Pickering, S J, Ambridge, B, & Wearing, H (2004). The structure of working memory from 4 to 15 years of age. *Developmental Psychology, 40*, 177-190.

Gathercole, S E, Alloway, T P, Willis, C, & Adams, A M (2006). Working memory in children with reading disabilities. *Journal of Experimental Child Psychology, 93*, 265-281.

Gayán, J & Olson, R K (2003). Genetic and environmental influences on individual differences in printed word recognition. *Journal of Experimental Child Psychology, 84*, 97-123.

Gevins, A & Cutillo, B (1993). Spatiotemporal dynamics of component processes in human working memory. *Electroencephalography and Clinical Neurophysiology., 87*, 128-143.

Hawke, J L, Wadsworth, S J, & DeFries, J C (2006). Genetic influences on reading difficulties in boys and girls: the Colorado twin study. *Dyslexia, 12*, 21-29.

Jansma, J M, Ramsey, N F, Coppola, R, & Kahn, R S (2000). Specific versus nonspecific brain activity in a parametric n-back task. *Neuroimage, 12*, 688-697.

Kane, M J, Hambrick, D Z, Tuholski, S W, Wilhelm, O, Payne, T W, & Engle, R W (2004). The generality of working memory capacity: a latent-variable approach to verbal and visuospatial memory span and reasoning. *Journal of Experimental Psychology: General, 133*, 189-217.

Kercher, A C & Sandoval, J (1991). Reading disability and the differential ability scales. *Journal of School Psychology, 29*, 293-307.

Livingston, R B, Jennings, E, Reynolds, C R, & Gray, R M (2003). Multivariate analyses of the profile stability of intelligence tests: high for IQs, low to very low for subtest analyses. *Archives of Clinical Neuropsychology, 18*, 487-507.

Moelands, F, Kamphuis, F, & Verhoeven, L (2008). Verantwoording Drie-Minuten-Toets (DMT) [Jusitfication Three-Minutes-Task (DMT)].

Neale, M., Boker, S.M., Xie, G., & Maes, H.H. (2006) *Mx: statistical modelling.* Richmond, VA: Department of Psychiatry, Medical College of Virginia

Pennington, B F, Gilger, J W, Olson, R K, & DeFries, J C (1992). The external validity of age- versus IQ-discrepancy definitions of reading disability: Lessons from a twin study. *Journal of Learning Disabilities.Vol 25(9),* 562-573.

Peper, J S, Brouwer, R M, Schnack, H G, Van Baal, G C M, Van Leeuwen, M, Van den Berg, S M, Delemarre-van de Waal, H A, Janke, A L, Collins, D L, Evans, A C, Boomsma, D I, Kahn, R S, & Hulshoff Pol, H E (in press). Cerebral white matter in early puberty is associated with luteinizing hormone concentrations. *Psychoneuroendocrinology.*

Petrill, S A (1997). Molarity Versus Modularity of Cognitive Functioning? A Behavioral Genetic Perspective. *Current Directions in Psychological Science, 6*, 96-99.

Bayne J. and Byrd M.L. Plomin, R., DeFries, J.C., McClearn, G.E., & McGuffin, P. (2001) *Behavioral genetics.* New York: Worth Publisher

Polderman, T J C, Stins, J F, Posthuma, D, Gosso, M F, Verhulst, F C, & Boomsma, D I (2006). The phenotypic and genotypic relation between working memory speed and capacity. *Intelligence, 34*, 549-560.

Price, T S, Eley, T C, Dale, P S, Stevenson, J, Saudino, K, & Plomin, R (2000). Genetic and environmental covariation between verbal and nonverbal cognitive development in infancy. *Child Development, 71*, 948-959.

Snowling, M J (1980). The development of grapheme-phoneme correspondence in normal and dyslexic readers. *Journal of Experimental Child Psychology, 29*, 294-305.

Stanovich, K E (1993). A Model for Studies of Reading Disability. *Developmental Review, 13*, 225-245.

Sternberg, R J & Grigorenko, E L (2002). Difference Scores in the Identification of Children with Learning Disabilities It's Time to Use a Different Method. *Journal of School Psychology, 40*, 65-83.

Swanson, H L (2003). Age-related differences in learning disabled and skilled readers' working memory. *Journal of Experimental Child Psychology, 85*, 1-31.

Swanson, H L & Ashbaker, M H (2000). Working memory, short-term memory, speech rate, word recognition and reading comprehension in learning disabled readers: does the executive system have a role? *Intelligence, 28*, 1-30.

Swanson, H L & Berninger, V (1995). The role of working memory in skilled and less skilled readers' comprehension. *Intelligence, 21*, 83-108.

Swanson, H L & Jerman, O (2007). The influence of working memory on reading growth in subgroups of children with reading disabilities. *Journal of Experimental Child Psychology, 96*, 249-283.

Thompson, L A, Detterman, D K, & Plomin, R (1991). Associations between cognitive abilities and scholastic achievement: Genetic overlap but environmental differences. *Psychological Science, 2*, 158-165.

Tiu, R D, Jr., Thompson, L A, & Lewis, B A (2003). The Role of IQ in a Component Model of Reading. *Journal of Learning Disabilities, 36*, 424-436.

Tiu, R D, Jr., Wadsworth, S J, Olson, R K, & DeFries, J C (2004). Causal Models of Reading Disability: A Twin Study. *Twin Research, 7*, 275-283.

Van der Sluis, S, Van der Leij, A, & De Jong, P F (2005). Working memory in Dutch children with reading- and arithmetic-related LD. *Journal of Learning Disabilities, 38*, 207-221.

Van Leeuwen, M, Van den Berg, S M, Hoekstra, R A, & Boomsma, D I (2007). Endophenotypes for intelligence in children and adolescents. *Intelligence, 35*, 369-380.

Van Leeuwen, M, Van den Berg, S M, & Boomsma, D (2008). A twin-family study of general IQ. *Learning and Individual Differences, 18*, 76-88.

Wadsworth, S J, DeFries, J C, Fulker, D W, & Olson, R K (1995). Reading performance and verbal short-term memory: A twin study of reciprocal causation. *Intelligence, 20*, 145-167.

Wechsler, D., Kort, W., Compaan, E.L., Bleichrodt, N., Resing, W.C.M., Schittekatte, M., Bosmans, M., Vermeir, G., & Verhaeghe, P. (2002) *Handleiding WISC-III^{NI} [Manual WISC-III^{NI}].* London: The Psychological Corporation Limited, Nederlands Instituut van Psychologen Dienstencentrum

A twin-family study of general IQ

In this paper we asses the presence of assortative mating, gene-environment interaction and the heritability of intelligence in childhood using a twin family design with twins, their siblings and parents from 112 families. We evaluate two competing hypotheses about the cause of assortative mating in intelligence: social homogamy and phenotypic assortment, and their implications for the heritability estimate of intelligence. The Raven Progressive Matrices test was used to assess general intelligence (IQ) and a person's IQ was estimated using a Rasch model. There was a substantial correlation between spouses for IQ (r = .33) and resemblance in identical twins was higher than in first-degree relatives (parents and offspring, fraternal twins and siblings). A model assuming phenotypic assortment fitted the data better than a model assuming social homogamy. The main influence on IQ variation was genetic. Controlled for scale unreliability, additive genetic effects accounted for 67% of the population variance. There was no evidence for cultural transmission between generations. The results suggested that an additional 9% of observed IQ test variation was due to gene-environment interaction, with environment being more important in children with a genetic predisposition for low intelligence.

This chapter is published as:

Van Leeuwen, M., Van den Berg, S. M., & Boomsma, D. (2008). A twin-family study of general IQ. *Learning and Individual Differences, 18,* 76-88.

Individual differences in intelligence tend to cluster in families (for reviews see e.g. Bouchard, & McGue, 2003; Bouchard & McGue, 1981; Boomsma, 1993; Deary, Spinath, & Bates, 2006). The resemblance between relatives can be due to genetic relatedness, environmental similarities, cultural transmission from one generation to the next, social interactions between family members, or a combination of these mechanisms. When one wants to study causes of this resemblance and only first-degree relatives, such as parents and their offspring, or siblings are included in the study design, it is not possible to disentangle shared genetic from shared environmental effects. However, in a twin design such a distinction can be made, because monozygotic (MZ; identical) twins share all, or nearly all of their DNA, while dizygotic twins (DZ; fraternal) share on average 50% of their segregating genes (Boomsma, Busjahn, & Peltonen, 2002; Plomin, DeFries, Craig, & McGuffin, 2002). A larger resemblance of MZ than of DZ twins therefore is suggestive of genetic influences on twin resemblance.

Numerous studies of young and adult twins have explored the etiology of resemblance in intelligence between family members. Twin studies in children estimate the contribution of genetic effects to the variability in intelligence at 25% to 50%. Part of the remaining variance is due to environmental factors shared by children who grow up in the same family (Bartels, Rietveld, Van Baal, & Boomsma, 2002; Plomin et al., 2002; Turkheimer, Haley, Waldron, D'Onofrio, & Gottesman, 2003). Heritability appears to increase with age and the influence of shared environment disappears in early adolescence (see e.g. Bouchard and McGue, 2003; Cherny & Cardon, 1994; Plomin et al, 2002; Plomin and Spinath, 2004; Plomin, Pedersen, Lichtenstein, & McClearn, 1994; Posthuma, De Geus, & Boomsma, 2001; Scarr & Weinberg, 1983).

The classical twin design in which data from MZ pairs are compared to data from DZ pairs relies on several assumptions. It is often assumed that the phenotypes of parents are uncorrelated (i.e., one assumes random mating), that there is no genotype-environment (GE) *interaction*, and no GE *correlation*. GE *interaction* refers to the phenomenon that the influence of a particular genotype may depend on the environment (or vice versa: that the influence of the environment depends on genotype). GE *correlation* refers to the non-random distribution of genotypes over environments and may for instance occur when parents transmit not only their genes but also their environment to their children.

When these assumptions are not met, results from twin studies may be biased. For example, if there is genotype by *common* environment interaction then heritability will be overestimated (Lynch & Walsh, 1998). If there is genotype by *unique* environment interaction then, heritability will be underestimated. If random mating between parents is assumed, while there is non-random mating in the population, this will bias heritability estimates downwards and increases estimates of shared environmental influences. If the classical twin design is extended by including the twins' parents (Fulker, 1982) assortative mating and some forms of GE correlation can be assessed. Additionally, by looking at the association between MZ sum and difference scores, it is possible to detect and estimate GE interaction. In this paper we use such an extended twin design which includes MZ and DZ twins, their siblings and their parents, to study to what extent assortative mating, cultural inheritance and GE interaction and correlation are present for general intelligence (IQ). Data on general IQ were collected in both generations with the Raven Progressive Matrices test.

Spousal resemblance

Spouse and family studies show that spouses resemble each other in IQ scores and traits correlated to IQ, such as educational attainment. Spousal correlations for performance on the Raven Progressive Matrices are around 0.30 (Guttman, 1974; Watkins & Meredith, 1981). For the Wechsler Adult Intelligence Scale (WAIS), spousal correlations are between 0.37 and 0.61 (Mascie-Taylor, 1989; Watson et al., 2004; Williams, 1975). In the Colorado Adoption Project (CAP), the correlation between spouses on the first unrotated principal component derived from a battery of 13 cognitive tests was 0.11, and in the Hawaii Family Study of Cognition (HFSC) this correlation was 0.20 (Phillips, Fulker, Carey, & Nagoshi, 1988). These and other studies clearly show a resemblance in intelligence between spouses. This resemblance, or non-random mating, may be due to marital interaction, phenotypic assortment, or social homogamy.

The hypothesis of marital interaction or convergence states that spousal correlations arise because spouses spend time together. Spouses would tend to become more similar the longer they are together, because they either influence each other or because they share similar experiences. The few studies that tested this hypothesis found no

indications of convergence for intelligence (Gilger, 1991; Mascie-Taylor, 1989; Watson et al., 2004).

Under phenotypic assortment it is assumed that spouses choose each other based on observable characteristics (Reynolds, Baker, & Pedersen, 1996), in this case based on intelligence or a trait related to it: individuals would tend to mate with partners with an intelligence level resembling their own. Most models of assortative mating assume phenotypic assortment (Fulker, 1982; Fulker & DeFries, 1983; Rice, Carey, Fulker, & DeFries, 1989; Wadsworth, DeFries, Fulker, & Plomin, 1995). Mascie-Taylor and Vandenberg (1988) tried to estimate the role of personal preference in mate selection by correcting for variables representing proximity such as social class, locality, family size, birth order and type and years of education. After correcting for these variables, there still was a significant correlation between spouses' IQs, suggesting that that this correlation could be ascribed to direct phenotypic assortment.

Social homogamy refers to assortment based on solely environmental similarities. Spousal phenotypes become correlated because spouses meet each other within a particular environment (Reynolds et al., 1996). In the case of intelligence, the social homogamy hypothesis states that people with the same intelligence level live in the same social environment. Within a particular social environment, partners do not choose each other on the basis of intelligence, but since they live in the same environment, they tend to mate with people with a similar IQ. Spousal correlations in the general population occur when social environment is correlated with intelligence.

When there is mate resemblance for intelligence, it may be necessary to include its effects in the genetic model. When resemblance is caused by phenotypic assortment, this induces genetic similarity between parents, which affects the genetic similarity between parents and offspring and among siblings and dizygotic twins. Under random mating, genetic effects are uncorrelated in parents. The correlation between a parental and a child's genotype, and among siblings and in DZ twins is then ½. If there is positive phenotypic assortment, these genetic correlations increase and heritability will be underestimated if this effect is not taken into account (Cavalli-Sforza & Bodmer, 1971). When spousal resemblance is purely due to environmental effects that are not correlated with genetic effects, there are no genetic consequences.

GE interaction

One approach to detect and estimate GE interaction is by looking at the association between MZ intrapair sum (or average) and difference scores (Jinks & Fulker, 1970). Genetic and shared environmental effects add to the similarity of MZ pairs and unique environment to the differences between MZ pairs. When there is a positive correlation between intrapair sum and absolute differences, less intelligent individuals are more similar than more intelligent individuals, and thus more intelligent people are more susceptible to unique environmental influences (Finkel & Pedersen, 2001).

Jinks and Fulker reported intrapair sum/ intrapair difference correlations for IQ of -.10 and -.13, based on data from 19 MZ twin pairs. Jensen (1970) reported a correlation for IQ of -.15 in MZ twins reared apart. And Finkel and Pedersen (2001) reported a correlation of -.11 in MZ twins reared apart and of -.09 in MZ twins reared together. Although these correlations were all non-significant, all correlations were of similar magnitude and negative, suggesting that the environment might have a greater influence in less intelligent people.

Parent-offspring resemblance

Including parents in a twin design adds extra information about the origins of individual differences. The resemblance between parents and offspring may reflect genetic transmission, cultural transmission, or both. In the case of genetic transmission, resemblance between parents and offspring is caused by the genes which are transmitted from the parents to their children. In an ordinary family design genetic transmission is confounded with cultural influences of parents on their offspring. Cultural transmission will increase parent-offspring correlations, as well as correlations between siblings and twins who grow up in the same home environment. In the classical twin design, cultural transmission will show up as shared (or common) environmental variance.

Parents may create a particular kind of environment that is correlated with their genotype or their phenotype, for example, bright parents might stimulate their children with schoolwork or provide them with more intelligence-boosting toys. Whenever there is cultural transmission in the presence of genetic transmission, environmental influences become correlated with genetic influences.

In an adoption design, genetic and cultural transmission can be disentangled because then the adopted child's environment is uncorrelated with the intelligence levels of its biological parents. In the CAP study mentioned earlier, IQ data from adopted and non-adopted children were collected at ages 1, 2, 3, 4, 7, 9, 10 and 12 years. When analyzing the IQ data from the adoptive and non-adoptive children up until the age of 12, no significant shared environmental influence was found: all variance could be explained by additive genetic factors and environmental factors that are not shared by children raised in the same family (Bishop, Cherney & Hewitt, 2003).

The CAP study also collected data on the biological and, if they were adopted, the adoptive parents of these children. Significant genetic transmission for intelligence was found at all ages (Fulker et al., 1983; Humphreys & Davey, 1988; Rice et al., 1989). The CAP data also showed significant cultural transmission from foster parent to offspring but only before the age of 4 years (Fulker et al., 1983; Humphreys et al., 1988; Rice et al., 1989). Alarcón, Plomin, Corley, and DeFries (2003) also showed that there was no cultural transmission for specific cognitive abilities at ages 7 and 12: assuming phenotypic assortment, all variance was due to additive genetic effects and random environmental effects. Similar findings were reported by another adoption study (Scarr & Weinberg, 1983) showing that in adolescence, the impact of the family environment on IQ disappears.

The present study

Up until now only the CAP-study (Alarcón et al., 2003; Fulker et al., 1983; Humphreys et al., 1988; Rice et al., 1989) examined the genetic and environmental transmission of intelligence from parents to their children in the presence of spousal resemblance. Other studies using twins sometimes take assortative mating into account when interpreting their results (e.g. Wainwright, Wright, Geffen, Luciano, & Martin, 2005), but do not assess or model assortative mating directly. In the CAP study different measures of IQ were used across generations. Parental intelligence was estimated based on an unstandardised measure of IQ (see above) whereas in the children intelligence was measured, depending on age, using the Bayley Mental Development Index, the Stanford-Binet Intelligence Scale or the Wechsler Intelligence Scale for Children. The measure used in adults resulted in a relatively low spousal correlation when compared to studies using full scale IQ tests (Mascie-Taylor, 1989; Watson et al., 2004; Williams, 1975). There are studies using

comparable IQ tests in parents and children but these studies do not report heritability estimates, since the samples studied were not suitable for this purpose (Guttman,1974; Guttman & Shoham, 1983; Williams, 1975).

In the present study, we collected data on intelligence using Raven's Progressive Matrices (Raven, 1960; Raven, Raven, & Court, 1998), in MZ and DZ twins, one of their siblings and both of their parents. With this design, cultural and genetic transmission can be studied while taking into account spousal resemblance. The inclusion of additional siblings increases the power to detect additive and non-additive genetic effects (Keller, Coventry, Heath, & Martin, 2005; Posthuma and Boomsma, 2000). Raven IQ measures were estimated based on a Rasch model (Rasch, 1966). This way the intelligence measure is not dependent on the particular items that are included in the test and has no a priori distribution.

We expect that additive genetic effects will explain a large part of the individual differences in IQ. We also explore the presence of non-additive genetic influences, or genetic dominance, on IQ variation. Genetic non-additivity has been suggested in studies on inbreeding (Agrawal, Sinha & Jensen, 1984; Bashi, 1977), reflecting recessive effects of rare alleles that might not contribute much to the variation in the general population. Genetic dominance has at times been suggested in twin and other studies (Chipuer, Rovine, & Plomin, 1990; Fulker, 1979; Jinks & Fulker, 1970). Dominance effects can be masked by assortative mating and cultural transmission in studies with only MZ and DZ twins.

We fitted two models, one assuming phenotypic assortment and one assuming social homogamy to determine which of both model fits the data best. To assess GE interaction, we tested whether there is an association between absolute difference scores in MZ twins (reflecting non-shared environmental effects) and average scores (reflecting familial effects).

Material and methods

Participants

The study was approved by the Central Committee on Research involving Human Subjects (CCMO). Twins were recruited form the Netherlands Twin Registry (NTR), established by the Department of Biological Psychology at the Vrije Universiteit (VU) in Amsterdam

(Boomsma, Orlebeke, & Van Baal, 1992; Boomsma et al., 2002; Boomsma et al., 2006). Twin families with an extra sibling between 9 and 14 years were selected from two birth cohorts (1995-1996). Because the twins and siblings also took part in an MRI study, there were several exclusion criteria such as a pacemaker and metal materials in the head. Families with children with a major medical history, psychiatric problems (as reported by the parents), participation in special education, or physical or sensory disabilities were also excluded. A total of 214 families were invited by letter, which was sent out one to two months before the ninth birthday of the twins. Two weeks after receiving the letter, the families were contacted by phone. Of these families 52% (112) agreed to participate. There was no significant difference between the educational level of mothers who did participate and who did not participate in the study (F (1,195) = .68, p = .41). Of the 112 families, 103 had full siblings who wanted to participate. Parents signed informed consent forms for the children and themselves. Children also signed their own consent forms. Parents were compensated for their travel expenses and children received a present.

The 112 families came from all over the Netherlands. Mean age of the twins at time of cognitive assessment was 9.1 years, ranging from 8.9 to 9.5 years. There were 23 MZ male, 23 DZ male, 25 MZ female, 21 DZ female and 20 DZ pairs of opposite sex. Zygosity was based on DNA polymorphisms and questionnaire items. Mean age of the sibs (N = 103; 59 female) was 11.9 years ranging from 9.9 to 14.9. The mean age of the fathers was 43.7 (N = 94, SD = 3.7 years), and of the mothers 41.9 (N = 95, SD = 3.4 years). Only data from biological parents were included in analyses.

Testing procedures

This study collected cognitive, behavioral and hormonal data, pubertal status and structural Magnetic Resonance Imaging (MRI) brain data. Data collection took place on two different days. Cheek swabs, for DNA isolation, were collected at home by parents and children. For cognitive testing, families arrived between nine and eleven o'clock in the morning. Children were tested in separate rooms with a cognitive test battery including the Raven's Standard Progressive Matrices (SPM; Raven, 1960). Parents completed the Raven Advanced Progressive Matrices (APM; Raven et al., 1998). The whole protocol took approximately five hours, including two short breaks and one longer lunch break.

Materials

Children were individually tested with the Standard Progressive Matrices (Raven, 1960), which they completed at their own pace after verbal instruction. The test consists of 60 problems divided into five sets of twelve. In each set the first problem is as nearly as possible self-evident. The problems within a set become progressively more difficult. The test is intended to cover the whole range of intellectual development from the time a child is able to grasp the idea of finding a missing piece to complete a pattern, and to be sufficiently long to assess a child's maximum capacity to form comparisons and reason by analogy. The test provides an index of general intelligence. For children retest reliability is .88 (Raven, 1960).

Parents were given the Advanced Progressive Matrices (Raven et al., 1998), since the SPM is too easy for most adults. They received written instructions and made the test at their own pace. The APM is comparable to the SPM with the main difference being the level of difficulty. The APM consists of two sets. The first set contains twelve practice items, to familiarize Ss with the test. The second set consists of 36 items, which are identical in presentation and argument with those in Set I. They only increase in difficulty more steadily and become considerably more complex. Reported retest reliability for adults is .91 (Raven et al., 1998).

Zygosity determination

In 110 twin pairs, zygosity was determined at the VU Medical Centre with eight highly polymorphic di-, tri- and tetranucleotide genetic markers. The zygosity testing included a multiplex PCR of markers D2S125, D8S1130, D1S1609, D5S816 and a second multiplex reaction of markers 15 ActC, D21S1437, D7S2846, and D10S1423. These two multiplex PCR reactions were performed by the protocol provided in the website of the Marshfield Institute (http:/ /www.marshmed.org/genetics). Results of the zygosity test were sent to the parents. In the remaining two twin pairs zygosity was based on questionnaire items (Rietveld et al., 2000).

Statistical analysis

Rasch score

IQ measures in parents and offspring were estimated based on the Rasch model (Rasch, 1966). In this model, every person is represented by a person parameter θ that reflects that person's ability. Every test item is represented by a difficulty parameter β. The probability that a person *j* answers item *i* correctly is parameterized by the logistic function $p(Y_{ij} = 1) = \Psi(\theta_j - \beta_i)$, where θ_j is the person parameter, β_i is the difficulty parameter for that particular item, and $\Psi(x) = \exp(x) / (1 + \exp(x))$ see also (Van den Berg, Glas, & Boomsma, 2007). Thus, for example, the probability that person *j* with ability θ_j answers item *i* with difficulty level β_i correctly, equals $e^{\theta j-\beta i} / [1 + e^{\theta j-\beta i}]$. When $\theta_j - \beta_i = 0$, the probability of a correct answer is exactly 50%, as $e^0 = 1$. When ability dominates the difficulty, $\theta_j > \beta_i$, then the probability is higher than 50%, becoming 100% when ability is infinitely higher than the difficulty. When ability is lower than the difficulty of the item, $\theta_j < \beta_i$, then the probability of a correct answer is lower than 50%, becoming 0% when the ability is infinitely lower than the difficulty. Note that the values for θ and β, the ability of a person and the difficulty of an item, are on the same scale.

The rationale for the Rasch model can be presented by analogy to the success of an athletic hurdle jumper: some people jump higher than others do. For each jumper there might exist a hurdle with a certain height where only 50% of the attempts is successful. If the hurdle's height increases, the probability of a successful jump decreases whereas it increases when the hurdle's height decreases. If a hurdle is very low, the probability of a successful jump approaches one; when the hurdle is very high, the probability of a successful jump approaches zero. In the Rasch model, a person's ability is defined as the difficulty level where the probability of a correct answer (or jump) is 50%. The model assumes local independence. This means that the probability of a success is entirely explained by the θ and β parameter: given θ and β, the probability of a correct answer is not dependent on whether other items are answered correctly or whether other people answered the same item correctly. This assumption is for example also used in the common factor model, where only one factor explains all correlations among the indicator variables (Spearman, 1927). Thus, an assumption in the Rasch model is unidimensionality of ability.

The Rasch model has a number of nice properties. The most important is the property of invariant comparison or separability of person and item parameters: the comparison between two persons is independent of the particular measurement instrument and other persons being measured at the same time. The estimated difference in ability measures between two persons is the same regardless whether we use all items from a test or any possible subset of the items (if all items measure the same ability). This is for example not true when we merely use the number of correct answers. Similarly, the estimated difference in difficulty level between two items is the same regardless which people are used to measure the difficulty of the items. It does not matter whether we take 20 persons with an ability of 80 and 30 persons with an ability of 100, or we take 50 people with ability scores uniformly distributed between 70 and 100. This is related to a second property of Rasch scales: the estimation of ability and difficulty needs no assumption about their distribution. There is for example no need for a constraint on the distribution of the ability parameters, such as a normal distribution. The distribution is an empirical question. If the Rasch model fits the data, then the estimates of the θ parameters can be regarded as interval level measures of ability on the logit scale and one can check whether on that scale, the distribution of the ability parameters is normal. This is not true for sum scores: the distribution is a direct consequence of the difficulty levels of the items in the test.

An important point is that the ability measures based on the Rasch model are estimates, just as a sum score is an estimate of the true score in classical test theory. A Rasch estimate for ability is more reliable when the test contains many items with difficulty levels comparable to the true ability score. Therefore, and in contrast to classical test theory, the reliability of an ability measure may vary across the scale. For more on Rasch modeling, see Smith and Smith (2004) and Bond and Fox (2001).

Studies have shown that the Raven is largely unidimensional (Rost & Gebert, 1980), but there are also indications that the Raven test might be multidimensional (Lynn, Allik, and Irwing, 2004; Van der Ven & Ellis, 2000; Vigneau & Bors, 2005). Multidimensionality is often noticed in tests with items varying widely in difficulty. Linear factor models then usually show several factors, one for each difficulty level, a phenomenon generally attributed to non-linearity (Gibson, 1959). In the case of the Raven, the dimensions are highly correlated. Lynn et al. (2004) showed for the Standard Raven that all three factors they found loaded highly onto one second-order factor. The correlations

between the three factors and the second-order factor were .95, .80 and .90. Thus, the use of a unidimensional Rasch model leads to only very limited bias. There are also indications that the Standard Raven is biased across gender (Abad, Colom, Rebollo & Escorial. 2004; Mackintosch & Bennett, 2005) and that there are sex by age interactions (Lynn et al., 2004). Despite these indications of suboptimal fit of the Rasch model, imperfect scaling is to be preferred over no scaling at all. The bias due to multiple highly intercorrelated factors is negligible.

The Rasch based intelligence scores were estimated using the Gibbs sampler as implemented in the BUGS software (http://www.mrc-bsu.cam.ac.uk/bugs) by taking the mean of each individual's posterior distribution. The estimation procedure used no assumptions regarding the distribution of the intelligence scores or item difficulties. Extreme scores (like no item correct or all items correct) are inestimable in the Rasch model. Therefore, individuals who had extreme scores were assigned a value half a logit higher than the second highest scoring individuals.

Extended twin design

In the classical twin study, the relative influence of variation in genes and environment is estimated by comparing MZ and DZ correlations, or covariances. The more similar MZ twins are relative to DZ twins, the more variability in phenotype is caused by genetic variability. When DZ twins resemble each other and are as alike as MZ twins, the resemblance between twins is caused by shared environment, and therefore it can be concluded that part of the variability in intelligence is caused by variability in shared environment. A distinction can be made between variation caused by additive genetic effects (A; caused by the additive effects of alleles at multiple loci), dominance genetic effects (D; non-additive effects of alleles), and environmental effects (E). Environmental effects might be correlated in offspring since they share potentially important environmental factors such as SES. The covariance of E is often denoted as the shared or common environmental variance component (C). The assumption is that MZ twins have the same DNA sequence and therefore A and D are perfectly correlated in MZ twins. DZ twins and siblings share on average half of their segregating genes, therefore the genetic correlation between their additive genetic values (A) is ½ (this correlation is higher in the presence of phenotypic assortment). The genetic correlation between the dominance

deviations (D) is ¼. Formally, stated as a random effects model the phenotypes of twins and siblings are modeled as:

$$P_{sibling1} = h*A_1 + d*D_1 + e*E_1, \text{ and}$$

$$P_{sibling2} = h*A_2 + d*D_2 + e*E_2,$$

where A, D and E are standardized to have unit variance. Corr(A_1 A_2) = 1 for MZ twins, Corr(A_1 A_2) = ½ for DZ twins and full siblings; Corr(D_1 D_2) = 1 for MZ twins, Corr(D_1 D_2) = ¼ for DZ twins and siblings and Corr(E_1 E_2) is to be estimated. The variance in P due to A, D and E is given by the square of *h*, *d* and *e*, respectively, so that Var(P) = $h^2 + d^2 + e^2$. The variance attributable to Common environment (environment shared by siblings from the same family) is obtained as: Var(C) = Corr(E_1 E_2) * e^2. Note that e^2 also contains variance due to measurement error. When only data from twins and siblings reared together are available, it is only possible to estimate Corr(E_1 E_2) under the assumption that *d* is zero or any other specified value, and vice versa, since a model including free parameters for both Corr(E_1 E_2) and *d* is not identified. For a parent, we have

$$P_{parent} = h*A_p + d*D_p + e*E_p.$$

In the absence of assortative mating, the expectation for Corr(A_p A_1) = Corr(A_p A_2) is ½. When there is no cultural transmission, or any other shared environment between parents and offspring, the expectation for Corr(E_p E_1) = Corr(E_p E_2) equals 0. Regardless of cultural transmission and assortative mating, the expectation for Corr(D_p D_1) = Corr(D_p D_2) is 0. When data from both twins and parents are available, the effects of cultural transmission and genetic dominance can be estimated at the same time.

Two different approaches were evaluated to model assortment between the parents of twins. The first model assumed that spousal resemblance was due to phenotypic assortment. The second model assumed that spousal correlation was caused by social homogamy. Figure 5.1 shows a path diagram of the model assuming phenotypic assortment. It is based on Fulker (1982) with the addition of dominance genetic variance. The phenotypes of the parents and one child are represented by IQ_f, IQ_m and IQ_o (father, mother and offspring). Variability in intelligence is caused by variation in A, E and D, and these are represented as latent factors in the model and have unit variance. The factor loadings on the latent factors are represented by, *h* (for A), *e* (for E), and *d* (for D). Parents pass their genes to their children, which is represented by arrows going from A of the parents to A of the child, with the factor loading ½. In the children, part of the genetic

variance is explained by transmission from the parents. The remaining residual additive genetic variance represents the variance that results from recombination. Because dominance effects are not transmitted from parents to offspring there are no paths going from the parental Ds to the child's D (Cavalli-Sforza et al., 1971).

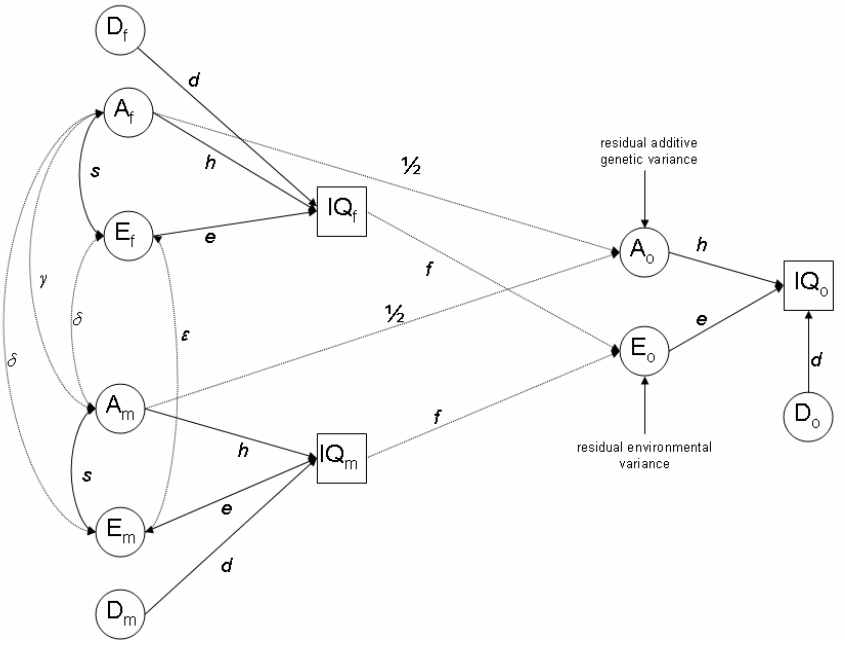


Figure 5.1. Path model for the spouse and parent-offspring correlations under the assumption of phenotypic assortment with γ representing genotypic correlation between parents, ε environmental correlation between parents, δ correlation between environment of one parent with genotype of other parent, *f* cultural transmission, *s* genotype environment correlation, f father, m mother, o offspring, A additive genetic value (*h* its factor loading), E environmental value (*e* its factor loading), D dominance variation (*d* its factor loading). Twins and sibling are not drawn in this figure for clarity reasons; however they mirror the drawn components (the relationship between twins are drawn in Fig 5.2).

The Greek letters on the left of the diagram in Figure 5.1 represent the correlations induced by phenotypic assortment. Coefficient γ represents the genotypic correlation between the parents, ε the environmental correlation between the parents, and δ represents the correlation of the environment of one parent with the genotype of the other parent. There is no dominance correlation between the parents, since in the case of polygenic inheritance this correlation is negligible (Cavalli-Sforza et al., 1971). All three correlations are induced by phenotypic assortment that can be represented as a parameter μ equal to the spousal correlation. This spousal correlation can be drawn as a co-path (Cloninger, 1980)

between the phenotypes of the parents instead of the paths which are represented by the Greek letters. Parameter μ can be written as a function of γ, ε, or δ (cf Fulker, 1982):

$$\mu = \gamma / (h + s\,e)^2 = \varepsilon / (e + h\,s)^2 = \delta / (e + h\,s)\,(h + e\,s).$$

Cultural transmission is represented by *f*, the regression of the child's environment on the parents' phenotypes. If *f* is not equal to 0, genotype and environment in the offspring generation become correlated (GE correlation). It is assumed that the system is at equilibrium, thus stable over generations, and therefore genotype and environment are correlated to the same extent in the parents as in the offspring. This GE correlation, *s*, is represented by the double-headed arrow between A and E of the parents. The correlation is implied in the offspring generation and at equilibrium equals $(1 + \mu)\,f\,h / [1 - (1 + \mu)\,f\,e]$ (Eaves, Eysenck & Martin, 1989). The residual environmental variance represents environmental effects not transmitted by the parents.

Figure 5.2 represents the effects of phenotypic assortment on the twin and sibling correlations. The phenotypes of the children are indicated by IQ_{T1}, and IQ_{T2} (oldest twin and youngest twin). The sibling data are omitted from the figure for clarity, but the expectations for twin-sib resemblance are the same as for DZ twin resemblance. Variation in intelligence is caused by variation in A, D and E, and the factor loadings for these variance components are represented by *h*, *d*, and *e*. Since dominance variation is not transmitted from the parents to their offspring, spousal resemblance does not influence correlations between dominance deviations in siblings and DZ twins (Cavalli-Sforza et al., 1971). Since A is transmitted from parents to their offspring, mate resemblance influences the twin and sibling correlations. MZ twins share the same DNA regardless whether phenotypic assortment takes place or not; therefore the genetic correlation in A between MZ twins stays 1. For DZ twins and sibs the correlation in A depends on the genotypic correlation between the parents. On average DZ twins share half their DNA, but the correlation between the genotypic values changes as a function of the genotypic correlation between the parents, γ.

