Attention-deficit hyperactivity disorder (ADHD) is the most common, highly heritable childhood-onset psychiatric disorder. A high degree of inattention with or without hyperactive-impulsive behaviour results in impaired social and academic functioning (American-Psychiatric-Association, 1994). It is generally accepted that there are three behavioural subtypes which are an inattentive subtype, a hyperactive/impulsive subtype and a combined subtype (Wood et al., 2009). The estimated worldwide prevalence of ADHD is 5.3% in children and adolescents (Polanczyk et al., 2007). The frequency of ADHD declines with age but persists into adulthood, affecting between 2.5% and 4.4% of adults (Fayyad et al., 2007; Kessler et al., 2005; Simon et al., 2009). Boys are more likely to have ADHD than girls (Pastor and Reuben, 2008), and similarly 4.1% of men compared with 2.7% of women have adult ADHD (Fayyad et al., 2009).

ADHD in childhood overlaps with the diagnosis of childhood conduct disorder (CD) and there is considerable epidemiological and family data which shows increased risks for substance abuse (Faraone et al., 2007), unipolar affective disorder (Biederman et al., 1991b, 2009), bipolar disorder (Biederman et al., 1997, 1991a, 2003, 1987; Jaideep et al., 2006; Masi et al., 2006), autism (Reiersen et al., 2007), unipolar affective disorder (Biederman et al., 1997, 1991a, 2003, 1987; Jaideep et al., 2006; Masi et al., 2006), autism (Reiersen et al., 2007) and dis-social personality disorder in the relatives of ADHD probands (Hofvander et al., 2009).

There is a high degree of comorbidity between ADHD and other disorders. Anti-social personality and affective disorders are very strongly associated with alcoholism and substance abuse independently of ADHD (Merikangas and Gelernter, 1990; Merikangas et al., 1985; Ohlmeier et al., 2008; Pottenger et al., 1978; Preisig et al., 2001). Both of these disorders are also associated with ADHD and it is no surprise that childhood ADHD predicts adult alcoholism and substance dependence (Clure et al., 1999; Mannuzza et al., 1991; Milin et al., 1991; Ohannessian et al., 1995; Ohlmeier et al., 2008). There is accumulating evidence that one subgroup of childhood ADHD is likely to be a variant of bipolar disorder (Henin et al., 2007; Hirshfeld-Becker et al., 2006; Rubino et al., 2009) and that another subtype may be related to CD and anti-social personality disorder (Zhou et al., 2008a). Reading disability also appears to share the same genetic cause as a proportion of ADHD cases (Loo et al., 2004). The association of ADHD with autistic traits in twins (Reiersen et al., 2007) and in families seem to define...
a genetic subgroup of ADHD associated with increased neuro-developmental and oppositional defiant and conduct disorders (Mulligan et al., 2009).

Adoption, twin and family studies (Kuntsi et al., 2004; Lasky-Su et al., 2008a; Mick et al., 2009; Ouelet-Morin et al., 2008; Rietveld et al., 2003, 2004; Sprich et al., 2000; van’t Ent et al., 2007) show that there is a strong genetic aetiology for ADHD. Twenty twin studies of ADHD have been consistent with an average heritability rate of 76% (Faraone et al., 2005). The twin studies are able to differentiate subgroups of ADHD. One of these studies (Coolidge et al., 2000) examined the heritability and comorbidity of ADHD with CD, oppositional defiant disorder (ODD) and executive function (inattention) deficits. Heritabilities were high, 82% for ADHD, 74% for CD, 61% for ODD and 77% for executive function. Multivariate twin analyses showed that ADHD shared genetic liability with CD, ODD, and executive function deficits and that additional genetic influences underlying CD, ODD, and executive function that are independent of ADHD may also exist. Another twin study showed that oppositionality overlapped with hyperactivity-impulsivity (r = 0.95) more than with inattentiveness (r = 0.52). The study found that aetiological influences on hyperactivity-impulsivity were shared with those on oppositionality, by contrast inattention as a dimension tended to be more self contained (Wood et al., 2009).

McLoughlin and others (McLoughlin et al., 2007) studied twins and found that hyperactive-impulsivity and inattention substantially shared genetic overlap but that there were also significant independent genetic effects. They predicted that many genes associated with the hyperactivity-impulsivity dimension would also be found to be associated with inattention but that there would be significant genetic heterogeneity.

