Chapter 12

Discussion[1]

[1] This chapter is based on: D Posthuma, EJC de Geus, DI Boomsma. Genetic contributions to anatomical, behavioural, and neurophysiological indices of cognition. To appear in: R. Plomin, J.C. deFries, I.C. Craig, & P. McGuffin (Eds.). *Behavioral genetics in the postgenomic era.* Washington, DC. APA Books. (2002).

This final Chapter summarizes and discusses the results that have been described in Chapters 3 to 11 in the light of existing literature. It is divided in three main sections: the extended twin design, cognitive ability and its endophenotypes, and follow-up research.

The extended twin design

Results from this thesis

This thesis comprises the first large study that employed a twin design with additional non-twin siblings. Effects of adding one or two non-twin siblings to a twin pair on the sample size required to detect additive genetic influences, non-additive genetic influences and shared environmental with a power of 80%, was investigated in Chapter 2. A summary of the results is given in Table 12.1.

Table 12.1
Indication of the effects of adding one or two non-twin siblings to a twin pair on sample size required to detect additive genetic influences (A), non-additive genetic influences (D) or shared environmental influences (C), with a power of 80%.

	Estimating A	*Estimating D*	*Estimating C*
Adding one additional sibling	Slight decrease in sample size	Large decrease in sample size	Large decrease in sample size
Adding two additional siblings	Slight increase in sample size	Large decrease in sample size	Large decrease in sample size

Compared to a design in which only MZ and DZ twins are included, adding one additional sibling leads to an average decrease of 9% (depending on the magnitude of the genetic influences) in sample size needed to detect additive genetic influences (with a power of 80% and alpha level of 0.05). The impact on the detection of shared environmental influences is even larger: on average 50% of the sample size is needed when one sibling is added to the classical twin design. For most complex traits it is found that genetic influences are of greater importance than shared environmental influences (Plomin *et al.*, 2000; Lynch and Walsh, 1996). Small contributions of shared environmental factors may easily go undetected in the classical twin design, which leads to an overestimation of the contribution of genetic factors to the overall variance of a trait. With the same sample size, but with a different design, namely the extended twin design, small contributions of shared environmental factors are less likely to go undetected. Thus, adding an additional sibling to a twin pair enhances the power to decompose familial influences on complex traits into genetic and shared environmental components. In the light of future QTL studies an extended twin design is also desirable as large sibship sizes are known to greatly enhance the power to detect QTL influences on a trait (Dolan, Boomsma and Neale, 1999).

Besides having a positive effect on statistical power, extended twin designs allow the evaluation of certain assumptions made in twin studies. Heritability estimates

derived from twin samples are sometimes criticized for their non-generalizability to the general (non-twin) population. A sub optimal intrauterine environment may have adverse effects later in life (Barker, 1998; Philips *et al.* 2000; Hack *et al.*, 2002). As twins share a womb at the same time, they generally experience a less optimal intrauterine environment than singletons. This is also reflected in lower birth weights in twins and lower birth weights-for-gestational-age in twins as compared to singletons. Studies comparing intellectual abilities of twins and singletons have found that during childhood, twins score significantly less on IQ tests than singletons (e.g. Record *et al.*, 1970). Previous studies, however, compared twins and singletons raised in different families. As was outlined in chapters 3 and 4 this may not be an optimal comparison as twin families and non-twin families may not be perfectly matched. The extended twin design provides an optimal matched singleton to the twins: the non-twin sibling. Non-twin siblings are raised in the same home as the twins; they even shared the same womb, although not at the same time. I found that at adult age, twins and singletons do not differ in mean scores on an IQ test. In Chapters 8 and 9 I also explicitly tested whether the covariance (and correlation) in non-twin sibling pairs differs from the covariance (and correlation) in DZ twins, since this – rather than the mean- is the basis of all heritability estimates. No differences in (co) variance structures of twins and singletons were found. Heritability estimates for IQ derived from twin studies can thus be generalized to the general population.

Using the same optimally matched twin-singleton design no twin-singleton differences in means and variances of intracranial volume, total brain volume, grey matter volume, white matter volume, lateral ventricular volume and third ventricle volume were found, implying that heritability estimates for brain volumes derived from twin studies can be generalized to the general population. We did find that second born twins have a smaller intracranial volume than their first-born co-twins (and siblings), reflecting the less optimal intrauterine environment for second born twins as compared to first born twins. This difference in intracranial volume, however, did not correspond to a difference in intellectual ability later in life.

Chapters 6 to 10 do not explicitly discuss the generalizability of estimates derived from twin samples. However, they all incorporate tests for homogeneity of (co-) variances across twins and their non-twin siblings. A summary of these tests is given in Table 12.2.