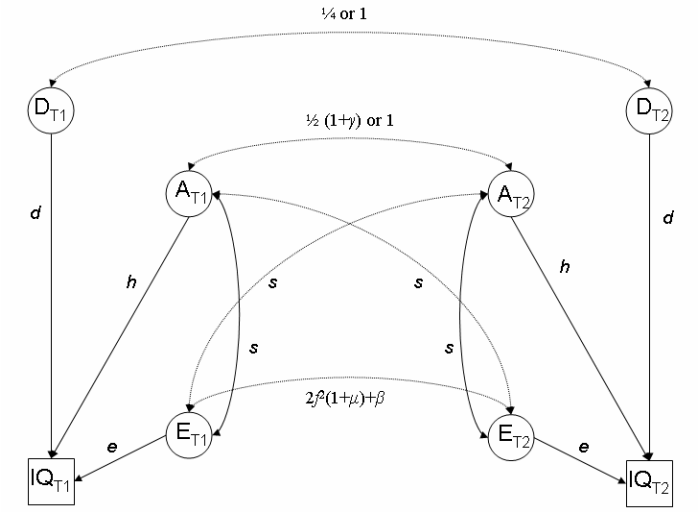


Figure 5.2. Path model for the twin correlations under the assumption of phenotypic assortment with γ representing genotypic correlation between parents, μ spousal correlation, β residual environmental covariance not explained by cultural transmission, *f* cultural transmission, *s* genotype environment correlation, A additive genetic value (*h* its factor loading), E environmental value (*e* its factor loading), D dominance variation (*d* its factor loading).T1 oldest twin, T2 youngest twin. The sibling is not drawn in this figure for clarity reasons; however the relationship between twins and sibling is similar to the relationship between dizygotic twins.

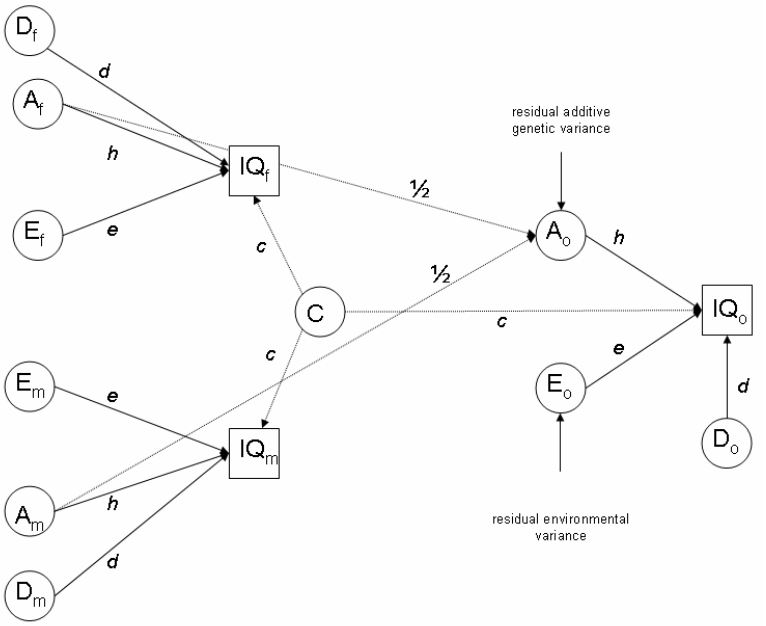


Figure 5.3. Path model for the spouse and parent-offspring correlations under the assumption of social homogamy, with f representing father, m mother, o offspring, A additive genetic value (*h* its factor loading), E unique environmental value (*e* its factor loading), D dominance variation (*d* its factor loading), C common environmental value (*c* its factor loading). Twins and sibling are not drawn in this figure for clarity reasons; however they mirror the drawn components (the relationship between twins and sibling are drawn in Fig 5.4).

The environmental correlation among offspring as the result of cultural transmission depends not only on *f*, but also on the phenotypic correlation between the parents, μ. Var(C), the environmental variance in the classical twin model that is shared by offspring, is now represented as the variance in the phenotype due to cultural transmission, $2\,e^2\,f^2\,(1+\mu)$, plus $e^2\,\beta$, the residual environmental variance shared by siblings (see Figure 5.2, cf. Boomsma et al. 1987). GE correlation is represented by parameter *s*, both within and across twins. Table 5.1 presents the derived expected correlations between family members in this model. Estimation of both Var(D) and β is not possible.

Figure 5.3 presents an alternative model that assumes that spousal correlation is due to social homogamy. Here, phenotypic resemblance in IQ in parents is only accounted for by a common environmental effect, C, that is uncorrelated with genotype A. This environmental effect is assumed to be the same in their children with an equal influence on the phenotype, *c*. Since the child's environment does not depend on the phenotypes of the parents, there is no GE correlation. Figure 5.4 gives the implications of these assumptions for the resemblance between twins and their siblings. In this model it is possible to estimate both Var(C) and Var(D). Table 5.1 gives the derived expected correlations between family members under the assumption of social homogamy.

Table 5.1. *Expected correlations between family members based on two genetic models.*

	Correlation	Expectation
Phenotypic assortment	MZ	$h^2 + e^2\,(2f^2(1+\mu) + \beta) + d^2 + 2hse$
	DZ / siblings	$\frac{1}{2}\,h^2\,(1+\gamma) + e^2\,(2f^2(1+\mu) + \beta) + \frac{1}{4}d^2 + 2hse$
	Parent-child	$\frac{1}{2}\,h\,(h+se)\,(1+\mu) + ef\,(1+\mu)$
	Spouse	μ
Social homogamy	MZ	$h^2 + d^2 + c^2 + e^2$
	DZ / siblings	$\frac{1}{2}\,h^2 + \frac{1}{4}\,d^2 + c^2 + e^2$
	Parent-child	$\frac{1}{2}\,h^2 + c^2 + e^2$
	Spouse	c^2

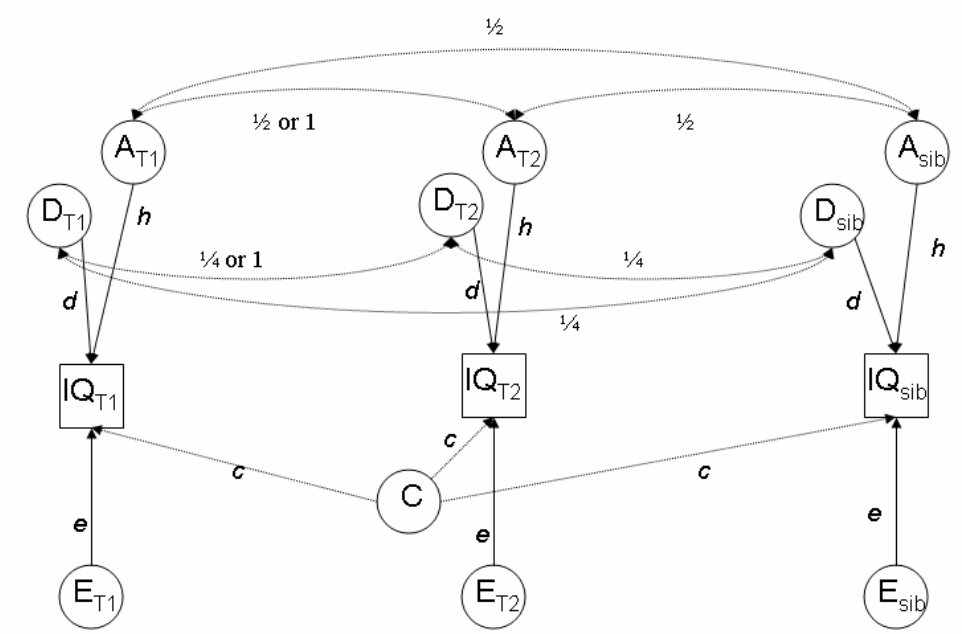


Figure 5.4. Path model for the twin and sibling correlations under the assumption of social homogamy with T1 representing the oldest twin, T2 the youngest twin, sib the sibling, A additive genetic value (*h* its factor loading), E unique environmental value (*e* its factor loading), D dominance variation (*d* its factor loading), C common environmental value (*c* its factor loading).

Model fitting

The Rasch-based IQ measures were first analyzed by fitting a general covariance matrix to the data from DZ and MZ twin families. In these general models, several assumptions were tested, such as equality of means and variances between MZ and DZ twins and between twins and siblings. The model was also used to test for sex and age effects on the means. Within the models best fitting model was chosen based on likelihood-ratio tests. Next, the Rasch IQ measures were fit to both the phenotypic assortment model and the social homogamy model using the statistical modeling package Mx (Neale, Boker, Xie, & Maes, 2003). Between models the best fitting model was chosen by minimizing Akaike's Information Criteria (AIC; $X^2 - 2*df$). The scripts can be downloaded from www.psy.vu.nl/mxbib.

Because the general model indicated that the MZ twin correlation was about twice the DZ twin correlation, we chose to include dominance genetic variance, and therefore not to estimate β in the model which assumed phenotypic assortment; β was fixed to zero.

Heritabilities in parents and their offspring were assumed to be equal (see Rijsdijk, Vernon, & Boomsma, 2002; Reynolds et al., 1996).

The best fitting genetic model resulting from the Mx analyses on the Rasch IQ measures (which are estimates) was also estimated using the raw item data directly in BUGS (Van den Berg, Glas, & Boomsma, under review; Van den Berg, Beem & Boomsma, 2006). For clarity we report about the credibility regions as confidence intervals, although in Bayesian statistics one generally speaks of credibility regions instead of confidence intervals. The results on the estimated Rasch IQ measures are somewhat biased for two reasons: the precision of the estimates is not equal across generations, since the estimates in the parents were based on 36 items from the Advanced version and the estimates in the offspring were based on 60 items from the Standard version. The reliabilities for the scales might be different and by modeling the observed item data directly, one adjusts the model parameters for attenuation effects due to scale unreliability that might be different across test version. Secondly, scale reliability does not only differ across test versions, but is also dependent on the location on the scale: estimation precision is usually better for people with average scores than people at the extremes of the scales. By modeling the item data directly one gets parameter estimates that take all these scale effects into account, yielding results that are corrected for attenuation effects (see also Van den Berg et al., under review).

To test for GE interaction, the average scores of MZ twin pairs were correlated with the absolute differences within a pair. Differences within MZ twin pairs can be attributed to the environment, and differences between MZ twin pairs can be attributed to genotype and environmental effects shared in twins. Thus, if the averages and differences are correlated - and there are no shared environmental effects - this suggests that people with a certain genotype are more sensitive to environmental influences than people with another genotype. Since the scores are estimates, and the precision of a Rasch IQ estimate is dependent of the location on the scale (see above), the correlation estimate and its *p*-value might be incorrect. Therefore the correlation between the latent trait average and latent absolute difference was estimated by modeling the observed item data in BUGS and applying the Rasch model.

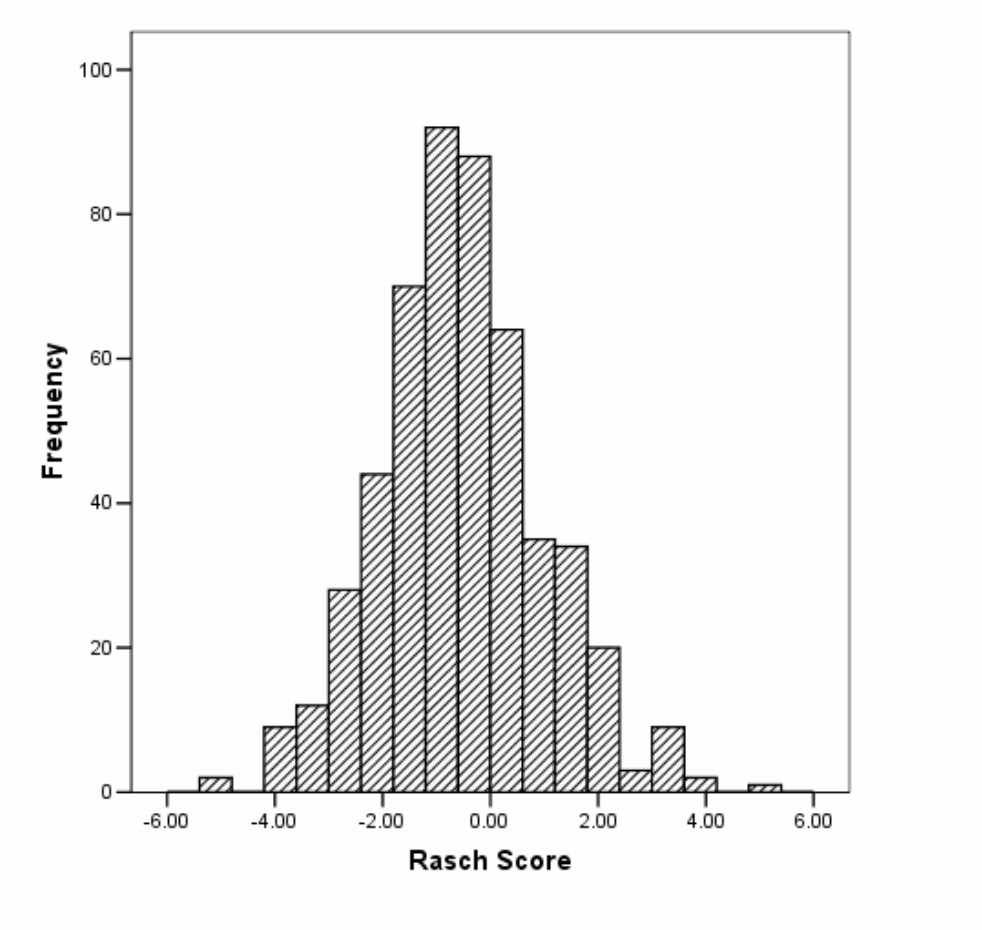


Figure 5.5. The distribution of estimated, Rasch-based, IQ scores for parents, siblings and twins.

Results

Descriptive statistics of the Raven IQ sum scores are in Table 5.2. For the estimated IQ measures based on the Rasch scaling, no significant sex differences were observed: neither in the total group, nor within groups (parents, siblings, twins). There was no age by sex interaction in the offspring. The variance in the siblings was significantly larger than in the twins, which could partly be explained by age differences. Therefore the age effect was retained in all models. Phenotypic correlations, variances and covariances estimated in the general reference (non-genetic) model are given in Table 5.3 and model fit is in Table 5.4. The distribution of Rasch IQ scores looked more or less normal (see Figure 5.5). The

distribution of the estimated measures in twins showed a slight negative skew and in the sibling and parental data a slight positive skew.

Table 5.2. *Descriptives for all subjects of the sum IQ score on the Raven Progressive Matrices. Parents received the Advanced Progressive Matrices Test (maximum achievable score = 36) and offspring received Standard Progressive Matrices (maximum achievable score = 60).*

	N	Min	Max	Mean	*SD*
Fathers	94	4	36	27.0	6.5
Mothers	95	9	36	25.9	6.0
Male siblings	44	24	56	43.8	7.8
Female siblings	57	30	59	46.4	6.5
Male twins	114	13	50	36.7	8.6
Female twins	110	19	50	36.6	7.1

In the genetic analyses, the larger variance in the siblings was modeled using a scalar effect in addition to the age effect to account for their variance, assuming that the components of genetic and environmental variance were proportional to those observed in twins. Fitting of the model assuming phenotypic assortment (see Fig. 5.1 and 5.2) showed that including dominance variation in this model does not lead to a significantly better fit (*d*; -Δ2LL = .88; 95% confidence interval 0, .75). There was also no significant contribution of cultural transmission (*f*; -Δ2LL = .86; 95% confidence interval -.30, .44) and therefore no GE correlation (*s*). A simple model with only additive genetic effects and non-shared environmental effects explained the data best. The expected phenotypic correlations, variances and covariances are given in Table 5.3; the model fit is in Table 5.4. In this model genetic variation contributes 58% to the variation in intelligence in children as well as adults. The remaining 42% is explained by unique environmental variation.

Table 5.3. *Expected phenotypic correlations, variances and covariances for the general reference model and for the genetic models with phenotypic assortment and social homogamy.*

	Reference model			Phenotypic Assortment			Social Homogamy		
	Var	Cov	*r*	Var	Cov	*r*	Var	Cov	*r*
Twin	1.18			1.18			1.20		
Sibling	1.82			1.79			1.81		
Spouse	2.60	.85	.33	2.64	.84	.32	2.56	.70	.27
Twin MZ		.74	.63		.69	.58		.74	.61
Twin DZ		.30	.25		.41	.34		.48	.39
Twin-sibling		.55	.37		.50	.34		.58	.39
Parent-twin		.62	.35		.68	.38		.61	.35
Parent-sibling		.83	.38		.83	.38		.75	.35

Table 5.4. *Fit indices for the general (non-genetic) reference model, best fitting phenotypic assortment genetic model and best fitting social homogamy genetic model.*

Model	*-2LL*	# free parameters	# *df*	AIC
Reference	1633.95	13	500	633.95
Phenotypic Assortment	1635.71	9	504	627.71
Social Homogamy	1636.08	10	503	630.08

In the model assuming social homogamy (see Fig. 5.3 and 5.4) there is a significant contribution of dominance variance (fixing *d* to 0 leads to a significantly worse fit Δ-2LL = 2.93), and social environment (fixing *c* to 0 leads to a significantly worse fit Δ-2LL = 8.16). Additive genetic variance could however be dropped (Δ-2LL = 0.73). As a model with only dominance genetic variance is a priori not sensible, this additive genetic component was retained. Tables 5.3 and 5.4 present the expected variances, covariances, correlations and model fit indices. In this model additive genetic variation contributes 15% to the variation in intelligence in children as well as adults, dominance deviation explains 19% in variation in IQ, and shared environment explains 27%. The remaining 39% is

explained by non-shared environmental variation. Comparing the phenotypic assortment model and the social homogamy model, the model assuming phenotypic assortment appears superior as it showed a higher likelihood while having fewer parameters.

The phenotypic assortment model was also estimated in BUGS, this time on the raw item data. The estimate for *h* was a bit higher, leading to a heritability estimate of 67% (95% confidence interval: 52%, 79%). Similar to the analyses on the Rasch estimates, the parameter for the effect of a sib's age (in years) was not significantly different from zero (.18, 95% confidence interval: -0.02, 0.37). Estimated variance of the unobserved intelligence scores was 1.99 in the parents, 1.08 in the twins and 1.46 in the siblings (after age correction). The 67% point estimate can be regarded as the estimate for the heritability that we would get with an infinite number of similar test items, that is, corrected for attenuation effects (cf. Van den Berg et al., under review).

The estimate for the correlation between average intelligence and difference between MZ twins is -.30, which is significantly different from 0, $p < .05$ (95% confidence interval: -.08, -.52). This suggests that the environment is relatively more important in explaining individual differences for low IQ groups than for high IQ groups. This GE interaction effect explains 9% of the variance in the scores (Jinks & Fulker, 1970). In the models fitted above, the G*E variance is attributed to environmental effects not shared by family members.

Discussion

In this study several quantitative genetic models to study the heritability of intelligence were evaluated using data from twins, one of their siblings and both parents. With a Rasch measurement model, a measure of IQ based on the Raven Progressive Matrices test was estimated in all participants.

Correlations were higher in MZ twins than in first-degree relatives (siblings, DZ twins and parent-offspring pairs). The spousal correlation for the Rasch IQ estimates was significant and moderately high (0.33). A model assuming that this correlation is due to phenotypic assortment proved superior to a model assuming that the correlation was due to purely environmental factors that are transmitted from generation to generation. Corrected for scale unreliability effects, additive genetic effects account for 67% of the variation in intelligence and the remainder is explained by random environmental factors, including

measurement error. Non-additive genetic effects (D) and cultural transmission effects (*f*) were not significant. Some other studies have suggested that non-additive genetic effects plays a role in the heredity of intelligence (Chipuer, Rovine, & Plomin, 1990; Fulker 1979; Jinks & Fulker, 1970). We did not find evidence for genetic non-additivity, which seems consistent with most of the behavior genetics literature on IQ (though we recognize that studies that explicitly addressed this issue are scarce).

The absence of common environmental effects shared by family members is in line with the findings from for instance the CAP study where an adoptive parent's IQ does not predict the IQ of the adopted child (Phillips & Fulker, 1989). Prior studies on intelligence in children have reported environmental influences that are shared by siblings, a finding that we did not replicate. Usually familial environmental effects are only seen in children and tend to disappear in adolescence (e.g., Posthuma, et al., 2001; Scarr & Weinberg, 1983). There may be several reasons why other studies found such effects and we did not. First of all, our final model assumed phenotypic assortment. When phenotypic assortment is not controlled for, an analysis based on only MZ and DZ twin correlations overestimates shared environmental influences and underestimates additive genetic variance. Therefore, in studies where only twins are used, part of the variance that is labeled shared environmental influences may actually include genetic variance due to assortative mating. Secondly, the absence of shared environmental influences may be related to the IQ measure that was used. IQ was assessed with the Raven Progressive Matrices, a test conceptually more related to performal IQ than verbal IQ. Thus, the findings of our relatively high heritability estimate relative to other studies in children and the absence of shared environmental influences may be due to the measure that was used in addition to modeling the effects of assortative mating.

One important assumption in the modeling was that heritability was equal across generations, and the same genes are expressed. Regarding the first assumption, the heritability estimate based on the estimated scores, uncorrected for reliability, (58%), is comparable to the 64% reported by Rijsdijk et al. (2002), who collected Raven data in Dutch 16-year-old twin pairs, and also comparable to the heritability observed in adults by Reynolds et al. (1996). Regarding the second assumption, it is known that intelligence scores are highly stable and that in children, this stability is partly due to a common genetic factor explaining IQ at different ages: the genes that influence IQ in early childhood are

largely the same genes that influence IQ at later ages (e.g., Bartels et al. 2002). Also in adulthood, stability in intelligence is largely due to the same set of genetic factors (e.g. Plomin et al., 1994; Van den Berg, Posthuma & Boomsma, 2004; see also DeFries, Plomin, & LaBuda, 1987; Plomin, Fulker, Corley & DeFries, 1997). Thus, although there is some evidence that the genetic correlation across age is not perfect, the conclusions from our models are not likely to be severely biased.

Our conclusions are based on the assumption of phenotypic assortment. A model with phenotypic assortment provided a more parsimonious explanation of the present data than a model with social homogamy. This finding is in contrast to that obtained by Reynolds et al., (1996) who studied twins born between 1911 and 1935. They reported that social homogamy could explain spousal similarity and that phenotypic assortment was not significant. However, their analysis was based on the (unlikely) assumption that there is no correlation between genotypes and the environment in which prospective partners meet. Alternatively, it is possible that nowadays, social homogamy plays a less important role than in the early 20th century.

There was evidence for GE interaction, suggesting that the environment is relatively more important in explaining individual differences for low IQ groups than for high IQ groups. Similar findings were reported by Jinks & Fulker (1970), Jensen (1970) and Finkel and Pedersen (2001), although their effect sizes were smaller. The GE interaction effect is in agreement with findings from Turkheimer et al. (2003) who showed that the relative influence of genotype is larger for children from parents of high social-economic status (SES) than for children from low SES parents.

We found that the mean IQ score in the older siblings was higher and also that there was more variance in siblings than in twins, even though the same test was used. This could not be fully explained by age differences among the siblings. The finding is, however, consistent with results obtained by Thurstone (1928) who showed a positive relationship between group mean and group variance with scaled intelligence scores. Such a phenomenon cannot be observed in normed IQ scores by definition. Future research should determine whether it is merely a scaling effect or whether it perhaps reflects increased variability due to individual differences in the timing of puberty.

Variability in fluid intelligence as measured by the Raven is largely explained by additive genetic effects that are transmitted from parents to offspring. In accordance with

adoption studies (Alarcón et al., 2003; Fulker et al., 1983; Humphreys et al., 1988; Rice et al., 1989; cf. Scarr & Weinberg, 1978, 1983), we found no evidence for cultural transmission: all influence from parents on their children's IQ was explained by the transmission of genes. However, in the approach that was taken, cultural transmission was modeled as a direct effect of parental IQ on offspring environment. Although this model does not seem unreasonable for IQ, it might be that Raven IQ does not capture those aspects of the parental phenotype that are most salient in determining the child's Raven IQ.

The present study design is not suited to uncover GE correlations other than one resulting from simultaneous genetic and cultural transmission. But what we can conclude is that if there is GE correlation, the role of parents seems limited to responding to the needs and interests as indicated by the child. We found no indication that intelligent parents provide their offspring with intelligence promoting circumstances. More likely, children with a genetic predisposition for either a low or a high IQ ask for a specific type of stimulation. In other words, an evocative gene-environment correlation (where individuals are reacted to on the basis of their genetically influenced phenotype) or an active GE correlation (where individuals seek or create environments correlated with their genetic inclinations) seems a more probable mechanism than a passive GE correlation (Scarr & McCartney, 1983). Only the last type of correlation could in principle have been detected by our extended twin family design.

In conclusion, individual differences in intelligence are largely accounted for by genetic differences. Environmental factors are significantly more important in children with a genetic predisposition for low IQ than in children with a genetic predisposition for high IQ. Environmental factors influencing IQ are generally not shared among siblings.

For future research we recommend to implement extended twin designs similar to the one used in this study. Although our study consisted of a high number of participants (516), we only included a limited number of families. In our sample we had only limited power to detect effects of genetic dominance and perhaps they will reach significance in a larger sample. Measures of cognition that include aspects of e.g. verbal cognition, correlates of IQ such as brain volume and function, and inclusion of twins and sibs of different ages should shed more light on, for instance, the presence of cultural transmission for verbal IQ, how genetic effects on IQ are mediated, and the extent to which results generalize to younger and older children,. Moreover, we recommend that future genetic

research on intelligence focuses on the exact nature of the GE interaction and the possible existence of evocative and active GE correlation.

Reference List

Abad, F., Colom, R., Rebollo, I., & Escorial, S. (2004). Sex differential item functioning in the Raven's Advanced Progressive Matrices: evidence for bias. *Personality and Individual Differences, 36,* 1459-1470.

Agrawal, N., Sinha, S.N., & Jensen, A.R. (1984). Effects of inbreeding on Raven matrices. *Behavior Genetics*, *14*, 579-585.

Alarcón, M., Plomin, R., Corley, R.P., and DeFries, J.C. (2003). Multivariate parent-offspring analyses of specific cognitive abilities. In: S.A. Petrill, Plomin, R., J.C. DeFries, J.K. Hewitt (Eds). *Nature, nurture, and the transition to early adolescence.* Oxford: Oxford University Press.

Bartels, M., Rietveld, M. J. H., Van Baal, G. C. M., & Boomsma, D. I. (2002). Genetic and environmental influences on the development of intelligence. *Behavior Genetics, 32,* 237-249.

Bashi, J. (1977). Effects of inbreeding on cognitive performance. *Nature*, *266*, 440-442.

Bishop, E.G., Cherney, S.S. & Hewitt, J.K. (2003). Developmental analysis of IQ. In: S.A. Petrill, Plomin, R., J.C. DeFries, J.K. Hewitt (Eds). *Nature, nurture, and the transition to early adolescence*. Oxford: Oxford University Press.

Bond, T.G. & Fox, C. M. (2001) Applying the Rasch model: fundamental measurement in the human sciences. Mahwah, N.J.: Erlbaum.

Boomsma, D., Busjahn, A., & Peltonen, L. (2002). Classical twin studies and beyond. *Nature Reviews Genetics, 3,* 872-882.

Boomsma, D. I. (1993). Current status and future prospects in twin studies of the development of cognitive abilities, infancy to old age. In T.J.Bouchard & P. Propping (Eds.), *Twins as a Tool of Behavioral Genetics* (pp. 67-82). Chichester: John Wiley & Sons.

Boomsma, D. I., De Geus, E. J. C., Vink, J. M., Stubbe, J. H., Distel, M. A., Hottenga, J. J. et al. (2006). Netherlands Twin Register: from twins to twin families. *Twin Research and Human Genetics*, *9*, 849-857

Boomsma, D. I. & Molenaar, P. C. (1987). Constrained maximum likelihood analysis of familial resemblance of twins and their parents. *Acta Geneticae Medicae Gemellogiae, 36,* 29-39.

Boomsma, D. I., Orlebeke, J. F., & Van Baal, G. C. (1992). The Dutch Twin Register: growth data on weight and height. *Behavior Genetics, 22,* 247-251.

Boomsma, D. I., Vink, J. M., Van Beijsterveldt, T. C., De Geus, E. J., Beem, A. L., Mulder, E. J. et al. (2002). Netherlands Twin Register: A focus on longitudinal research. *Twin Research, 5,* 401-406.

Bouchard, T. J., Jr. & McGue, M. (1981). Familial studies of intelligence: a review. *Science, 212,* 1055-1059.

Bouchard, T. J., Jr. & McGue, M. (2003). Genetic and environmental influences on human psychological differences. *Journal of Neurobiology, 54,* 4-45.

Cavalli-Sforza, L. L. & Bodmer, W. F. (1971). Quantitative characters, polygenic inheritance, and environmental interactions. In *The Genetics of Human Populations* (pp. 508-633). San Francisco: W.H. Freeman and Company.

Cherny, S. S. & Cardon, L. R. (1994). General cognitive ability. In J.C.DeFries, R. Plomin, & D. W. Fulker (Eds.), *Nature and nurture during middle childhood* (pp. 46-56). Oxford: Blackwell.

Chipuer, H.M, M.J. Rovine, & R. Plomin. 1990. LISREL modeling: genetic and environmental influences on behavior revisited. *Intelligence*, *14*, 11-29.

Cloninger, C. R. (1980). Interpretation of intrinsic and extrinsic structural relations by path analysis: theory and applications to assortative mating. *Genet. Res. 36,* 135–145.

Deary, I. J., Spinath, F. M., & Bates, T. C. (2006). Genetics of intelligence. *European Journal of Human Genetics., 14,* 690-700.

DeFries, J. C., Plomin, R., & LaBuda, M. C. (1987). Genetic stability of cognitive development from childhood to adulthood. *Developmental Psychology*, 23, 4-12.
Eaves, L.J., Eysenck, H.J. & Martin, N.G. (1989). Genes, culture and personality: an empirical approach. London: Academic.
Finkel, D. & Pedersen, N. L. (2001). Sources of environmental influence on cognitive abilities in adulthood. In E.L.Grigorenko & R. J. Sternberg (Eds.), *Family environment and intellectual functioning* (pp. 173-194). Mahwah: Lawrence Erlbaum Associates.
Fulker, D. W. (1979). Nature and nurture: heredity. In H. J. Eysenck (ed.) *The Structure and Measurement of Intelligence.* Berlin: Springer.
Fulker, D. W. (1982). Extensions of the classical twin method. In B.C.Weir, E. J. Eisen, M. M. Goodman, & G. Namkoong (Eds.), *Proceedings of the second international conference on quantitative genetics* (pp. 395-406). Sunderland, MA: Sinauer.
Fulker, D. W. & DeFries, J. C. (1983). Genetic and environmental transmission in the Colorado Adoption Project: path analysis. *British Journal of Mathematical and Statistical Psychology, 36 (Pt 2),* 175-188.
Gibson, W. A. (1959). Three multivariate models: factor analysis, latent structure analysis, and latent profile analysis. *Psychometrika*, *24*, 229–252
Gilger, J. W. (1991). Differential assortative mating found for academic and demographic variables as a function of time of assessment. *Behavior Genetics, 21,* 131-150.
Guttman, R. (1974). Genetic analysis of analytical spatial ability: Raven's progressive matrices. *Behavior Genetics, 4,* 273-284.
Guttman, R. & Shoham, I. (1983). Parent-offspring resemblance and parental education. *Behavior Genetics, 13,* 99-105.
Humphreys, L. G. & Davey, T. C. (1988). Continuity in intellectual growth from 12 months to 9 years. *Intelligence, 12,* 183-197.
Jensen, A. R. (1970). IQ's of identical twins reared apart. *Behavior Genetics, 1,* 133-148.
Jinks, J. L. & Fulker, D. W. (1970). Comparison of the biometrical genetical, MAVA, and classical approaches to the analysis of human behavior. *Psychological Bulletin, 73,* 311-349.
Keller, M. C., Coventry, W. L., Heath, A. C., & Martin, N. G. (2005). Widespread evidence for non-additive genetic variation in Cloninger's and Eysenck's personality dimensions using a twin plus sibling design. *Behavior Genetics, 35,* 707-721.
Lynch, M. & Walsh, B. (1998). *Genetics and analysis of quantitative traits*. Sunderland: Sinauer Associates.
Lynn, R., Allik, J. & Irwing, P. (2004). Sex differences on three factors identified in Raven's Standard Progressive Matrices. *Intelligence, 32,* 411-424.
Mackintosh, N. & Bennett, T. (2005) What do Raven's matrices measure? An analysis in terms of sex differences. *Intelligence*, 33, 663-674.
Mascie-Taylor, C. G. (1989a). Spouse similarity for IQ and personality and convergence. *Behavior Genetics, 19,* 223-227.
Mascie-Taylor, C. G. & Vandenberg, S. G. (1988). Assortative mating for IQ and personality due to propinquity and personal preference. *Behavior Genetics, 18,* 339-345.
Neale, M., Boker, S. M., Xie, G., & Maes, H. H. (2003). *Mx: statistical modelling*. (6 ed.) Richmond, VA: Department of Psychiatry, Medical College of Virginia.
Phillips, K. & Fulker, D. W. (1989). Quantitative genetic analysis of longitudinal trends in adoption designs with application to IQ in the Colorado Adoption Project. *Behavior Genetics, 19,* 621-658.
Phillips, K., Fulker, D. W., Carey, G., & Nagoshi, C. T. (1988). Direct marital assortment for cognitive and personality variables. *Behavior Genetics, 18,* 347-356.
Plomin, R., DeFries, J. C., Craig, I. W., & McGuffin, P. (2002). Behavioral Genetics. In R.Plomin, J. C. DeFries, I. W. Craig, & P. McGuffin (Eds.), *Behavioral Genetics in the Post Genomic Era* (pp. 3-15). Washington D.C.: American Psychological Association.

Plomin, R., Fulker, D.W., Corley, R. & DeFries, J.C. (1997) Nature, nurture and cognitive development from 1 to 16 years: A Parent-Offspring Adoption Study *Psychological Science, 8,* 442–447

Plomin, R., Pedersen, N., Lichtenstein, P., & McClearn, G. E. (1994). Variability and stability in cognitive abilities are largely genetic later in life. *Behavior Genetics, 24,* 207-215.

Plomin, R & Spinath, F. (2004). Intelligence: genetics, genes, and genomics. *Journal of Personality and Social Psychology*, *86*, 112-129.

Posthuma, D. & Boomsma, D. I. (2000). A note on the statistical power in extended twin designs. *Behavior Genetics, 30,* 147-158.

Posthuma, D., De Geus, E. J., & Boomsma, D. I. (2001). Perceptual speed and IQ are associated through common genetic factors. *Behavior Genetics, 31,* 593-602.

Rasch, G. (1966). An item analysis which takes individual differences into account. *Br.J Math.Stat.Psychol, 19,* 49-57.

Raven, J., Raven, J. C., & Court, J. H. (1998). *Raven manual: Section 4 advanced progressive matrices*. Oxford: Oxford Psychologists Press.

Raven, J. C. (1960). *Guide to the Standard Progressive Matrices*. London: H.K. Lewis & Co. Ltd.

Reynolds, C. A., Baker, L. A., & Pedersen, N. L. (1996). Models of spouse similarity: applications to fluid ability measured in twins and their spouses. *Behavior Genetics, 26,* 73-88.

Rice, T., Carey, G., Fulker, D. W., & DeFries, J. C. (1989). Multivariate path analysis of specific cognitive abilities in the Colorado Adoption Project: conditional path model of assortative mating. *Behavior Genetics, 19,* 195-207.

Rietveld, M. J., van der Valk, J. C., Bongers, I. L., Stroet, T. M., Slagboom, P. E., & Boomsma, D. I. (2000). Zygosity diagnosis in young twins by parental report. *Twin Research, 3,* 134-141.

Rijsdijk, F. V., Vernon, P. A., & Boomsma, D. I. (2002). Application of hierarchical genetic models to Raven and WAIS subtests: a Dutch twin study. *Behavior Genetics, 32,* 199-210.

Rost, D. H. & Gebert, A. (1980). Zum Problem der Faktoreninterpretation bei Raven's Coloured Progressive Matrices [On the problem of factor interpretation in Raven's Coloured Progressive Matrices]. *Zeitschrift für Differentielle und Diagnostische Psychologie*, *1*, 255-273.

Scarr, S. & McCartney, K. (1983). How people make their own environments: a theory of genotype - environment effects. *Child Development*, *54*, 424-435.

Scarr, S. & Weinberg, R. A. (1978). The influence of "family background" on intellectual attainment. *American Sociological Review*, *43*, 674-692.

Scarr, S. & Weinberg, R. A. (1983). The Minnesota Adoption Studies - Genetic-Differences and Malleability. *Child Development, 54,* 260-267.

Smith E. & Smith, R. (Eds) (2004). Introduction to Rasch Measurement: theory, models and applications. Maple Grove, MN: JAM Press.

Spearman, C. (1927). The abilities of man: their nature and measurement. New York: MacMillan.

Thurstone, L.L. (1928). The absolute zero in intelligence measurement. *Psychological Review*, *35*, 175-197.

Turkheimer, E., Haley, A., Waldron, M., D'Onofrio, B., & Gottesman, I. I. (2003). Socioeconomic status modifies heritability of IQ in young children. *Psychological Science, 14,* 623-628.

Van den Berg, S. M., Beem, L., & Boomsma, D. I. (2006). Fitting genetic models using Markov Chain Monte Carlo algorithms with BUGS. *Twin Research and Human Genetics, 9,* 334-342.

Van den Berg, S. M., Glas, C. A. W., & Boomsma, D. I. (under review). Variance decomposition using an IRT measurement model. *Behavior Genetics*.

Van der Ven, A. H. G. S. & Ellis, J. L. (2000). A Rasch analysis of Raven's standard progressive matrices. *Personality and Individual Differences, 29,* 45-64.

Vigneau, F. & Bors, D.A. (2005). Items in context: assessing the dimensionality of Raven's Advanced Progressive Matrices. *Educational and Psychological Measurement*, *65*, 109-123.