At least 25% of adults with a history of hyperactivity are the biologic parent of a child with hyperactive symptoms (Biederman et al., 1990; Zametkin et al., 1990). The fact that ADHD has a high population frequency, that twin studies show clinical heterogeneity and that linkage analyses in families has proven heterogeneity of linkage shows that multiple genes must have major or minor effects on a variety of genetic subtypes.

2. Genetic linkage studies

Linkage analysis is a robust and elegant method for identifying the presence of susceptibility genes for a genetic disorder within regions of chromosome of up to forty million bases of DNA possibly containing thousands of genes. The presence of linkage is usually expressed as log₁₀ of the odds (lod) score for the probability of observing marker alleles cosegregating with the disorder in multiple affected families compared to the null hypothesis of no cosegregation or 50% recombination between marker alleles and the disease. In another approach called the sib pair linkage method, marker alleles are observed in affected siblings to test the hypothesis that they are shared in affected cases more than by chance. In disorders which are heterogeneous, such as ADHD, small or medium sized families have the power to detect different genetic subtypes, as defined by positive lods at linkage hotspots, whereas the affected sib pair linkage method has little or no power to detect heterogeneity. In order to identify which gene is involved in ADHD once a linked region has been confirmed using the lod or sib pair method, it is necessary to make use of evolutionarily determined patterns of allelic association (linkage disequilibrium) between disease mutations and closely linked genetic markers in order to narrow down the actual susceptibility gene. Methods which combine tests of linkage and allelic association to do both linkage and fine mapping in the same family sample have been popular because they are immune to population stratification effects. However, restrictions on the size of samples, and the ability of microarray genotyping to detect genetic stratification between cases and controls has now focussed attention on getting the very large case control samples that are needed to overcome the problem of genetic heterogeneity.

The first systematic genome-wide linkage scan for loci influencing ADHD employed 126 affected sib pairs and identified chromosomal regions on 2q24, 5p12, 10q26, 12p13, 12q23, and 16p as possibly harbouring genes increasing susceptibility with lod scores greater than 1.00 (Fisher et al., 2002). A second genome-wide linkage study using affected sib pairs from 203 families confirmed linkage to chromosome 16p13 (Smalley et al., 2002). In a follow-up study of 270 affected sib pairs significant linkage at 5p13, 6q14, 11q25, 17p11, and 20q13 was found (Ogdie et al., 2003). In another whole-genome linkage scan of 164 white Dutch affected sib pairs, chromosomes 5p13, 7p13, and 9q33 were implicated in ADHD (Bakker et al., 2003). A linkage study of 308 affected ADHD sib pairs provided further evidence that the 5p13, 6q12, and 17p11 chromosomal regions harboured susceptibility genes for ADHD (Ogdie et al., 2004). Linkage on chromosomes 4q13, 5q33, 8q11, 11q22, and 17p11 were found in a Cumbrian study, along with suggestive evidence for a new locus on 8q11.23 (Acosta et al., 2004). Confirmation across linkage studies was found for the 11q22 and 17p11 loci (Acosta et al., 2004; Bakker et al., 2003; Fisher et al., 2002; Ogdie et al., 2003).

A further genome-wide linkage scan (Loo et al., 2004) for susceptibility loci for ADHD in a sample of 233 affected sib pairs suggested linkage to four chromosomal regions, three of which replicated previous linkages on 10q26, 16p13 and 17q22, and also implicated 2p24 not far from a locus at 2p16 which has previously been linked with reading disorder (DYX3 (Fagerheim et al., 1999)). In addition, reading measures of individuals with ADHD showed linkage to putative reading disability susceptibility regions on chromosomes 8p and 15q (Loo et al., 2004). Variations in genes on 2q24 (Fisher et al., 2002), 15q15 (Bakker et al., 2003), or 16p13 (Fisher et al., 2002; Smalley et al., 2002) may contribute to common deficits found in genetically overlapping disorders, including both ADHD and autism.