Table 12.2
Tested homogeneity in means, variances and covariances across twins and non-twin siblings

		TWIN-SIBLING COMPARISON		
		Mean	Variance	Covariances
COGNITIVE ABILITY	Block design	ok	ok*	ok*
	Letter-number sequencing	ok	ok*	ok*
	Information	ok	ok*	ok*
	Matrix reasoning	ok	ok*	ok*
	Similarities	ok	ok*	ok*
	Picture completion	ok	ok*	ok*
	Arithmetic	twins < non-twin sibling	ok*	ok*
	Vocabulary	ok	ok*	ok*
	Digit symbol coding	ok	ok*	ok*
	Digit symbol free recall	DZ/non-twin siblings < MZ	ok*	ok*
	Digit symbol pairing	ok*	ok	ok
	WAIS-IIIR Full Scale IQ	ok*	ok	ok
	WAIS-IIIR Verbal IQ	ok*	ok	ok
	WAIS-IIIR Performance IQ	ok*	ok	ok
	WAIS-IIIR Verbal Comprehension	ok*	ok	ok
	WAIS-IIIR Working Memory	ok*	ok	ok
	WAIS-IIIR Perceptual Organization	ok*	ok	ok
	WAIS-IIIR Processing Speed	ok*	ok	ok
BRAIN VOLUME	Intracranial volume	2nd born twin < non-twin sibling	ok	ok
	White matter volume	ok	ok	ok
	Grey matter volume	ok	ok	ok
	Lateral ventricular volume	ok	ok	ok
	Third ventricle volume	ok	ok	ok
	Cerebellar volume	ok*	ok	ok
SPEED	Alpha Peak Frequency	ok*	ok	ok
	Perceptual speed	ok*	ok	ok
	Speed of premotor selective response activation	ok*	ok	ok
	Speed of motor selective response activation	ok*	ok	ok
	Decision time	ok*	ok	ok
INHIBITION	Speed of premotor response activation with stimulus-response incongruency	ok*	ok	ok
	Speed of motor response activation & SR incongruency	ok*	ok	ok
	Decision time & SR incongruency	ok*	ok	ok
	Flanker Task Performance & SR incongruency	ok*	ok	ok

* not reported on in this thesis. SR = stimulus-response

Cognitive ability and its endophenotypes

Results from this thesis

Biological, neurophysiological, electrophysiological and behavioural indices of the pathways that connect genes and cognitive ability are called *endophenotypes* of cognitive ability. A summary of the heritability of cognitive ability and its anatomical, electrophysiological and behavioural indices as investigated in this thesis is given in Table 12.3.

Table 12.3
Overview of heritability estimates across two age cohorts of cognitive ability and neurophysiological indices, as investigated in this thesis.

	MEASURE	Young Dutch adults (26 yrs)	Middle aged Dutch adults (50 yrs)
		Heritability	Heritability
COGNITIVE ABILITY	WAIS-IIIR Full Scale IQ	0.86	0.86
	WAIS-IIIR Verbal IQ	0.85	0.85
	WAIS-IIIR Performance IQ	0.69	0.69
	WAIS-IIIR Verbal Comprehension	0.83	0.83
	WAIS-IIIR Working Memory	0.71	0.67
	WAIS-IIIR Perceptual Organization	0.68	0.68
	WAIS-IIIR Processing Speed	0.66	0.66
*BRAIN VOLUME	Intracranial volume	0.89	-
	White matter volume	0.87	-
	Grey matter volume	0.82	-
	Cerebellar volume	0.89	-
SPEED	Alpha Peak Frequency	0.71	0.83
	Perceptual speed	0.46	0.46
	Speed of premotor selective response activation	0.62	0.00
	Speed of motor selective response activation	0.39	0.39
	Decision time	0.43	0.43
INHIBITION	Speed of premotor response activation with stimulus-response incongruency	0.00	0.00
	Speed of motor response activation with stimulus-response incongruency	0.00	0.45
	Decision time with stimulus-response incongruency	0.48	0.48
	Flanker Task Performance (incorrect responses) with stimulus-response incongruency	0.54	0.41

*assessed in the sample (aged 30) of dr. Baaré, which partly overlaps with the young adult age cohort.

The first goal of this thesis was to investigate the heritability of cognitive ability in young and middle aged Dutch adults. Heritability estimates were similar for both age cohorts and were very high. The highest heritability was found for Full scale IQ (86%) and somewhat lower (66-83%) for the dimensions of cognitive ability. These heritability estimates of cognitive ability are among the highest reported for cognitive ability (Bouchard and McGue, 1981).

Combining these results with studies on cognitive ability in Dutch children and adolescents nicely shows the increasing heritability of cognitive ability with increasing age (see Figure 12.1).

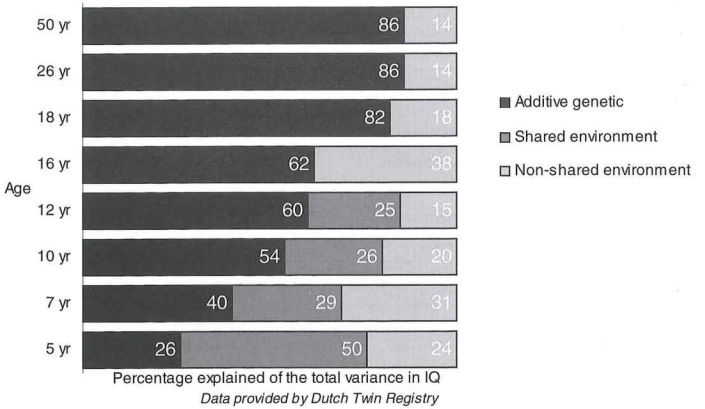

Figure 12.1:
Decomposition of the variance in Full Scale IQ into additive genetic variance, shared environmental variance, and non-shared environmental variance, at different ages in the Dutch population. Significance of additive genetic influences is .03 at age 5; of shared environmental influences it is .06 at age 7 , .09 at age 10 and .08 at age 12.