Wadsworth, S. J., DeFries, J. C., Fulker, D. W., & Plomin, R. (1995). Cognitive ability and academic achievement in the Colorado Adoption Project: a multivariate genetic analysis of parent-offspring and sibling data. *Behavior Genetics, 25,* 1-15.

Wainwright, M. A., Wright, M. J., Geffen, G. M., Luciano, M., & Martin, N. G. (2005). The genetic basis of academic achievement on the Queensland Core Skills Test and its shared genetic variance with IQ. *Behavior Genetics, 35,* 133-145.

Watkins, M. P. & Meredith, W. (1981). Spouse similarity in newlyweds with respect to specific cognitive abilities, socioeconomic status, and education. *Behavior Genetics, 11,* 1-21.

Watson, D., Klohnen, E. C., Casillas, A., Simms, E. N., Haig, J., & Berry, D. S. (2004). Match makers and deal breakers: analyses of assortative mating in newlywed couples. *Journal of Personality, 72,* 1029-1068.

Williams, T. (1975). Family resemblance in abilities: the Wechsler scales. *Behavior Genetics, 5,* 405-409.

A genetic analysis of brain volumes and IQ in children

In a population-based sample of 112 nine-year old twin pairs, we investigated the association among total brain volume, gray matter and white matter volume, intelligence as assessed by the Raven IQ test, verbal comprehension, perceptual organization and perceptual speed as assessed by the Wechsler Intelligence Scale for Children – III. Phenotypic correlations between the brain volumes and intelligence traits ranged between .20 and .33. Processing speed and brain volume did not correlate. The relation between brain volume and intelligence was entirely explained by a common set of genes influencing both sets of phenotypes.

6

This chapter is published as:

Van Leeuwen, M., Peper, J. S., Van den Berg, S. M., Brouwer, R. M., Hulshoff Pol, H. E., Kahn, R. S., & Boomsma, D.I. (in press). A genetic analysis of brain volumes and IQ in children. *Intelligence*.

Sir Sir Francis Galton (Galton, 1869) was the first to systematically attempt to investigate the effect of heredity on intellectual abilities using normal distribution and pedigree analysis. Later Galton suggested the use of twin studies to disentangle nature from nurture and developed, based on his work on inheritance, regression and correlation coefficients. Like many of his contemporaries Galton thought that head circumference and intelligence are positively related. In 1901, with Pearson he used their newly developed correlational techniques to compute the correlation between head size and academic record. This correlation turned out to be low (.1; Fancher, 1983; Tredoux, 2007).

Since 1901, the relationship between total brain volume (TBV) and intelligence in adults (e.g. Andreasen et al., 1993; Egan et al., 1994; Thoma et al., 2005; Wickett, Vernon, & Lee, 2000; Witelson, Beresh, & Kigar, 2006) and children (e.g. Frangou, Chitins, & Williams, 2004; Reiss, Abrams, Singer, Ross, & Denckla, 1996) has been well established. In a review McDaniel (2005) estimated a population correlation between intelligence and TBV of .33. To study whether this relationship is a function of nerve conduction velocity or the number of neurons, a number of studies also examined the correlation between intelligence and *white matter volume* (WMV) and *gray matter volume* (GMV) separately. Gray matter consists of neural cell bodies, dendrites and synapses, whereas white matter consists of myelinated axons. The myelination hypothesis (Miller, 1994) states that thicker myelin sheaths will result in larger brain volume. Increased myelination in turn leads to increased speed of cortico-cortical connections, which is in concordance with research evidence showing that people with faster brains, as expressed by shorter reaction time and inspection time, are more intelligent (Posthuma, De Geus, & Boomsma, 2002). Nevertheless, the relation between WMV and intelligence is unclear. Andreasen et al. (1993) and Reiss et al. (1996) did not find a relation between intelligence and WMV in children and adults, while Posthuma et al. (2002) and Thoma et al. (2005) observed a significant positive relation in adults. One possible explanation for this discrepancy can lie in the subtle differences in WMV responsible for differences in cognitive function: (Pujol et al., 2004) showed a significant difference of only 1.6% in WMV between a group of children (N =100) with cognitive decline and a group of controls (N = 50). This observed difference was equivalent to a 3.2-year myelination delay. So, to detect the effect of WMV on intelligence in the normal population samples of sufficiently large size are required. In contrast, in both children and adults there is consistent evidence for a positive relation

between GMV (Andreasen et al., 1993; Frangou et al., 2004; Posthuma et al., 2002; Reiss et al., 1996; Thoma et al., 2005; Wilke, Sohn, Byars, & Holland, 2003) and cortical thickness (Shaw et al., 2006) and intelligence.

These earlier studies on brain volume and intelligence often report on samples of above average IQ and/ or range restricted samples (e.g. Egan et al., 1994; Frangou et al., 2004; Reiss et al., 1996; Wickett et al., 2000; Wilke et al., 2003; Witelson et al., 2006). The studies in children used groups of subjects which typically ranged from five to eighteen years (Frangou et al., 2004; Reiss et al., 1996; Wilke et al., 2003). Brain volumes are age dependent (Giedd et al., 1999;(Lenroot et al., 2007). TBV peaks at age eleven for girls and age fifteen for boys, after which it slowly decreases (Giedd et al., 1999). GMV follows an inverted U-shaped developmental pattern which peaks between nine and fifteen years depending on brain area and sex. WMV on the other hand increases linearly with age (Lenroot et al., 2007; see also Lenroot & Giedd (2006) for a review on brain development in children and adolescents). Therefore, the wide age range may blur the relation between brain volumes and intelligence. This may explain for instance the finding in the study of Wilke et al. (2003), where there was no relationship between WMV and IQ and where the relation between GMV and IQ was present in the older age group (mean age 15 years) only.

To disentangle the etiology of variation in brain volume and intelligence twin studies are useful. These studies can separate variation caused by differences in human DNA sequence and other genetic sources of variation and variation caused by differences in environment (Plomin & Kosslyn, 2001). The proportion of genetic variance over the total variance is defined as *heritability*. Environmental variance can be decomposed into variance shared by family members (*shared environment*) and variance which is unique for each individual (*unique environment*). Heritability estimates are obtained by comparing the resemblance between *monozygotic twins* (MZ) with the resemblance between *dizygotic twins* (DZ). When MZ twin correlations are higher than DZ twin correlations, part of the twin resemblance in the phenotype is caused by genetic effects. When DZ twin correlations are more than half the size of MZ correlations, the resemblance between twins is at least partly caused by shared environmental effects. The importance of unique environment is reflected by differences between MZ twins.

Using a multivariate genetic design one can establish the etiology of the relation between IQ and brain structure. The association may be caused by genetic factors which

influence brain structure as well as IQ, and / or by environmental factors influencing both traits. These types of studies can lead to new insights not only about how genes affect intelligence, but also about how the brain works. As stated by Plomin and Kosslyn (2001) and Peper, Brouwer, Boomsma, Kahn, and Hulshoff Pol (2007) genetic studies on brain and intelligence will help dissecting pathways relating genes, brain and intelligence. Currently the nature of the genetic polymorphisms involved in intelligence and brain volume are unclear (Deary, Spinath, & Bates, 2006).

The relation between brain structure and cognition has been studied in genetic informative samples in adults (Carmelli, Swan, DeCarli, & Reed, 2002; Schoenemann, Budinger, Sarich, & Wang, 2000; Thompson et al., 2001; Tramo et al., 1998; Wickett et al., 2000). Posthuma et al. (2002), Hulshoff Pol et al., (2006) and Posthuma et al. (2003) showed in a sample of 135 individuals (existing of twins and siblings coming from 60 twin families) that the relationship between full scale IQ and WMV and GMV is completely mediated by genetic factors, as well as the relationship between verbal IQ and performal IQ and several focal GM and WM areas. A similar genetic origin was found for the association between working memory, GMV and WMV (Posthuma et al, 2003). In addition, for the other three dimensions of the Wechsler Adult Intelligence Scale – III (WAIS-III), it was found that processing speed was genetically related to WMV, whereas perceptual organization and verbal comprehension were neither related to WMV nor to GMV. In another study in young adults and adolescents, 80% of the correlation between TBV and IQ could be explained by genetic factors common to TBV and IQ (Pennington et al., 2000). However, roughly two third of the 66 twin pairs in this sample consisted of twin pairs with reading disabilities (DeFries, 1985). This may make it hard to generalize the findings of this study to the normal population. And since brain structure is known to change during adolescence (Giedd et al., 1999) it is possible that some of these genetic associations could have been partly mediated by age-related changes.

Here we report a genetic study in a population-based sample of 112 nine-year-old healthy twin pairs (Peper et al., 2008; Peper et al., in press; Van Leeuwen, Van den Berg, & Boomsma, 2008). In this important period of cognitive development and (structural) brain maturation (Blakemore & Choudhury, 2006), we examine whether TBV, GMV, and WMV are associated with intelligence, verbal comprehension, perceptual organization and perceptual speed. If a significant association is found, we investigate whether genetic

and/or environmental factors mediate the association, and the direction of causation in this association.

Material and Methods

Participants

The group of children participating in intelligence testing consisted of 112 nine-year-old twin pairs (M = 9.1, SD = .10). There were 23 monozygotic male (MZM), 23 dizygotic male (DZM), 25 monozygotic female (MZF), 21 dizygotic female (DZF) and 20 dizygotic pairs of opposite sex (DOS). For the same sex twin pairs, zygosity determination was based on DNA polymorphisms (90 twin pairs), or on questionnaire items (2 pairs; Rietveld et al., 2000). From the 112 families, 107 (N=214) underwent magnetic resonance (MR) scanning. This group consisted of 22 MZM, 22 DZM, 23 MZF, 21 DZF and 19 DOS twin pairs.

Average time between intelligence testing and magnetic resonance imaging (MRI) was 43 days (with psychological testing before MRI; SD = 35). The study was approved by the Central Committee on Research involving Human Subjects (CCMO). Parents signed informed consent statements for the children as well as themselves. Children also signed consent. Parents were financially compensated for their travel expenses and children received two presents worth €10,-, one after a testing day (see also Van Leeuwen et al., 2008).

Measurements

One day children were tested at the VU University (VU) in Amsterdam and one day a MRI scan was made at University Medical Center in Utrecht (UMCU). At the VU all children underwent cognitive testing. After arriving between nine and eleven o'clock in the morning, children were individually tested in separate rooms by experienced test administrators. Children completed as part of a larger test battery the Wechsler Intelligence Scale for Children – III (WISC-III; Wechsler et al., 2002) and the Raven Standard Progressive Matrices (Raven, 1960). The whole protocol took approximately five hours, including two short breaks and one long lunch break.

Intelligence testing

IQ scales were assessed with the Dutch adaptation of the WISC-III (Wechsler et al., 2002). For the analysis we used the three dimensions described in the WISC-III guidelines: Verbal Comprehension (VC; information, similarities, vocabulary, and comprehension), Perceptual Organization (PO; block design, picture completion, picture arrangement, and object assembly), and Perceptual Speed (PS; digit-symbol substitution and symbol search). Children also completed the Raven Standard Progressive Matrices (SPM; Raven, 1960) at their own pace after verbal instruction. The test consists of 60 problems divided into five sets of twelve, which become progressively more difficult. The test provides an index of general intelligence. For children, retest reliability is .88 (Raven, 1960). Raven-SPM scores were estimated based on the Rasch model (Rasch, 1966). In the Rasch model, every person is represented by a person parameter θ that reflects that person's ability. Every test item is represented by a difficulty parameter β. The probability that a person *j* answers item *i* correctly is parameterized by the logistic function $p(Y_{ij} = 1) = \Psi(\theta_j - \beta_i)$, where θ_j is the person parameter, β_i is the difficulty parameter for that particular item, and $\Psi(x) = \exp(x) / (1 + \exp(x))$ (see also Van den Berg, Glas, & Boomsma, 2006). Thus, for example, the probability that person *j* with ability θ_j answers item *i* with difficulty level β_i correctly, equals $e^{\theta j-\beta i} / [1 + e^{\theta j-\beta i}]$. When $\theta_j - \beta_i = 0$, the probability of a correct answer is exactly 50%, as $e^0 = 1$. When ability dominates the difficulty, $\theta_j > \beta_i$, then the probability is higher than 50%, becoming 100% when ability is infinitely higher than the difficulty. When ability is lower than the difficulty of the item, $\theta_j < \beta_i$, then the probability of a correct answer is lower than 50%, becoming 0% when the ability is infinitely lower than the difficulty. Note that the values for θ and β, the ability of a person and the difficulty of an item, are on the same scale. Rasch scores were estimated using the Gibbs sampler as implemented in the BUGS software (http://www.mrc-bsu.cam.ac.uk/bugs) by taking the mean of each individual's posterior distribution. The estimation procedure used no assumptions regarding the distribution of the intelligence scores or item difficulties. Extreme scores (like no item correct or all items correct) are inestimable in the Rasch model. Therefore, individuals who had extreme scores were assigned a value half a logit higher than the second highest scoring individuals (for further details see Van Leeuwen et al., 2008).

MR image acquisition and processing

A three-dimensional T1-weighted coronal spoiled-gradient echo scan of the whole head (256 × 256 matrix, TE = 4.6 ms, TR = 30 ms, flip angle = 30^{o}, 160–180 contiguous slices; 1 × 1 × 1.2 mm^{3} voxels, Field-of-View = 256 mm / 70%) was acquired. Furthermore, a single-shot EPI (Echo Planar Imaging) scan was made as part of a diffusion tensor imaging (DTI)-series (SENSE factor 2.5; flip angle 90^{o}; 60 transverse slices of 2.5 mm; no gap; 128 x 96 acquisition matrix; FOV 240 mm; TE=78 ms) together with a magnetization transfer imaging (MTI) scan (60 transverse slices of 2.5 mm; no gap;128 x 96 acquisition matrix; FOV 240 mm; flip angle 8^{o}; TE=4.5 ms; TR=37.5 ms), which were used for segmentation of the intracranial volume (see Peper et al., 2008) for details on MR acquisition and processing).

The scans were coded to ensure blindness for subject and zygosity identification. The T1-weighted images were automatically put into Talairach orientation (Talairach & Tournoux, 1988) without scaling, by registering them to a model brain in Talairach orientation. The translation and rotation parameters of this registration were then applied to the images (Maes, Collignon, Vandermeulen, Marchal, & Suetens, 1997). After linear registration to the T1-weighted image, the intracranial segment served as a mask for all further segmentation steps. The T1-weighted images were corrected for field inhomogeneities using the N3 algorithm (Sled, Zijdenbos, & Evans, 1998). Our automatic image processing pipeline was used for segmentation of total brain volume, and gray and white matter of the cerebrum. The software included histogram analysis, mathematical morphology operations, and anatomical knowledge based rules to connect all voxels of interest, as was validated before (Schnack et al., 2001). The intracranial and total brain segments were all visually checked and edited if necessary. Ten brains from the cohort were randomly selected and analyzed by two independent raters to estimate inter-rater reliability. Intra-class Correlation Coefficients (ICC) were above 0.97.

Four individuals (coming from 4 DZ pairs) dropped out during the MR scanning, leading to a total number of 210 children who successfully completed the protocol. Due to motion artifacts, separation of gray and white matter tissue was not possible in 14 subjects (6 MZ, 8 DZ). These subjects were included in the analyses of total brain volume. One outlier was excluded (DZ) since he had extremely large ventricles. Consequently, the total

number of individuals included in total gray and white matter analyses was 195 (84 MZ, 111 DZ), whereas for total brain volume the number of subjects was 209 (90 MZ, 119 DZ).

Statistical Analyses

All data analyses were performed using the software package Mx (Neale, Boker, Xie, & Maes, 2006). First, general covariance matrices, means and sex regressions on the means were estimated in a saturated model. Means and covariance matrices for the *phenotypic* (observed) variation in the seven measures (TBV, GMV, WMV, IQ, VC, PO and PS) were estimated separately for MZ twins and DZ twins. In addition, the saturated model supplied within family covariance matrices for MZ twin pairs and DZ twin pairs. Standardization of these covariance matrices gives respectively MZ and DZ correlations. Since a large number of parameters was estimated, this model yielded a good description of the data.

By fitting nested models in which the means and variances between MZ and DZ twins were equated, several assumptions were tested such as equality of means between MZ and DZ twins. These models were also used to test for sex effects on the means. We continued equating parameters until the most parsimonious model with still acceptable fit was established. The choice for the best fitting model was based on likelihood-ratio tests. The difference between minus twice the log likelihoods (*-2LL*) of two nested models, asymptotically follows a χ^2 distribution. The degrees of freedom are given by the difference in the number of parameters estimated in the two nested models. A high increase in χ^2 against a low gain of degrees of freedom denotes a worse fit of the sub model compared to the full model. All data were analyzed, including data from incomplete twin pairs using the raw data option in Mx.

Genetic Modeling

Univariate analysis

In the classic twin design MZ and DZ twin correlations contain the information on the relative influence of genetic and environmental factors on the variability in traits. When MZ twin correlations are higher than DZ twin correlations, part of the twin resemblance in the phenotype is caused by genetic factors (comprising of *additive effects* of alleles at one or more loci (A) and *non-additive effects* of alleles (D)). When DZ twin correlations are more than half the size of MZ correlations, the resemblance between twins is at least partly

caused by shared environmental factors (C; common environmental factors shared between siblings brought up in the same family). Differences between MZ twins reflect the importance of unique environment (E). Large sample sizes are required to have sufficient power to detect D or C (Boomsma, Busjahn, & Peltonen, 2002; Plomin, DeFries, McClearn, & McGuffin, 2001).

The phenotype for an individual can be represented as:

$$P_{ij} = a*A_{ij} + c*C_{ij} + d*D_{ij} + e*E_{ij},$$

where i = 1,2, …or 112 (families) and j = 1 or 2 (twin 1 and twin 2) and A, C, D and E are latent variables (factors) standardized to have unit variance. The variance in P due to A, C and E is given by the square of *a, c* and *e*, respectively, so that Var (P) = $a^2 + c^2 + d^2 + e^2$. The observed variance in a population thus is attributed to variance caused by genes and variance caused by environment. Note that e^2 also contains variance due to measurement error. MZ twins have the same DNA sequence and therefore genetic factors are perfectly correlated in MZ twins. DZ twins share on average half of their segregating genes, so that the expected correlation between their additive genetic factors (A) is ½. The genetic correlation between the dominance deviations (D) is ¼. By definition the correlation between common environmental factor (C) is one, and between unique environmental factors (E) is zero. Therefore the covariance within MZ twin pairs is: Cov (MZ) = $a^2 + c^2 + d^2$, and within DZ twin pairs: Cov (DZ) = ½ a^2 + ¼ d^2 + c^2. When only data from twins reared together are available, it is only possible to estimate *c* under the assumption that *d* is zero or any other specified value, and vice versa, since a model including free parameters for both *c* and *d* is not identified.

Multivariate analysis

To determine to what extent the covariation between the seven measures was due to genetic and environmental effects, multivariate genetic factor analysis was applied. In a multivariate analysis the *cross twin - cross trait correlations* (e.g. the correlation between GMV in twin 1 and VO in twin 2) for MZ and DZ twins and siblings contain information on the etiology of the association between traits. If MZ cross correlations are larger than the DZ cross correlations, this indicates genetic factors play a role in the covariation between the two traits.

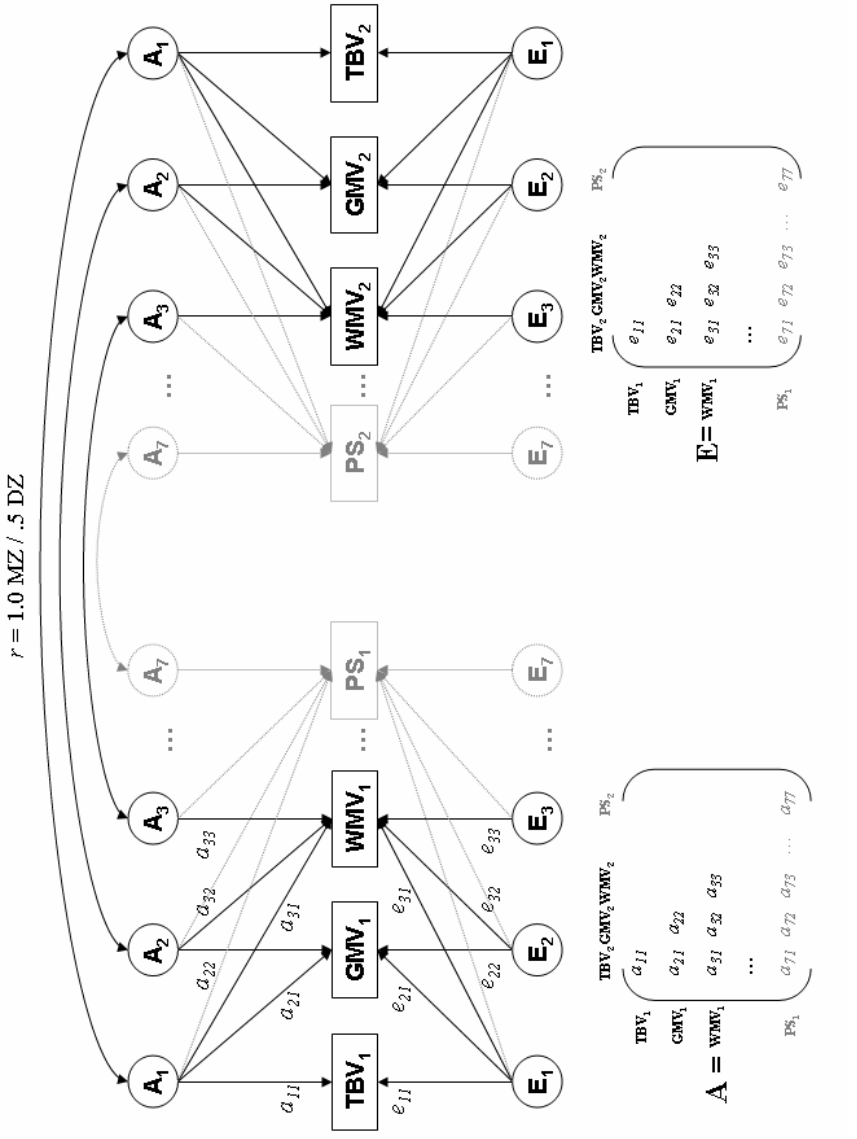


Figure 6.1. Path model and matrices of the seven variate saturated AE model, with on the left side phenotypes measured in twin 1 and on the right side phenotypes in twin 2. For clarity reasons only total brain volume (TBV), white matter volume (WMV), gray matter volume (GMV), and processing speed (PS) are included (A = genetic factor, E = environmental factor, *a* = genetic factor loading, e = environmental factor loading)

Based on the sample size and on inspection of the MZ and DZ correlations estimated in the saturated models a genetic model in which the relative contributions of A end E were estimated was fitted to the data. Figure 6.1 represents the seven variate

saturated AE model as applied in the multivariate genetic analyses. For clarity reasons only TBV, WMV, GMV, and PS are included in the figure. In a seven-variate saturated AE model the factor loadings of the A and E factors are modeled in lower triangular matrices of dimensions 7 × 7 (for seven variables: three brain measures and four intelligence measures), where matrix **A** contains the genetic factor loadings and **E** the unique environmental factor loadings. The model is than represented as follows:

$$\mathbf{p}_{ij} = \mathbf{A} \times \mathbf{a}_{ij} + \mathbf{E} \times \mathbf{e}_{ij}$$

where i = 1,2, …or 112 (families) and j = 1 and 2 (twin 1 and twin 2), vector **p** denotes the 7 phenotypes and has the dimension 7 × 1. Vectors $\mathbf{a}_{ij}$ and $\mathbf{e}_{ij}$ have the dimensions 7 × 1 and contain the genetic and environmental factors. The random factors are standardized to have unit variance. The variance in **p** due to **a** and **e** is than given by:

$$\mathbf{V}_P = \mathbf{A} \times \mathbf{A}' + \mathbf{E} \times \mathbf{E}'$$

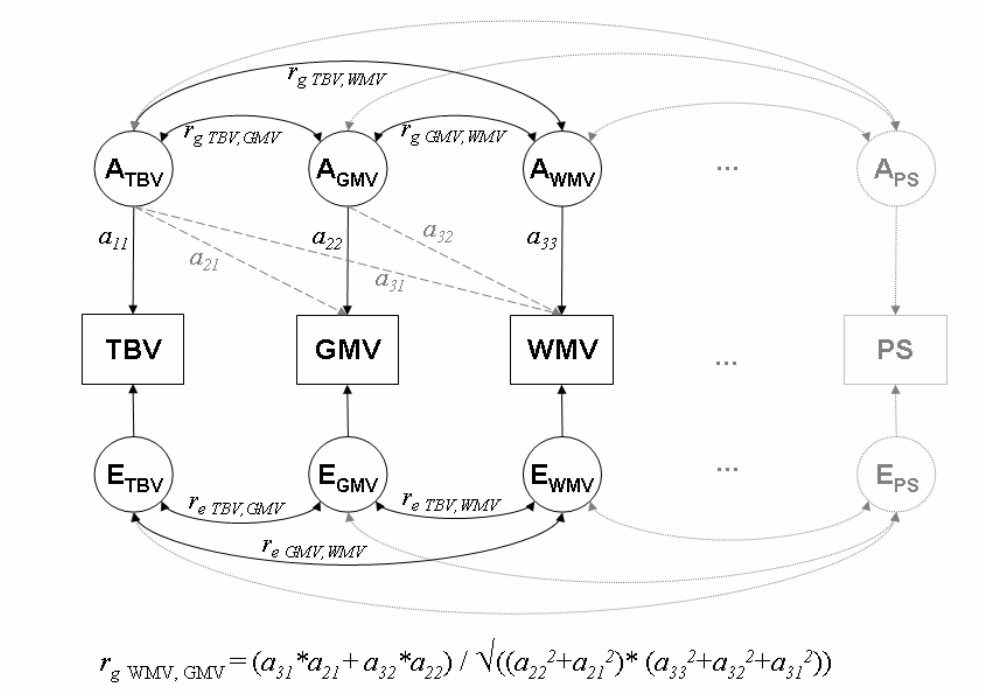
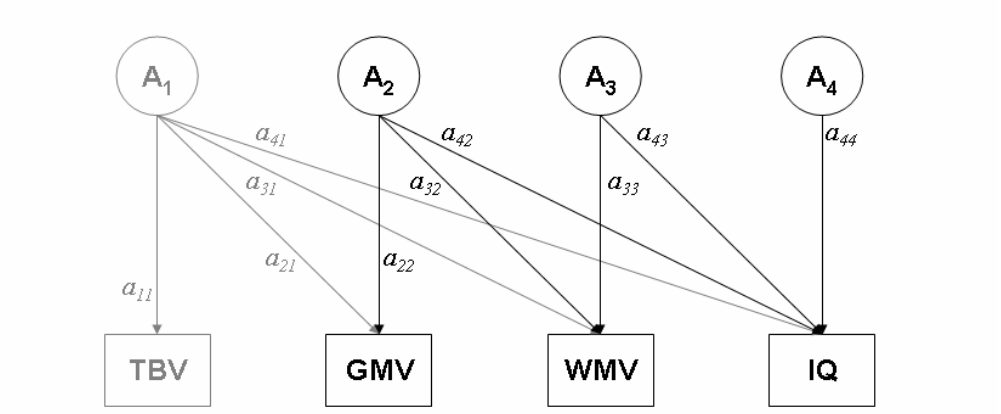


Figure 6.2. Genetic correlation (rg) between WMV and GMV. (TBV = total brain volume, PS = procesing speed, A = genetic factor, E = environmental factor, re = environmental correlation)

where matrix $\mathbf{V_P}$ is a symmetric matrix of 7 × 7, **A** and **E** are lower triangular matrices of 7 × 7, and ' indicates transposition. This seven-variate AE model is a completely saturated model; and was used to test whether variation in genes contributed significantly to the variability in brain volume and intelligence by assessing the deterioration in model fit after the A factors were dropped from the model.

The extent to which genetic factors on one trait correlate with the genetic factors on another trait is expressed in the genetic correlation (r_g; see also Figure 6.2). The genetic correlation matrix is equal to the standardized genetic covariance matrix (**A** × **A**'). The size of the genetic correlations is independent of the influence of the genetic variance on the traits. Therefore, this correlation still can be high in case there is hardly any genetic variance. The same applies for the matrix of the environmental correlations (see Boomsma & Molenaar, 1986; Martin & Eaves, 1977; Neale & Cardon, 1992).



$$r_{g\ WMV,IQ} = (a_{41}*a_{31} + a_{42}*a_{32} + a_{43}*a_{33}) / \sqrt{((a_{33}^2 + a_{32}^2 + a_{31}^2)* (a_{44}^2 + a_{43}^2 + a_{42}^2 + a_{41}^2))}$$

$$r_{g\ WMV,IQ-TB} = (a_{42}*a_{32} + a_{43}*a_{33}) / \sqrt{((a_{33}^2 + a_{32}^2)* (a_{44}^2 + a_{43}^2 + a_{42}^2))}$$

Figure 6.3. The genetic correlation between white matter volume (WMV) and IQ uncorrected and corrected for total brain volume (TBV; GMV = gray matter volume, A = genetic factor, *a* = genetic factor loading)

To determine whether differences in intelligence were driven by proportion WMV and GMV, we corrected in subsequent analyses the phenotypic (using partial correlation) and genetic correlations between GMV and WMV and the intelligence measures for TBV (Lenroot et al., 2007). The corrected genetic correlation was derived by estimating the

covariances after correction for genetic effects of TBV. This is illustrated in Figure 6.3, where the correlation between WMV and IQ serves as an example. Because GMV and WMV corrected for TBV are in fact the proportions GMV and WMV (i.e. when one proportion increases, the other decreases), the correlations between corrected GMV and WMV and intelligence have opposite signs.

Table 6.1. *Maximum likelihood estimates of means for girls and boys, and SD of the seven variables*

Variable	*N*	Mean girls	Mean boys	*SD*
TBV	209	1291	1421	117
GMV	195	705	780	65
WMV	195	429	470	47
IQ	223	-1.30*	-1.30*	1.07
VC	224	100	100	15
PO	224	100	100	12
PS	223	105	95	14

Note. *based on Rasch score, the mean of total number correct items is 36.70. TBV = total brain volume, GMV= gray matter volume of the cerebrum, WMV = white matter volume of the cerebrum, VC = verbal comprehension, PO = perceptual organization, PS = processing speed

Results

Maximum likelihood estimates of means and standard deviation are presented in Table 6.1. There were significant effects of sex on the means of TBV, GMV, WMV and PS; girls had smaller TBV, GMV, and WMV, and performed better on PS.

Table 6.2. *Phenotypic correlations between the seven variables corrected for sex.*

Variable	TBV	GMV	WMV	IQ	VC	PO
GMV	.89*					
WMV	.85*	.54*				
IQ	.20*	.22*	.13			
VC	.33*	.27*	.32*	.53*		
PO	.28*	.25*	.24*	.52*	.52*	
PS	.12	.06	.14	.16*	.12	.28*

Note. TBV = total brain volume, GMV= gray matter volume of the cerebrum, WMV = white matter volume of the cerebrum, VC = verbal comprehension, PO = perceptual organization, PS = processing speed, *significant $\alpha = .05$

Table 6.2 presents the phenotypic correlations between the seven variables. Most phenotypic correlations were significant, except for the correlation between WMV and IQ, between the three brain volumes and PS and between PS and VC. Phenotypic correlations amongst the brain measures ranged from .5 to .9, amongst intelligence measures from .1 to .5, and between the brain and intelligence measures from .1 to .3.

Table 6.3. *MZ and DZ correlations*

Variable	TBV	GMV	WMV	IQ	VC	PO	PS
TBV	.94 / .47	0.43	.38	.10	.30	.18	.01
GMV	.83	.84 / .49	.29	.18	.24	.17	.00
WMV	.81	.62	.82 / .40	.00	.31	.13	-.01
IQ	.21	.24	.12	.61 / .33	.40	.30	.16
VC	.33	.28	.29	.51	.78 / .64	.36	.11
PO	.25	.21	.23	.41	.37	.60 / .22	.11
PS	.13	.04	.17	.15	.12	.22	.62 / .21

Note. On the diagonal on the left side the MZ correlations and on the right the DZ correlations, below the diagonal MZ cross correlations and above the diagonal DZ cross correlations. TBV = total brain volume, GMV= gray matter volume of the cerebrum, WMV = white matter volume of the cerebrum, VC = verbal comprehension, PO = perceptual organization, PS = processing speed

Table 6.3 displays the correlations in MZ twins (first figure on diagonal), and in DZ twins (second figure on diagonal). For all measures MZ correlations were higher than DZ correlations, indicating genetic influence on the variance of the seven traits. For the majority of the twin cross-correlations (off-diagonal of Table 6.3), the MZ correlations are larger than the DZ correlations, suggesting that the covariance between the measures is influenced by genetic factors. Table 6.4 presents the estimates of heritability (proportion of variance explained by genetic factors), and the proportions of variance explained by D or C and E, based on univariate genetic analyses of the 7 phenotypes. Based on these results, we decided to limit the multivariate model analyses on an AE model.

The first impression that genetic factors influence the variance and covariance between the seven phenotypes was confirmed in the seven-variate analysis: dropping the A component in the saturated AE model led to a significant deterioration of fit ($\Delta\chi^2$ = 388.547, Δdf = 28, p = .00). Therefore it can be concluded that additive genetic factors

Table 6.4. *Univariate analyses: variance component estimates*

Variable	a^2	d^2	c^2	e^2
TBV	.94 (.62-.96)	-	.00 (.00-32)	.06 (.04-.09)
GMV	.77 (.40-.90)	-	.07 (.00-.43)	.15 (.09-.25)
WMV	.84 (.50-.90)	-	.00 (.00-.32)	.16 (.10-.27)
Raven	.50 (.00-.73)	-	.10 (.00-.52)	.40 (.27-.58)
VC	.28 (.00-.63)	-	.51 (.17-.74)	.21(.14-.33)
PO	.25 (.00-.72)	.35 (.00-.73)	-	.40 (.27-.58)
PS	.26 (.00-.72)	.34 (.00-.74)	-	.40 (.26-.62)

Note. TBV = total brain volume, GMV= gray matter volume of the cerebrum, WMV = white matter volume of the cerebrum, VC = verbal comprehension, PO = perceptual organization, PS = processing speed

Table 6.5. *Unstandardised estimates of the genetic and environmental (co)variances that contribute to the variance and covariances in and between the seven phenotypes.*

Variable	TBV	GMV	WMV	IQ	VC	PO	PS
TBV	60.39 / 3.58	3.94	4.51	.02	.60	2.63	-.16
GMV	56.52	61.19 / 10.95	-8.2	-3.61	.01	3.89	1.34
WMV	89.38	71.95	158.04 / 32.44	5.81	3.09	1.28	-3.61
IQ	16.11	22.32	11.33	64.53 / 48.11	2.27	14.97	-1.33
VC	35.81	30.04	58.34	79.61	175.36 / 47.18	27.99	.23
PO	25.17	21.91	39.79	52.32	65.64	87.21 / 68.39	7.43
PS	13.69	6.75	29.16	22.97	21.32	38.93	100.11 / 71.53

Note. On the diagonal on the left the genetic variances, and on the right the environmental variances. Below the diagonal the genetic covariances and above the environmental covariances. TBV = total brain volume, GMV= gray matter volume of the cerebrum, WMV = white matter volume of the cerebrum, VC = verbal comprehension, PO = perceptual organization, PS = processing speed

contribute significantly to the variance and covariance in the three brain measures and the four intelligence measures.

The unstandardised genetic and environmental (co)variances which contribute to the phenotypic variance in and covariances between the three brain measures and the four intelligence measures are presented in Table 6.5. On the diagonal on the left the genetic variances, and on the right the environmental variances are presented. Genetic variances are larger than environmental variances, indicating that variance in genotype is more important than variance in environment to explain differences between children in brain volume and intelligence. On the lower off-diagonal the genetic covariances and on the upper off-diagonal the environmental covariances are displayed. The genetic covariances are larger than environmental covariances. Thus, the relationship between brain volumes and amongst brain and intelligence measures is mainly explained by genetic factors. In Table 6.6 the heritabilities (percentage of total variation explained by genetic variation) of the seven traits are presented. The traits were moderately to highly heritable. The heritabilities of the brain

measures and IQ will not be thoroughly discussed, since they are discussed elsewhere (Peper et al., in press, Van Leeuwen et al., 2008).

Table 6.6. *Heritabilities of the seven variables.*

Variable	heritability
TBV	.94 (.91-.96)
GMV	.85 (.76-.90)
WMV	.83 (.73-.89)
IQ	.57 (.40-.70)
VC	.79 (.69-.86)
PO	.56 (37-.70)
PS	.58 (.37-.73)

Note. Between brackets 95% confidence intervals. TBV = total brain volume, GMV= gray matter volume of the cerebrum, WMV = white matter volume of the cerebrum, VC = verbal comprehension, PO = perceptual organization, PS = processing speed

Table 6.7 shows the genetic and environmental correlations, below and above the diagonal, respectively. Amongst brain measures the genetic as well as the environmental correlations are significant, showing that correlations between genetic and environmental factors both contribute to the phenotypic correlations amongst the three brain measures. The same applies for the phenotypic correlations between PO and *g*, and PO and VC; common genetic as well environmental factors contribute to the phenotypic correlations between these intelligence measures. In contrast, the phenotypic correlations between brain and intelligence measures and among intelligence measures are explained by correlations between genetic factors only.