In a whole-genome scan of 102 German families with two or more offspring who fulfilled diagnostic criteria for ADHD, linkage to chromosome 5p17 was shown (lod score 2.59). The previous linkage studies were for an ADHD locus at 5p13 near the solute carrier 6A3 (SLC6A3; dopamine transporter 1; DAT1) at 5p15.33. There was also suggestive evidence for linkage to chromosomes 6q, 7p, 9q, 11q, 12q, and 17p (Hebebrand et al., 2006).

A recent meta-analysis of seven ADHD linkage studies (Arcos-Burgos et al., 2004; Asherson et al., 2008; Bakker et al., 2003; Faraone et al., 2008; Hebebrand et al., 2006; Ogdie et al., 2003; Romanos et al., 2008) confirms genome-wide significance for a region on chromosome 16, between 16q21–16q24, and ADHD (Zhou et al., 2008b). Furthermore, 10 chromosomal regions on 5, 6, 7, 8, 9, 15, 16, and 17 with nominal evidence for linkage with ADHD were identified (Zhou et al., 2008b).

3. Allelic association studies and susceptibility genes

Allelic association studies are able to pin down aetiological genes for ADHD from a large group of genes in a region showing linkage with ADHD derived from family data. This employs the phenomenon of linkage disequilibrium in unrelated cases of ADHD and compares frequencies of neutral SNP base pair changes in cases of ADHD to those in comparison subjects. Genetic association detects SNP markers that have an evolutionarily created linkage disequilibrium relationship with an unknown disease mutation that is very close to the genetic marker. In practice this means less than one million base pairs away and usually within
a few hundred thousand bases of the associated SNP markers. Genome-wide association studies with microarrays able to geno-
type up to 1 million SNP markers in a single person in large
samples in a short time have now begun in ADHD (Franke et al.,
2009). Pharmacologic, neuroimaging studies and animal-models
suggest an imbalance of dopaminergic, serotonergic, and norad-
renergic neurotransmission in ADHD. The aetiology of ADHD is
strongly genetic therefore monoamine genes were strong a priori
candidates.

3.1. The dopamine transporter (DAT1)

In retrospect it was no surprise that the dopamine transporter
DAT1 has been conclusively implicated in the genetics of ADHD by
both linkage and association studies. DAT1 is localised to a recur-
rent linkage hotspot on chromosome 5p. One further linkage study
of DAT1 haplotypes segregating in affected ADHD families also
suggested the involvement of DAT1 (Barr et al., 2001b). The positive
genetic association studies are numerous and five of the most
commonly studied polymorphisms have been subjected to meta-
analyses (Asherson et al., 2007; Barkley et al., 2006; Brookes et al.,
2006b; Carrasco et al., 2006; Cook et al., 1995; Curran et al., 2001;
Daly et al., 1999; Das and Mukhopadhyay, 2007; Feng et al., 2005b;
Genro et al., 2008, 2007; Hawi et al., 2003; Hebebrand et al., 2006;
Kustanovich et al., 2004; Payton et al., 2001) suggests that this polymorphism may have
association with this polymorphism however, meta-analysis of the
VNTRs in the 3′ untranslated region (UTR) and intron 8 are the most
widely studied DAT1 polymorphisms. Meta-analysis of the VNTRs
and two 3′ UTR SNPs were significantly associated with ADHD (3′
UTR VNTR fixed effects P = 0.002, random effects P = 0.028; intron
VNTR fixed effects P = 0.002, random effect P = 0.034; rs27072
deviation tests of ADHD (P = 0.066). Proband with short alleles of the repeat performed
significantly worse on a test of attention and a ‘dose effect’ was
observed with increasing repeat size. Presence of between two and
seven repeats was accompanied by a reduced performance on the
attention test (Manor et al., 2002). Lynn et al. (2005) found that
a novelty-seeking temperament in adults with either a lifetime
history of ADHD, but not a current diagnosis of adult ADHD, was
associated with the 7R allele in 171 parents from 96 families with
ADHD-affected sib pairs. Neuropsychological testing of children
revealed that possession of the 7R allele is associated with an
inaccurate, impulsive response style in neuropsychological tasks
that was not explained by ADHD symptom severity. Children with
the 7R allele had significantly more incorrect responses, shorter
mean reaction times for incorrect responses, and displayed higher
activity levels compared to children without the allele. The children
with and without allele 7R did not differ significantly in the number
of ADHD symptoms when the symptoms were split into the areas of
attention and hyperactivity/impulsivity (Langley et al., 2004).