A second goal of this thesis was to gain more insight in the biological pathways connecting genes and cognitive ability. The heritability of several endophenotypes has been investigated (Table 12.3) as well as their relation with cognitive ability (Table 12.4). Investigated endophenotypes were brain volumes, alpha peak frequency, perceptual speed, speed of premotor and motor response selection, decision time, and inhibitory control.

Table 12.4
Phenotypic correlation between cognitive ability and biological indices, and the percentage of the correlation explained by common genetic factors (homogeneous across both age cohorts, unless specified otherwise).

		FSIQ	VIQ	PIQ	VC	WM	PO	PS
BRAIN VOLUMES	White matter volume	0.24 100%	-	-	ns	0.28 100%	ns	0.25 100%
	Grey matter volume	0.25 100%	-	-	ns	0.27 100%	0.20 100%	ns
	Cerebellar volume	-	-	-	ns	0.27 100%	0.18 66%	ns
SPEED	Alpha Peak Frequency	-	-	-	ns	ns	ns	ns
	Perceptual speed	-	0.19 100%	0.27 100%	-	-	-	-
	Speed of premotor response activation	-	ns	ns	-	-	-	-
	Speed of motor response activation	-	ns	- 0.54[1]	-	-	-	-
	Decision time	-	ns	0.14[3]	-	-	-	-
INHIBITION	Speed of premotor response activation & SR incongruency	-	ns	ns	-	-	-	-
	Speed of motor response activation & SR incongruency	-	- 0.35[2]	- 0.41[1]	-	-	-	-
	Decision time & SR incongruency	-	ns	ns	-	-	-	-
	Number of incorrect responses & SR incongruency	-	-0.35 100%	-0.35 100%	-	-	-	-

[1] only for older females
[2] only for young males
[3] only for older males
FSIQ = Full Scale IQ; VIQ = verbal IQ; PIQ = performance IQ; VC = verbal comprehension; WM = working memory; PO = perceptual organization; PS = processing speed; SR = stimulus-response; ns = not significantly different from zero.
Note: Endophenotypes were either analysed in a multivariate design including Full scale IQ, in a multivariate design including verbal and performance IQ, or in a multivariate design including the four WAIS-IIIR dimensions.

Integrating our results with the recent literature[1]

Brain size

An obvious source of individual differences in cognitive abilities is the size of the brain. Since the second half of the 19th century positive relations between head size and intelligence have been observed. Correlations generally range around 0.20 (Jensen, 1994; Posthuma *et al.*, 2001b), but have been reported as high as 0.44 (van Valen, 1974). Head size is usually measured with a measuring tape as circumference of the head. A more accurate measure of the size of the brain can be obtained through Magnetic Resonance Imaging (MRI).

Willerman *et al.* (1991) correlated brain size as measured through MRI with IQ (measured with the revised Wechsler Adult Intelligence Scale, WAIS-R) in a sample of 40 unrelated subjects. They found a correlation of 0.51, which was higher in men (0.65) than in women (0.35). In a follow-up study, Willerman *et al.* (1992) suggested that, in men, a relatively larger left hemisphere better predicted verbal IQ than it predicted performance IQ, whereas in women the opposite was true. Since then, several studies have provided confirmative evidence that brain volume and IQ correlate around 0.40 (e.g. Egan at al. 1994; Andreasen *et al.*, 1993; Raz *et al.*, 1993; Storfer, 1999; Wickett *et al.*, 2000)

In a large MRI study including 112 twin pairs and 34 additional siblings the heritability of volumes of several brain structures was investigated (Baaré *et al.*, 2001). Heritability estimates for intracranial volume, total brain volume, grey matter volume, and white matter volume were all between 80-90%. Genetic intercorrelations between these measures were all very high indicating that a largely overlapping set of genes is responsible for individual differences in each of these measures.

Two early multivariate genetic studies have been conducted to investigate whether the relation between IQ and brain volumes is mediated through a common genetic pathway or through a common environmental pathway. The first study, often cited although only published as an abstract so far (Wickett *et al.*, 1997), was based on MRI and IQ data from 68 adult males from 34 sibships, and compared within-family correlations with between-family correlations of brain volume and IQ. A within-family correlation of 0.24 and a between-family correlation of 0.50 were reported, suggesting that some, but not all, of the phenotypic correlation between brain volume and IQ is due to a common underlying set of genes.