Table 6.7. *Genetic and environmental correlations between the seven variables.*

Variable	TBV	GMV	WMV	IQ	VC	PO	PS
TBV		**.63 (.42-.78)**	**.42 (.15-.63)**	.00 (-.28-.28)	.05 (-.22-.31)	.17 (-.14-.44)	-.01 (-.30-.29)
GMV	**.93 (.89-.96)**		**-.44 (-64--.17)**	-0.16 (-.40-.11)	.00 (-.28-.28)	.14 (-.15-.41)	.05 (-.24-.32)
WMV	**.92 (.86-.96)**	**.73 (.59-.87)**		.15 (-.13-.40)	.08 (-.21-.35)	.03 (-.26-.31)	-.07 (-.34-21)
IQ	**.26 (.05-.45)**	**.36 (.13-.56)**	.11 (-.12-.34)		.05 (-.20-.29)	**.26 (.02-.47)**	-.02 (-.27-.23)
VC	**.35 (.18-.50)**	**.29 (.10-.46)**	**.35 (.16-.52)**	**.75 (.58-.90)**		**.49 (.26-.67)**	.00 (-.26-.26)
PO	**.35 (.14-.54)**	**.30 (.07-.52)**	**.34 (.11-.56)**	**.70 (.49-.87)**	**.53 (.34-.69)**		.11 (-.17-.37)
PS	.18 (-.04-.38)	.09 (-.15-.31)	.23 (-.01-.45)	**.29 (.00-.57)**	.16 (-.07-.39)	**.42 (.12-.71)**	

Note. Genetic and environmental correlations are presented below and above the diagonal respectively. In bold significant correlations and heritabilities. TBV = total brain volume, GMV= gray matter volume of the cerebrum, WMV = white matter volume of the cerebrum, VC = verbal comprehension, PO = perceptual organization, PS = processing speed

The phenotypic and genetic correlations between WMV and GMV and the intelligence measures corrected for TBV are shown in Table 6.8. The table shows that when corrected for TBV the correlation between GMV and WMV and the intelligence measures disappears. However, these genetic correlations should be interpreted with caution, since there are no phenotypic correlations. As follows from Table 6.7 and 6.8 a large part of the genetic correlation between WMV and GM, and VC and PO was explained by genetic factors WMV and GMV have in common with TB. For example, the genetic correlation between WMV and VC is .35 (see Table 6.7). This genetic correlation is partly mediated by genes which have WMV and VC in common with TBV. In Table 6.8 the genetic factor which is common to TBV, WMV and VC is removed (see factor A_1, Figure 3), which lowers the correlation between WMV and VC to .08. In the case of the relation between proportion GMV/WMV and Raven, this relation appears also to be explained by genetic factors specific for the proportion GMV/WMV. A higher proportion GMV is related to better performance on the Raven, and this relation seems to be mediated by genes.

Table 6.8. *Phenotypic and genetic correlations between gray matter volume (GMV) and white matter volume (WMV) and intelligence measures corrected for total brain volume.*

	Phenotypic correlation		Genotypic correlation	
Variable	GMV	WMV	GMV	WMV
IQ	.09	-.08	.32	-.32
VC	-.06	.08	-.10	.08
PO	.00	.00	-.07	.06
PS	-.10	.07	-.21	.18

Note. VC = verbal comprehension, PO = perceptual organization, PS = processing speed

Discussion

We analyzed in 9-year old children the relation between brain volume and intelligence. We showed that there is a significant association among measures of brain volume as assessed by sMRI and cognitive traits and also showed that the relation among brain volumes and intelligence measures is entirely explained by a set of genes common to both sets of variables. Correlations between the different measures of brain volume and intelligence ranged between .20 and .33 (the non-significant correlation between IQ and WMV excluded). Processing speed and the brain volumes did not correlate. There was no indication that IQ, PO and VC correlated differently to each of the brain volumes.

Several features distinguish this study from the other studies on the relation between brain volume and intelligence done in children. First of all, all children were the same age, and therefore the reported relation between brain volumes and intelligence was relatively unaffected by age-related changes in brain structure demonstrated earlier (Giedd et al., 1999; Lenroot et al., 2007; Paus et al., 1999; Sowell, Trauner, Gamst, & Jernigan, 2002). Moreover, this is the largest study until now on the relation between brain volumes and intelligence in a group of children that is representative for the general population.

The correlation between TBV and VC and PO ranged between .28 and .33 replicating previous research on the relation between intelligence and TBV (McDaniel, 2005). We also reproduced the relation between GMV and intelligence (Andreasen et al., 1993; Frangou et al., 2004; Posthuma et al., 2002; Reiss et al., 1996; Thoma et al., 2005). In

contrast to Wilke et al. (2003) we found a correlation between GMV and TBV and IQ in nine-year-olds. Possibly this can be explained by smaller sample size and wider age range in the study of Wilke et al. (1 year in the subgroups vs. 1 month in our sample).

We also reported partial correlations, which indicated that intelligence was not related to proportion WMV/GMV. The association between intelligence and WMV and GMV disappears once corrected for TBV (consisting of WMV, GMV, cerebellum volume and stem volume), since TBV and WMV and GMV are highly correlated. Nevertheless, we can not merely conclude from the partial correlation analyses, that intelligence is only influenced by TBV and that WMV and GMV separately do not influence intelligence.

Our study is in agreement with previous studies in children showing that variance in IQ test performance is for 25 to 70% accounted for by genetic variation between individuals (Bartels, Rietveld, Van Baal, & Boomsma, 2002; Hoekstra, Bartels, & Boomsma, 2007; Jacobs et al., 2001; Plomin, 2003; Rietveld, Dolan, Van Baal, & Boomsma, 2003; Turkheimer, Haley, Waldron, D'Onofrio, & Gottesman, 2003). Genetic factors entirely explain the significant phenotypic correlations between the three brain volumes and IQ, PO and PS. This finding is in concordance with the findings by Posthuma et al. (2002) in adults. Possibly, these genetic factors come into play already early in development. Gale, O'Callaghan, Bredow, and Martyn (2006) and Gale, O'Callaghan, Godfrey, Law, and Martyn (2004) showed –measuring head circumference- that growth in brain volume during infancy predicts intelligence in eight- and nine-years-olds, while brain size at birth and brain growth later in life is not associated with intelligence in both these age groups. After infancy children could not compensate for poor brain growth earlier in life. This shows that the relation between brain volume and intelligence already is established between birth and one year of age.

Genetic and environmental correlations can give an indication for the direction of causation for the association between intelligence and brain volume (De Moor, Boomsma, Stubbe, Willemsen, & De Geus, 2008). If intelligence causally influences brain volume, all genetic and environmental factors that influence intelligence will also, through the causal chain, influence brain volume. Under the causal hypothesis both genetic and environmental correlations should be significant, whereas a significant genetic correlation in the absence of an environmental correlation falsifies the hypothesized causal effect of intelligence. However, when traits are highly heritable (in the range of 90-100%), as is the case in brain

volumes, causality (brain volume causes intelligence) can not be distinguished from pleiotropy (the same set of genes affects brain volume as well as intelligence).

The heritability estimates for the brain volumes are around 90%. In contrast, variability in intelligence is for about 60% caused by differences in genotypes. If intelligence causally influences brain volumes, this would also be reflected in the genetic and environmental correlations: all genetic and environmental factors that influence intelligence would, through the causal chain, influence brain volume. However, our study shows that only the genetic correlations are significant. In fact 85% to 100% of the covariation between brain volume and intelligence are caused by shared genetic factors. This leaves two options: 1) the relation between brain volume and intelligence is caused by a set of genes which influences variation in brain volume and this variation in turn leads to variation in intelligence 2) pleiotropy: there is a set of genes which influence brain volume as well as intelligence.

Future studies should aim to dissect the pathways relating genes, brain and intelligence (Plomin & Kosslyn, 2001), using genome wide association (Kruglyak, 2008), gene expression (DeRisi, Iyer, & Brown, 1997; Tang, 2006), proteomics and metabolomics approaches (Petrella, Mattay, & Doraiswamy, 2008). We can speculate about the nature of the genes involved in the association between intelligence and brain volume. As mentioned before the association between brain volume and intelligence is established between birth and one year of age. Moreover, genes which influence brain volume, influence intelligence via a causal pathway or pleiotropy. Therefore, genes responsible for myelination and the proliferation and organization of synapses could possibly explain the relation between intelligence and brain volume, since both these processes predominantly occur before birth until early childhood (Lenroot & Giedd, 2006).

Based on linkage studies probable candidate genes for the association between brain volume and intelligence are the genes for prion protein (PrP; Rujescu, Hartmann, Gonnermann, Moller, & Giegling, 2003), brain-derived neurotrophic factor (BDNF; Miyajima et al., 2007; Savitz, Solms, & Ramesar, 2006; Tsai, Hong, Yu, & Chen, 2004) and the synaptosomal associated protein of 25 kD (SNAP-25; Gosso et al., 2006; Gosso et al., 2007): A mutation in the PrP gene has been implied in white matter reduction and a decline in intelligence (Rujescu et al., 2003). BDNF exerts amongst others long-term effects on neuronal survival, migration, and dendritic and axonal growth (Pang & Lu, 2004)

and intelligence (Miyajima et al., 2007; Tsai et al., 2004). Finally, SNAP-25 is amongst others implicated in axonal growth and IQ, and most strongly performal IQ (Gosso et al., 2006; Gosso et al., 2007).

Limitations of the current research and directions for future research

One limitation of this study is that, in spite of the large sample size, we did not have sufficient power to test for sex differences in the relation between brain volumes and intelligence. The study of Lenroot et al. (2007) showed different trajectories of brain development between boys and girls for TBV, GMV, and WMV. We applied a linear correction for sex differences in brain volume and PS, which seems plausible since we did not measure a developmental curve. And declines in TBV and GMV only start after age nine in boys as well as girls (Lenroot et al., 2007). However, to test whether genetic and / or environmental factors have different effects in boys than in girls or some factors have an effect on one sex and not on the other, one should evaluate scalar and non-scalar sex limitation models in sufficiently large samples and take into account potential problems with fitting these models (see Neale, Roysamb, & Jacobson (2006).

We only looked at gross brain volumes. A next step should be to investigate whether specific brain areas contribute to the relationship between brain volumes and intelligence. Voxel-based morphometry (VBM) analyses in adolescents and young adults showed positive correlations between IQ and gray matter density in the orbifrontal cortex and cingulated gyrus, the cerebellum, and thalamus and negative correlations in caudate nucleus; areas known to be involved in executive control (Frangou et al., 2004). In an adult sample Haier, Jung, Yeo, Head, and Alkire (2004) found a positive correlation between intelligence and gray matter density within all four lobes of the cerebrum (i.e. frontal (Brodmann areas (BA) 10, 46, 9), temporal (BA 21, 37, 22, 42), parietal (BA 43 and 3) and occipital (BA 19) lobe), and with white matter density in the right parietal area near BA 39. In adults, (Hulshoff Pol et al., 2006) showed that the phenotypic correlation (up to .35) between intelligence and white matter of the superior occipitofrontal, callosal, and left optical radiation and gray matter of the frontal and occipital lobes and the parahippocampal gyrus could be explained by a common set of genes. Future studies should investigate if in children the same brain areas contribute to the relation between intelligence and brain structure and if the relation between these areas and intelligence stems from common

genetic and / or environmental factors. Moreover, it would be interesting to follow the developmental trajectories between intelligence and brain structure, as Shaw et al. (2006) showed that more intelligent children follow a different developmental trajectory than less intelligent children. We intend to follow the children who were tested at age 9 years in the upcoming years, so as to be able to track these trajectories and elucidate the mechanisms underlying these trajectories.

To summarize, at 9 years of age, variation in brain structure is associated with individual differences in intelligence measures. This relation is entirely explained by genetic factors common to both sets of traits. The genes which influence brain volume, probably influence intelligence via a causal pathway or via pleiotropy. A causal chain from genes, to IQ to brain size is less likely.

Reference List

Andreasen, N C, Flaum, M, Swayze, V, O'Leary, D S, Alliger, R, Cohen, G, Ehrhardt, J, & Yuh, W T (1993). Intelligence and brain structure in normal individuals. *American Journal of Psychiatry, 150*, 130-134.

Bartels, M, Rietveld, M J H, Van Baal, G C M, & Boomsma, D I (2002). Genetic and Environmental Influences on the Development of Intelligence. *Behavior Genetics, 32*, 237-249.

Blakemore, S J & Choudhury, S (2006). Brain development during puberty: state of the science. *Developmental Science, 9*, 11-14.

Boomsma, D, Busjahn, A, & Peltonen, L (2002). Classical twin studies and beyond. *Nature Reviews Genetics, 3*, 872-882.

Boomsma, D I & Molenaar, P C (1986). Using LISREL to analyze genetic and environmental covariance structure. *Behavior Genetics, 16*, 237-250.

Carmelli, D, Swan, G E, DeCarli, C, & Reed, T (2002). Quantitative genetic modeling of regional brain volumes and cognitive performance in older male twins. *Biological Psychology, 61*, 139-155.

De Moor, M H M, Boomsma, D I, Stubbe, J H, Willemsen, G, & De Geus, E J C (2008). Testing causality in the association between regular exercise and symptoms of anxiety and depression. *Archives of General Psychiatry, 65,* 897-905

Deary, I J, Spinath, F M, & Bates, T C (2006). Genetics of intelligence. *European Journal of Human Genetics, 14*, 690-700.

DeFries, J.C. (1985). Colorado Reading Project. In: D.B. Gray & J.F. Kavanaugh *Biobehavioral measures of dyslexia* (pp. 123-135) Parkton, MD: York Press

DeRisi, J L, Iyer, V R, & Brown, P O (1997). Exploring the metabolic and genetic control of gene expression on a genomic scale. *Science, 278*, 680-686.

Egan, V, Chiswick, A, Santosh, C, Naidu, K, Rimmington, J E, & Best, J J K (1994). Size isn't everything: A study of brain volume, intelligence and auditory evoked potentials. *Personality and Individual Differences, 17*, 357-367.

Fancher, R E (1983). Biographical origins of Francis Galton's psychology. *Isis, 74*, 227-233.

Frangou, S, Chitins, X, & Williams, S C R (2004). Mapping IQ and gray matter density in healthy young people. *Neuroimage, 23*, 800-805.

Gale, C R, O'Callaghan, F J, Bredow, M, & Martyn, C N (2006). The influence of head growth in fetal life, infancy, and childhood on intelligence at the ages of 4 and 8 years. *Pediatrics, 118*, 1486-1492.

Gale, C R, O'Callaghan, F J, Godfrey, K M, Law, C M, & Martyn, C N (2004). Critical periods of brain growth and cognitive function in children. *Brain, 127*, 321-329.

Galton, F. (1869) *Hereditary Genius. An inquiry into its laws and consequences.* London: Macmillan and co.

Giedd, J N, Blumenthal, J, Jeffries, N O, Castellanos, F X, Liu, H, Zijdenbos, A, Paus, T, Evans, A C, & Rapoport, J L (1999). Brain development during childhood and adolescence: a longitudinal MRI study. *Nature Neuroscience, 2*, 861-863.

Gosso, M F, De Geus, E J, Polderman, T J, Boomsma, D I, Heutink, P, & Posthuma, D (2007). Common variants underlying cognitive ability: further evidence for association between the SNAP-25 gene and cognition using a family-based study in two independent Dutch cohorts. *Genes Brain and Behavior,*

Gosso, M F, De Geus, E J, van Belzen, M J, Polderman, T J, Heutink, P, Boomsma, D I, & Posthuma, D (2006). The SNAP-25 gene is associated with cognitive ability: evidence from a family-based study in two independent Dutch cohorts. *Molecular Psychiatry, 11*, 878-886.

Haier, R J, Jung, R E, Yeo, R A, Head, K, & Alkire, M T (2004). Structural brain variation and general intelligence. *Neuroimage, 23*, 425-433.

Hoekstra, R A, Bartels, M, & Boomsma, D I (2007). Longitudinal genetic study of verbal and nonverbal IQ from early childhood to young adulthood. *Learning and Individual Differences, 17*, 97-114.

Hulshoff Pol, H E, Schnack, H G, Posthuma, D, Mandl, R C, Baare, W F, Van Oel, C, Van Haren, N E, Collins, D L, Evans, A C, Amunts, K, Burgel, U, Zilles, K, De Geus, E, Boomsma, D I, & Kahn, R S (2006). Genetic contributions to human brain morphology and intelligence. *Journal of Neuroscience, 26*, 10235-10242.

Jacobs, N, Van Gestel, S, Derom, C, Thiery, E, Vernon, P, Derom, R, & Vlietinck, R (2001). Heritability estimates of intelligence in twins: effect of chorion type. *Behavior Genetics, 31*, 209-217.

Kruglyak, L (2008). The road to genome-wide association studies. *Nature Reviews Genetics, 9*, 314-318.

Lenroot, R K, Gogtay, N, Greenstein, D K, Wells, E M, Wallace, G L, Clasen, L S, Blumenthal, J D, Lerch, J, Zijdenbos, A P, Evans, A C, Thompson, P M, & Giedd, J N (2007). Sexual dimorphism of brain developmental trajectories during childhood and adolescence. *Neuroimage, 36*, 1065-1073.

Lenroot, R K & Giedd, J N (2006). Brain development in children and adolescents: Insights from anatomical magnetic resonance imaging. *Neuroscience & Biobehavioral Reviews, 30*, 718-729.

Maes, F, Collignon, A, Vandermeulen, D, Marchal, G, & Suetens, P (1997). Multimodality image registration by maximization of mutual information. *IEEE Transactions on Medical Imaging, 16*, 187-198.

Martin, N G & Eaves, L J (1977). The genetical analysis of covariance structure. *Heredity, 38*, 79-95.

McDaniel, M A (2005). Big-brained people are smarter: A meta-analysis of the relationship between in vivo brain volume and intelligence. *Intelligence, 33*, 337-346.

Miller, E M (1994). Intelligence and brain myelination: A hypothesis. *Personality and Individual Differences, 17*, 803-832.

Miyajima, F, Ollier, W, Mayes, A, Jackson, A, Thacker, N, Rabbitt, P, Pendleton, N, Horan, M, & Payton, A (2007). Brain-derived neurotrophic factor polymorphism Val66Met influences cognitive abilities in the elderly. *Genes Brain and Behavior,*

Neale, M.C. & Cardon, L.R. (1992) *Methodology for Genetic Studies of Twins and Families.* Dordrecht, The Netherlands: Kluwer Academic

Neale, M C, Roysamb, E, & Jacobson, K (2006). Multivariate genetic analysis of sex limitation and G x E interaction. *Twin Research and Human Genetics, 9*, 481-489.

Neale, M., Boker, S.M., Xie, G., & Maes, H.H. (2006) *Mx: statistical modelling.* Richmond, VA: Department of Psychiatry, Medical College of Virginia

Pang, P T & Lu, B (2004). Regulation of late-phase LTP and long-term memory in normal and aging hippocampus: role of secreted proteins tPA and BDNF. *Ageing Research Reviews, 3*, 407-430.

Paus, T, Zijdenbos, A, Worsley, K, Collins, D L, Blumenthal, J, Giedd, J N, Rapoport, J L, & Evans, A C (1999). Structural maturation of neural pathways in children and adolescents: in vivo study. *Science, 283*, 1908-1911.

Pennington, B F, Filipek, P A, Lefly, D, Chhabildas, N, Kennedy, D N, Simon, J H, Filley, C M, Galaburda, A, & DeFries, J C (2000). A twin MRI study of size variations in human brain. *Journal of Cognitive Neuroscience, 12*, 223-232.

Peper, J S, Brouwer, R M, Boomsma, D I, Kahn, R S, & Hulshoff Pol, H E (2007). Genetic influences on human brain structure: a review of brain imaging studies in twins. *Human Brain Mapping, 28*, 464-473.

Peper, J S, Brouwer, R M, Schnack, H G, Van Baal, G C M, Van Leeuwen, M, Van den Berg, S M, Delemarre-van de Waal, H A, Janke, A L, Collins, D L, Evans, A C, Boomsma, D I, Kahn, R S, & Hulshoff Pol, H E (2008). Cerebral white matter in early puberty is associated with luteinizing hormone concentrations. *Psychoneuroendocrinology. 33,* 909-15

Peper, J S, Schnack, H G, Brouwer, R M, Van Baal, G C M, Pjetri, E, Székely, E, Van Leeuwen, M, Van den Berg, S M, Collins, D L, Evans, A C, Boomsma, D I, Kahn, R S, & Hulshoff Pol, H E (in press). Heritability of regional and global brain volume at the onset of puberty: a Magnetic Resonance Imaging study in 9-year old twin-pairs. *Human Brain Mapping.*

Petrella, J R, Mattay, V S, & Doraiswamy, P M (2008). Imaging genetics of brain longevity and mental wellness: the next frontier? *Radiology, 246*, 20-32.

Plomin, R (2003). Genetics, genes, genomics and g. *Molecular Psychiatry, 8*, 1-5.

Plomin, R & Kosslyn, S M (2001). Genes, brain and cognition. *Nature Neuroscience, 4*, 1153-1154.

Bayne J. and Byrd M.L. Plomin, R., DeFries, J.C., McClearn, G.E., & McGuffin, P. (2001) *Behavioral genetics.* New York: Worth Publisher

Posthuma, D, Baare, W F, Hulshoff Pol, H E, Kahn, R S, Boomsma, D I, & De Geus, E J (2003). Genetic correlations between brain volumes and the WAIS-III dimensions of verbal comprehension, working memory, perceptual organization, and processing speed. *Twin Research, 6*, 131-139.

Posthuma, D, De Geus, E J, Baare, W F, Hulshoff Pol, H E, Kahn, R S, & Boomsma, D I (2002). The association between brain volume and intelligence is of genetic origin. *Nature Neuroscience, 5*, 83-84.

Posthuma, D., De Geus, E.J.C., & Boomsma, D.I. (2002). Genetic contributions to anatomical , behavioral, and neurophysiological indices of cognition. In: R. Plomin, J.C. DeFries, I.W. Craig, & P. McGuffin *Behavioral Genetics in the Post Genomic Era* (pp. 141-161) Washington D.C.: American Psychological Association

Pujol, J, Lopez-Sala, A, Sebastian-Galles, N, Deus, J, Cardoner, N, Soriano-Mas, C, Moreno, A, & Sans, A (2004). Delayed myelination in children with developmental delay detected by volumetric MRI. *Neuroimage, 22*, 897-903.

Rasch, G (1966). An item analysis which takes individual differences into account. *British Journal of Mathematical and Statistical Psychology, 19*, 49-57.

Raven, J.C. (1960) *Guide to the Standard Progressive Matrices.* London: H.K. Lewis & Co. Ltd.

Reiss, A L, Abrams, M T, Singer, H S, Ross, J L, & Denckla, M B (1996). Brain development, gender and IQ in children. A volumetric imaging study. *Brain, 119 (Pt 5)*, 1763-1774.

Rietveld, M J, Van der Valk, J C, Bongers, I L, Stroet, T M, Slagboom, P E, & Boomsma, D I (2000). Zygosity diagnosis in young twins by parental report. *Twin Research, 3*, 134-141.

Rietveld, M J H, Dolan, C V, Van Baal, G C M, & Boomsma, D I (2003). A twin study of differentiation of cognitive abilities in childhood . *Behavior Genetics, 33*, 367-381.

Rujescu, D, Hartmann, A M, Gonnermann, C, Moller, H J, & Giegling, I (2003). M129V variation in the prion protein may influence cognitive performance. *Molecular Psychiatry, 8*, 937-941.

Savitz, J, Solms, M, & Ramesar, R (2006). The molecular genetics of cognition: dopamine, COMT and BDNF. *Genes, Brain, and Behavior, 5*, 311-328.

Schnack, H G, Hulshoff Pol, H E, Baare, W F, Staal, W G, Viergever, M A, & Kahn, R S (2001). Automated separation of gray and white matter from MR images of the human brain. *Neuroimage, 13*, 230-237.

Schnack, H G, Hulshoff, H E, Baare, W F, Viergever, M A, & Kahn, R S (2001). Automatic segmentation of the ventricular system from MR images of the human brain. *Neuroimage, 14*, 95-104.

Schoenemann, P T, Budinger, T F, Sarich, V M, & Wang, W S (2000). Brain size does not predict general cognitive ability within families. *Proceedings of the National Academy of Sciences, 97*, 4932-4937.

Shaw, P, Greenstein, D, Lerch, J, Clasen, L, Lenroot, R, Gogtay, N, Evans, A, Rapoport, J, & Giedd, J (2006). Intellectual ability and cortical development in children and adolescents. *Nature, 440*, 676-679.

Sled, J G, Zijdenbos, A P, & Evans, A C (1998). A nonparametric method for automatic correction of intensity nonuniformity in MRI data. *IEEE Transactions on Medical Imaging, 17*, 87-97.

Sowell, E R, Trauner, D A, Gamst, A, & Jernigan, T L (2002). Development of cortical and subcortical brain structures in childhood and adolescence: a structural MRI study. *Developmental Medicine and Child Neurology, 44*, 4-16.

Talairach, J. & Tournoux, P. (1988) *Co-planar stereotaxic atlas of the human brain: 3-Dimensional proportional system: an approach to cerebral imaging.* New York: Thieme

Tang, B L (2006). Molecular genetic determinants of human brain size. *Biochemical and Biophysical Research Communications, 345*, 911-916.

Thoma, R J, Yeo, R A, Gangestad, S W, Halgren, E, Sanchez, N M, & Lewine, J D (2005). Cortical volume and developmental instability are independent predictors of general intellectual ability. *Intelligence, 33*, 27-38.

Thompson, P M, Cannon, T D, Narr, K L, Van Erp, T, Poutanen, V P, Huttunen, M, Lonnqvist, J, Standertskjold-Nordenstam, C G, Kaprio, J, Khaledy, M, Dail, R, Zoumalan, C I, & Toga, A W (2001). Genetic influences on brain structure. *Nature Neuroscience, 4*, 1253-1258.

Tramo, M J, Loftus, W C, Stukel, T A, Green, R L, Weaver, J B, & Gazzaniga, M S (1998). Brain size, head size, and intelligence quotient in monozygotic twins. *Neurology, 50*, 1246-1252.

Tredoux, G (2007). Sir Francis Galton F.R.S. (1822-1911). *http://galton.org/,*

Tsai, S J, Hong, C J, Yu, Y W, & Chen, T J (2004). Association study of a brain-derived neurotrophic factor (BDNF) Val66Met polymorphism and personality trait and intelligence in healthy young females. *Neuropsychobiology, 49*, 13-16.

Turkheimer, E, Haley, A, Waldron, M, D'Onofrio, B, & Gottesman, I I (2003). Socioeconomic status modifies heritability of iq in young children. *Psychological Science, 14*, 623-628.

Van den Berg, S M, Glas, C A W, & Boomsma, D I (2006). Variance decomposition using an IRT measurement model.

Van Leeuwen, M, Van den Berg, S M, & Boomsma, D (2008). A twin-family study of general IQ. *Learning and Individual Differences, 18*, 76-88.

Wechsler, D., Kort, W., Compaan, E.L., Bleichrodt, N., Resing, W.C.M., Schittekatte, M., Bosmans, M., Vermeir, G., & Verhaeghe, P. (2002) *Handleiding WISC-III^{Nl} [Manual WISC-III^{Nl}]*. London: The Psychological Corporation Limited, Nederlands Instituut van Psychologen Dienstencentrum

Wickett, J C, Vernon, P A, & Lee, D H (2000). Relationships between factors of intelligence and brain volume. *Personality and Individual Differences, 29*, 1095-1122.

Wilke, M, Sohn, J H, Byars, A W, & Holland, S K (2003). Bright spots: correlations of gray matter volume with IQ in a normal pediatric population. *Neuroimage, 20*, 202-215.

Witelson, S F, Beresh, H, & Kigar, D L (2006). Intelligence and brain size in 100 postmortem brains: sex, lateralization and age factors. *Brain, 129*, 386-398.

7

Effects of twin separation in primary school

We studied the short- and long-term effects of classroom separation in twins on behavior problems and academic performance. Short-term effects were studied at age 7 in twins separated at age 5 and long-term effects at age 12 in twins who had been separated or together most of the time at school. Behavior problems were rated by mothers (Child Behavior Checklist at ages 3, 7 and 12) and teachers (Teacher Report Form at ages 7 and 12). Academic achievement was measured at age 12 using a national academic achievement test (CITO).

At age 7, twins from separated pairs had more internalizing and externalizing problems than non-separated twins, as rated by both mothers and teachers. Only for the maternal ratings of internalizing problems, however, could these effects be attributed to the separation itself and not pre-existing problems (at age 3) between separated and non-separated twins. Long-term effects of separation were significant for maternal and teacher ratings of internalizing and externalizing problems, but these effects could be explained by pre-existing differences between separated and non-separated groups. There were no differences in academic achievement between the separated and non-separated group. These results suggest that the decision to separate twins when they go to school is based in part on the existing behavioral problems of the twins at preschool and that, in the long run, separation does not affect problem behavior or academic achievement. The findings were the same for monozygotic and dizygotic twins.

This chapter is published as:
Van Leeuwen, M., Van den Berg, S. M., Van Beijsterveldt, T. C., & Boomsma, D. I. (2005). Effects of twin separation in primary school. Twin Research and Human Genetics, 8, 384-391.

In the Netherlands, the majority of children go to primary school at the age of 4 (Statistics Netherlands, Centraal Bureau voor de Statistiek, n.d.) and this is when parents and teachers of twins decide whether or not to put the children into the same classroom. As there is hardly any research comparing the adjustment of twins who are separated versus those kept together at school, this decision is presently not evidence-based (Hay, 2004). The Dutch Society for Parents of Multiples (Nederlandse Vereniging voor Ouders van Meerlingen; NVOM) advises parents to base their decision of whether or not to separate twins on what they think is best for their children, though generally NVOM believes separation to be best for the individualization of the twins (Geluk & Hol, 2001).

Because of the importance of this question for parents of multiples, Tully et al. (2004) investigated the effects of classroom separation on twins' behavior, progress at school, and reading abilities. They studied a sample of 878 same-sex twin pairs from the United Kingdom (UK). The children were first assessed at the age of 5 years and were tested again approximately 18 months later. The assessment was done by the teacher and included externalizing and internalizing problems, prosocial behavior, Attention Deficit Hyperactivity Disorder symptoms, standard reading scores, how hard twins worked and how much they learned. The sample was divided into three groups: not separated (at both assessments twins were together in the same class, 552 pairs), separated early, (twins were separated at age five and were still separated at the second assessment 18 months later, 162 pairs), and separated late (twins were together at the first assessment but had been separated by the second assessment 18 months later, 164 pairs). When compared with non-separated pairs, twins who were separated early had significantly more internalizing problems and twins separated later showed more internalizing problems and lower reading scores. Monozygotic (MZ) twins suffered more from separation than dizygotic (DZ) twins. Tully and colleagues did not find any effects on the other variables.

It is not known whether these UK findings generalize to other countries and cultures where the grounds for separation may differ. The Tully et al. (2004) results were based on teacher ratings of behavior at school. Behavior in the home situation, as rated by the parents, was not studied. The effects of separation on behavior were analyzed at ages 5 and 7, but no data were available at later ages. We carried out a replication study in a large sample of Dutch twins from the Netherlands Twin Registry (NTR), looking at behavior ratings from mothers and teachers and at a national test of academic achievement (CITO).

As mother as well as teacher ratings are used in this study, information on the behavior of the twins at school as well as at home was collected. Looking at behavioral problems in the home as well as in the school situation may give a better understanding of the association between problem behavior and separation at school. Furthermore, we have information on problem behavior at the age of 3 years; problem behavior at an early age may be one of the reasons for parents and educators to separate the twins. We also could control for any differences in the twins' behavior before separation by including maternal ratings at age 3 into the analyses. We distinguished between the short- and long-term effects of separation on both maternal and teacher ratings. Short-term effects were defined as effects showing up at the age of 7 years as the result of separation at age 5, and long-term effect as effects showing up at the age of 12 years as the result of separation for the entire schooling up until that age. We concentrated on internalizing and externalizing problems. In addition, we looked at academic performance at age 12 using the CITO, a national test of educational achievement administered in the last grade of primary school in order to determine high school entrance level (Bartels et al., 2002). This study thus addresses the following questions:

- Are there pre-existing differences between twins who attend separate classes and twins who are in the same class when they enter primary school?
- Are there any short-term effects of separation on maternal ratings of problem behavior at age 7, when controlling for pre-existing differences?
- Are there any short-term effects of separation on teacher ratings at age 7?
- Are there any pre-existing differences between twins who are in the same classes and those who are in different classes for their entire schooling?
- Is there an effect of separation for the entire schooling on maternal ratings at age 12, when controlled for pre-existing differences?
- Is there an effect of separation at school on teacher ratings at age 12?
- Is there an effect of separation at school on academic performance at age 12?

Like Tully et al. (2004), whether MZ and DZ twins differed in the way they reacted to separation was examined.

Method

Sample

All subjects were registered with the Netherlands Twin Registry (NTR), established by the Department of Biological Psychology at the Vrije Universiteit in Amsterdam. Around 40% to 50% of all multiple births in the Netherlands are registered by the NTR (Boomsma, 1998; Boomsma et al., 1992; Boomsma et al., 2002). Data of twins from the 1986 – 1996 birth cohorts were used in this study. Surveys have been collected longitudinally at the ages of 1, 2, 3, 5, 7, 10 and 12 years. For this study, information from surveys collected at the ages of 3 and 5 years (completed by mothers), and 7 and 12 years (completed by mothers and teachers) was used.

Surveys sent out at the ages of 3, 7, 10 and 12 years contained the Child Behavior Checklist (CBCL; Achenbach, 1991a), to be filled out by both parents. All questionnaires were mailed within 3 months of the twins' birthday. Reminders were sent 2 to 3 months after the mailing and, if finances permitted, persistent non-responders were contacted by phone 4 months after the initial mailing. Families for whom the addresses were no longer available were included in the non-response group. Response rates at ages 3, 7, 10 and 12 years were 72%, 66%, 64%, and 64% respectively (Note that if a family did not participate at a particular age, they were approached again for the next mailing. So a response rate of 66% at age 7 means that 66% of all the registered families with a twin pair that reached this particular age returned the questionnaire). Teacher ratings were assessed using the Teacher Report Form (TRF; Achenbach, 1991b) and collected at ages 7, 10 and 12 years. After the parents' consent, the TRF was sent to the teachers of the twins. Response rates were 78%, 77%, and 75% at age 7, 10 and 12 years respectively. The NTR only started collecting TRF data in 1999 so that TRF data collected at age 7 are not available for the 1986 – 1992 cohort.

The short- and long-term effects of separation were studied in two overlapping samples. For studying the short-term effect, questionnaires completed for twins of ages 3 to 7 years were available for 7595 twin pairs. Twin pairs were excluded when one or both twins had a disease or handicap at age 7 or younger that interfered severely with daily functioning (N = 263 pairs). Data from 594 pairs, of whom at least one twin received special education, were also excluded. So for short-term effects on maternal CBCL ratings,

data from 6738 twin pairs were used for analysis. Short-term effects of separation on TRF ratings were studied in 5686 pairs.

For studying the long-term effects, data from 2359 twin pairs were available from which another 175 pairs were excluded as one or both children were attending special education, resulting in a sample of 2184 twin pairs. The long-term effects on TRF ratings were studied in 284 twin pairs. Academic achievement was measured in 843 twin pairs.

Zygosity was determined by DNA or blood group polymorphisms for 859 twin pairs. For the remaining same-sex pairs, zygosity was determined from questionnaire items (Rietveld et al., 2000).

Measures

Data on socioeconomic status (SES) from the survey mailed out when the twins were 3 years old, were analyzed to address the question as to whether classroom separation is associated with SES. SES was based on a full description of the occupation of the parents and classified using a 5-point scale, according to the system used by Statistics Netherlands (Fengler et al., 1997). The higher of the two parents' SES scores determined the SES of the twin pair.

Externalizing and internalizing problems were assessed with the two broad band scales of the CBCL/ 4-18 (Achenbach, 1991a; Verhulst et al., 1996) at the ages of 7 and 12 years, and the TRF (Achenbach, 1991b; Verhulst et al., 1997) at the ages of 7 and 12 years. The CBCL and TRF were developed for parents and teachers to score the behavioral and emotional problems of 4- to 18-year-old children. They consist of 120 and 118 items respectively, scored on a 3-point scale based on the occurrence of the behavior during the preceding 6 months. The internalizing scale consists of the Anxious/Depressed, Somatic Complaints and Withdrawn subscales. The Externalizing scale consists of the Aggressive and Rule Breaking Behavior subscales.

At the age of 3 years, the CBCL/ 2-3 (Achenbach, 1992; Koot et al., 1997) was used. The CBCL/ 2-3 is modeled on the CBCL/4-18 and consists of 99 items. The internalizing scale consists of the Anxious and the Withdrawn/ Depressed subscales, and the externalizing scale consists of the Aggression, Oppositional and Overactive subscale.

Educational achievement was assessed by the Dutch CITO-elementary test. The CITO consists of 240 multiple-choice items assessing four different intellectual skills: Language, Mathematics, Information Processing, and World Orientation. Each performance scale contains 60 multiple-choice questions. In 2001, the test was changed slightly with respect to the distribution of questions, resulting in 60 questions for Mathematics and World Orientation, 90 questions for Language, and 30 questions for Information Processing. Together, the performance scales result in a standardized score of between 501 and 550 (Bartels et al., 2002). In the surveys sent to the parents and teachers when the twins were 12 years of age, parents as well as teachers were asked to fill in this standardized score.