A second wave of studies has found correlations between endo-
phenotypes such as cognitive function (Bellgrove and Mattingley,
2008; Boonstra et al., 2008), EEG variation in ADHD (Loo et al.,
2009, 2003), MRI volumetric changes (Durston et al., 2005) and
drug treatment response (Cheon et al., 2005; Gruber et al., 2009;
Lott et al., 2005) which are all correlated with DAT1 marker poly-
morphisms. Not all studies of ADHD are positive for DAT1 (Bakker
et al., 2005; Banoe et al., 2008; Bobb et al., 2005; Cheuk et al., 2006;
Friedel et al., 2007; Galli-Weisstub and Segman, 2003; Guan et al.,
2009; Hebebrand et al., 2005; Kim et al., 2005; Kustanovich et al.,
2004; Langley et al., 2005; Lunetta et al., 2000; Maher et al., 2002;
Qian et al., 2004; Simsek et al., 2006; Smith et al., 2003; Swanson
et al., 2000; Todd et al., 2001; Wang et al., 2008). This can be
explained by hidden population stratification between cases and
controls, by lack of power due to small sample sizes and also by
extensive genetic heterogeneity even within European populations
(Gizer et al., 2009). The DAT1 findings are supported by brain
imaging changes in ADHD which find abnormalities in the fronto-
striatal circuits (Durston, 2003). The finding of behavioural changes
in DAT1 mice heterozygous and homozygous for DAT1 hypofunction,
such as neophoria, behavioural inflexibility, anxiety-reduction,
increased novelty seeking and increased stereotypical perseveration
are all compatible with human ADHD.

3.2. Other monoamine related genes associated with ADHD

Several other monoamine related genes dopamine beta
hydroxylase (DBH), D4 dopamine receptor (DRD4), D5 dopamine
receptor (DRD5), serotonin 1B receptor (HTR1B) and synaptosomal-
associated protein 25 isoform (SNAP25) have been associated with
ADHD (Brophy et al., 2002; Daly et al., 1999; Hawi et al., 2002; Lowe
et al., 2004a; Sheehan et al., 2005).

3.3. The dopamine D4 receptor (DRD4)

Ebstein et al. (1996) first reported an association between an
exon 3 VNTR in the DRD4 gene and novelty seeking in a comparison
population sample. Novelty seeking has been hypothesised to be on
a continuum with anti-social personality disorder and impulsivity
which are traits that are comorbid with ADHD. The VNTR is made
up of two to eleven 48 bp repeats. The most commonly observed
alleles comprise 2, 4 and 7 repeats. Alleles with six or fewer repeats
have been defined as short and those with seven or more repeats as
long. The seven repeat (7R) allele has been reported to be func-
tionally different from the common shorter alleles (Asghari et al.,
1995). This VNTR has been studied in a number of systematic meta-
analyses of ADHD (Faraone et al., 2001, 2005; Gizer et al., 2009; Li
et al., 2006a). The results from all of these have shown evidence for
association of allele 7R with ADHD. The most recent meta-analysis
showed a fixed effects significance of P < 0.00001 (random effects
P = 0.00007) with evidence of substantial heterogeneity (Gizer et al.,
2009). There is also a very low prevalence of the 7R allele in Asian
populations (Leung et al., 2005). TDT (transmission disequi-
librium test) analysis shows preferential transmission of short
alleles of the VNTR in 178 Israeli triads in association with ADHD
(P = 0.006). Proband with short alleles of the repeat performed
significantly worse on a test of attention and a ‘dose effect’ was
observed with increasing repeat size. Presence of between two and
seven repeats was accompanied by a reduced performance on the
attention test (Manor et al., 2002). Lynn et al. (2005) found that
a novelty-seeking temperament in adults with either a lifetime
history of ADHD, but not a current diagnosis of adult ADHD, was
associated with the 7R allele in 171 parents from 96 families with
ADHD-affected sib pairs. Neuropsychological testing of children
revealed that possession of the 7R allele is associated with an
inaccurate, impulsive response style in neuropsychological tasks
that was not explained by ADHD symptom severity. Children with
the 7R allele had significantly more incorrect responses, shorter
mean reaction times for incorrect responses, and displayed higher
activity levels compared to children without the allele. The children
with and without allele 7R did not differ significantly in the number
of ADHD symptoms when the symptoms were split into the areas of
attention and hyperactivity/impulsivity (Langley et al., 2004).