The second study (Pennington *et al.*, 2000), specifically addressed the relation of reading disorder with brain volume, but also included measures of IQ (WISC and WAIS3-R scores). In this study both a reading disorder sample (25 MZ, 23 DZ) and a healthy sample (9 MZ, 9 DZ) were included. The MZ and DZ correlations in the

[1] See Appendix IX for a summary of the multivariate genetic studies investigating the association between putative endophenotypes and cognitive ability discussed in this chapter.

reading disorder sample and the healthy sample were comparable and suggested high heritability of brain volume (90%), which is in line with the larger study on heritability of brain volumes of Baaré *et al.* (2001). Phenotypic correlations between cerebral brain volume and IQ were 0.31 in the healthy sample and 0.42 in the reading disorder sample. The genetic correlation, as calculated from the cross-twin correlations, was 0.48 in the combined sample. This indicates that about half of the genetic influence on either cerebral brain volume or IQ is due to genetic factors influencing both. Put differently, 80% of the phenotypic correlation is explained by genetic mediation.

In a recent study by Thompson *et al.* (2002), voxel based MRI techniques were used on a dataset of 10 MZ twins and 10 DZ twins. They reported high heritability of grey matter volume in several cortical regions (80-90%), specifically for the frontal, parieto-occipital and linguistic regions. In addition they reported a correlation between the size of these regions and cognition, and suggested that, although their study did not have the statistical power to estimate the relative contributions of genetic and environmental correlations, this correlation was likely to be mediated by an underlying set of genes.

In Chapter 7a of this thesis (Posthuma *et al.*, 2002) it was determined that the observed correlation between brain volume and intelligence is completely mediated by an underlying set of common genes. In a data set of 24 MZ pairs, 31 DZ pairs, and 25 additional siblings a correlation of 0.25 (0.24) between cerebral grey (white) matter volume and Full Scale IQ was found and a correlation of 0.29 between the Working Memory dimension of the WAISIII-R and grey or white matter volume. Results from multivariate genetic modelling showed that these correlations were completely determined by a genetic correlation between the genes that influence brain volume and the genes that influence IQ. In Chapter 7b we included cerebellar volume in the analyses as well as all four dimensions of the WAISIIR. It was found that cerebellar volume was related to Working Memory (0.27) and Perceptual Organization (0.18). One hundred percent of the correlation between cerebellar volume and Working Memory was explained by common genetic factors, while 66% of the correlation between cerebellar volume and Perceptual Organization was explained by common genetic factors.

Alpha peak frequency

Electroencephalographic (EEG) recording is a non-invasive technique to measure electrical activity of the brain. EEG activity can be analysed according to the frequency spectrum that is obtained when a Fourier transformation is performed on an EEG time-series. Generally five frequencies are distinguished in the EEG power spectrum: delta (0.5-4 cycles per second), theta (4-8 cycles per second), alpha (8-13 cycles per second), beta (13-30 cycles per second), and gamma (> 30 cycles per second). In the past decade the underlying biological mechanisms of the different

frequencies, especially the alpha and beta rhythms, are well understood and have been described in the literature (Steriade *et al.*, 1990; Lopes da Silva, 1991).

The dominant frequency in an adult human EEG spectrum lies in the alpha range around 10 cycles per second. The alpha peak frequency has been related to cognitive abilities in general and to (working) memory in particular. Lebedev (1990, 1994) proposed a functional role for the human alpha peak frequency in stating that 'cyclical oscillations in an alpha rhythm determine the capacity and speed of working memory. The higher the frequency the greater the capacity and the speed of memory'. In addition, Klimesch (1997) argued that thalamo-cortical feedback loops oscillating within the alpha frequency range allow searching and identification of encoded information. He speculated that faster oscillating feedback loops would correspond to faster access to encoded information. These theories are supported by the results of some recent studies; Klimesch (1997) found that the alpha peak frequency of good working memory performers lies about 1 Hz. higher than that of bad working memory performers. Anokhin and Vogel (1996) reported a correlation of 0.35 between alpha peak frequency and verbal abilities. In addition it is found that within the same subject alpha peak frequencies increase with increasing cognitive load of the task in which they are measured (Klimesch, 1999).

Results from a few small twin studies have suggested that alpha peak frequency is influenced by genetic factors (Christian *et al.*,1996), and it has also been speculated that its relation with IQ is due to a genetic basis (e.g. Vogel, 2000, page 117). In only one multivariate genetic study, however, the nature of the relation between alpha peak frequency and cognitive abilities is formally investigated. Including 102 MZ pairs and 525 DZ/sib pairs from two age cohorts (mean ages 26 and 50 years).

Chapter 8 of this thesis reports on a study from which it was concluded that that alpha peak frequency is highly heritable (Posthuma *et al.* (2001b). In young adults (mean age 26 years) heritability was estimated at 71%, and in older adults (aged 50 years) heritability was somewhat higher at 83%. Heritabilities for the WAIS-3R dimensions ranged from 66 to 83%. Surprisingly, no correlation was found between alpha peak frequency and IQ (WAIS-IIIR), thereby dismissing alpha peak frequency as a valuable electrophysiological substrate of cognitive ability as measured with the WAIS-IIIR.

Inspection Time

Inspection time is a measure of central nervous system processing and is defined as the minimum display time a subject needs for making an accurate perceptual discrimination on an obvious stimulus. It is distinct from reaction time since there is no need to make the discrimination quickly all that is required is an accurate response. Visual inspection time can easily be measured in a computerized version of the Π-paradigm in which subjects are asked to decide which leg of the Π-figure is longest. Visual inspection time is generally thought to reflect speed of apprehension

or perceptual speed. A meta-analysis conducted by Kranzler and Jensen (1989) including virtually all studies until 1989 investigating the relation between inspection time and intelligence indicated that inspection time and IQ correlate around –0.50: the less time a person needs to make an accurate decision on an obvious stimulus, the higher his score on an IQ test. The overall consensus on the relation between inspection time and IQ is given by Deary and Stough (1996): "inspection time accounts for approximately 20% of intelligence-test variance".