The questionnaires sent to the parents of twins at ages 5 and 12 years contained questions on whether the twins were in the same class. In the Netherlands, most children start primary school at the age of 4 years; compulsory education, however, starts at the age of 5 years. Nearly all children attend primary school for 8 years and go to secondary school at the age of 12 years. The separation of twin pairs can occur when children first start school or during primary school. When the twins were 5 years old, the parents were asked whether "the twins are now a) together in the same school in the same classroom b) together in the same school but not in the same classroom and c) at different schools". The answers were coded as *together* (same school, same classroom) and *separated* (different or same school, different classroom).

Parents of twins who were 12 years of age were asked "which statement applies best to the school history of your twins a) same school, same classroom b) same school, parallel classes c) same school, different levels d) different schools e) partly same class, partly separated". Answers were coded as *together* (same school, same classroom), *separated* (parallel class or different level or different school) and *partly* (partly same class, partly separated; there is no information about when and how long these twins were separated). At the ages of 5, 7, 10 and 12 years, mothers were asked whether the twins were in a school for special education.

Data Analysis

First we explored whether the percentage of the twin pairs separated at age 5 differed as a function of birth cohort and what percentage of twin pairs separated or together at the age of 5 years stayed separated or together. Next, the following analyses were performed to test the short- and long-term effects of separation on problem behaviour and academic achievement:

To test whether separation at the age of 5 was associated with SES, internalizing and externalizing problems at the age of 3 years or within–twin pair differences in externalizing and internalizing problems at the age of 3 years, a logistic regression analysis was carried out. Separation at age 5 was the dependent variable and SES, internalizing and externalizing problems at age 3, and within–twin pair differences in externalizing and internalizing problems at age 3, were predictors.

To test the short-term effect of separation at the age of 5 years on internalizing and externalizing problems as rated by the mother, a MANOVA with repeated measures was carried out. The within-subject factor was age of testing (ages 3 and 7 years), the between-subject factor was separation of the twin pair at age 5 and the dependent variables were maternal CBCL internalizing and externalizing ratings at ages 3 and 7. We chose repeated-measures analysis to correct for any pre-existing differences between the separated and non-separated twins. A main effect of separation indicates that that there is an overall difference between children separated and children not separated. Such a difference may already exist before separation. Only when an interaction effect between the age of testing and separation is found, can the difference between separated and non-separated twins be attributed to the separation.

To test the effect of separation at age 5 on internalizing and externalizing problems at age 7 as observed by the teacher, a MANOVA was carried out. TRF internalizing and externalizing problems at age 7 were the dependent variables, and separation at age 5 the between factor.

A multinomial regression analysis tested whether SES, preschool behavioral problems, or within–twin pair differences in problem behavior were associated with separation for their entire schooling. In this analysis, the dependent variable was separation (together, separated or partly), with *together* as the reference group and SES, problem

behaviors at age 3, and within–twin pair differences in problem behavior as continuous predictors.

To test the long-term effect on problem behavior of going to school together or apart at the age of 12 years, a MANOVA with repeated measures was performed. The within-subject factor was the age of testing (3 years and 12 years) and the between-subject factor was separation for the entire schooling (together, separated, or partly). Dependent variables were maternal CBCL internalizing and externalizing problems at age 3 and 12.

To test the effect of separation for the entire school period on teacher-rated problem behavior, a MANOVA was performed with TRF internalizing and externalizing ratings at age 12 as dependent variables and separation (together, separated, or partly) as a between factor. Pair-wise comparisons were performed to see which of the three groups differed from each other.

To test the long-term effect of going to school together on academic achievement, an ANOVA was performed with the CITO scores as the dependent variable and separation as a between factor. Pair-wise comparisons were performed to see which of the three groups differed from each other. CITO data were corrected for SES differences.

If post hoc pair-wise comparisons were performed, Bonferroni correction for multiple testing was used.

As CBCL and TRF data were not normally distributed, scores were square-root transformed. After transformation, all skewness and kurtosis indices were between -1.0 and 1.0, implying that not much distortion is to be expected (Muthén & Kaplan, 1985). For MANOVA and ANOVA, CBCL, TRF and CITO data were corrected for SES for each child by taking at each age the difference between his/ her score and the average score in his/ her SES group, as these scores are associated with SES (Van Beijsterveldt et al., in press).

As twin data consist of non-independent observations, one child from each twin pair was selected randomly to be included in the study. Data from the non-selected twins were used in a replication in which the same pattern of results were found (for details, contact the first author). If an effect was found, all analyses were repeated with zygosity as an additional between factor to test if MZ and DZ twins react differently to separation.

Results

Most twins are in the same classroom at school; however in recent years there is an increase in the number of twins who attend separate classrooms. In 1988, 72% of the twin pairs at the age of 5 years were in the same classroom, but by 1998 this rate had dropped to 52%. The decision to separate twins seems to change during their schooling in 37% of the cases: of the 1006 twin pairs who were in the same classroom at the age of 5, 77% reported being together for (most of) the entire school period, 16% being separated, and 7% being partly separated and partly together, by the age of 12 years. Of the 500 twin pairs who went to separate classes, 64% reported being separated for (most of) their schooling, 26% reported being together and 9% reported being partly separated, partly together.

Classroom separation at the age of 5 was significantly associated with externalizing problems at age 3, $\chi^2(1) = 19.13$, $p < .01$ and with SES, $\chi^2(1) = 58.96$, $p < .01$. The higher the score on the externalizing scale at age 3, and the higher the SES, the more likely that twins were in separate classrooms at age 5 (see Table 7.1.). Internalizing problems at age 3, $\chi^2(1) = 1.50$, $p = .22$, within–twin pair differences in externalizing problems at age 3, $\chi^2(1) = 1.90$, $p = .17$, and within–twin pair differences in internalizing problems at age 3, $\chi^2(1) = .02$, $p = .89$, did not predict separation.

Table 7.1. *Number of twin pairs together or separated at age 5 as a function of SES*

SES	Together	Separated
1 (lowest)	35 (66)	18 (34)
2	721 (74)	255 (26)
3	1612 (68)	762 (32)
4	765 (61)	486 (39)
5 (highest)	271 (57)	203 (43)
total	3404 (66)	1724 (34)

Note. percentages in parentheses

Short-term effects

Table 7.2 shows average maternal CBCL ratings at ages 3 and 7 and the TRF ratings at age 7. Untransformed and uncorrected mean ratings are given for the separated and non-separated twins (at age 5). Additionally, ratings are given for MZ and DZ twins separately. MANOVA with repeated measures tested for differences in maternal ratings at the

internalizing and the externalizing scale at age 7 between separated and non-separated twins. Separated twins scored significantly higher on problem behavior than non-separated twins, $F(2, 4854) = 18.40$, $p < .01$. There was a significant interaction between age of testing and classroom separation ($F[2, 4854] = 7.53$, $p < .01$). Separated twins were more dissimilar from non-separated twins at age 7 than at age 3. This means that there is a difference between separated and non-separated twins that can not be explained by pre-existing differences at age 3. Univariate tests showed significant main effects of separation for internalizing, $F(1, 4855) = 18.53$, $p < .01$, and externalizing problems, $F(1, 4855) = 35.50$, $p < .01$. The interaction between age and separation was significant for the internalizing scale only: internalizing $F(1, 4855) = 14.77$, $p < .01$; externalizing $F(1, 4855) = 0.97$, $p = .33$, with an effect size of 0.14 *SD*. Thus, as a consequence of separation, separated twins at age 7 have more internalizing problems than non-separated twins.

An extra analysis was carried out to see whether DZ and MZ twins reacted differently to classroom separation. To take pre-existing differences in maternal ratings at age 3 into account, the interaction between age of testing, separation and zygosity was taken. Only when there is an interaction between age of testing, separation and zygosity it can be concluded that zygosity influences the way twins react to separation. No significant difference was found, $F(2, 4852) = 0.88$, $p = .42$. Thus, MZ and DZ twins do not react differently to separation.

The MANOVA carried out to see whether there are differences in teacher ratings at age 7 in separated and non-separated twins showed that separated twins were rated significantly higher by the teacher on problem behavior at age 7 than the non-separated twins ($F(2, 1495) = 3.09$, $p = .05$). Univariate analyses showed only a significant difference on internalizing problems (internalizing: $F[1, 1496] = 6.00$, $p = .01$; externalizing: $F[1, 1496] = 1.13$, $p = .29$). An extra analysis performed to test for MZ DZ differences showed no interaction effect between zygosity and separation ($F[2, 1730] = 0.45$, $p = .64$).

Table 7.2. *Separation at age 5 and mean untransformed and uncorrected maternal CBCL ratings at age 3 and 7 and TRF ratings at age 7*

measure	separated at age 5	Zygosity	*N*	*M (SD)*
Internalizing problems age 3 mother	separated	DZ	1060	4.41 (3.81)
		MZ	636	4.75 (3.78)
		Total	1696	4.54 (3.81)
	together	DZ	2125	4.37 (3.70)
		MZ	1192	4.59 (3.85)
		Total	3317	4.45 (3.76)
Externalizing problems age 3 mother	separated	DZ	1059	15.77 (9.49)
		MZ	636	17.18 (10.35)
		Total	1695	16.30 (9.84)
	together	DZ	2124	15.18 (9.62)
		MZ	1190	15.54 (9.90)
		Total	3314	15.31 (9.72)
Internalizing problems age 7 mother	separated	DZ	1190	4.82 (4.61)
		MZ	728	5.09 (4.49)
		Total	1918	4.92 (4.57)
	together	DZ	2417	4.52 (4.49)
		MZ	1361	4.17 (4.23)
		Total	3778	4.39 (4.40)
Externalizing problems age 7 mother	separated	DZ	1211	7.81 (6.90)
		MZ	740	8.53 (7.08)
		Total	1951	8.08 (6.97)
	together	DZ	2453	7.24 (6.47)
		MZ	1382	7.21 (6.57)
		Total	3835	7.23 (6.50)
Internalizing problems age 7 teacher	separated	DZ	413	5.08 (5.40)
		MZ	273	4.49 (4.99)
		Total	686	4.85 (5.25)
	together	DZ	685	4.59 (5.23)
		MZ	365	3.42 (4.14)
		Total	1050	4.18 (4.91)
Externalizing problems age 7 teacher	separated	DZ	432	4.80 (7.67)
		MZ	287	4.28 (6.68)
		Total	719	4.59 (7.29)
	together	DZ	706	4.37 (7.00)
		MZ	370	3.53 (5.92)
		Total	1076	4.08 (6.66)

Long-term effects

Multinominal regression analysis was carried out to test for pre-existing differences between separated and non-separated twins at school. Results showed that SES, problem behavior at age 3 and within–twin pair differences in internalizing problems at age 3 did not predict separation at school (SES: χ^2 (2)= 3.24, p = .20; internalizing: χ^2 (2) = .60, p = .74; externalizing: χ^2 (2) = 2.71, p = .26; within–twin pair difference internalizing: χ^2 (2) = 3.46, p = .18). Nevertheless, the data were corrected for SES ratings to maintain uniformity across analyses. Within–twin pair differences at age 3 in externalizing problems predicted separation at school, χ^2 (2) = 8.34, p = .02

The untransformed and uncorrected maternal and teacher ratings at age 12 are given in Table 7.3. Twins in the *partly* group scored highest on the maternal ratings, followed by *separated* and *together* twins consecutively. To test whether these differences could be explained by separation itself, a MANOVA with repeated measures was done. Results of this analysis revealed a significant effect of separation, F(4, 3294) = 5.92, p < .01, on maternal CBCL ratings. Subsequent univariate testing showed that the main effect was significant for the internalizing, F(2, 1647) = 10.11, p < .01, and externalizing, F(2, 1647) = 8.29, p < .01, scales. However, there was no significant interaction effect between age of testing and separation, F(4, 3294) = 1.17, p = .32, meaning that after controlling for pre-existing differences at age 3, the difference between separated and non-separated twins could not be attributed to separation itself.

On the TRF *separated* twins scored highest on the internalizing and externalizing scale, followed by the *together* and the *partly* group, respectively. A MANOVA performed to test whether these differences were significant revealed there was a main effect of separation at school on TRF ratings at age 12, F(4, 1646) = 4.25, p < .01. Univariate testing showed this effect was significant for the internalizing, F(2, 823) = 7.29, p < .01, and the externalizing scale, F(2, 823) = 9.84, p = .02. Post hoc pair-wise comparisons revealed for the internalizing scale as well as the externalizing scale that there was only a significant difference (p < .05) between the *together* group and the *separated* group. An extra analysis performed to test whether MZ and DZ twins reacted differently to separation showed no interaction between zygosity and separation, F(4, 1640) = 1.28, p = .28.

Table 7.3.: *Separation at school and mean untransformed and uncorrected maternal CBCL and TRF ratings at age 12*

measure	separated entire schooling	zygosity	*N*	*M (SD)*
		DZ	919	4.17 (4.76)
	together	MZ	362	3.90 (4.17)
		Total	1281	4.09 (4.60)
Internalizing		DZ	410	4.45 (4.43)
problems age 12	separated	MZ	202	4.97 (6.09)
mother		Total	612	4.62 (5.04)
		DZ	111	5.96 (6.05)
	partly	MZ	49	4.98 (4.72)
		Total	160	5.66 (5.68)
		DZ	926	5.60 (5.96)
	together	MZ	363	5.18 (5.70)
		Total	1289	5.49 (5.89)
Externalizing		DZ	420	6.35 (6.40)
problems age 12	separated	MZ	206	6.78 (7.58)
mother		Total	626	6.49 (6.81)
		DZ	111	6.09 (5.83)
	partly	MZ	49	6.96 (5.61)
		Total	160	6.36 (5.76)
		DZ	412	4.23 (5.11)
	together	MZ	181	3.26 (3.89)
		Total	593	3.93 (4.79)
Internalizing		DZ	170	5.11 (5.49)
problems age 12	separated	MZ	80	5.86 (6.68)
teacher		Total	250	5.35 (5.90)
		DZ	47	3.87 (5.42)
	partly	MZ	23	4.30 (6.980
		Total	70	4.01 (5.93)
		DZ	419	4.47 (7.850
	together	MZ	183	3.98 (6.03)
		Total	602	4.32 (7.34)
Externalizing		DZ	183	5.51 (7.76)
problems age 12	separated	MZ	92	6.03 (10.31)
teacher		Total	275	5.69 (8.68)
		DZ	49	3.92 (6.80)
	partly	MZ	27	4.26 (5.56)
		Total	76	4.04 (6.35)

The average score on the CITO was 538.4 (*SD* = 8.61). Twins in the *partly separated* group scored highest (*M* = 541.6, *SD* = 5.83), followed by the *together* (*M* = 538.1, *SD* = 8.69) and *separated* (*M* = 537.8, *SD* = 8.95) groups respectively. Separation had a significant effect on CITO scores, $F(2, 840) = 4.25$, $p = .02$. Post hoc pair wise comparisons showed that these differences were only significant between the *partly separated* and *together* group, and the *partly* and *separated* group of twins. Thus, there was no difference in academic performance between the separated and non-separated twins, but the partly separated twins scored higher on academic performance. Additional analyses performed to test whether MZ and DZ twins react differently to separation showed no interaction between CITO-ratings, zygosity and separation, $F(2, 837) = 0.07$, $p = .93$. Thus, MZ and DZ twins do not differ in academic performance as a consequence of separation.

Discussion

Like Tully and colleagues (2004), a difference at age 7 was found between separated and non-separated twins on the internalizing scale of the TRF. It was also found that twins who were separated and were in different classrooms at the age of 5 years generally scored higher on maternal ratings of internalizing and externalizing problems than non-separated twins. In addition, we found that twins separated for almost their entire schooling scored significantly higher on teacher and mother ratings of internalizing and externalizing problems than non-separated twins.

As twins had been rated on internalizing and externalizing problems by their mother when they were 3 years old, it was possible to look for pre-existing differences in behavior between separated and non-separated twins. Interestingly, differences in externalizing problems already existed before separation. Externalizing problems predicted separation at ages 5 and 12, and within–twin pair differences in externalizing problem behavior predicted separation for the entire schooling, but not separation at age 5. This suggests that the decision to separate twins when they go to school is based in part on their externalizing problems at a young age, but not on any internalizing problems at age 3.

When pre-existing differences in externalizing problems at age 3 were taken into account, separation of the twins had no significant effect on externalizing problems as rated by the mother at age 7. The significant differences between the separated and non-separated twins at age 7 on the externalizing scale already existed before separation at age 5 and

separation at age 5 had no additional effect on externalizing problems at age 7. A different result was obtained for internalizing problems: twins separated at age 5 had more internalizing problems at age 7 than non-separated twins, a result that could not be explained by pre-existing problems. For maternal ratings of problem behavior at age 12, both for internalizing and externalizing, separated twins did not score higher than non-separated twins after correction for these problems at age 3. So, the differences we found at age 12 between twins who were separated or non-separated for almost their entire schooling already existed at age 3. This indicates that separation for the entire schooling has no additional effect on problem behavior at the age of 12. Separation at an early age only seems to have a short-term effect on internalizing problems at age 7.

The finding of Tully and colleagues (2004) that MZ twins suffer more from separation than DZ twins was not replicated in this study. We did not find that MZ and DZ twins reacted to separation in a different way at either age 7 or 12. The study had a large sample, and it is unlikely that a lack of statistical power caused these results. One possible explanation is that the findings regarding the separation of twins in the UK do not generalize to Dutch settings, as the decision to separate twins in both countries is based on different grounds and has different consequences. This interpretation is supported by the observation that Tully and co-workers (2004) found no relationship between familial social class and separation, whereas a relationship was found in this study.

The effect of separation on behavioral, emotional problems and academic performance was studied. A limitation of this study is that the effect of separation on identity formation was not investigated. Identity formation is often given as the major justification to separate a twin pair, as this can be more problematic for twins who are often treated and judged as one of a pair and not as an individual (Akerman & Suurvee, 2003; Geluk & Hol, 2001). The children's own point of view was not taken into account. Twins may experience their separation as positive, as they no longer have to share attention with their co-twin.

Based on the findings of this study, it can be concluded that for behavioral problems at the age of 7 years, it does matter whether twins are separated or not. The separation of twins at school leads to internalizing problem behavior. However, it is important to note that all findings represent small effect sizes. And furthermore, at the age of 12 years, this effect has disappeared. When these last two points are taken into

consideration, it seems that it makes no difference whether twins are separated or not. The recommendation that the classroom separation of twins decision be based upon what parents think is best for their twins and for themselves, still seems sensible (Geluk & Hol, 2001).

Reference List

Achenbach, T. M. (1991a). Manual for the Child Behavior Checklist / 4-18 and 1991 Profile. Burlington, VT: University of Vermont, Department of Psychiatry.

Achenbach, T. M. (1991b). Manual for the Teacher's Report Form and 1991 Profile. Burlington, VT: University of Vermont, Department of Psychiatry.

Achenbach, T. M. (1992). Manual for the Child Behavior Checklist/2–3 and 1992 profile. Burlington, VT: University of Vermont, Department of Psychiatry.

Akerman, B. A., & Suurvee, E. (2003). The cognitive and identity development of twins at 16 years of age: A follow-up study of 32 twin pairs. *Twin Research, 6*, 328-333.

Bartels, M., Rietveld, M. J., Van Baal, G. C., & Boomsma, D. I. (2002). Heritability of educational achievement in 12-year-olds and the overlap with cognitive ability. *Twin Research, 5*, 544-553.

Boomsma, D. I. (1998). Twin registers in Europe: An overview. *Twin Research, 1*, 34-51.

Boomsma, D. I., Orlebeke, J. F., & Van Baal, G. C. (1992). The Dutch Twin Register: growth data on weight and height. *Behavior Genetics, 22*, 247-251.

Boomsma, D. I., Vink, J. M., van Beijsterveldt, T. C., de Geus, E. J., Beem, A. L., Mulder, E. J., Derks, E. M., Riese, H., Willemsen, G. A., Bartels, M., van den Berg, M., Kupper, N. H., Polderman, T. J., Posthuma, D., Rietveld, M. J., Stubbe, J. H., Knol, L. I., Stroet, T., & van Baal, G. C. (2002). Netherlands Twin Register: A focus on longitudinal research. *Twin Research, 5*, 401-406.

Fengler, M., Joung, I., & Mackenbach, J. (1997). Sociaal-demografische kenmerken en gezondheid: Hun relatief belang en onderlinge relaties [Social demographic characteristics and health: Their relative importance and mutual relations]. In J. P. Mackenbach & H. Verkleij (Eds.), Volksgezondheid Toekomst Verkenning 1997, II Gezondheidsverschillen (pp. 49-67). Maarssen, The Netherlands: Elsevier/De Tijdstroom.

Geluk, A., & Hol, J. (2001). Samen of Apart? Meerlingkinderen naar school, peuterspeelzaal of kinderopvang [Together or Apart? Multiples to school, playground or daycare]. NVOM [Dutch Society for Parents of Multiples] ref type: pamphlet

Hay, D. A. (2004). Together or apart? *Twin Research, 7(2)* iii-iv.

Koot, H. M., van den Oord, E. J., Verhulst, F. C., & Boomsma, D. I. (1997). Behavioral and emotional problems in young preschoolers: Cross-cultural testing of the validity of the Child Behavior Checklist/2-3. *Journal of Abnormal Child Psychology, 25*, 183-196.

Muthén, B., & Kaplan, D. (1985). A comparison of some methodologies for the factor analysis of non-normal Likert variables. *British Journal of Mathematical and Statistical Psychology, 38*, 171-189.

Rietveld, M. J., Der Valk, J. C., Bongers, I. L., Stroet, T. M., Slagboom, P. E., & Boomsma, D. I. (2000). Zygosity diagnosis in young twins by parental report. *Twin Research, 3*, 134-141.

Statistics Netherlands, Centraal Bureau voor de Statistiek. (n.d.). Statline Webselector. Retrieved November, 2004, from http://statline.cbs.nl/StatWeb

Tully, L. A., Moffitt, T. E., Caspi, A., Taylor, A., Kiernan, H., & Andreou, P. (2004). What effect does classroom separation have on twins' behavior, progress at school, and reading abilities? *Twin Research, 7*, 115-124.

Van Beijsterveldt, C. E. M., Hudziak, J. J., & Boomsma, D. I. (2005). Short- and Long-term Effects of Child Care on Problem Behaviors in a Dutch Sample of Twins. *Twin Research and Human Genetics, 8,* 250-258.

Verhulst, F. C., Van der Ende, J., & Koot, H. M. (1996). Handleiding voor de CBCL/4-18 [Manual for the CBCL/ 4-18]. Rotterdam: Department of Child and Adolescent Psychiatry, Sophia Children's Hospital / Academic Hospital Rotterdam/ Erasmus University Rotterdam.

Verhulst, F. C., Van der Ende, J., & Koot, H. M. (1997). Handleiding voor de teacher's report form (TRF) [Manual for the teacher's report form (TRF)]. Rotterdam: Department of Child and

Adolescent Psychiatry, Sophia Children's Hospital / Academic Hospital Rotterdam/
Erasmus University Rotterdam.

8

Summary and discussion

I have described the first part of a longitudinal study on cognition, brain structure, and hormonal levels during adolescence. Data were collected in nine-year-old twins and their nine-to-fourteen year old siblings who are registered with the Netherlands Twin Register (NTR). This thesis focuses on cognition and its relation to brain structure. This last chapter first summarizes the results from the previous chapters and then discusses these with reference to other publications that resulted from this study.

Chapter 2: Endophenotypes for intelligence in children.

In adults, a small set of endophenotypes for intelligence is already available. In children, however, much less is known about the suitability of these cognitive measures as endophenotypes for intelligence. Chapter 2 identified promising endophenotypes for intelligence in children and adolescents for future genetic studies in cognitive development. Based on the available set of endophenotypes for intelligence in adults, cognitive tasks were chosen covering the domains of working memory, processing speed, and selective attention. This set of tasks was assessed in a test-retest design and their correlation with intelligence was examined in children and in adolescents. The test battery included the *n*-back task, Eriksen flanker task, and the π-inspection time task.

All test-retest correlations in children exceeded .60, except for accuracy and stimulus congruency effects of the flanker task. For the adolescents the same holds true, with an exception of the 2-back (r = .16) and the π-inspection task (r = .58). In both children and adolescents *n*-back performance was significantly related to IQ. Better performance on the *n*-back task was related to higher IQ-scores. Reaction time on the congruent and incongruent trials of the flanker was significantly related to IQ in children only; the longer the reaction time, the lower the IQ. Incongruency effects on reaction time, accuracy on the congruent and incongruent trials, as well as incongruency effects on accuracy were not related to IQ in children or in adolescents. Inspection time was related to IQ in children, the shorter the inspection time the higher the IQ, but was not significantly related to IQ in adolescents.

Working memory capacity seems a good endophenotype for intelligence in children and adolescents: it can be reliably assessed using the *n*-back and it correlates with intelligence. Processing speed is not an optimal endophenotype for intelligence in children and adolescents. Once corrected for working memory, it contributes only a very small part

to the variance of intelligence. Selective attention, at least when measured as the flanker incongruency effect on reaction time and accuracy, is not a suitable endophenotype for either age group.

Table 8.1. *Heritability of cognitive measures*

Measure	*heritability*
IQ (Raven)[a]	.67
IQ (WISC-III)[a]	.75
Reading ability (OMRT)[a]	.83
Verbal IQ (WISC-III)[b]	.81
Verbal comprehension (WISC-III)[a]	.79
Perceptual organization (WISC-III)[a]	.56
Processing speed (WISC-III)[a]	.58
Verbal learning (AVLT)[b]:	.46
Learning speed[c]	.43
Forgetting speed[c]	.20*
Letter Fluency (COWA)[b]	.40
Category Fluency (COWA)[b]	.29
Verbal STM (DSF)[a]	.47
Visuospatial STM (Corsi)[a]	.47
Verbal WM (DSB)[a]	.35
Visuospatial WM (2-back)[a]	.47

Note *heritability in twins only, for siblings .30; [a]based on data-analysis in this thesis; [b]Hoekstra et al., in revision); [c]Van Soelen et al. (in revision)

Chapter 3: Genetic architecture of memory

Chapters 3 and 4 examined the heritability of various cognitive measures, including working memory which had proven in chapter 2 to be a reliable endophenotype in children and adolescents. Table 8.1 offers a summary of the heritabilities that were observed for these measures, as well as for psychometric IQ based on the analyses described in this

thesis and as published by others from the same study (Hoekstra, Bartels, Van Leeuwen, & Boomsma, submitted; Van Soelen et al., in revision). Heritability estimates ranged from .20 to .83, with the highest estimates seen for IQ, either total or verbal IQ, and for reading ability.

Chapter 3 examined the heritability of verbal and visuospatial working memory (WM) and short-term memory (STM) in a developmental study on the genetic and environmental relationship between these four measures. Although a wealth of studies on individual differences has focussed on the relation between STM and WM in adults (e.g. Conway, Cowan, Bunting, Therriault, & Minkoff, 2002; Kane et al., 2004) as well as children (e.g. Alloway, Gathercole, & Pickering, 2006; Bayliss, Jarrold, Baddeley, & Gunn, 2005; Kail & Hall, 2001), the extent to which verbal and visuospatial WM and STM tests measure the same or multiple constructs is still unclear. Likewise the relationship between WM and STM across development is not known. These questions were addressed studying the current cohort and a cohort of 186 families of young adult twins and siblings. Verbal and visuospatial WM and STM were measured using the Corsi block tapping task (Corsi, 1974), *n*-back task (Gevins & Cutillo, 1993), and the digit span forward and backwards task (Wechsler, 1997; Wechsler et al., 2002).

In the young adult cohort the relationship between the four measures was best captured by a model consisting of two correlated factors for verbal and visuospatial memory explaining all genetic variance, one common environmental factor for the visuospatial memory tasks, and one specific environmental factor for each variable. In the child cohort most of the phenotypic correlations were explained by a genetic factor for verbal and a genetic factor for visuospatial memory. However, the results in the children also indicated significant differences in the genetic structure of cognition in children as opposed to young adults: STM and WM in children were also influenced by specific genetic factors. Thus, from a genetic viewpoint one could say that WM and STM are part of the same system, and verbal and visuospatial information are processed using two partly overlapping memory pathways. Second, during the course of development the specific genetic factors, which create differences between the four abilities, disappear. This suggests that with aging these cognitive abilities start to become part of two genetic systems, one for verbal memory and one for visuospatial memory.

Chapter 4: Genetic architecture of reading ability

Chapter 4 investigated the genetic relationship between reading ability, intelligence and verbal and visuospatial WM and STM. The study used WISC IQ, performance on the One Minute Reading Test (OMRT; Cito, 1995) as a measure of reading ability, and measures of verbal and visuospatial WM and STM.

The relationship between reading ability and IQ has been well established in non-affected and in groups affected with reading disability (Tiu, Jr., Thompson, & Lewis, 2003). The association between reading disability and memory is still subject of debate (Cohen-Mimran & Sapir, 2007; Gathercole, Alloway, Willis, & Adams, 2006; Kercher & Sandoval, 1991; Swanson & Jerman, 2007) and literature on the genetic relationship between memory and reading abilities is scarce. Resolving the etiology of the relationship between IQ, memory and reading abilities, may give information whether impairments in memory and IQ in children with reading disability is a sign of the severity of the reading disability or a symptom of reading disability per se (Bishop, 2006).

The phenotypic correlations between reading ability and the other measures ranged between .24 and .44. The phenotypic correlations between IQ, WM, STM and reading ability were completely explained by common sets of genes. The model which explained these phenotypic correlations best consisted of: a common genetic factor for all variables; a common genetic factor for visuospatial STM and verbal and visuospatial WM; a common genetic factor for verbal memory and reading ability; a specific genetic factor for visuospatial WM; and a specific genetic factor for reading ability. Forty-seven percent of variation in additive genetic variance in reading ability was specific for reading ability.

Chapter 5: Intelligence

In Chapter 5 the presence of assortative mating (non-random mating), Gene-Environment (GE) interaction (people with a certain genotype are more vulnerable to a certain environment) and the heritability of intelligence in childhood was assessed using a twin family design with twins, their siblings and parents. With this design, cultural and genetic transmission can be studied while taking into account spousal resemblance. Two competing hypotheses about the causes of assortative mating in intelligence were evaluated: social homogamy (spouses meet each other within an environment which is correlated with intelligence) and phenotypic assortment (spouses choose each other based on intelligence or

a trait related to it), and their implications for the heritability estimate of intelligence. Intelligence was assessed using the Raven's Progressive Matrices (Raven, Raven, & Court, 1998; Raven, 1960) in both the parental and the offspring generation. IQ scores were estimated based on a Rasch model (Rasch, 1966).

The spousal correlation was .33, monozygotic (MZ) correlation was .63, and dizygotic (DZ) correlations, twin-sibling correlations and parent offspring correlations varied between .25 and .38. A simple model with only additive genetic effects and non-shared environmental effects explained the correlations between family members best. Comparing the phenotypic assortment model and the social homogamy model, the model assuming phenotypic assortment appeared superior. Thus spouses choose each other based on intelligence or on a related trait.

There was no significant contribution of cultural transmission and therefore no passive Genotype-Environment (GE) correlation. The study design of Chapter 5 was not suited to uncover GE correlations other than one resulting from simultaneous genetic and cultural transmission (i.e. reactive or active GE correlation). However, if there is GE correlation, it is more likely that either reactive or active GE correlation are of importance and that the role of parents is limited to responding to the needs and interests as indicated by the child. Such correlations, which are "part of the genotype of the child" are embedded in the heritability estimates in the current study.

To detect and estimate GE interaction the association between MZ intrapair sum and difference scores was examined (Jinks & Fulker, 1970). The estimate for the correlation between intelligence sum and difference between MZ twins was -.30. When there is a negative correlation between intrapair sum and absolute differences, less intelligent individuals are less similar than more intelligent individuals (Finkel & Pedersen, 2001). This suggests that the environment is relatively more important in explaining individual differences for low IQ groups than for high IQ groups.

Chapter 6: Intelligence and brain volumes

Chapter 6 employed a multivariate twin design to investigate the association between total brain volume, gray matter and white matter volume and intelligence as assessed by the Raven IQ test and verbal comprehension, perceptual organization and processing speed as assessed by the WISC-III. Phenotypic correlations between the brain volumes and

intelligence traits ranged between .20 and .33. Processing speed and brain volume did not correlate. The relation between brain volume and intelligence was entirely explained by a set of genes influencing both intelligence and brain volume.

The phenotypic correlations between WMV and GMV and the intelligence measures corrected for TBV indicated that intelligence was not related to proportion WMV/GMV. The association between intelligence and WMV and GMV disappeared once corrected for TBV. Nevertheless from the partial correlation analyses it can not be concluded that intelligence is only influenced by TBV and that WMV and GMV by themselves do not influence intelligence.

Genetic and environmental correlations gave an indication for the direction of causation for the association between intelligence and brain volume. The heritability estimates for the brain volumes are around 90%. In contrast, variability in intelligence is for about 60% caused by differences in genotypes. If intelligence causally influences brain volumes, this would be reflected in genetic as well as environmental correlations: all genetic and environmental factors that influence intelligence would, through the causal chain, influence brain volume (De Moor, Boomsma, Stubbe, Willemsen, & De Geus, in press). However, Chapter 6 showed that only the genetic correlations are significant. In fact 85% to 100% of the covariation between brain volume and intelligence are caused by shared genetic factors.

Chapter 7: Twin separation in primary school

At present there is hardly any research comparing the adjustment of twin pairs who are separated versus those kept together at school. Therefore, in Chapter 7 the short- and long-term effects of classroom separation in twins on behavior problems and academic performance were studied in twin pairs selected from the NTR. Short-term effects were studied at age 7 in twins separated at age 5 and long-term effects at age 12 in twins who had been separated or together most of the time at school. Behavior problems were rated by mothers (Child Behavior Checklist at ages 3, 7 and 12; Achenbach & Dumenci, 2001) and teachers (Teacher Report Form at ages 7 and 12; Achenbach, 1991). Academic achievement was measured at age 12 using a national academic achievement test (CITO; Bartels, Rietveld, Van Baal, & Boomsma, 2002a).

At age 7, twins from separated pairs had more internalizing and externalizing problems than non-separated twins, as rated by both mothers and teachers. However, only

for the maternal ratings of internalizing problems these effects could be attributed to the separation itself and not to pre-existing problems (at age 3) between separated and non-separated twins. Long-term effects of separation were significant for maternal and teacher ratings of internalizing and externalizing problems, but these effects could be explained by pre-existing differences between separated and non-separated groups. Thus, for behavioral problems at the age of 7 years, it may matter whether twins are separated or not. The separation of twins at school leads to internalizing problem behavior. However, all findings represent small effect sizes. Furthermore, at age of 12, this effect has disappeared. There were no differences in academic achievement between the separated and non-separated group.

Discussion

Early adolescence (the gradual transition between childhood and adulthood) is the focus of this thesis since this is a critical period in cognitive and brain development with important changes in brain structure and cognitive abilities (e.g. Durston et al., 2001; Spear, 2000). Both these developmental changes may be essential for optimal adult functioning. Diseases that affect the integrity of the brain at a young age, such as schizophrenia are likely to display their first symptoms during this period (Van Oel, Sitskoorn, Cremer, & Kahn, 2002). To get a better understanding of the development of these diseases, it is important to learn more about the genetic and environmental processes underlying the transition from childhood into adulthood in healthy children (Luna & Sweeney, 2001).

Studies generated from the first wave of data collection in this longitudinal twin study investigated the relationship among cognitive measures, brain volumes and intelligence, and hormonal levels and brain structure (this thesis; Hoekstra, Bartels, Van Leeuwen, & Boomsma, in revision; Peper et al., 2008; Peper et al., in press). These studies showed that in preadolescence the relationship among these measures are mainly caused by overlapping sets of genes, rather than by environment. This finding regularly returns in the literature on individual differences: genetic influences tie together diverse measures of cognitive functioning, whereas environmental effects drive wedges between different dimensions of cognitive processing (Luo, Petrill, & Thompson, 1994; Pedersen, Plomin, & McClearn, 1994). This applies also to the relation between brain structure and cognition, as shown in adults by e.g. Posthuma et al. (2002, 2003) and Hulshoff Pol et al. (2006). This

thesis shows that in children genes also are the binding factor for the coherence between various cognitive traits and between cognition and brain structure.

Further, the developmental studies (this thesis; Hoekstra, et al., in revision) on cognitive measures showed that during the course of development genetic correlations between these measures increase. However, this conclusion was based on cross-sectional studies. By following this sample longitudinally, it can be confirmed whether this truly is the case. At this moment the first follow-up measurement is taking place, now that the twins are almost twelve years old.

The relationship between intelligence and brain volumes might be caused by overlapping sets of genes, as suggested by the results in Chapter 6. However, nested on the genetic pleiotropic model, are several phenotypic causation models which might also explain the association between intelligence and brain volume. Recent evidence showed that for specific traits, like juggling or knowing your way around London as a taxi driver, a causal relation from training to increased local gray matter volume can be inferred (Draganski et al., 2004; Maguire, Woollett, & Spiers, 2006). For the relation between intelligence and brain volume in children a direction-of-causation model seems less likely. Environmental stimulation to increase intelligence does not influence total brain volumes. As put forward by De Moor et al. (2008) such a model requires that all genetic and environmental factors that influence intelligence would, through the causal chain, also influence brain volume.