Recently two large twin studies have investigated whether the relation between inspection time and IQ is mediated by shared genetic factors or by shared environmental factors (Luciano *et al.*, 2001a; Posthuma *et al.*, 2001; Chapter 9 of this thesis). These two studies were also the first to report on the heritability of inspection time per se. Using 184 monozygotic (MZ) pairs and 206 dizygotic (DZ) pairs aged 16, Luciano *et al.* (2001a) reported a heritability estimate of inspection time of 36% and of IQ-measures between 73-81%. In chapter 9 (Posthuma *et al.* 2001a) we reported a slightly higher heritability estimate of inspection time (46%) and similar heritability estimates of IQ measures (WAIS-IIIR) ranging from 69-85%. The latter sample consisted of 102 MZ pairs and 525 DZ/sib pairs belonging to two age cohorts (mean ages 26 and 50).

Luciano *et al.* (2001a) reported a correlation between inspection time and performance IQ of -0.35 and between inspection time and verbal IQ of -0.26. In this thesis slightly lower correlations were reported; -0.27 and -0.19 respectively (Posthuma *et al.*, 2001a). Both studies unanimously found that the phenotypic correlations between inspection time and performance IQ/verbal IQ were completely mediated by common genetic factors. This meant that in the study by Luciano *et al.* (2001a) the genetic correlation between inspection time and performance IQ was -0.65 and between inspection time and verbal IQ was -0.47 . In our own study (Posthuma *et al.*, 2001) the genetic correlations were -0.47 and -0.31 respectively. Thus, the genes shared with inspection time are, across studies, estimated to explain between 10 and 42% of the total genetic variance in IQ.

Speed of premotor and motor response selection activation

Speed of premotor and motor response selection activation can be measured with the lateralized readiness potential (LRP). The LRP is mathematically derived from the readiness potential (RP, Kornhuber and Deecke 1965), an evoked potential that can be observed in an EEG registration. The LRP onset is considered to reflect the output of the response selection stage (Coles, 1989; Eimer, 1998) and to be closely time-related to central decision processes. The time of maximal LRP amplitude, LRP peak latency, is thought to additionally reflect central motor processes that take place after response selection has taken place (Falkenstein *et al.*, 1994). The LRP can be calculated from the EEG registration during the execution of any task that requires the selection of either right or left hand responses.

In a dataset of 102 MZ pairs and 525 DZ/sib pairs from two age cohorts (mean ages 26 and 50 years), we determined LRP onset and peak latencies (see Chapter 10 of this thesis and Posthuma *et al.*, (2002b). An inconsistent pattern of heritabilities (ranging from 0 – 62%) was found, and no correlations between these and IQ as measured with the WAIS-IIIR.

Reaction times

Galton (1883) was the first one to propose that reaction time is correlated with general intelligence and may be used as a measure of it. His observations and the results of empirical studies afterwards led to the general belief in the speed of processing theory of intelligence; the faster the accomplishment of basic cognitive operations the more intelligent a person will be (Eysenck, 1986; Vernon, 1987). Since then reaction times have consistently been negatively related to intelligence (e.g. Vernon, 1987; Deary *et al.*, 2001), i.e. a shorter reaction time corresponds to a higher IQ. Correlations with IQ generally range between –0.20 and –0.40, but can be as high as –0.60 (Fry and Hale, 1996). Increasing the information processing load of a task results in prolonged reaction times within in the same subject (Hick, 1952; Eriksen and Eriksen, 1974) and decreases performance. Higher correlations between reaction times and IQ, therefore, are more likely to be found when more complex reaction time tasks are used, although this effect is not unequivocally confirmed in empirical studies (Mackintosh, 1986).

Results from twin studies suggest heritabilities for reaction time of the same magnitude as those for IQ. McGue and Bouchard (1989) observed heritabilities of 54 and 58% for basic and spatial speed factors in a sample of MZ (N=49) and DZ (N=25) twins reared apart. For a general speed factor based on eight complex reaction time tests Vernon (1989) found a heritability of 49% in 50 MZ and 52 DZ twins. In the same study it was also found that reaction time tests requiring more complex mental operations show higher heritabilities. A bivariate analysis of these data with IQ in 50 MZ and 32-SS DZ pairs (15 to 57 years) was reported by Baker *et al.* (1991). Phenotypic correlations of Verbal and Performance IQ with general speed were both -0.59 and were entirely mediated by genetic factors. Genetic correlations were estimated at -0.92 and -1.00. This is in line with results from an earlier study in which phenotypic correlations between reaction time (measured as the total number of correct responses on a timed task) and IQ ranged between 0.37 and 0.42, from which 70-100% was attributed to genetic factors influencing both reaction time and IQ (Ho *et al.*, 1988).

More recently, Rijsdijk *et al.* (1998) conducted a multivariate genetic analysis on reaction time data and IQ data, using 213 twin pairs measured at ages 16 and 18. Heritabilities were reported for age 16 of 58%, 57%, and 58% for simple reaction time, choice reaction time and IQ (RAVEN) respectively. Phenotypic correlations of simple reaction time and choice reaction time with IQ were –0.21 and –0.22 respectively and were completely mediated by common genetic factors. Virtually the same picture was shown at age 18 where the same reaction time battery was correlated with IQ as measured with the WAIS.

Finkel and Pedersen (2000) investigated the underlying covariance structure of measures of speed and measures of cognition in a sample of 292 reared together and reared apart MZ and DZ twins (aged 40-84). Speed was measured by oral versions of the Digit Symbol and Picture Identification subtests of the WAIS. A cognitive factor was constructed based on several standard IQ tests. The phenotypic correlation between the speed factor and the cognitive factor was 0.66, of which 61% was due to correlated genetic factors between the two. Also, they reported that 70% of the genetic variance in the cognitive factor was shared with the speed factor.

Neubauer *et al.* (2000) reported heritability estimates of reaction time data and IQ (RAVEN) ranging from 11-61% and 39-81% respectively. Phenotypic correlations between reaction time data and IQ (RAVEN) data were between –0.08 to –0.50, where higher correlations with IQ were found for more complex reaction time tasks. Common genetic influences on reaction time and IQ accounted for 65% of the observed phenotypic correlation.

Evidence for a genetic mediation between reaction time and IQ also emerges from a recent large twin study by Luciano *et al.* (2001b). Using reaction time data from a two-choice reaction time (2CRT) task, a four-choice reaction time task (4CRT), an eight-choice reaction time task (8CRT) and IQ data from 166 MZ pairs and 190 DZ pairs Luciano *et al.* (2001b) report high heritabilities for reaction times (CRT: 52%, 4CRT: 59%, 8CRT: 70%) and IQ (81%), and moderately high phenotypic (–0.32 to –0.55. The genetic contributions to the observed correlations were all 100% (i.e. were as high as the observed correlations; from –0.32 to –0.49) except for the 4CRT where the genetic contribution covered 89% of the observed correlation. In other words, common genetic influences explained at least 89% of the observed phenotypic correlation between reaction times and IQ.

In Chapter 10 of this thesis the heritability of reaction time (as indexed by decision time) was investigated in a large sample of twins and siblings (see also Posthuma *et al.*, 2002b). To be comparable to reaction times measures used in previous studies, reaction times are best studied in the congruent condition of the Eriksen Flanker Task that was described earlier. The heritability of reaction time was moderate (43%), but no correlation with cognitive ability was found on the phenotypic, genetic or environmental level.

In summary, non-genetic studies have shown a consistent and stable relation between reaction time and IQ; interindividual variance in reaction time seems to explain at most 30% of IQ test variance. Results from previous genetic studies, have suggested that between 65 and 100% of this covariance is explained by a common underlying genetic mechanism. Results from our own, relatively large, study show no

evidence of a covariance between reaction time and IQ, possibly because accuracy may have been stressed more than speed.

Indices of components of working memory

Working memory is considered a central component of cognitive functioning (e.g. Kyllonen and Chrystal, 1990; Baddely, 1986). Some behavioural measures of working memory functioning have already been investigated in a multivariate genetic design simultaneously with a measure of IQ. For instance, Ando *et al.*, (2001) used a spatial and verbal working memory task which were revised versions of the working memory tasks developed by Shah and Miyake (1996). These two tasks generate performance scores on four conditions: spatial storage (Ss), spatial executive (Se), verbal storage (Vs) and verbal executive (Ve). IQ was assessed with the Kyodai NX15 Japanese intelligence scale, form which two components were calculated: Verbal cognitive ability (VCA) and Spatial cognitive ability (SCA). Their data set consisted of 143 MZ's and 93 DZ's aged 16 – 29 years. The working memory performance measures were all moderately heritable (Ss 45%; Se 49%; Vs 48%; Ve 43%) and heritability of IQ was slightly higher (VCA: 65%; SCA 65%). VCA correlated around 0.30 to the working memory measures, whereas SCA correlated around 0.40 to the working memory measures. Genetic contributions predominantly (>85%) explained the observed correlation between IQ and working memory performance.

Luciano *et al.* (2001) determined the heritability and relation with IQ of accuracy on a delayed response task. This task has been adapted from animal research and is widely used as an index for working memory (Goldman-Rakic, 1996). Accuracy on this task was moderately heritable (48%), correlated around 0.20 with IQ, and 100% of this correlation was explained by common genetic factors. Similar results were recently obtained by Wright *et al.*, (2002) after this dataset had been extended by many newly added twin pairs.