This thesis did not find evidence of influence of shared environment on any of the cognitive measures that were included in the protocol. This is in contrast with previous findings in children which reported a contribution of shared environmental influences on aspects of intelligence (Bartels, Rietveld, Van Baal, & Boomsma, 2002b; Rietveld, Dolan, Van Baal, & Boomsma, 2003; Scarr & Weinberg, 1983). In adults, shared environment does not contribute to the variability in cognition (Bouchard, Jr., Lykken, McGue, Segal, & Tellegen, 1990; Posthuma, 2002). There may be several reasons why we did not observe any influence of C. First, it should be recognized that with 112 families, the statistical power to detect shared environmental is not large (Martin & Eaves, 1977; Purcell, Cherny, & Sham, 2003; Visscher, Gordon, & Neale, 2008). However, for most cognitive measures twin correlations did not suggest an influence of shared environment. The influence of shared environment in childhood may be confined to traits like vocabulary and general knowledge and may be so small that we did not detect it. A second

hypothesis is suggested by Guiso, Monte, Sapienza, and Zingales (2008) who observed that girls close the gender gap (differences between girls' and boys' scores on math and reading) by becoming better in both math and reading in countries where women are equal in economic and political opportunities, education, and well-being. This may also suggest that when the opportunities provided by society are adequate, genotypes can come fully to expression. Thus, maybe in recent years, environment in the Netherlands has changed in a way, e.g. equal educational opportunities for all children, which made it possible for children to reach the cognitive level in accordance with their genotype at an earlier age than before.

There was no evidence for cultural transmission or shared environmental influence on general IQ. This does not imply that environment does not influence cognition. It merely means that environmental variation, which is shared between siblings and influences these siblings in the same way, does not play a role in variability in cognition. Shared environment is not the same as for instance parental style or stimulation. Parents do not treat all their offspring exactly in the same way independent of the phenotype (or genotype) of their children. Chapter 5 even shows that depending on the genotype of the child the environment plays a more or less important role. This study showed that in children with a genetic predisposition to be less intelligent, environmental stimulation is more influential. Moreover, Chapter 5 shows that genetic studies on intelligence should take GE interaction into account, because else heritability estimates will be inflated.

Combining the results of Chapter 3 and 4 gives some insight what kind of processes are captured by the two common genetic factors involved in the relation between verbal and visuospatial WM and STM. In Chapter 4 three factors were involved in the relation among the memory measures: 1) a genetic factor common to the memory measures and intelligence and reading ability; 2) a genetic factor common to visuospatial STM and verbal and visuospatial WM; 3) a genetic factor common to verbal memory and reading ability. The first factor seems to involve general intelligence, the second factor probably involves an ability which is essential for complex memory, and the third factor seems to embody the ability to code information phonemically or verbally. Therefore, the overlap between the two memory factors which were derived from the genetic analysis of WM and STM seem to represent general intelligence and processing of complex memory tasks. The specific genetic factor involved in verbal memory most likely represents verbal coding.

Chapter 7 showed that putting twin pairs in separate classes does not lead to extra environmental variation between separated and non-separated twin pairs in variability in intelligence (at least at age twelve). This has important consequences for twins and their parents. Schools in the Netherlands often have a policy of separating twins. Our study shows, that if anything, this policy is harmful because it leads to an increase in internalizing problems. The effects of separation into different classrooms did not show up for educational attainment. This implies that for cognitive traits, data from twins can be generalized to the non-twin population.

Several traits studied in this thesis seem suitable endophenotypes for intelligence: all traits are reasonably reliable, heritable, genetically associated with intelligence, and these genetic associations seem theoretically meaningful (De Geus & Boomsma, 2001). For the relationship between brain volumes and intelligence, the theoretically meaningfulness of this relation is obvious, but also memory measures are theoretically meaningful endophenotypes. Memory performance has been included in theories of intelligence since the beginning of the development of psychometric IQ (Ackerman, Beier, & Boyle, 2005). The greater an individual's STM capacity and therefore its WM capacity, the more information the individual has simultaneously available for use in solving problems (Fry & Hale, 2000; Just & Carpenter, 1992). The question is however, whether these traits are no too complex by themselves. One of the complications with identifying genes affecting complex traits is that they are influenced by many genes, and therefore each gene is likely to have a relatively small effect (Plomin, DeFries, McClearn, & McGuffin, 2001) and therefore difficult to identify. Further research, like genome wide association studies (Kruglyak, 2008), should point out if these traits indeed can aid the search for genes involved in intelligence.

Several relationships among brain structure, cognition and hormonal levels remain to be elucidated. Chapter 6 showed that intelligence is related to brain volume; however this thesis did not look in to the relationship between specific brain regions and intelligence. Are the same brain regions implicated in the relationship between intelligence and brain structure in children as in adults?

Peper et al. (in press) showed that children, who are more advanced in puberty stage, are also more advanced in brain development. Decreases were found in frontal and parietal gray matter density, areas involved in higher level cognition. It is unclear which

implications these decreases in gray matter have for cognition and cognitive development. Shaw et al. (2006) showed that it is rather the trajectory of change in the thickness of the cerebral cortex, rather than cortical thickness itself, which is most closely related to level of intelligence. Extremely intelligent children seemed to be delayed in brain development compared to average and high intelligent children. Is the trajectory of change in thickness of the cortex related to physical maturation? Or are the specific brain regions which development is related to puberty different from the specific brain regions implicated in intelligence? Further longitudinal research in this sample should point this out.

Another area, which remained unstudied, is the relation between brain structure and other domains of cognition besides intelligence. For instance the study on brain volumes and intelligence did not reveal a relationship between processing speed and brain volumes. A possible explanation for this finding is that processing speed as measured by the WISC is confounded with motor speed. Another possibility is that processing speed is only related to specific white matter areas. Using inspection time as measure of processing speed and VBM should point out whether processing speed indeed is related to increased speed of cortico-cortical connections by means of increased myelination (Miller, 1994).

Reference List

Achenbach, T.M. (1991) *Manual for the Teacher's Report Form and 1991 Profile.* Burlington, VT: University of Vermont, Department of Psychiatry

Achenbach, T M & Dumenci, L (2001). Advances in Empirically Based Assessment: Revised Cross-Informant Syndromes and New DSM-Oriented Scales for the CBCL, YSR, and TRF: Comment on Lengua, Sadowksi, Friedrich, and Fisher (2001)*1. *Journal of Consulting and Clinical Psychology, 69*, 699-702.

Ackerman, P L, Beier, M E, & Boyle, M O (2005). Working memory and intelligence: the same or different constructs? *Psychological Bulletin, 131*, 30-60.

Alloway, T P, Gathercole, S E, & Pickering, S J (2006). Verbal and visuospatial short-term and working memory in children: are they separable? *Child Development, 77*, 1698-1716.

Bartels, M, Rietveld, M J, Van Baal, G C, & Boomsma, D I (2002a). Heritability of educational achievement in 12-year-olds and the overlap with cognitive ability. *Twin Research, 5*, 544-553.

Bartels, M, Rietveld, M J H, Van Baal, G C M, & Boomsma, D I (2002b). Genetic and environmental influences on the development of intelligence. *Behavior Genetics, 32*, 237-249.

Bayliss, D M, Jarrold, C, Baddeley, A D, & Gunn, D M (2005). The relationship between short-term memory and working memory: complex span made simple? *Memory, 13*, 414-421.

Bishop, D V M (2006). Developmental cognitive genetics: How psychology can inform genetics and vice versa. *The Quarterly Journal of Experimental Psychology, 59*, 1153-1168.

Bouchard, T J, Jr., Lykken, D T, McGue, M, Segal, N L, & Tellegen, A (1990). Sources of human psychological differences: the Minnesota Study of Twins Reared Apart. *Science, 250*, 223-228.

Cito. (1995) *Drie Minuten Test.* Arnhem: Cito

Cohen-Mimran, R & Sapir, S (2007). Deficits in working memory in young adults with reading disabilities. *Journal of Communication Disorders, 40*, 168-183.

Conway, A R A, Cowan, N, Bunting, M F, Therriault, D J, & Minkoff, S R B (2002). A latent variable analysis of working memory capacity, short-term memory capacity, processing speed, and general fluid intelligence. *Intelligence, 30*, 163-183.

Corsi, P M (1974). Human memory and the medial temporal region of the brain. *Dissertation Abstracts International, 34*, 819B (University Microfilms No. AAI05-77717)-

De Geus, E J & Boomsma, D I (2001). A genetic neuroscience approach to human cognition. *European Psychologist, 6*, 241-253.

De Moor, M H M, Boomsma, D I, Stubbe, J H, Willemsen, G, & De Geus, E J C (2008). Testing causality in the association between regular exercise and symptoms of anxiety and depression. *Archives of General Psychiatry, 65,* 897-905.

Draganski, B, Gaser, C, Busch, V, Schuierer, G, Bogdahn, U, & May, A (2004). Neuroplasticity: changes in grey matter induced by training. *Nature, 427*, 311-312.

Durston, S, Hulshoff Pol, H E, Casey, B J, Giedd, J N, Buitelaar, J K, & Van Engeland, H (2001). Anatomical MRI of the developing human brain: what have we learned? *Journal of the American Academy of Child and Adolescent Psychiatry, 40*, 1012-1020.

Finkel, D. & Pedersen, N.L. (2001). Sources of environmental influence on cognitive abilities in adulthood. In: E.L. Grigorenko & R.J. Sternberg *Family environment and intellectual functioning* (pp. 173-194) Mahwah: Lawrence Erlbaum Associates

Fry, A F & Hale, S (2000). Relationships among processing speed, working memory, and fluid intelligence in children. *Biological Psychology, 54*, 1-34.

Gathercole, S E, Alloway, T P, Willis, C, & Adams, A M (2006). Working memory in children with reading disabilities. *Journal of Experimental Child Psychology, 93*, 265-281.

Gevins, A & Cutillo, B (1993). Spatiotemporal dynamics of component processes in human working memory. *Electroencephalography and Clinical Neurophysiology., 87*, 128-143.

Guiso, L, Monte, F, Sapienza, P, & Zingales, L (2008). Diversity. Culture, gender, and math. *Science, 320*, 1164-1165.

Hoekstra, R A, Bartels, M, Van Leeuwen, M, & Boomsma, D I (in revision). Genetic architecture of verbal abilities in children and adolescents. *Developmental Science.*

Hulshoff Pol, H E, Schnack, H G, Posthuma, D, Mandl, R C, Baare, W F, Van Oel, C, Van Haren, N E, Collins, D L, Evans, A C, Amunts, K, Burgel, U, Zilles, K, De Geus, E, Boomsma, D I, & Kahn, R S (2006). Genetic contributions to human brain morphology and intelligence. *Journal of Neuroscience, 26*, 10235-10242.

Jinks, J L & Fulker, D W (1970). Comparison of the biometrical genetical, MAVA, and classical approaches to the analysis of the human behavior. *Psychological Bulletin, 73*, 311-349.

Just, M A & Carpenter, P A (1992). A Capacity Theory of Comprehension: Individual Differences in Working Memory. *Psychological Review, 99*, 122-149.

Kail, R & Hall, L K (2001). Distinguishing short-term memory from working memory. *Memory and Cognition, 29*, 1-9.

Kane, M J, Hambrick, D Z, Tuholski, S W, Wilhelm, O, Payne, T W, & Engle, R W (2004). The generality of working memory capacity: a latent-variable approach to verbal and visuospatial memory span and reasoning. *Journal of Experimental Psychology: General, 133*, 189-217.

Kercher, A C & Sandoval, J (1991). Reading disability and the differential ability scales. *Journal of School Psychology, 29*, 293-307.

Kruglyak, L (2008). The road to genome-wide association studies. *Nature Reviews Genetics, 9*, 314-318.

Luna, B & Sweeney, J A (2001). Studies of brain and cognitive maturation through childhood and adolescence: a strategy for testing neurodevelopmental hypotheses. *Schizophrenia Bulletin, 27*, 443-455.

Luo, D, Petrill, S A, & Thompson, L A (1994). An exploration of genetic g: Hierarchical factor analysis of cognitive data from the western reserve twin project. *Intelligence, 18*, 335-347.

Maguire, E A, Woollett, K, & Spiers, H J (2006). London taxi drivers and bus drivers: a structural MRI and neuropsychological analysis. *Hippocampus, 16*, 1091-1101.

Martin, N G & Eaves, L J (1977). The genetical analysis of covariance structure. *Heredity, 38*, 79-95.

Miller, E M (1994). Intelligence and brain myelination: A hypothesis. *Personality and Individual Differences, 17*, 803-832.

Pedersen, N L, Plomin, R, & McClearn, G E (1994). Is there G beyond g? (Is there genetic influence on specific cognitive abilities independent of genetic influence on general cognitive ability?). *Intelligence, 18*, 133-143.

Peper, J S, Brouwer, R M, Schnack, H G, Van Baal, G C M, Van Leeuwen, M, Van den Berg, S M, Delemarre-van de Waal, H A, Janke, A L, Collins, D L, Evans, A C, Boomsma, D I, Kahn, R S, & Hulshoff Pol, H E (2008). Cerebral white matter in early puberty is associated with luteinizing hormone concentrations. *Psychoneuroendocrinology, 33, 909-915*

Peper, J S, Schnack, H G, Brouwer, R M, Van Baal, G C M, Pjetri, E, Székely, E, Van Leeuwen, M, Van den Berg, S M, Collins, D L, Evans, A C, Boomsma, D I, Kahn, R S, & Hulshoff Pol, H E (in press). Heritability of regional and global brain volume at the onset of puberty: a Magnetic Resonance Imaging study in 9-year old twin-pairs. *Human Brain Mapping*.

Plomin, R., DeFries, J.C., McClearn, G.E., & McGuffin, P. (2001) *Behavioral genetics.* New York: Worth Publisher

Posthuma, D, Baare, W F, Hulshoff Pol, H E, Kahn, R S, Boomsma, D I, & De Geus, E J (2003). Genetic correlations between brain volumes and the WAIS-III dimensions of verbal comprehension, working memory, perceptual organization, and processing speed. *Twin Research, 6*, 131-139.

Posthuma, D, De Geus, E J, Baare, W F, Hulshoff Pol, H E, Kahn, R S, & Boomsma, D I (2002). The association between brain volume and intelligence is of genetic origin. *Nature Neuroscience, 5*, 83-84.

Posthuma, D (2002). Genetic variation and cognition.1-246.

Purcell, S, Cherny, S S, & Sham, P C (2003). Genetic Power Calculator: design of linkage and association genetic mapping studies of complex traits. Bioinformatics., 19, 149-150.
Raven, J., Raven, J.C., & Court, J.H. (1998) Raven manual: Section 4 advanced progressive matrices. Oxford: Oxford Psychologists Press
Raven, J.C. (1960) Guide to the Standard Progressive Matrices. London: H.K. Lewis & Co. Ltd.
Rietveld, M J H, Dolan, C V, Van Baal, G C M, & Boomsma, D I (2003). A twin study of differentiation of cognitive abilities in childhood . Behavior Genetics, 33, 367-381.
Scarr, S & Weinberg, R A (1983). The Minnesota Adoption Studies: Genetic differences and malleability. Child Development, 54, 260-267.
Shaw, P, Greenstein, D, Lerch, J, Clasen, L, Lenroot, R, Gogtay, N, Evans, A, Rapoport, J, & Giedd, J (2006). Intellectual ability and cortical development in children and adolescents. *Nature, 440*, 676-679.
Spear, L P (2000). The adolescent brain and age-related behavioral manifestations. *Neuroscience & Biobehavioral Reviews, 24*, 417-463.
Swanson, H L & Jerman, O (2007). The influence of working memory on reading growth in subgroups of children with reading disabilities. *Journal of Experimental Child Psychology, 96*, 249-283.
Tiu, R D, Jr., Thompson, L A, & Lewis, B A (2003). The Role of IQ in a Component Model of Reading. *Journal of Learning Disabilities, 36*, 424-436.
Van Oel, C J, Sitskoorn, M M, Cremer, M P, & Kahn, R S (2002). School performance as a premorbid marker for schizophrenia: a twin study. *Schizophrenia Bulletin, 28*, 401-414.
Van Soelen, I L C, Van den Berg, S M, Dekker, P H, Van Leeuwen, M, Peper, J S, Hulshoff Pol, H E, & Boomsma, D I (in revision). Individual differences in verbal short term memory in 9-year-old twin pairs and their older siblings. *Learning and Individual Differences.*
Visscher, P M, Gordon, S, & Neale, M C (2008). Power of the classical twin design revisited: II detection of common environmental variance. *Twin Research and Human Genetics, 11*, 48-54.
Wechsler, D. (1997) *Wechsler Adult Intelligence Scale-Third edition, Dutch Version.* Amsterdam: The Psychological Corporation Ltd. Harcourt Publishers
Wechsler, D., Kort, W., Compaan, E.L., Bleichrodt, N., Resing, W.C.M., Schittekatte, M., Bosmans, M., Vermeir, G., & Verhaeghe, P. (2002) *Handleiding WISC-III^{NL} [Manual WISC-III^{NL}].* London: The Psychological Corporation Limited, Nederlands Instituut van Psychologen Dienstencentrum

Nederlandse samenvatting

Een studie naar cognitie in pre-adolescente tweelingen

Woordenlijst

constructen eigenschappen die niet direct zichtbaar en dus niet direct te meten zijn
culturele transmissie ouders geven omgeving door aan hun kinderen *endohenotype* een deelcomponent of –element die bijdraagt aan de variabiliteit van een trek waar veel genen bij betrokken zijn
endofenotypering het bestuderen van cognitieve deelcomponenten of –elementen (dit kunnen ook fysiologische eigenschappen zijn) die bijdragen aan de variabiliteit van trekken waar zeer veel genen bij zijn betrokken zoals intelligentie
fenotypische selectie actieve partnerselectie: partners kiezen elkaar uit gebaseerd op elkaars intelligentie of een eigenschap die daar aan gerelateerd is
genetische transmissie ouders geven genen door aan hun kinderen
gen-omgevingsinteractie mensen met een bepaald genotype zijn meer kwetsbaar voor een bepaalde omgeving
kortetermijngeheugen het vermogen om informatie voor een korte periode te onthouden in de afwezigheid van andere aandachtvragende cognitieve processen
partnerselectie mensen gaan actief of passief op zoek naar een partner met dezelfde dan wel tegenovergestelde eigenschappen
passieve GE-correlatie ouders geven passief ervaringen door aan hun kinderen die gecorreleerd zijn met hun eigen genetische eigenschappen: ouders die intelligent zijn, geven intelligentie via hun genen door aan hun kinderen, maar door hun intelligentie zouden ze ook een omgeving kunnen creëren die de intelligentie van hun kinderen stimuleert
psychometrisch IQ intelligentie bepaald met behulp van een intelligentietest
selectieve aandacht het besteden van aandacht aan relevante stimuli in de aanwezigheid van afleidende stimuli
sociale homogeniteit passieve partnerselectie: partners ontmoeten elkaar in een omgeving die gecorreleerd is met intelligentie
stimulus incongruentie effecten verlies van tijd en/ of accuratesse als gevolg van conflicterende stimuli
test-hertest onderzoek een onderzoeksopzet om de stabiliteit van een test over de tijd te bepalen. Als uit een test twee tot drie weken na afname iets heel anders komt, is de test niet betrouwbaar
unieke omgevingseffecten omgevingseffecten die niet hetzelfde zijn voor kinderen die in hetzelfde gezin opgroeien
verwerkingssnelheid de tijd die nodig is om bepaalde informatie te verwerken
werkgeheugen het systeem dat nodig is om simultaan informatie op te slaan en te bewerken

In mijn proefschrift heb ik de eerste fase beschreven van een longitudinale tweelingstudie naar cognitie, hersenstructuur en hormoonwaardes gedurende de adolescentie. De data zijn verzameld in negen jaar oude tweelingen en hun negen tot veertien jaar oude broers en zussen die ingeschreven staan bij het Nederlands Tweelingen Register (NTR). Dit proefschrift behandelt de resultaten van het cognitieonderzoek en de relatie tussen cognitieve parameters en hersenstructuur. Hieronder worden de bevindingen van de voorgaande hoofdstukken samengevat en besproken.

Hoofdstuk 2: Endofenotypen voor intelligentie in kinderen

Hoofdstuk 2 beschrijft een onderzoek waarin een strategie wordt aangereikt voor het vinden van genen die verschillen in intelligentie tussen kinderen veroorzaken. Uit eerder onderzoek is gebleken dat verschillen in intelligentie voor een groot gedeelte worden veroorzaakt door verschillen in genotype. Voor kinderen is dat voor 25-50% het geval en voor volwassenen voor zo'n 70%. Tot op heden is het echter nog niet gelukt genen aan te wijzen die betrokken zijn bij verschillen in intelligentie in de algemene populatie. Een mogelijk reden hiervoor is dat bij intelligentie zoveel genen zijn betrokken, dat ieder gen maar een klein effect sorteert. Hierdoor zijn deze genen moeilijk op te sporen. Een manier om dit probleem te ondervangen, is het concept van intelligentie op te delen in componenten die bijdragen aan intelligentie. Hierbij moet men bijvoorbeeld denken aan geheugen en reactiesnelheid. De achterliggende aanname hiervoor is dat hoe beter het geheugen en / of hoe hoger de reactiesnelheid is, hoe intelligenter de persoon is. Als een kleienere set genen bijdraagt aan verschillen in deze eigenschappen, is het mogelijk dat genen voor deze eigenschappen makkelijker op te sporen zijn. Deze strategie heet *endofenotypering*. In hoofdstuk 2 worden een aantal *endofenotypen* onderzocht op hun bruikbaarheid in genonderzoek naar intelligentieverschillen in kinderen.

In volwassenen is er al een kleine verzameling van endofenotypen voor intelligentie beschikbaar. In kinderen is er minder bekend over de bruikbaarheid van deze cognitieve maten als endofenotypen voor intelligentie. Hoofdstuk 2 heeft voor toekomstige genetische studies in kinderen en adolescenten veelbelovende endofenotypen voor intelligentie geïdentificeerd. De cognitieve taken zijn gekozen op basis van een voor volwassenen al beschikbare verzameling van endofenotypen voor intelligentie en omvatten de domeinen *werkgeheugen*, *verwerkingssnelheid* en *selectieve aandacht*.

In kinderen en adolescenten is de verzameling taken afgenomen in een *test-hertest onderzoek*. Vervolgens is de relatie van deze taken tot intelligentie onderzocht. De testbatterij bestond uit de '*n*-back task' (2 en 3 terug; voor werkgeheugen), de Eriksen flanker taak (voor selectieve aandacht) en de π-inspectie taak (voor verwerkingssnelheid). In kinderen waren alle test-hertest correlaties 0,60 of hoger, behalve voor accuratesse en *stimulus incongruentie effecten* gemeten met de flanker taak. Voor adolescenten gold hetzelfde, met uitzondering van de 2-back (r = 0,16) en de π-inspectie taak (r = 0,58). In zowel kinderen als adolescenten waren prestaties op de n-back significant gerelateerd aan IQ: betere prestaties op de n-back waren gerelateerd aan hogere IQ scores. Tevens werd gevonden dat alleen in kinderen reactiesnelheid op de congruente en incongruente sessies van de flanker significant gerelateerd was aan IQ: hoe langer de reactietijd, hoe lager het IQ. Daarnaast bleken, zowel in kinderen als adolescenten, incongruentie effecten op reactietijd, accuratesse gedurende de congruente en incongruente sessies, en ook de incongruentie effecten op accuratesse niet gerelateerd aan IQ. Ten slotte was inspectietijd in kinderen, maar niet in adolescenten, gerelateerd aan IQ: hoe korter de inspectietijd, hoe hoger het IQ.

Hiermee lijkt werkgeheugen in kinderen en adolescenten geschikt als endofenotype voor intelligentie: het kan betrouwbaar gemeten worden met de *n*-back taak en correleert met intelligentie. In kinderen en adolescenten is verwerkingssnelheid geen optimaal endofenotype voor intelligentie. Als verwerkingssnelheid gecorrigeerd wordt voor werkgeheugen, is er slechts een kleine bijdrage aan de variabiliteit in intelligentie. In geen van beide leeftijdsgroepen is selectieve aandacht, tenminste zoals gemeten aan de hand van incongruentie-effecten op reactietijd en accuratesse op de flanker taak, geschikt als endofenotype voor intelligentie.

Tabel 1 *Erfelijkheid van cognitieve maten*

Maat	erfelijkheid
IQ (Raven)[a]	0,67
IQ (WISC-III)[a]	0,75
Leesvaardigheid (OMRT)[a]	0,83
Verbaal IQ (WISC-III)[b]	0,81
Verbaal begrip (WISC-III)[a]	0,79
Perceptuele organisatie (WISC-III)[a]	0,56
Verwerkingssnelheid (WISC-III)[a]	0,58
Verbaal leren (AVLT)[b]:	0,46
Leersnelheid[c]	0,43
Vergeetsnelheid[c]	0,20*
Lettervloeiendheid (COWA)[b]	0,40
Categorievloeiendheid (COWA)[b]	0,29
Verbaal STM (DSF)[a]	0,47
Visuospatieel STM (Corsi)[a]	0,47
Verbaal WM (DSB)[a]	0,35
Visuospatieel WM (2-back)[a]	0,47

Voetnoot. tussen haakjes de gebruikte testen, WISC = Wechsler intelligentie schalen voor kinderen; OMRT = één minuut leestest; AVLT = auditieve verbale leertaak; COWA = gecontroleerde mondelinge woordassociatie; DSF = cijferreeksen vooruit; DSB = cijferreeksen achteruit; *erfelijkheid alleen voor tweelingen, voor broers en zussen 0,30; [a]gebaseerd op data-analyse in dit proefschrift; [b]Hoekstra et al., (in revisie); [c]Van Soelen et al. (in revisie)

Hoofdstuk 3: De genetische structuur van geheugen

In hoofdstuk 3 en 4 wordt de erfelijkheid van verscheidene cognitieve maten onderzocht. In tabel 1 staan de erfelijkheidsschattingen voor deze maten en *psychometrisch IQ* weergegeven, gebaseerd op de analyses in dit proefschrift en gepubliceerd door anderen op grond van deze longitudinale studie. Erfelijkheidsschattingen varieerden tussen 0,20 en 0,83. Dus verschillen tussen mensen in prestatie op deze cognitieve taken werden voor 20-

83% verklaard door verschillen in genotype. De hoogste erfelijkheidsschattingen werden gevonden voor IQ (totaal en verbaal) en leesvaardigheid.

In hoofdstuk 3 werd de oorsprong van de samenhang tussen werkgeheugen (WM) en kortetermijngeheugen (STM) bestudeerd door in een ontwikkelingsstudie de genetische en de omgevingsrelaties tussen verbaal en visuospatieel WM en STM te onderzoeken. Wordt de samenhang verklaard doordat er genen zijn die zowel STM als WM beïnvloeden, zijn er omgevingsinvloeden die op beide eigenschappen invloed uitoefenen, of wordt de samenhang verklaard door een combinatie van genen en omgevingsinvloeden? Verder is er in deze studie gekeken of de invloeden van genen en omgeving veranderen gedurende de loop van de ontwikkeling.

Ondanks dat veel studies op het gebied van individuele verschillen zich gericht hebben op WM en STM in zowel volwassenen als kinderen, is de mate waarin verbaal en visuospatieel WM en STM verschillende of dezelfde constructen meten nog onduidelijk. Daarnaast is onduidelijk hoe de relatie tussen WM en STM gedurende de ontwikkeling verloopt. Deze vragen zijn beantwoord door WM en STM te bestuderen in de negenjarige tweelingen en hun broers en zussen en in een cohort van 186 families van jongvolwassen tweelingen en hun broers en zussen. Daarvoor zijn de Corsi blokkentaak, de ‘n-back’ taak en cijferreeksen voor- en achteruit afgenomen.

In het jong-volwassen cohort werd de relatie tussen de vier variabelen het best beschreven door een model bestaande uit twee gecorreleerde factoren die alle genetische variatie verklaarden (één voor verbaal en één voor visuospatieel geheugen) en een specifieke omgevingsfactor voor iedere variabele. Dus twee groepen van genen zijn verantwoordelijk voor de samenhang tussen verbaal en visuospatieel WM en STM: één groep van genen beïnvloedt verbaal geheugen en één groep beïnvloedt visuospatieel geheugen. Deze twee groepen genen hangen met elkaar samen. Ook in het kindercohort werd de samenhang tussen verbaal en visuospatieel WM en STM verklaard door een genetische factor voor verbaal en een genetische factor voor visuospatieel geheugen. Desondanks lieten de resultaten van de kinderen ten opzichte van het jong volwassenen cohort ook significante verschillen in de genetische structuur zien: in kinderen werden STM en WM ook beïnvloed door specifieke genetische factoren. Dus bij kinderen spelen ook genen een rol die elk van de vier geheugen maten afzonderlijk beïnvloeden. Gezien vanuit een genetisch standpunt, kan men zeggen dat STM en WM deel uitmaken van eenzelfde

systeem, en dat verbale en visuopatiële informatie door gedeeltelijk overlappende geheugenpaden worden verwerkt. Bovendien verdwijnen genetische factoren die specifiek zijn voor bepaalde eigenschappen in de loop van de ontwikkeling.

Hoofdstuk 4: De genetische structuur van leesvaardigheid

Hoofdstuk 4 onderzocht de genetische relatie tussen leesvaardigheid, intelligentie en verbaal en visuospatieel WM en STM. Intelligentie werd gemeten met behulp van de Wechsler Intelligentie Schalen voor Kinderen (WISC). Als maat voor leesvaardigheid werd prestatie op de één minuut leestaak gebruikt. Als maat voor verbaal en visuospatieel WM en STM werden de geheugentaken beschreven in hoofdstuk 3 gebruikt.

De relatie tussen leesvaardigheid en intelligentie is aangetoond in groepen kinderen met en zonder leesproblemen. De relatie tussen leesproblemen en geheugen is echter nog onderwerp van discussie. Literatuur over de genetische relatie tussen geheugen en leesvaardigheid is zeldzaam. Als de herkomst van de relatie tussen intelligentie, geheugen en leesvaardigheid wordt gevonden, geeft dit duidelijkheid of geheugen- en intelligentieproblemen in kinderen met leesvaardigheidproblemen een teken zijn van de ernst van de leesproblemen of slechts een symptoom van de leesproblemen op zichzelf.

De geobserveerde correlatie tussen leesvaardigheid en intelligentie was ,42 en tussen leesvaardigheid en de geheugenmaten varieerde deze tussen de 0,24 en de 0,44. De geobserveerde correlaties tussen IQ, STM, WM en leesvaardigheid werden geheel verklaard door genen: genen zorgen ervoor dat IQ, STM, WM en leesvaardigheid met elkaar samenhangen, omgevingsinvloeden spelen geen rol in deze samenhang. Het model dat de onderlinge relaties het beste verklaarde, bestond uit een gedeelde genetische factor voor alle variabelen; een gedeelde genetische factor voor visuospatieel STM en verbaal en visuospatieel WM; een gedeelde genetische factor voor verbaal geheugen en leesvaardigheid; een specifieke genetische factor voor visuospatieel WM; en een specifieke genetische factor voor leesvaardigheid. De specifieke genetische factor verklaarde ongeveer de helft van de genetische variantie. In andere woorden, de helft van de genetische verschillen die gevonden werden voor leesvaardigheid, waren ook van invloed op IQ, WM en STM. De andere helft van de genetische verschillen in leesvaardigheid waren alleen van invloed op leesvaardigheid.

Deze studie toont aan dat leesvaardigheid is gerelateerd aan intelligentie, WM en STM en dat deze relatie volledig gemedieerd wordt door genetische invloeden: er is een set genetische factoren die leesvaardigheid en intelligentie, WM en STM beïnvloedt en een set genen die alleen leesvaardigheid en verbaal geheugen beïnvloedt. Dit suggereert dat er ten minste drie groepen kinderen zijn met leesvaardigheidproblemen: kinderen die minder intelligent zijn en daardoor meer moeite hebben met lezen; kinderen die gemiddeld of hoger scoren op een IQ-test, maar problemen hebben met verbaal geheugen en daardoor ook problemen met lezen ondervinden; en tenslotte kinderen met een lager IQ en problemen met verbaal geheugen. Deze groep kinderen ondervindt de meeste leesproblemen.

Hoofdstuk 5: Intelligentie

In hoofdstuk 5 werd de aanwezigheid van *partnerselectie*, *gen-omgevingsinteractie* (GE-interactie) en de erfelijkheid van intelligentie bij kinderen onderzocht door gebruik te maken van een studieopzet die bestond uit 9-jarige tweelingenparen, hun broers en zussen en hun ouders. Met behulp van deze onderzoeksopzet konden *culturele* en *genetische transmissie* worden bestudeerd terwijl er rekening gehouden werd met gelijkenis tussen partners (de ouders van de tweelingen). Twee concurrerende hypotheses over de herkomst van partnergelijkenis werden geëvalueerd: *sociale homogeniteit* en *fenotypische selectie*. De gevolgen van deze twee typen van partnerselectie voor de erfelijkheidsschatting van intelligentie werden ook onderzocht. Intelligentie werd gemeten met behulp van de Raven IQ test in zowel de ouders als de kinderen.

De correlatie voor Raven IQ tussen de partners was 0,33, de correlatie tussen de eeneiige tweelingparen 0,63 en de correlaties tussen de twee-eiige tweelingparen, tussen tweelinghelften en broer/zus en tussen ouders en kinderen varieerden tussen 0,25 en 0,38. Een model met alleen genetische effecten en *unieke omgevingseffecten* verklaarde de correlatie tussen de familieleden het beste: verschillen in intelligentie worden verklaard door verschillen in genen en omgevingsinvloeden die voor ieder lid van een gezin uniek zijn. Verder bleek dat partners elkaar uitkiezen op grond van elkaars intelligentieniveau of een eigenschap die daaraan gerelateerd is.

Daarnaast liet het onderzoek zien dat ouders intelligentie niet via de omgeving aan hun kinderen doorgaven, maar alleen via de genen: culturele transmissie droeg niet significant bij aan verschillen in intelligentie. Doordat culturele transmissie afwezig is, is er

ook geen sprake van *passieve gen-omgevingscorrelatie* (GE-correlatie). De studieopzet van dit hoofdstuk was niet geschikt om reactieve of actieve GE-correlatie te detecteren. Wat we echter wel weten is, dat als er sprake is van GE-correlatie, het waarschijnlijker is dat reactieve of actieve GE correlatie van belang is. Dit zou betekenen dat de rol van de ouders is beperkt tot reageren op behoeftes en interesses zoals die aangegeven worden door het kind.

Om GE-interactie te detecteren en te schatten werd in eeneiige tweelingen de relatie tussen de som in intelligentiescores van een tweelingpaar en het verschil tussen de scores binnen een tweelingpaar onderzocht. De schatting van de correlatie tussen de som van de intelligentiescores en het verschil daartussen was -0,30. Een negatieve correlatie tussen somscores en absolute verschillen in intelligentiescores suggereert dat de omgeving relatief belangrijker is om individuele verschillen te verklaren in groepen mensen met een erfelijke aanleg voor een lager IQ dan in groepen mensen met een erfelijke aanleg voor een hoger IQ.

Hoofdstuk 6: Intelligentie en hersenvolume

In de studie van hoofdstuk 6 werd onderzocht in hoeverre verschillen in hersenvolumes samenhangen met verschillen in intelligentie bij kinderen. Verder is er gekeken of deze samenhang verklaard wordt door genetische factoren die zowel hersengrootte en intelligentie beïnvloeden of door omgevingsfactoren die beide maten beïnvloeden. Daarvoor is in hoofdstuk 6 gebruik gemaakt van multivariate tweelinganalyse waarin de relatie tussen totaal hersenvolume, grijze en witte stof volume enerzijds en intelligentie (gemeten met de Raven IQ test), verbaal begrip, perceptuele organisatie en verwerkingssnelheid (alle drie gemeten met de WISC) anderzijds onderzocht werd.

De geobserveerde correlaties tussen de hersenvolumes en de intelligentiematen varieerden tussen de 0,20 en de 0,33. Verwerkingssnelheid en hersenvolume correleerden niet met elkaar. De relatie tussen hersenvolume en intelligentie werd volledig verklaard door genen die zowel intelligentie als hersenvolume beïnvloeden.

Als de geobserveerde correlaties tussen witte stof volume en grijze stof volume en de intelligentiematen gecorrigeerd werden voor totaal hersenvolume, verdween de relatie tussen witte en grijze stof volume en intelligentie. Dit toont aan dat er geen relatie is tussen intelligentie en globale grijze stof/witte stof verhouding.

De genetische en omgevingscorrelaties in hoofdstuk 6 geven een aanwijzing in welke richting een oorzakelijk verband tussen intelligentie en hersenvolume gezocht moet worden bij kinderen. De erfelijkheidsschattingen voor totaal hersenvolume, grijze en witte stof volume bevinden zich rond de 90%. De variabiliteit in intelligentie wordt voor ongeveer 60% door genetische variatie verklaard. Als intelligentie hersenvolume beïnvloedt, zou dit zowel in de genetische als de omgevingscorrelaties tussen de hersenvolumes en intelligentie gereflecteerd moeten worden: alle genetische en omgevingsfactoren die intelligentie beïnvloeden, zouden ook via de oorzakelijke keten hersenvolume moeten beïnvloeden. Hoofdstuk 6 toonde echter aan dat alleen de genetische correlaties tussen de hersenvolumes en intelligentie significant zijn. Feitelijk wordt 85-100% van de covariatie tussen hersenvolume en intelligentie door gedeelde genetische factoren veroorzaakt bij kinderen. Daarom is het minder waarschijnlijk dat intelligentie invloed heeft op hersenvolume. Meer waarschijnlijk is dat grotere hersenvolumes leiden tot een hogere intelligentie, of dat er genen zijn die zowel hersenvolume als intelligentie beïnvloeden (pleiotropie).