In Chapter 10 we reported on the heritability of latency of response selection, reaction times and performance on the incongruent condition of the Flanker task. This task requires a left or right hand response depending on the stimulus. The stimulus can either be congruent with the response or incongruent with the response. Stimulus-response incongruency slows response times and LRP latencies and increases the number of incorrect responses. (Turken and Swick, 1999: Botvinick *et al.*, 1999; Awh and Gehring, 1999). Latencies and performance in the incongruent condition are often thought to reflect inhibition ability which is one of the major function of the frontal executive component of working memory (Baddeley, 1996). Latencies in the incongruent condition showed no or low heritability, whereas decision time was moderately heritable (48%). LRP latencies and decision time in the incongruent condition did not correlate with cognitive ability. However, performance on the incongruent trials was moderately heritable (54% in young adults aged 26; 41% in older adults aged 50) and correlated well to IQ (around 0.30 – 0.40). This correlation was completely explained by common genetic factors.

Follow up research

Endophenotypes of cognitive ability

Summarizing the above it can be stated that genetic variability in brain volume, perceptual speed and frontal inhibitory control is related to genetic variability in cognitive ability.

Insight into the pathways between genes and cognitive ability is not only important in understanding individual differences in normal cognitive functioning, but may also provide clues into the underlying mechanisms of impaired cognitive ability. Diverging conditions as reading disorder, schizophrenia, ADHD, depression, alcoholism, and dementia all share significant deficits in cognitive ability (Willcutt *et al.*, 2001; Harvey 2001; Bray and Owen, 2001; Austin, Mitchell and Goodwin, 2001: Braver, Barch, Cohen, 1999; Goldman-Rakic, 1999).

Another important advantage of identifying pathways between genes and cognitive ability is that they may aid in the detection of the actual genes or quantitative trait loci (QTL's) that influence cognitive ability. Although cognitive ability shows very high heritability, it is still a complex trait, likely to be influenced by a number of QTL's (Plomin and Crabbe, 2000). Its high heritability may reflect the summed genetic effects of a number of QTL's which each exert only small effects (in the order of explaining less than 2% of the total variance in cognitive ability). Biological substrates that are genetically correlated with cognitive ability each explain part of the variance in cognitive ability (10-20%), and may thus each mediate the influence of a small subset of the genes that influence cognitive ability. These subsets of genes will explain a large part of the variance in the biological substrate or endophenotype and a relatively smaller part of the genetic variance in cognitive ability (De Geus and Boomsma, 2002). Including these endophenotypes in studies aimed at the detection of genes that influences cognitive ability may therefore enhance the chances of detecting these genes (Boomsma *et al.*, 1997; de Geus & Boomsma, 2001; Leboyer, 1998).

Especially when a genetic correlation between the genes that influence the endophenotype and cognitive ability reflects an underlying causality from endophenotype to cognitive ability, (as opposed to reflecting pleiotropic effects of genes), QTL effects on endophenotypes will be greater than effects of these QTLs on cognitive ability, as the latter are a function of the QTL effects on the endophenotype and the effect of the endophenotype on cognitive ability. As stated in Chapter 2, genetic correlations do not provide information on causal relations. Direction of causation, however, can be assessed using longitudinal designs, but also with cross-sectional twin designs, provided the two traits show different heritabilities, and provided the validity and reliability of the measurements are known (Heath *et*

al., 1993; Neale *et al.*, 1994; Duffy and Martin, 1994). For example, perceptual speed showed moderate heritability while cognitive ability was highly heritable. Direction of causation between these measures has not been resolved yet. Combining the datasets on perceptual speed and cognitive ability from the Australian researchers (Luciano *et al.*, 2001) and ourselves (Posthuma *et al.*, 2001) will provide an excellent opportunity to resolve the direction of causation between perceptual speed and cognitive ability.

Candidate genes for cognitive ability

Results from human linkage and associations studies for IQ have not yet identified "genes for IQ", although results from the IQ-QTL project (Plomin *et al.*, 1994, 1995; Daniels *et al.*, 1998) have initially provided some evidence for the association of cognitive ability with the alcohol dehygrenase 5 marker (ADH5), nerve growth factor beta polypeptide marker (NGFB), dystrophia myotonica marker (DM) (Petrill *et al.*, 1996), insulin-like growth factor-2 receptor marker (IGF2R) (Chorney *et al.*, 1998), and markers D4S2943, MSX1 and D4S1607 on chromosome 4 (Fisher *et al.*, 1999). Recently, however, Plomin *et al.*, 2001, conducted a genome scan for cognitive ability, using 1842 markers across the genome. They employed an extremely conservative approach (i.e. a five stage design with three samples) to guard against false positives and false negatives. Such a conservative approach seems necessary given the large number of tests that are employed and the problems in replicating QTL association studies (Cardon and Bell, 2001). Using these criteria, Plomin *et al.*, (2001) could not replicate any of the previously found QTL associations, and did not detect new QTL associations.

An alternative approach for screening the genome for possible QTL's is to pre-select candidate genes (de Geus *et al.*, 2001). The results from this thesis suggest that genes important for cerebral grey matter volume, cerebral white matter volume, cerebellar volume, perceptual speed and frontal inhibition may also be important genes for cognitive ability. The genetic connection between brain volumes, neural speed and cognitive ability fits very well in the myelination hypothesis as formulated by Miller (1994). According to this hypothesis, generally, the relation between speed and intelligence can be explained if part of the interindividual variance in cognitive ability can be ascribed to interindividual variance in the degree of myelination of cortico-cortical connections. If true, this could explain why more intelligent brains show faster nerve conduction, faster reaction times and faster inspection times. And, all other things equal, thicker myelin sheaths will result in larger brain volume, thus explaining the positive relation between brain size and IQ (Miller, 1994).

Although it is unlikely that the myelination hypothesis explains all observed anatomical, behavioural and neurophysiological relations with cognitive functioning (e.g. it does not directly explain the link with frontal inhibition), it may provide theoretical guidance in the choice of candidate genes for cognition. As also became

evident from Chapters 7 and 9, genes important for myelination and development of the brain may also be of importance for cognitive ability. Candidate genes important for myelination as mentioned in those Chapters were the *Plp* gene (Boison and Stoffel, 1994; Griffiths *et al*, 1995, Ikenaka and Kagawa, 1995; Lemke, 1993), the *cgt*-gene (Stoffel and Bosio, 1997), the *MAG* gene (Fujita *et al.*, 1998, Sheikh *et al.*, 1999), and the *tn-r* gene (Weber *et al.*, 1999).

An important new finding in this thesis was that human cerebellar volume is highly heritable and that its relation to cognitive ability is mainly mediated through a common genetic pathway. Some genes known to be important for cerebellar development derived from mouse studies were reported in Chapters 6 and 7; the Pax2 locus and the En-2 locus. Favor *et al.* (1996) showed that in mice, functioning of the Pax2 locus, which has its counterpart in the human PAX2 locus, is absolutely necessary for the normal development of the cerebellum. Millen *et al.* (1994) reported a reduction in cerebellar volume in mice due to dysfunctioning of the En-2 locus.

More recently Airey, Lu, and Williams (2001) conducted a full genome screen for cerebellar size in mice and reported linkage with five QTL's. They proposed a set of candidate genes lying within the linkage regions, which also confirmed the role of the Pax2 gene in cerebellar development. Human homologous chromosomal regions of the five QTL's in mice as reported by Airey, Lu, and Williams (2001), are 1q23-43, 10q11-23, 9q13-q24, 11q12-q13, 10q23-qter, 16q12-22.

Genes that may be important for frontal inhibitory control cannot be directly derived from genes investigated with the help of animal models, as there are no animal behavioural models of frontal inhibition. The COMT gene (Lachman *et al.*, 1996) has repeatedly been linked to frontal executive functioning in humans (Egan *et al.*, 2001; Weinberger *et al.*, 2001 for a review), which encompasses frontal inhibition functioning. This gene has also been associated with increased risk of schizophrenia (see Weinberger *et al.*, 2001) which is characterized by impaired executive functioning.

Morley and Montgomery (2001) extensively reviewed the existing (human and animal) literature for candidate genes for human cognition. They indexed published results from *Drosophila melanogaster*, mice and human studies in four phenotypic categories: memory, learning, cognition, and mental retardation. Over 150 candidate genes were derived that may all influence aspects of human cognition. Morley and Montgomery (2001) selected 36 candidate genes for which the literature review provided strongest evidence. Most of these candidate genes are associated with aspects of memory or learning. Very promising candidate genes are the N-methyl-D-aspartate (NMDA) receptor genes NMDA1, NMDA2A, NMDA2B. NMDA receptors are widely distributed in the brain and play a major role in long term potentiation (LTP) (Bear and Malenka, 1994), which is thought to be a cellular substrate of memory (Miller and Mayford, 1999; Eichenbaum and Harris, 2000).

Several studies have shown that mice lacking NMDA induced LTP show substantial learning deficits and spatial memory impairments (Tsien, Huerta en Tonegawa, 1996; McHugh *et al.*, 1996). Alternatively, Tang *et al.*, (1999) showed that overexpression of NMDA2B in the forebrains of transgenic mice results in a superior ability in learning and memory.

Other studies have recently identified genes or found evidence for linkage for human abilities related to IQ: speech and dyslexia. Lai *et al.*, (2001) found that a point mutation in a coding region within the FOXP2 gene on chromosome 7 is related to severe disruption of speech and language. In fact, Lai *et al.* (2001) suggested that this mutation is *causally* related to development of the neural substrate that underlies language and speech. The FOXP2 gene may also be related to aspects of IQ, or at least reveal genetic pathways important for e.g. verbal abilities.

In summary, candidate genes may be selected using an endophenotype approach, using animal models, or may be derived from human studies on traits related to cognitive ability. Whatever candidate genes are selected, they need to be analysed using optimal statistical methods. In Chapter 11 an existing powerful method for the simultaneous analysis of linkage and association (Fulker *et al.*, 1999) was extended to include variable sibship sizes, estimates of spurious and non-spurious dominance effects, and situation where parental genotypes are unavailable. This method also allows fine mapping, as the effect of linkage will be reduced when estimated in the presence of association, thereby providing information on the distance between the marker and the QTL (Cardon and Abecasis, 2000). An explicit test for the effects of population stratification is also incorporated in this method, thereby allowing to distinguish spurious allele effects from genuine allele effects. In Chapter 11 the translation of this method from the theoretical level to the practical implementation in structural equation modelling software was discussed.

With our current armament of behavioural and biological indices of cognitive ability, obtained in a dataset optimal for linkage and association analyses, we are well equipped to start searching for genes for cognition.