Hoofdstuk 7: Scheiding van tweelingparen in het basisonderwijs

Op dit moment is nog nauwelijks onderzoek gedaan waarin gekeken is wat het effect is van apart naar school gaan van tweelingen in de basisschooltijd. Het is niet duidelijk of tweelingparen die gescheiden naar school gaan meer of minder probleemgedrag en betere of slechtere schoolprestaties vertonen dan tweelingparen die in dezelfde klassen naar school gaan, ondanks het feit dat het beleid van scholen vaak is om tweelingen van elkaar te scheiden. Daarom zijn in hoofdstuk 7 de korte- en langetermijneffecten van klassenscheiding van tweelingen op gedragsproblemen en schoolprestaties onderzocht. De tweelingparen kwamen uit het Nederlands Tweelingen Register. Kortetermijneffecten werden bekeken op leeftijd zeven in tweelingen die op vijfjarige leeftijd apart of samen naar school gingen, en lange termijneffecten op leeftijd twaalf in tweelingen die voor het merendeel van hun schoolperiode samen of apart naar school waren gegaan. Gedragsproblemen werden door moeders en leerkrachten op de CBCL en TRF vragenlijsten gescoord. Schoolprestaties werden gemeten aan de hand van de score op de Cito-toets.

Op leeftijd zeven vertoonden gescheiden tweelingen volgens zowel de moeders als de leerkrachten meer internaliserende (zoals angst en depressie) en externaliserende (bijv. agressie en gedragsstoornissen) problemen dan tweelingen die niet gescheiden waren. Alleen in het geval van de internaliserende problemen die aangegeven werden door de moeder, konden deze problemen toegeschreven worden aan de scheiding zelf en niet aan al bestaande gedragsproblemen. Op 3-jarige leeftijd. Ook op lange termijn waren er significant meer internaliserende en externaliserende problemen bij kinderen die gescheiden naar school gegaan waren ten opzichte van kinderen die samen naar school gegaan waren. Deze kunnen echter worden toegeschreven aan verschillen die al voor de scheiding bestonden tussen de gescheiden en niet-gescheiden tweelingparen. Dus voor internaliserende gedragsproblemen op zevenjarige leeftijd kan het er toe doen of een tweelingpaar apart of samen naar school gaat. Scheiding van tweelingparen leidt op zevenjarige leeftijd tot meer internaliserende problemen. Alle bevindingen laten echter slechts kleine effecten van tweelingscheiding zien. Verder zijn deze effecten op twaalfjarige leeftijd verdwenen. Er waren geen verschillen in schoolprestatie tussen tweelingen die wel en niet samen naar school gingen.

List of publications

Van Leeuwen, M., Peper, J. S., Van den Berg, S. M., Brouwer, R. M., Hulshoff Pol, H. E., Kahn, R. S., & Boomsma, D. I. (in press). A genetic analysis of brain volumes and IQ in children. *Intelligence*.

Van Leeuwen, M., Van den Berg, S. M., Hoekstra, R. A., & Boomsma, D. I. (2007). Endophenotypes for intelligence in children and adolescents. *Intelligence, 35,* 369-380.

Van Leeuwen, M., Van den Berg, S. M., Hoekstra, R. A., & Boomsma, D. I. (in revision). The genetic and environmental structure of working and short-term memory in young adults and children. *Neuropsychology*.

Van Leeuwen, M., Van den Berg, S. M., Peper, J. S., Hulshoff Pol, H. E., & Boomsma, D. I. (submitted). The genetic relationship between reading ability, intelligence and memory in children. *Behavior Genetics*

Van Leeuwen, M., Van den Berg, S. M., Van Beijsterveldt, T. C., & Boomsma, D. I. (2005). Effects of twin separation in primary school. *Twin Research and Human Genetics, 8,* 384-391.

Van Leeuwen, M., Van den Berg, S. M., & Boomsma, D. (2008). A twin-family study of general IQ. *Learning and Individual Differences, 18,* 76-88.

De Bruin, J. P., Feenstra, M. G., Broersen, L.M., **Van Leeuwen, M**., Arens, C., De Vries, S., & Joosten, R. N. (2000). Role of the prefrontal cortex of the rat in learning and decision making: effects of transient inactivation. *Progress in Brain Research*, *12,* 103-13

Hoekstra, R. A., Bartels, M., **Van Leeuwen, M**., & Boomsma, D. I. (in revision). Genetic architecture of verbal abilities in children and adolescents. *Developmental Science*.

Peper, J.S., Brouwer, R.M., Schnack, G.H., Van Baal, G.C.M., **Van Leeuwen, M**, Van den Berg, S. M., Boomsma, D. I., Kahn, R. S., & Hulshoff Pol, H. E. (in press). Sex steroids and brain structure in pubertal boys and girls. *Psychoneuroendocrinology.*

Peper, J. S., Brouwer, R. M., Schnack, H. G., Van Baal, G. C. M., **Van Leeuwen, M**., Van den Berg, S. M., Delemarre-van de Waal, H.A., Janke, A.L., Collins, D.L., Evans, A.C., Boomsma, D. I., Kahn, R. S., & Hulshoff Pol, H. E. (2008). Cerebral white matter in early puberty is associated with luteinizing hormone concentrations. *Psychoneuroendocrinology, 33,* 909-915

Peper, J. S., Brouwer, R. M., Van Baal, G. C. M., Schnack, H. G., **Van Leeuwen, M**., Boomsma, D. I., Kahn, R.S., & Hulshoff Pol, H. E. (to be submitted). Does a twin-brother make for a bigger brain? *Psychological Medicine.*

Peper, J. S., Schnack, H. G., Brouwer, R. M., Van Baal, G. C. M., Pjetri, E., Székely, E., **Van Leeuwen, M.,** Van den Berg, S.M., Collins, D.L., Evans, A.C., Boomsma, D. I., Kahn, R. S. & Hulshoff Pol, H. E. (in press). Heritability of regional and global brain volume at the onset of puberty: a Magnetic Resonance Imaging study in 9-year old twin-pairs. *Human Brain Mapping*.

Polderman, T. J. C., De Geus, E. J. C., Hoekstra, R. A., Bartels, M., **Van Leeuwen, M**., Verhulst, F. C., Posthuma, D., & Boomsma, D. I. (in revision) Attention problems, inhibitory control, and intelligence index overlapping genetic factors: A study in 9, 12 and 18 years old twins. *Neurospychology*

Van Soelen, I. L. C., Van den Berg, S. M., Dekker, P. H., **Van Leeuwen, M**., Peper, J. S., Hulshoff Pol, H. E., & Boomsma, D.I. (in revision). Individual differences in verbal short term memory in 9-year-old twin pairs and their older siblings. *Learning and Individual Differences*.

Published Abstracts

Van Leeuwen, M., Van den Berg, S.M., & Boomsma, D.I. (2007). The genetic and environmental structure of memory in children. *Behavior Genetics, 37*, 770.

Van Leeuwen, M., Van den Berg S.M., Van Beijsterveldt, C.E.M., & Boomsma, D.I. (2005) Classroom separation of twins during primary school. *Twin research and Human Genetics, 8,* 421.

Peper, J.S., Kahn, R.S., **Van Leeuwen, M**., Van Baal, G.C.M., Schnack, H.G., Boomsma, D.I., & Hulshoff Pol, H.E. (2007). Quantitative Genetic Modelling of Brain Volume in Healthy (pre-) Puberty: a Magnetic Resonance Imaging study in 9-year old twin-pairs and their siblings. *European Neuropsychopharmacology*. *17 (S10):* 83.

Peper, J.S., **Van Leeuwen, M**., Schnack, H.G., Van Baal, G.C.M., Brouwer, R.M., Boomsma, D.I., Kahn, R.S., & Hulshoff Pol, H.E. (2007) Puberty related influences on human brain volume: a MRI study in healthy twin pairs and their siblings. *Neuroimage.35 (S57)*

Peper J.S., Zwiers, M.P., **Van Leeuwen, M.,** Van den Berg, S.M., Boomsma, D.I., Kahn, R.S., & Hulshoff Poll, H.E. (2004). Genetic Modeling of Brain Maturation in Puberty: a Longitudinal Study in Healthy Twins *Twin Research*, 7, 372.

Van den Berg, S.M., **Van Leeuwen, M.,** & Boomsma, D.I. (2007). Bayesian analysis of multilevel parent-offspring data in the presence of assortative mating. *Behavior Genetics, 37*, 802.

Van Soelen, I.L.C., Van den Berg, S.M., **Van Leeuwen, M**., & Boomsma, D.I. Genetic and environmental influences in verbal learning abilities in children, *Behavior Genetics, 37*, 804.

Dankwoord

Zonder de hulp van vele anderen was dit proefschrift niet tot stand gekomen. Nu is eindelijk de gelegenheid gekomen om al deze mensen (nogmaals) te bedanken. Allereerst gaat mijn dank uit naar alle tweelingfamilies die belangeloos aan dit onderzoek hebben meegedaan (zie einde van dit dankwoord). Dankzij hun hulp heb ik de data kunnen verzamelen aan de hand waarvan dit proefschrift is geschreven.

Daarnaast wil ik mijn promotoren prof. dr. Dorret Boomsma en prof. dr. Hilleke Hulshoff Pol en mijn co-promotor dr. Stéphanie van den Berg bedanken. Beste Dorret, bedankt voor al je hulp en steun, zeker ook in de beginmaanden van mijn AiO-schap. Dank ook voor je snelle en kundige commentaar op mijn manuscripten en je goede uitleg. Stéphanie, jij hebt mij geleerd zeer kritisch te denken (al viel dat niet altijd mee in eerste instantie). Je stond altijd voor mij klaar, of het nu voor praktische hulp bij een analyse of een luisterend oor was. Bedankt voor al je begeleiding, hulp, motivatie en gezelligheid! Beste Hilleke, bedankt voor al je enthousiastmerende woorden. En zonder jouw hulp was dit project nooit tot stand gekomen.

Ook wil ik graag de leden van de leescommissie bedanken voor de tijd en energie die zij gestoken hebben in het lezen en beoordelen van dit proefschrift. Dr. Eveline Crone, dr. Conor Dolan, dr. Sarah Durston, prof. dr. Chantal Kemner, dr. Tinca Polderman en prof. dr. Joseph Sergeant bedankt! Thank you prof. dr. Nick Martin for the time and effort you've spend to review my thesis.

Verder wil ik het secretariaat, Hannah, Michiel en Natascha, bedanken voor alle secretariële ondersteuning en meer dan dat. Ook gaat mijn dank uit naar de mensen van de helpdesk, voor hun hulp bij softwareproblemen. In het bijzonder wil ik Paul Groot bedanken voor het programmeren van en het actief meedenken over de cognitieve testen. Therèse en Louise wil ik bedanken voor hun hulp bij het uitdraaien van adressenbestanden. Toos, bedankt voor je hulp bij het aanleveren van databestanden. En verder gaat mijn dank uit naar alle stagiaires die hun bijdrage geleverd hebben aan het testen van alle families en schoolkinderen. Jetty, Mechteld, Marjory, Sigrid, Michiel, Rosa, Cornelie, Laura, Marlene, Thom, Marleen, Patricia, Manuela, Myrte, Noortje, Yuri, en Helena, bedankt!

Verder had ik het niet zo naar mijn zin gehad op mijn werk, als ik niet zulke leuke collega's had gehad. Ik wil de AiO's (waarvan er velen nu post-doc zijn) bedanken voor de gezelligheid en warmte tijdens de vele lunches en etentjes waarin ons dagelijks lief en leed werden besproken. Verder zijn er een aantal collega's die ik in het bijzonder wil bedanken:

Bedankt Jiska voor de fijne samenwerking. Ik had het niet beter kunnen treffen. Rosa, mijn langstzittende kamergenoot, bedankt voor de fijne tijd. Je was voor mij een ideale kamergenoot! Verder wil ik mijn andere kamergenoten Frederiek, Inge, Marjolein, Lot en Jenny bedanken voor hun gezelligheid, luisterend oor, en praktische adviezen. Tevens wil ik op deze plaats Inge veel succes wensen met het voortzetten van het project.

Ten slotte wil ik Hil en Gwen bedanken dat ze mij willen bijstaan in deze bijzondere gebeurtenis. Mijn ouders en Hil wil ik bedanken voor al hun liefde, steun en stimulatie. Zonder jullie was ik hier nooit gekomen. Mam, ik vind het ontzettend jammer dat je er deze bijzondere dag niet bij kan zijn. Maar als het enigszins mogelijk is, weet ik zeker, dat je op deze dag met mij meeleeft. En Haim en Daniël, jullie maken dat het elke dag een feest is om na mijn werk weer thuis te komen! Natuurlijk, heb ik nog een heleboel familie en vrienden niet genoemd die mij tot steun zijn geweest. Weet dat ik ook aan jullie denk, nu ik dit dankwoord schrijf.

Marieke, 17 juli 2008

Met dank aan alle tweelingen, hun broers, zussen en hun ouders

in volgorde van aanmelding:

Larissa Jesse Elise* Judith Susan Tim * Bodine Yoshi Sonny *Romy Jaimy Daisy * Emily Anika Lisa * Femke Sietse Jelle * Maaike Leonie Alex * Rick Bas * Glenn Melvin Jordy * Robbert Richard Harrold * Hanneke Chris * Daan Jorien * Luke Daan * Wouter Tjeerd Sjoerd * Christiaan Michiel Eline Matthijs David * Luuk Daan Nikki * Margot Simone Laura * Thomas Yorick Maike * Tom Koen Caroline * Jelle Tim Anouk * Yorrick Amber Boris * Michou Mignonne Maurice * Joyce Jill Britt * Daisy Joyce * Thomas Lukas Marlou * Roos Joep Loes * Hein Victoria Anna * Inge Jenny Rick * Carli Clair Oscar * Sytse Anneke Pytsje-Baukje * Lisa Sanne Rob * Simone Lieske Steven * Kaj Nils Britt * Koen Bart Chantal * Muriël Desirée Reinier * Sep Tara Niels * Gerben Ruben Manon * Douwe Thomas Gijs * Christiaan Manon Ron * Femke Steffie Geeske * Merel Jade Iris * Jesse Joris * Steve Jarl Kelly * Stijn Bram Merel * Paola Claudia Anouk * Muhammed Ali Gozdenur * Daphne Rosaly Kailey * Judith Anne Thijs * Lianne Marije Caroline * Denise Sabine Richard * Tim Mike Mark * Bryan Amy Brandon * Manon Chantal Iris * Ray Swen Renée * Rosanne Tijmen Myrthe * Martin Pascal Nathalie * Guus Dick Wouter * Laura Vera Wilma * Britt Romy Dennis * Thomas Gijs Stef * Joëlle Michelle Amy * Cynthia Debora Rebecca * Rosalie Eline Marjolein * Jannes Willem Klaas * Renneke Janneke Foeke-Jan * Ilse Iris Marije * Joep Dori Loes * Marieke Corine Niels * Simon Isabelle Julia * Mark Lars Niels * Rik Tom * Levi Juul Nino * Mattijs Friso Mirjam * Kas Sam Tess * Rico Koen Lars * Rembert Oscar Julia * Sharon Esmée * Janneke Marloes Femke * Roos Tessel Pol * Loes Emmy Gijs * Pernille Sacha Joep * Denise Nicole Stefanie * Marit Hester Heleen * Anouk Marloes Harm * Dylan Celine Denise *Carlot Jelmer Jolijn * Sterre Tijmen Romy * Rosanne Mattijs * Iris Bram Kim * Rick Coen Marly * Rick Jens Peter * Wilfred Chris Maikel * Jordi Dennis Robin * Thorsten Björn Sven * Melchert Jochem Brenda * Tamara Melanie Stefan * Sophie Lotte Anne * Mariska Vera Mirna * Marre Peter-Maarten Annemijn * Mike Dave Kevin * Mart Roel Annick * Edwin Stefan Kristel * Mandy Ylse Kevin * Sanne Tessa Roel * Rudy Maurice Naomi * Niels Steyn Lisa * Freya Quinta Zoë * Manon Mandy Larissa * Niek Jelle Babette * Casper Rick Ruben * Minouk Chantal Melissa.

Appendices

Appendix I Invitation letter to the parents

Geachte ouder en/of verzorger,

Zoals u weet staan uw kinderen ingeschreven bij het Nederlands Tweelingen Register (NTR). Wij stellen uw deelname aan het NTR-onderzoek zeer op prijs. Met uw hulp kan de bijdrage van erfelijke aanleg aan persoonlijkheid, groei, ontwikkeling, en ziekte worden onderzocht.

Wij benaderen u nu in verband met de start van een nieuw wetenschappelijk onderzoek waarbij het NTR van de Vrije Universiteit (VU) samenwerkt met het Universitair Medisch Centrum Utrecht (UMCU). U en uw kinderen wordt gevraagd mee te werken aan een onderzoek naar de normale ontwikkeling van hersenen en leervermogen bij kinderen. De titel van dit onderzoek luidt: "**Tweelingonderzoek naar de ontwikkeling van brein en cognitie tijdens de pre-adolescentie**".

Voor dit onderzoek zouden wij graag uw tweeling en hun eventuele broertjes en/of zusjes in de leeftijdsgroep 9 tot 14 jaar willen uitnodigen. Het onderzoek bestaat uit twee delen. Op de VU in Amsterdam zullen testen afgenomen worden om de intelligentie en het leervermogen van uw kinderen te bepalen en op het UMCU in Utrecht zal onderzoek worden gedaan naar de structuur van de hersenen. Dit gebeurt met een zogenaamde MRI-scanner. Daarnaast zouden wij het zeer op prijs stellen als uw kinderen genetisch materiaal (DNA, verkregen door middel van een monduitstrijkje), ochtendurine en speeksel zouden willen afstaan voor nader onderzoek. Ook van u zelf zouden wij graag door middel van een monduitstrijkje DNA willen verzamelen. Tevens willen wij u vragen een korte cognitieve test te doen die uw abstract redeneervermogen meet. **Uiteraard is bij dit onderzoek de privacy van u en uw kinderen gewaarborgd.**

In bijgaande folders kunt u meer over het onderzoek lezen:

- Ouderfolder: in deze folder kunt u meer lezen over het onderzoek zelf en de procedures
- Kinderfolder: in deze folder staat het onderzoek voor uw kinderen uitgelegd

Dit onderzoek heeft als doel de normale ontwikkeling bij jonge gezonde kinderen in kaart te brengen. Het is niet de bedoeling om ziektes en/of afwijkingen op te sporen. Mocht er tijdens het onderzoek desondanks toch informatie naar boven komen die aanleiding geeft tot medisch handelen dan wordt u daarvan op de hoogte gebracht. Wij zijn verplicht dit te doen; indien u dit niet wenst, kunt u niet meedoen aan het onderzoek. Het onderzoek zelf is onschadelijk voor de gezondheid. U en uw kinderen zijn uiteraard geheel vrij in uw keuze wat betreft deelname aan het onderzoek. Het onderzoek kan tevens op ieder moment door u of uw kinderen afgebroken worden.

Het onderzoek op de VU zal inclusief pauzes iets meer dan een dagdeel duren. De kinderen worden daar tegelijk getest. Het maken van een hersenscan op het UMCU duurt per kind 40 minuten en zal per familie minder dan 3 uur in beslag nemen (uw kinderen worden na elkaar

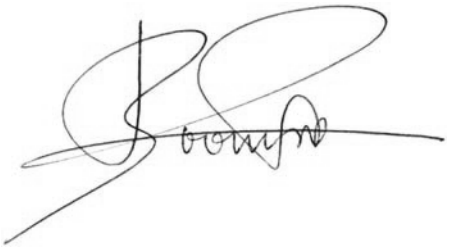
gescand, nadat ze eerst in een oefenapparaat vertrouwd zijn geraakt met de procedure). We zouden het prettig vinden wanneer u op een doordeweekse dag kunt komen, maar zaterdagen zijn ook mogelijk. Voor wetenschappelijk onderzoek kan vrij gekregen worden van school als de directeur van de school daar toestemming voor geeft. Verder zullen uw reiskosten worden vergoed en ontvangen de kinderen na afloop een kleine attentie.

Binnenkort zullen wij contact met u opnemen voor het beantwoorden van vragen die u mogelijk heeft en, indien u mee wilt doen aan het onderzoek, voor het maken van een afspraak. Mocht u na het lezen van de informatie direct al vragen hebben, kunt u zich tot de uitvoerende onderzoekers wenden:

Jiska Peper (UMCU), tel: 030-2507121.
Marieke van Leeuwen, (VU) tel: 020-5988992

Het is ook mogelijk dat u een deskundige wilt spreken die niet direct betrokken is bij dit onderzoek om een onafhankelijk advies te krijgen. Dhr. Prof. dr. Ph. Scheltens, arts, is bereid u daarvoor te woord te staan. U kunt via zijn secretaresse een afspraak maken, tel. 020-5983222.

Wij hopen u en uw kinderen bij dit onderzoek te mogen verwelkomen!
Met vriendelijke groet,



Prof. dr. D.I. Boomsma

Mede namens:
prof. dr. R.S. Kahn
dr. H.E. Hulshoff Pol
drs. Jiska Peper
drs. Marieke van Leeuwen

Bijlagen:

- Ouderfolder
- Kinderfolder

Appendix II Confirmation letter (1) to the parents

Geachte ouder en/of verzorger,

U heeft kort geleden telefonisch een afspraak gemaakt voor het “Tweelingonderzoek naar de ontwikkeling van brein en cognitie tijdens de pre-adolescentie.” Allereerst willen wij u hartelijk danken voor uw medewerking aan ons onderzoek. Tevens willen wij bij deze de door u gemaakte afspraken voor xxxxx om xxxx uur op de VU te Amsterdam en voor xxxx om xxxx uur in het UMC te Utrecht bevestigen. In de bijlage vindt u het programma voor deze dagen.

Voor deelname aan onderzoek is het altijd verplicht dat u een schriftelijke verklaring geeft dat u bereid bent om deel te nemen aan het onderzoek en dat u geheel bent ingelicht over de inhoud ervan. Dit wordt *informed consent* genoemd, of geïnformeerde toestemming. Voor kinderen onder de twaalf is het voldoende dat hun ouders toestemming geven voor deelname. Als kinderen twaalf jaar of ouder zijn, moeten ze tevens zelf toestemming geven. Wij verzoeken u en uw eventuele zonen en/of dochters van twaalf jaar en ouder de bijgevoegde informed consents te ondertekenen en naar ons terug te sturen in de bijgevoegde envelop.

Zoals u heeft kunnen lezen in de informatiefolders willen we voor het onderzoek ook graag over het DNA van u en uw kinderen beschikken. Dit kunt u afstaan door middel van een monduitstrijkje. Daarnaast zouden wij graag van uw kinderen ochtendurine en speeksel (zowel verzameld in buisjes als door middel van het kauwen op wattenrolletjes) ontvangen. Zodra wij van u de informed consent verklaringen hebben ontvangen, krijgt u de uitleg en de benodigdheden hiervoor thuisgestuurd. Wij verzoeken u vriendelijk de monduitstrijkjes, ochtendurine en speeksel mee te nemen naar de afspraak op de VU.

Mochten er nog vragen of onduidelijkheden zijn, dan kunt u altijd contact met ons opnemen. U kunt in het UMCU contact opnemen met Jiska Peper (030-2507121) en op de VU met Marieke van Leeuwen (020-4448992).
Met vriendelijke groet,

drs. Marieke van Leeuwen
drs. Jiska Peper
dr. H.E. Hulshoff Pol
prof. dr. D.I. Boomsma
prof. dr. R.S. Kahn

Bijlagen:

- Dagprogramma met checklist
- Routebeschrijving VU
- Routebeschrijving UMCU
- Retourenenvelop
- Informed consent ouders, 2
- Informed consent kinderen, 3

Appendix III Informed consent parents

tweelingonderzoek naar de ontwikkeling van brein en cognitie tijdens de pre-adolescentie

verklaring van toestemming na kennisneming voor de ouder

Wilt u hieronder tekenen en daarmee het volgende verklaren:

1) De onderzoeker heeft mij volledig ingelicht over de aard en het doel van het "**Tweelingonderzoek naar de ontwikkeling van brein en cognitie tijdens de pre-adolescentie**" en ik ben op de hoogte van de onderzoeksmethoden en procedures.
2) Ik heb de informatie over dit onderzoek, die in de folder en brief worden gegeven, begrepen.
3) Ik heb de gelegenheid gehad vragen te stellen over dit onderzoek.
4) Ik begrijp dat ik te allen tijde de medewerking aan dit onderzoek mag afbreken zonder dat dit ongenoegen zal geven.
5) Ik heb toegestemd om *zelf* deel te nemen aan het onderzoek:

Toestemming voor:
* deelname aan het cognitie-onderzoek ja / nee
* deelname aan het DNA-onderzoek en de opslag en analyse van het erfelijk materiaal ja / nee

Naam:……..... man/vrouw
Geboortedatum:……
Straat en huisnr.:……...
Postcode en plaats:……...
Telefoonnummer:…….

Handtekening:…… Datum: ……..…

Ik, ondergetekende, bevestig hierbij dat deze studie zowel mondeling als schriftelijk aan bovengenoemde deelnemer is uitgelegd.

Naam arts/onderzoeker:……..……

Handtekening:…….. Datum: ……..…….

Appendix IV Informed consent children

tweelingonderzoek naar de ontwikkeling van brein en cognitie tijdens de pre-adolescentie

verklaring van toestemming na kennisneming voor de tweeling / broer / zus

Wil je hieronder tekenen en daarmee het volgende verklaren:

1) De onderzoeker heeft mij volledig ingelicht over de aard en het doel van het "**Tweelingonderzoek naar de ontwikkeling van brein en cognitie tijdens de pre-adolescentie**" en ik ben op de hoogte van de onderzoeksmethoden en procedures.
2) Ik heb de informatie over dit onderzoek, die in de folder en brief worden gegeven, begrepen.
3) Ik heb de gelegenheid gehad vragen te stellen over dit onderzoek.
4) Ik begrijp dat ik te allen tijde de medewerking aan dit onderzoek mag afbreken zonder dat dit ongenoegen zal geven.
5) Ik heb toegestemd om deel te nemen aan de volgende onderzoeken:

Toestemming voor deelname aan:

* het hormoononderzoek op basis van speeksel en urine ja/ nee
* het MRI onderzoek (hersenfoto's) ja/ nee
* de cognitieve en neurospychologische testen ja/ nee
* opslag en analyse van het erfelijk materiaal (DNA) ja/ nee
* ik wil mijn testresultaten graag thuisgestuurd krijgen ja/ nee
* na afloop van dit onderzoek mag ik weer benaderd worden voor vervolgonderzoek ja/ nee

Naam kind:……..……..… jongen/meisje
Geboortedatum:……..……..…
Straat en huisnr.:……..……
Postcode en plaats:……..……
Telefoonnummer:……..…..……

Handtekening:…… Datum: ……
Naam ouder/vertegenwoordiger: ……..……
Handtekening:…… Datum: ….……..…..

Naam (eventuele) tweede ouder/vertegenwoordiger:……

Handtekening:…… Datum: ...……....…..
Ik, ondergetekende, bevestig hierbij dat deze studie zowel mondeling als schriftelijk aan de bovengenoemde deelnemer is uitgelegd.
Naam arts/onderzoeker:……

Handtekening:…….…… Datum: ……...

Appendix V Confirmation letter (2) to the parents

Geachte ouder en/of verzorger,

Allereerst willen wij u hartelijk bedanken voor uw medewerking aan ons onderzoek. Zoals u al heeft kunnen lezen in de vorige brief, ontvangen u en uw kinderen de aanwijzingen voor het verzamelen van het lichaamsmateriaal. In deze envelop treft u de gebruiksaanwijzing en de buisjes voor het verzamelen van DNA door middel van het monduitstrijkje aan.

Uw kinderen ontvangen ieder twee eigen enveloppen die, naast de buisjes en de gebruiksaanwijzing voor het verzamelen van DNA, ook gebruiksaanwijzingen en buisjes voor het verzamelen van speeksel en ochtendurine bevat. Wilt u zo vriendelijk zijn om ze hierbij te helpen?

Het is de bedoeling dat u en uw kinderen het lichaamsmateriaal verzamelen in de week voorafgaand aan uw afspraak op de VU, maandag XX XXXX om xxxx uur. Dit betekent dat u in principe op maandag XX XXXX kunt beginnen met het verzamelen van het materiaal.

Wij verzoeken u vriendelijk alle buisjes mee te nemen naar de afspraak op de VU. Mocht u met de auto komen kunt u hem parkeren op de parkeerplaats van de polikliniek (zie plattegrond van het VU terrein in de routebeschrijving).

Mocht u nog vragen hebben dan kunt u ons overdag bereiken op de volgende nummers:

- Marieke van Leeuwen: 020-598.8992 / 06-51071587 (als u in het weekend een afspraak heeft)
- Liza Lacet en Michiel Verburgh, secretariaat van het NTR: 020 598.8792

Ook kunt u vragen via e-mail sturen naar ntr@psy.vu.nl (onder vermelding van “cognitie-MRI onderzoek negenjarigen”).

Met vriendelijke groet,

Marieke van Leeuwen

Appendix VI Confirmation letter to the children

Beste ,

Allereerst heel erg bedankt dat je mee wilt helpen aan ons onderzoek.

Nu gaat het eerste gedeelte van het onderzoek beginnen. Je gaat speeksel, ochtendurine en DNA van jezelf verzamelen. In deze envelop vind je de gebruiksaanwijzingen, waarin staat uitgelegd waarom dit belangrijk is. Ook staat daarin uitgelegd hoe en wanneer je dat moet doen. Verder vind je in deze envelop de buisjes die je hiervoor nodig hebt.

Lees de aanwijzingen goed door. Als je iets niet helemaal begrijpt, vraag het dan aan je ouders of bel mij overdag (Marieke: 020-5988992). Het is de bedoeling dat je alles verzamelt in de week voordat je naar de VU gaat en het dan meebrengt naar Amsterdam.

Heel veel succes!

Met vriendelijke groet,

Marieke van Leeuwen

Appendix VII Information brochure for the parents

ALGEMENE INFORMATIE OVER HET TWEELINGONDERZOEK

Tweelingonderzoek is een belangrijk wetenschappelijk instrument binnen de geneeskunde en de psychologie. Dankzij onderzoek bij tweelingen kunnen we erachter komen in hoeverre verschillen in gedrag of het krijgen van bepaalde ziektes, worden beïnvloed door iemands genen of door iemands leefomgeving. Om goed onderzoek te kunnen doen hebben we gegevens nodig van veel tweelingen. Bovendien willen we graag die tweelingen volgen gedurende hun ontwikkeling. Daarom is in 1987 aan de Vrije Universiteit (VU) te Amsterdam het Nederlands Tweelingen Register (NTR) opgericht. Bij het NTR staan momenteel 30.000 tweelingen ingeschreven. De meeste gegevens van deze tweelingen zijn verkregen met gedragsvragenlijsten die door de tweelingen zelf of door hun ouders en leerkrachten zijn ingevuld. Verschillende wetenschappers maken gebruik van deze gegevens om zo beter te begrijpen welke invloed genen en omgeving hebben op onder andere de ontwikkeling van de hersenen, intelligentie, leervermogen, probleemgedrag van kinderen, gezondheid en leefgewoonten, angst, persoonlijkheid en veroudering. De belangeloze medewerking van tweelingen en hun families is daarbij van onschatbare waarde!

INFORMATIE OVER HET HUIDIGE ONDERZOEK

Voor dit onderzoek werken het NTR en het Universitair Medisch Centrum Utrecht (UMCU) samen. In het onderzoek wordt gekeken naar de erfelijkheid van hersenontwikkeling en het vermogen om te leren. Hierdoor hopen we in de eerste plaats meer te weten te komen over hoe het komt dat mensen verschillen in hun verstandelijke vermogens. Wordt dit vooral veroorzaakt door verschillen in de genen of vooral door verschillen in de omgeving? Tevens willen we meer te weten komen over de invloed van geslachtshormonen op de ontwikkeling van de hersenen, het verstandelijk vermogen, en over de samenhang tussen ontwikkeling van de hersenen en het vermogen om te leren. We hopen met dit onderzoek meer inzicht te krijgen in de normale ontwikkeling van de hersenen en het verstandelijk vermogen.
Wij willen in dit onderzoek zowel uw tweeling als een broertje of zusje van de tweeling onderzoeken. Wij willen graag eeneiige, twee-eiige tweelingen en hun broertjes en zusjes onderzoeken, omdat door deze combinatie het beste na te gaan is welke eigenschappen met name erfelijk zijn, welke vooral door gedeelde omgevingsfactoren worden veroorzaakt (in hetzelfde gezin opgegroeid) en welke eigenschappen bepaald worden door factoren die voor ieder mens uniek zijn. Dit kunnen we onderzoeken, doordat we weten dat eeneiige tweelingen uit één gezin dezelfde genen (erfelijk materiaal) hebben en in dezelfde omgeving zijn opgegroeid. Verschillen tussen helften van een eeneiige tweeling worden dus veroor-

zaakt door verschillen in de unieke omgeving. Van de broertjes en zusjes van de tweeling weten we dat ze in dezelfde omgeving zijn opgegroeid als de tweeling, maar dat gemiddeld slechts de helft van hun genen gelijk zijn aan die van de andere broers en zussen. Ditzelfde geldt ook voor twee-eiige tweelingen. Door nu zowel de gegevens van eeneiige en twee-eiige tweelingen en hun broertjes en zusjes met elkaar vergelijken, kunnen we berekenen hoe groot de invloed van genen, gedeelde omgeving en unieke omgeving op de ontwikkeling van hersenen en leervermogen is. Wij nodigen uw tweeling zowel op negen- als op elfjarige leeftijd uit om de ontwikkeling van de hersenen en de verstandelijke vermogens te kunnen volgen. Negen jaar is de leeftijd waarop de meeste kinderen de eerste tekenen van de puberteit vertonen en daarom een belangrijke periode in de ontwikkeling van kinderen. We willen ook graag de broertjes en/of zusjes van de tweeling in de leeftijdsgroep 9 tot 14 onderzoeken. Het onderzoek bestaat uit twee delen. In het eerste gedeelte zullen er op de VU testen afgenomen worden om de intelligentie en het leervermogen te bepalen. Op een andere dag zal er, na een kort interview, in het UMCU een hersenscan gemaakt worden. Naast de testresultaten en de hersenscan, willen we graag informatie over hoever de kinderen in de puberteit zijn. Daarom zal er een kort lichamelijk onderzoek plaatsvinden en willen we graag hormoonspiegels bepalen. Tenslotte willen we DNA verzamelen. Dit is enerzijds om te bepalen of de tweeling één- of twee-eiig is en anderzijds voor toekomstig onderzoek naar specifieke genen die ontwikkeling van de hersenen, cognitieve vaardigheden en leervermogen beïnvloeden. Materiaal voor hormoonspiegels en DNA bepalingen kan thuis worden verzameld en worden meegebracht naar de afspraak op de VU. Meer gedetailleerde informatie over de verschillende procedures vindt u hieronder.

INTELLIGENTIE

Iemands intelligentie wordt gemeten door middel van een IQ-test. Deze test bestaat uit een groot aantal vragen, waar zo snel mogelijk het juiste antwoord op moet worden gevonden. Sommige vragen testen het taalbegrip, andere testen ruimtelijk inzicht. De totaalscore op de verschillende onderdelen van de test resulteert in een bepaald getal, het IQ. In dit onderzoek zullen de WISC-III en de Raven Standard Matrices (deze test meet vooral abstract redeneren) worden opgenomen. Aan de ouders wordt ook gevraagd de Raven in te vullen, om te onderzoeken of er een relatie is tussen het IQ van ouders en kinderen en tussen de ouders onderling.

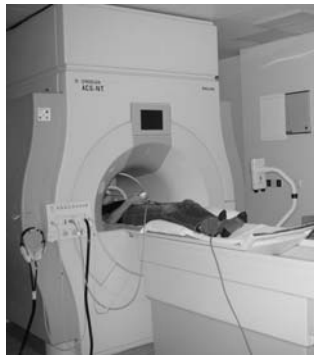
LEERVERMOGEN

We meten het leervermogen met behulp van een aantal taken. Deze taken meten aandacht, verwerkingssnelheid, verbaal vermogen, plannen, geheugen, herkennen van emoties, en reactievermogen. Zo zal er onderzocht worden hoe goed de kinderen een lijstje woorden kunnen onthouden, of hoeveel tijd ze nodig hebben om te zien welke van twee lijnstukken het langste is. Ook wordt er een test afgenomen om te kijken of er mogelijk sprake is van leesproblemen.

Een gedeelte van de taken zal op een computer afgenomen worden. Het andere deel zal door de onderzoekers afgenomen worden. Veel kinderen vinden het leuk om de taakjes zo snel en nauwkeurig mogelijk te volbrengen.

HERSENSCAN

De hersenscan zal gemaakt worden met behulp van een MRI-apparaat. De MRI-techniek is gebaseerd op radiogolven en magnetische velden en is daardoor niet schadelijk voor de gezondheid. Voordat er een MRI-scan wordt gemaakt van de hersenen, krijgen de kinderen een zorgvuldige uitleg over de scanprocedure en kunnen ze alle handelingen oefenen in de speciaal daarvoor bestemde oefenscanner. Vervolgens kunnen ze de eventuele metalen voorwerpen die zij nog bij zich dragen, afdoen in de hiervoor bestemde kleedruimte (persoonlijke eigendommen kunnen veilig worden opgeborgen). Om er zeker van te zijn dat de deelnemers geen metalen voorwerpen meer bij zich dragen op het moment dat zij de scanruimte ingaan, worden hierover door de onderzoeker nog enkele gerichte vragen gesteld. Ten slotte worden enkele meetelektroden op het lichaam geplakt om tijdens het scannen rekening te houden met de hart-slag en kan het scannen van de hersenen beginnen.
Tijdens het MRI-onderzoek liggen de kinderen op een bed. Dit bed wordt door een laborant gedeeltelijk een tunnel ingeschoven. Deze heeft een diameter van 60 cm en is aan beide kanten open (zie foto). Het duurt ongeveer 40 min. om de foto's te maken. Gedurende de scan mag uw kind zich niet bewegen en maakt het apparaat harde geluiden.



Tijdens de scan kan naar een video gekeken worden of naar zelf meegebrachte muziek worden geluisterd. Op ieder moment kunnen de kinderen aangeven te willen stoppen met het onderzoek.

PUBERTEIT

Om vast te stellen of en hoever de kinderen in de puberteit zijn, wordt er op het UMCU een onderzoek uitgevoerd waarbij de lichamelijke ont-wikkeling van uw kinderen vastgesteld wordt. Uw kinderen moeten zich daarvoor uitkleden. Daarnaast vragen we de kinderen thuis ochtendurine en speeksel te verzamelen om de hoeveelheid geslachtshormonen te kunnen bepalen.

CORTISOL

Cortisol is een hormoon dat vooral bekend staat als stresshormoon. Daarnaast is het ook van invloed op de werking van de hersenen. Om de hoeveelheid cortisol te bepalen, vragen we de kinderen twee dagen gedurende de ochtend een aantal keer op een wattenrolletje te kauwen.

DNA

Door middel van het vergelijken van tweelingen met elkaar en met hun broertjes en/of zusjes kan worden nagegaan hoe groot erfelijke en omgevingsinvloeden zijn op bepaalde eigenschappen. Het is voor dit gedeelte van het onderzoek nog niet nodig om het erfelijke materiaal zelf - het DNA -te bestuderen. Als we echter willen weten *welke* genen verantwoordelijk zijn voor verschillen in ontwikkeling van de hersenen en leervermogen, moeten we het DNA bestuderen.

DNA is een onderdeel van chromosomen. Iedere cel in het lichaam bevat 23 paar chromosomen. Van ieder paar wordt er echter maar 1 exemplaar doorgegeven aan het nageslacht. Een kind krijgt dus 1 chromosoom van vader en 1 van moeder. Welke van de twee chromosomen een ouder doorgeeft aan zijn of haar kind is toeval. Als ouders meerdere kinderen krijgen, dan zullen ze soms hetzelfde en soms een ander chromosoom aan beide kinderen doorgeven.

Als we willen onderzoeken welke genen een invloed hebben op een eigenschap zoals leervermogen, dan is het belangrijk om te weten of twee kinderen (bijvoorbeeld een twee-eiige tweeling, of gewone broers/zusjes) van hun vader en moeder hetzelfde chromosoom hebben gekregen, of verschillende. Als kinderen die hetzelfde chromosoom hebben gekregen meer op elkaar lijken dan kinderen die verschillende exemplaren hebben gekregen, dan ligt er op dat chromosoom waarschijnlijk een gen die de eigenschap beïnvloedt. Om de genen te vinden die de ontwikkeling van de hersenen en het leervermogen beïnvloeden, willen we daarom van zowel de tweeling, hun broers en/of zusjes *en* hun ouders DNA verzamelen.

Het DNA van de tweeling zal ook gebruikt worden om te bepalen of de tweeling één- of twee-eiig is. Bij eeneiige tweelingen is het DNA hetzelfde en daarom zien ze er ook precies hetzelfde uit. Bij twee-eiige tweelingen is het DNA net zo verschillend als bij andere broers en zussen die niet tegelijk geboren zijn, daarom lijken ze ook minder op elkaar dan eeneiige tweelingen. Bij een zygositeitstest wordt bekeken of het DNA van de tweeling hetzelfde of verschillend is. De tweelingen krijgen, als ze dat willen, de uitslag van dit onderzoek thuisgestuurd.

Met een monduitstrijkje kan op een eenvoudige en pijnloze manier DNA verzameld worden. Dit wordt gemaakt door met een wattenstaafje zachtjes langs de binnenkant van de mond te wrijven. (De cellen van het wangslijmvlies worden zeer vaak vernieuwd, daarom zijn deze cellen bij uitstek geschikt voor verzameling van DNA.) Dit kan gewoon thuis worden gedaan. De wattenstaafjes kunnen worden meegenomen naar de Vrije Universiteit.

PRIVACY

Voor alle duidelijkheid: alle gegevens zijn strikt vertrouwelijk en vallen onder de strenge privacyregels van de VU en het UMCU. Alle procedures en onderzoeksmethoden uit deze studie zijn getoetst door CCMO (Centrale Commissie Mensgebonden Onderzoek). Voor deelname aan het onderzoek zullen de tweelingen tevens ingeschreven worden in het UMCU. De gegevens die met dit onderzoek worden verzameld, worden altijd vertrouwelijk behandeld. De gegevens worden geregistreerd en verwerkt onder een nummer en dus niet onder een naam of andere persoonlijke gegevens. Ook in publicaties zijn namen niet terug te vinden. Tevens zullen de gegevens de VU en het UMCU niet verlaten. De gegevens kunnen wel worden ingezien door medewerkers die bij het onderzoek betrokken zijn, door de Gezondheidsinspectie en door de Medisch-Ethische Toetsingscommissie.

VERZEKERING

Deelname aan het onderzoek valt onder de aansprakelijkheidsverzekeringen van de VU en het UMCU. De verzekering van het VU medisch centrum loopt bij Onderlinge Waarborg maatschappij Centramed b.a. Bij letselschade kunt u contact opnemen met Dhr. Pardoen op telefoonnummer 070-3762185. Deze verzekering vergoed € 450.000 met een maximum van € 3.500.000 voor het hele onderzoek en een maximum van € 5.000.000 voor alle onderzoeken ver richt bij de VU. De verzekering van het UMCU loopt bij het Marketform Limited te London. De tussenpersoon van deze verzekering is de heer H. Melinga. Deze is te bereiken bij CenE verzekeringen b.v. op telefoonnummer 030-2560406. Deze verzekering vergoedt letselschade die zou kunnen ontstaan door deelname aan onderzoek. De verzekering biedt dekking voor schade tot een maximum bedrag van € 500.000 per deelnemer en € 6.900.000 voor de schade van alle proefpersonen tezamen die deelnemen aan dit onderzoek. Voor de totale schade die zich per verzekeringsjaar bij proefpersonen heeft geopenbaard bij alle onderzoeken die de opdrachtgever ver richt, is het verzekerd bedrag gelimiteerd tot € 9.100.000 per verzekeringsjaar. Voor beide verzekeringen geldt dat uitgesloten van dekking is schade:

a waarvan nagenoeg zeker was dat deze zich bij de proefpersoon zou voordoen;
b die zich bij nakomelingen openbaart als gevolg van een nadelige inwerking van het onderzoek op het genetisch materiaal van de proefpersoon;
c door aantasting van de gezondheid van de proefpersoon die zich ook zou hebben geopenbaard wanneer de proefpersoon niet aan dit onderzoek had deelgenomen;
d die het gevolg is van het niet, niet volledig of foutief opvolgen van de aanwijzingen en instructies die u door de onderzoeker zijn gegeven en welke in de proefpersooninformatiebrief zijn verwoord.

PRAKTISCHE INFORMATIE

- U bent geheel vrij in het al dan niet deelnemen aan (een gedeelte van) het onderzoek.
- Er kan altijd zonder opgaaf van redenen worden gestopt met deelname aan het onderzoek.
- De onderzoekers handelen volgens de gedragscode verzet van de Nederlandse Vereniging van Kindergeneeskunde. Als blijkt dat een van uw kinderen gedurende het onderzoek niet verder wil gaan met het onderzoek, zal het onderzoek met dit kind direct worden afgebroken.
- Na ongeveer twee jaar, als de tweeling de leeftijd van 11 jaar heeft bereikt, wordt er opnieuw contact opgenomen om de procedure te herhalen. Dit wordt gedaan om de veranderingen die in de tussenliggende periode zijn opgetreden in kaart te kunnen brengen. Het is daarom van groot belang om na twee jaar weer mee te doen aan het onderzoek.
- Gegevens die uit het cognitieve onderzoek naar voren komen en die voor eventuele behandeling van belang zijn, worden uiteraard besproken. Mochten er bijkomende bevindingen zijn naar aanleiding van het MRI-onderzoek, waarvoor medisch handelen noodzakelijk wordt geacht, dan wordt de betrokkene (of na toestemming eventueel ook de huisarts) hiervan op de hoogte gesteld.
- Als u een deskundige wilt spreken die niet direct betrokken is bij dit onderzoek om een onafhankelijk advies te krijgen, kunt u terecht bij Dhr. Prof. dr. Ph. Scheltens, arts. U kunt via zijn secretaresse een afspraak maken, tel. 020-4443222.
- De reiskosten die gemaakt zijn, zullen worden vergoed.
- Het is de bedoeling dat de tweeling (plus eventuele broertjes en zusjes) samen op dezelfde dag langskomen om te worden getest of gescand.
- Kinderen krijgen na afloop van het onderzoek een cadeautje.
- Indien u het op prijs stelt worden de belangrijkste resultaten van het wetenschappelijk onderzoek thuisgestuurd. Ditzelfde geldt voor de uitslagen van de testen en een plaatje van de hersenen.
- In de toekomst dus na het onderzoek dat over twee jaar plaatsvindt kunt u benaderd worden voor vervolgonderzoek, tenzij u aangeeft hier geen prijs op te stellen.

MEER INFORMATIE

Het Nederlands Tweelingen Register heeft een website. Surf naar: www.tweelingenregister.org en lees meer over het NTR en het onderzoek dat we doen.Voor praktische informatie over het hier gepresenteerde onderzoek kunt u bellen of e-mailen met de onderzoeksters die u en de kinderen op de VU en het UMCU zullen ontvangen:

Jiska Peper (UMCU):
030-2507121
J.S.Peper@psych.azu.nl

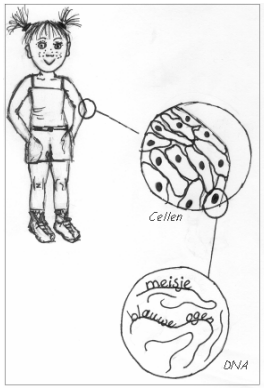
Marieke van Leeuwen (VU):
020-4448992
M.van.Leeuwen@psy.vu.nl

Appendix VIII Information brochure for the children

TWEELINGONDERZOEK

Bij de Vrije Universiteit Amsterdam en de Universiteit van Utrecht begint een nieuw onderzoek bij tweelingen en hun broers en zusjes. We willen graag weten waarom sommige kinderen beter kunnen leren dan andere kinderen en we willen onderzoeken of dit te maken heeft met de bouw en werking van de hersenen. In deze folder kun je lezen waar het onderzoek precies over gaat, en waarom we graag tweelingen met hun broertjes en zusjes willen onderzoeken.

Kinderen lijken op elkaar: de meeste hebben twee ogen, een neus en twee oren. Maar kinderen verschillen ook van elkaar: sommige kinderen hebben blauwe ogen, andere groene; sommige kinderen hebben een grote neus en andere een kleine. Sommige kinderen kunnen heel goed rekenen, en andere kunnen heel goed tekenen of sporten.



Hoe je eruit ziet hangt voor een gedeelte af van je DNA. Je lichaam bestaat uit heel veel kleine cellen en alle cellen bevatten een kern met DNA (zie plaatje). Dit DNA zorgt er bijvoorbeeld voor dat het ene kind blauwe ogen heeft en het andere bruine. Dit DNA komt van je ouders: van je moeder heb je de helft van haar DNA gekregen en van je vader heb ie de helft van zijn DNA gekregen. Daarom lijken broertjes en zusjes op elkaar: voor een gedeelte hebben ze hetzelfde DNA van hun ouders gekregen. Maar niet ieder kind krijgt dezelfde helften van het DNA van de moeder en van de vader en daarom verschillen broers en zussen van elkaar. Zo kan één broer blauwe ogen hebben en een andere broer bruine ogen.

Het bijzondere van eeneiige tweelingen is dat ze precies hetzelfde DNA hebben. Daarom lijken ze zoveel op elkaar. Twee-eiige tweelingen hebben niet precies hetzelfde DNA en ze lijken daarom net zoveel op elkaar als gewone broers en zusjes.

De kleur van je ogen hangt dus af van je DNA. Maar hoe zit het dan met dingen waar je wel of niet goed in bent? Sommige kinderen zijn heel goed in hoofdrekenen, en andere kinderen zijn heel goed in taal. Heeft dat ook te maken met je DNA?

Dat hoeft niet. Niet alle verschillen tussen kinderen hebben te maken met DNA. De verschillen tussen kinderen kunnen ook te maken hebben met dingen die ze meemaken in hun omgeving. Kinderen kunnen bijvoorbeeld andere vriendjes hebben, of een andere schooljuf. Sommige kinderen lezen veel boeken, of sporten liever in plaats van televisie te kijken. Om uit te zoeken of kinderen van elkaar verschillen omdat ze ander DNA hebben of omdat ze andere dingen meemaken, hebben we tweelingen en hun broers en zussen nodig. Als een-eiige tweelingen meer op elkaar lijken dan tweeeiige tweelingen en op hun broers en zusjes, dan heeft dat waarschijnlijk

te maken met het DNA dat bij hen hetzelfde is. Als er geen verschillen zijn in hoeveel eeneiige en twee-eiige tweelingen en broertjes en zusjes op elkaar lijken, dan weten we dat verschillen te maken hebben met wat kinderen meemaken. Verder willen we ook graag weten of tweelingen verschillen van hun broertjes en zusjes. We zouden dus heel blij zijn als jullie mee zouden doen aan dit onderzoek.

HET ONDERZOEK

Dit onderzoek gaat over waarom er verschillen zijn tussen kinderen in hoe goed ze kunnen leren en hoe kinderen veranderen als ze ouder worden. We willen daarom graag een aantal testjes bij tweelingen en hun broers en zussen doen om te zien hoe goed ze kunnen leren, onthouden en lezen. Bovendien willen we foto's maken van hun hersenen. Zo kunnen we zien hoe hersenen veranderen als je opgroeit. Als je nu meedoet is het dus belangrijk dat je over twee jaar nog een keertje meedoet aan het onderzoek, zodat we kunnen zien wat er allemaal is veranderd in de hersenen en in hoe goed je kunt leren. Als je wilt, kun je zelf ook foto's van je hersenen krijgen om misschien aan anderen te laten zien of boven je bed te hangen.

Voor het onderzoek kom je eerst een keer naar de Vrije Universiteit in Amsterdam en dan een keer naar het Medisch Centrum in Utrecht. In Amsterdam krijg je een aantal testjes en in Utrecht maken we foto's van je hersenen en krijg je een leestestje.

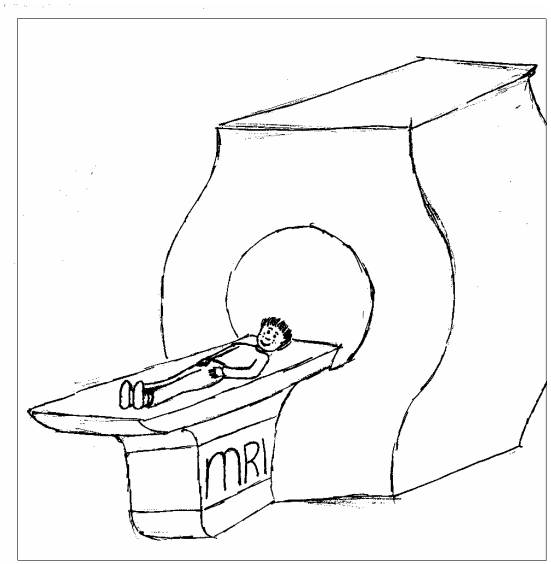
TESTJES IN AMSTERDAM

In Amsterdam krijg je verschillende testjes waarmee we onderzoeken hoe goed je kunt leren en onthouden. Sommige testjes bestaan uit vragen, bij andere gaat het erom dat je zo snel mogelijk reageert, en soms moet je een soort puzzel maken. Een aantal van deze testjes staat op een computer, maar soms worden de vragen ook door de onderzoeker gesteld. Andere testjes doe je weer met pen en papier. De meeste kinderen vinden de testjes leuk om te doen en vinden het ook leuk om zo snel mogelijk te reageren. Het testen duurt een paar uur met een aantal pauzes. Voor eten en drinken wordt gezorgd. Als je wilt, sturen we je later een overzicht van hoe goed je de testjes hebt gedaan.

FOTO'S VAN JE HERSENEN IN UTRECHT

In Utrecht worden foto's van je hersenen gemaakt in een MRI-apparaat, een scanner genoemd. Een scanner is een soort tunnel waar je helemaal in ligt. Als er foto's worden gemaakt voel je er niets van en het is ook niet ongezond. Het apparaat maakt wel wat lawaai. Voordat we de echte foto's gaan maken, kun je eerst uitproberen hoe het is om in een scanner te liggen. Daarom is er ook een oefenscanner die er precies zo uitziet als het echte apparaat. Pas als je helemaal gewend bent aan de oefenscanner gaan we foto's maken. Tijdens het maken van de foto's lig je op een bed dat door de onderzoeker een stukje de tunnel in wordt geschoven. Het is daarom belangrijk dat je niet bang bent voor kleine ruimtes (dat merk je vanzelf tijdens het oefenen). Tijdens het maken van de foto's kun je naar een video kijken of naar muziek luisteren

die je zelf hebt meegenomen. De bedoeling is dat je zo stil mogelijk blijft liggen. Het duurt iets langer dan een half uur om alle foto's te maken.



Je mag op ieder moment aangeven dat je wilt stoppen met het onderzoek. Dit geldt voor alleonderdelen van het onderzoek. Je hoeft niet te zeggen waarom je wilt stoppen

VOLWASSEN WORDEN

Als je volwassen wordt, verandert je lichaam. Dat hoort bij de puberteit. De puberteit begint als in je lichaam hormonen worden gemaakt. Hormonen zijn stoffen die tegen het lichaam zeggen dat het volwassen moet worden. Hierdoor krijgen jongens en meisjes bijvoorbeeld allebei haren onder de armen en rond de geslachtsdelen. Deze hormonen zitten ook in je speeksel en je urine. Om te kunnen meten hoeveel hormonen al aanwezig zijn, vragen we of je wat ochtendplas en speeksel wilt verzamelen. Aan je lichaam kunnen we nog beter zien of je al in de puberteit bent. Bij het bezoek in Utrecht zal daarom een kort lichamelijk onderzoek worden gedaan door een dokter waarvoor je je even uit moet kleden. Het is belangrijk te weten of je al in de puberteit bent, omdat er dan ook dingen veranderen in je hersenen.

DNA

Om te kijken of de tweelingen die meedoen een- of twee-eiig zijn en voor later onderzoek, willen we graag DNA verzamelen bij alle deelnemers. Aan de binnenkant van je wang zitten losse cellen. Als je zachtjes met een wattenstaafje langs de binnenkant van je wang gaat, blijven die cellen op het wattenstaafje achter. Uit deze cellen kunnen wij DNA halen. Voor het onderzoek vragen wij jullie met een wattenstaafje langs je wang te schrapen en dit mee te nemen naar Amsterdam.

TENSLOTTE

Als dank voor je medewerking krijgen jullie aan het eind van het onderzoek een cadeautje en, als jullie dat leuk vinden, krijgen jullie ook de uitslagen van de testen en een foto van jullie hersenen opgestuurd.

MEER INFORMATIE

Als jullie nog vragen hebben, kunnen jullie in de folder van jullie ouders kijken, of bellen of mailen naar de onderzoekers. De onderzoekster in Amsterdam die de testjes doet, heet Marieke van Leeuwen en de onderzoekster die in Utrecht de hersenfoto's maakt, heet Jiska Peper.

Jiska Peper (UMC Utrecht):
030-2507121 / email: J.S.Peper@psych.azu.nl
Marieke van Leeuwen (Vrije Universiteit Amsterdam):
020-4448992 / email: M.van.Leeuwen@psy.vu.nl

Als je liever je vragen wilt stellen aan iemand die niets met dit onderzoek te maken heeft, maar er wel veel van af weet, kun je je vragen stellen aan dokter **Scheltens**. Je kunt hem bereiken door te bellen met zijn secretaresse op telefoonnummer 020-4443222.

Appendix IX Day Program and Checklist

DAGPROGRAMMA

VU AMSTERDAM

Bij aankomst in het "Transitorium"-gebouw van de VU zult u opgewacht worden door een van de medewerkers van dit onderzoek., die u mee zal nemen naar de onderzoeksruimten. Hier krijgt u uitleg over het dagprogramma en kunt u de buisjes met speeksel, ochtendurine en de monduitstrijkjes die thuis zijn afgenomen inleveren. Vervolgens zal er met het onderzoek gestart worden. Afwisselend worden verschillende soorten taken gedaan. Sommige taken bestaan uit het maken van puzzels, andere taken bestaan uit het beantwoorden van vragen. De taken worden van tevoren allemaal duidelijk uitgelegd. Ook de ouders krijgen een korte test.
Na een pauze zal er verder gegaan worden met het leervermogenonderzoek. Afwisselend worden er taken op de computer of met pen en papier gedaan. Wederom worden de taken van tevoren allemaal duidelijk uitgelegd.
Na de lunch, die u door de VU krijgt aangeboden, zal er verder gegaan worden met het laatste deel van het leervermogenonderzoek. Na afloop van het onderzoek ontvangen de deelnemers een verrassing en reiskostenvergoeding.

UMC UTRECHT

Na aankomst bij de hoofdingang van het UMC-U kunt u zich melden bij de receptie. Hier zal men u de weg wijzen naar MRI-wachtruimte 9, waar u opgewacht zal worden door een van de onderzoekers. U krijgt dan een korte uitleg van het onderzoeksprogramma. In het eerste deel van dit programma worden een korte medische vragenlijst en een drie-minuten-durende leesvaardigheidstest afgenomen. Daarna wordt door een arts een kort fysiek onderzoek naar de lichamelijke ontwikkeling uitgevoerd (zie informatiefolder).
Voordat er een MR-scan wordt gemaakt van de hersenen, krijgen de kinderen een zorgvuldige uitleg over de scanprocedure en kunnen ze alle handelingen oefenen in de speciaal daarvoor bestemde oefenscanner. Dit is een oefenapparaat dat precies hetzelfde is als de echte scanner maar waar niet mee gescand wordt. De kinderen kunnen dan wennen aan de kleine ruimte en aan de geluiden. Vervolgens kunnen ze de eventuele metalen voorwerpen die zij nog bij zich dragen afdoen in de hiervoor bestemde kleedruimte (persoonlijke eigendommen kunnen veilig worden opgeborgen). Om er zeker van te zijn dat de deelnemers geen metalen voorwerpen meer bij zich dragen op het moment dat zij de scanruimte ingaan worden hierover door de onderzoeker nog enkele gerichte vragen gesteld. Tenslotte worden enkele elektroden op het lichaam geplakt en kan het scannen van de hersenen beginnen.
De scanprocedure zal per kind ongeveer 40 minuten in beslag nemen. Uw kind staat via een microfoon en een alarmbel in contact met de onderzoekers. Het is belangrijk te weten dat op ieder moment de kinderen kunnen aangeven te willen stoppen met het onderzoek. Ook is het mogelijk dat één van de ouders meegaat de onderzoeksruimte in. Gedurende deze tijd hoeven de kinderen geen taken uit te voeren en wordt hen gevraagd zo stil mogelijk te liggen (afgezien van enkele korte pauzes). Ter ontspanning kunnen ze naar een videofilm kijken of naar (eventueel zelf meegebrachte) muziek luisteren.
Het onderzoeksprogramma voor de tweeling en hun broertje of zusje zal in totaal minder dan 3 uur in beslag nemen. Na afloop kunnen de deelnemers nog een drankje drinken en worden de gemaakte reiskosten vergoed.

Z.O.Z.

CHECKLIST

VU AMSTERDAM

Heeft u het volgende bij u?

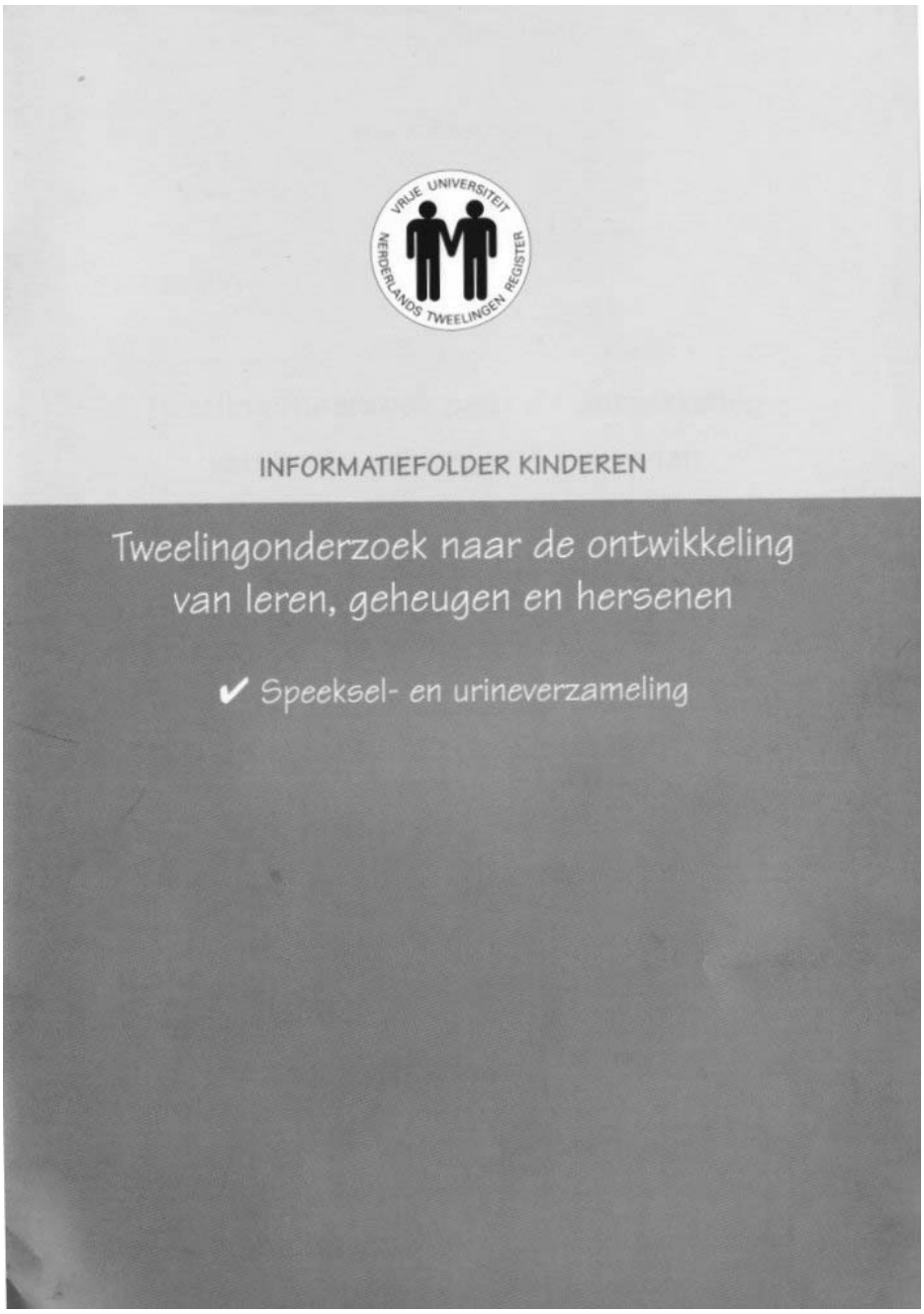
- Uw wanguitstrijkjes en die van uw kinderen
- Speeksel van uw kinderen
- Ochtendurine van uw kinderen

UMC UTRECHT

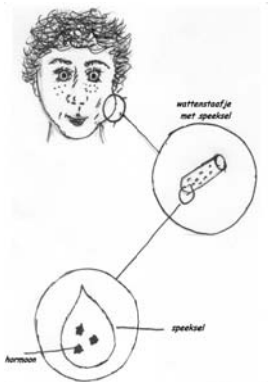
Als u één van de volgende vragen bevestigend beantwoordt, zou u dan contact met ons willen opnemen?

- Heeft uw kind een metalen beugel in de mond die niet te verwijderen is?
- Heeft uw kind een pacemaker-, of hartklepoperatie ondergaan
- Heeft uw kind clips in de bloedvaten van het hoofd?
- Heeft uw kind metaalsplinters (onverwijderd) in zijn/haar hoofd?
- Is uw kind angstig in nauwe ruimtes?
- Heeft uw kind een hydrocephalus- of een insulinepomp die niet te verwijderen is?
- Zijn er stents in het lichaam van uw kind geplaatst?
- Heeft uw kind metalen implantaties, oor of oogprotheses? (b.v. piercings, gewrichtsvervangingen, schroeven, cava filters)
- Heeft uw kind een gehoorapparaat dat niet te verwijderen is, of één of meer metalen oorbuisjes?
- Is er bij uw kind een port-a-cath catheter aangeprikt?

Appendix X Saliva and urine collection instructions



Deze hormonen zitten ook in je spuug (speeksel) en plas (urine). Voor het eerste gedeelte van dit onderzoek vragen wij je op wattenrolletjes te kauwen en in een potje te spugen. Met behulp van het speeksel dat in deze rolletjes achterblijft en wat je uitgespuugd hebt, kunnen wij onderzoeken hoeveel hormonen je in je lichaam hebt. Hiernaast staat uitgelegd hoe je het speeksel moeten verzamelen. Lees de folder goed en laat hem ook door je ouders lezen.



SPEEKSELVERZAMELING

Zoals je in de eerste folder al gelezen hebt, willen we in dit onderzoek weten of je al in de puberteit bent. Hier komen wij achter door te kijken hoeveel hormonen je in je lichaam hebt. Hormonen zorgen dat je lijf zo verandert dat je volwassen wordt.

GEBRUIKSAANWIJZING

WANNEER?

Verzamel het speeksel op twee **doordeweekse** dagen voorafgaand aan de dag dat je op de VU komt om de testen te gaan doen. Tussen de twee dagen dat je het speeksel gaat verzamelen, zit één dag waarop je niet meet. Daarnaast is het belangrijk dat jij en je broer(s) en/of zus(sen) ongeveer tegelijkertijd het speeksel verzamelen.

HOE LAAT?

1. direct als je wakker wordt, als je nog in bed ligt
2. 15 minuten na het wakker worden
3. 30 minuten na het wakker worden
4. 45 minuten na het wakker worden
   Nu kun je ontbijten
5. 12.00 uur (voor het middageten)

HOE?

1. Als je 's morgens wakker wordt, willen we dat je niet alleen op het wattenrolletje kauwt, maar ook dat je op een theelepeltje spuugt (alle andere keren van de dag hoef je alleen nog op het wattenrolletje te kauwen). Laat het speeksel uit je mond op het lepeltje lopen. Giet het speeksel van af het lepeltje in het buisje (zonder watje, met de juiste sticker) en doe het dopje er goed op. **Verzamel ten minste 2 cm speeksel!** (Zorg bij het bewaren dat het buisje rechtop staat.) Nadat je in het potje gespuugd hebt, kauw je op het wattenrolletje.

2. Voor het kauwen op het wattenrolletje willen we dat je op de hiervoor en op de buisjes aangegeven tijdstippen het buisje met het wattenrolletje uit de houder neemt. Het tijdstip dat op de sticker van de houder staat, moet hetzelfde zijn als het tijdstip waarop je het speeksel verzamelt.
3. Je draait de dop van de buis en haalt het wattenrolletje eruit.
4. Dan leg je het wattenrolletje ten minste 45 seconden in je mond. Het speeksel gaat door de gaatjes in het plastic hulsje in het wattenrolletje zitten. Het is erg belangrijk (!) dat het wattenrolletje goed nat wordt. Je kunt hierbij helpen door niet te slikken en goed op het wattenrolletje te blijven kauwen en het rolletje heen en weer te bewegen in je mond van links naar rechts en van boven naar beneden.
5. Stop het wattenrolletje terug in de genummerde buis en draai de dop erop.
6. Vul op het schema in de binnenkant van deze folder in hoe laat je verzameld hebt en of je 30 minuten of korter van te voren gegeten, gedronken (water mag wel) en of je je tanden gepoetst hebt. Vul ook in hoe laat je de dagen voor de verzameling bent opgestaan en hoe laat je gewoonlijk opstaat.
7. Bewaar de gebruikte genummerde buizen (zowel de buisjes met alleen speeksel, als de buisjes met de wattenrolletjes) in de koelkast.
8. Twee dagen later doe je precies hetzelfde.
9. Neem de buisjes mee naar de afspraak op de VU.

Haal het hele middenvel uit de folder, vul het schema (zie punt 6 op bladzijde 6 van de folder) in en neem het mee naar je afspraak op de VU.

Tijdstippen van:	Hoe laat heb je het speeksel verzameld?	Heb je 30 minuten van tevoren gegeten /gedronken/tanden gepoetst?
Speekselverzameling dag 1		
Direct na het ontwaken		
15 minuten na het ontwaken		
30 minuten na het ontwaken		
45 minuten na het ontwaken		
12.00 uur		
Speekselverzameling dag 2		
Direct na het ontwaken		
15 minuten na het ontwaken		
30 minuten na het ontwaken		
45 minuten na het ontwaken		
12.00 uur		

Naam: .....

Ntrid:.....

Op welke dagen heb je verzameld?

Dag 1 .....

Dag 2 .....

Hoe laat ben je op deze dagen opgestaan?

Dag 1 .....uur

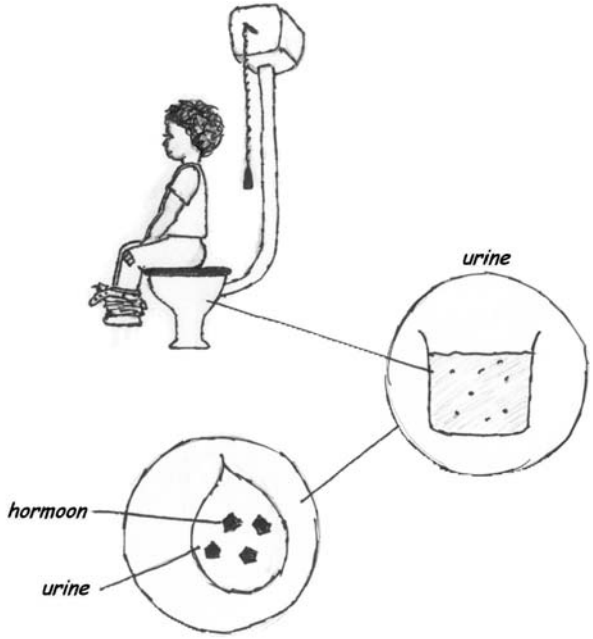
Dag 2 .....uur

Hoe laat sta je doordeweeks gewoonlijk op?

.....uur

ETEN EN DRINKEN

Verder is het erg belangrijk dat je een half uur voordat je op het watten-rolletje kauwt, niets hebt gegeten en geen koffie, thee of fris hebt gedronken. Het is het beste als je pas je ontbijt neemt als je klaar bent met het verzamelen van het ochtendspeeksel. Dit is omdat er anders broodkruimels en ander eten in het speeksel gaat zitten. Je kunt wel gewoon water drinken. Verder is het belangrijk om niet vlak voor het speeksel verzamelen je tanden te poetsen (anders kan er misschien bloed in je speeksel terecht komen). Het beste kun je elke keer voordat je speeksel verzamelt, even kort je mond spoelen met water. Mocht het nu echt niet lukken om tussendoor niet te eten, dan vragen we je tenminste tot nà de derde keer van het speeksel verzamelen te wachten met



OCHTENDURINE

Voor het tweede gedeelte van dit onderzoek vragen wij je om ochtendplas te verzamelen. Met behulp van je ochtendplas kunnen wij onderzoeken hoeveel hormonen je in je lichaam hebt. Hiernaast staat uitgelegd hoe je dat moet doen. Lees het onderstaande verhaal goed en laat het ook door je ouders lezen.

GEBRUIKSAANWIJZING

WANNEER?

Verzamel de ochtendplas op **twee** doordeweekse dagen in de week voorafgaand aan de afspraak op de VU. Doe dit op dezelfde dagen als je je speeksel verzamelt.

HOE?

De eerste keer dat je 's morgens moet plassen, plas je in plaats van in de wc-pot in een schoon potje (bijvoorbeeld een jampotje). Giet de urine over in het buisje met het juiste etiket. Vul het buisje voor 3/4. Draai daarna het dopje goed op het buisje. Bewaar de buisjes rechtop in de koelkast. Neem de buisjes mee naar de afspraak op de VU.

VRAGEN

Als je nog vragen hebt, kun je contact met ons opnemen. Onze gegevens staan op de achterzijde van deze folder.

Veel succes!