A family-based association analysis of the DRD4 120-bp inser-
tion/deletion promoter 1.2 kb upstream of the transcriptional start
site showed a significant association with ADHD in 372 ADHD cases
and their parents (McCracken et al., 2000). This finding was sup-
ported in an expanded ADHD sample (Kustanovich et al., 2004).
However, there have been a number of studies that have failed to
replicate these findings and a meta-analysis of the data from this
polymorphism found no evidence for association (Gizer et al.,
2009). SNP rs1800955 located 521 bp upstream of the DRD4 tran-
scription/deletion promoter 1.2 kb upstream of the transcriptional start
site showed a significant association with ADHD in 372 ADHD cases
and their parents (McCracken et al., 2000). This finding was sup-
ported in an expanded ADHD sample (Kustanovich et al., 2004).
However, there have been a number of studies that have failed to
replicate these findings and a meta-analysis of the data from this
polymorphism found no evidence for association (Gizer et al.,
2009). SNP rs1800955 located 521 bp upstream of the DRD4 tran-
scriptional start site has recently been found to show association
with ADHD in a Korean sample (P = 0.013, Yang et al., 2008).
Previous case control and TDT studies had not found evidence for
association with this polymorphism however, meta-analysis of the
data (Barr et al., 2001a; Kereszturi et al., 2007; Lowe et al., 2004b;
Payton et al., 2001) suggests that this polymorphism may have a role in
ADHD (fixed effects P = 0.007, Gizer et al., 2009). Alleles of both of these upstream polymorphisms have been reported to alter
promoter activity (D'Souza et al., 2004; Okuyama et al., 1999).
Over-transmission of paternal risk alleles with the DRD4 SNP rs17270373
was also observed in an Irish ADHD sample (Hawi et al., 2005).

3.4. The dopamine D5 receptor (DRD5)

A 146-bp dinucleotide repeat allele 18.5 kb from the DRD5 gene
has been associated with ADHD (P = 0.02). The estimated genotype

relative risk of 1.7 was modest (Kustanovich et al., 2004). However, the 148 bp allele of the DRD5 microsatellite was not found to be associated with ADHD in 236 Dutch families (Bakker et al., 2005). A meta-analysis of nine DRD5 studies shows that the 148 bp allele of the dinucleotide repeat is significantly associated with ADHD (fixed effects $P = 0.000095$; random effects $P = 0.0027$) with moderately significant heterogeneity in effect sizes across studies (Gizer et al., 2009).

3.5. The tachykinin receptor 1 (TACR1)

Another gene strongly related to dopaminergic function in the prefrontal cortex is the neurokinin (substance P) receptor (NK1R) also called the tachykinin receptor 1 (TACR1). In a knockout mouse lacking the TACR1 gene (NK1R$^{-/-}$), mice were found to be hyperactive (Yan et al., 2009). This was prevented by psychostimulants (o-amphetamine and methylphenidate). The mice had reduced (>50%) spontaneous dopamine efflux in the prefrontal cortex and displayed lack of the striatal dopamine response to o-amphetamine. These behavioural and neurochemical abnormalities in NK1R$^{-/-}$ mice, together with their atypical response to psychostimulants, are very similar to clinical characteristics of ADHD in humans (see: Stanford et al. (ibid)). An allelic association study of 450 ADHD cases and 600 controls found that four TACR1 SNPs previously associated with bipolar disorder and alcoholism were also associated with ADHD (Yan et al., 2009). If confirmed TACR1 may turn out to be another dopamine related gene which is more strongly associated with an affective disorder subgroup of ADHD rather than the CD subtype.

3.6. The serotonin 1B receptor (HTR1B)

Evidence of significant genetic association of the 861G allele of the HTR1B gene using parent offspring transmission tests has been shown in 273 European families ($P = 0.0065$, Hawi et al., 2002), as well as in 115 Canadian families ($P = 0.03$, Quist et al., 2003). Similarly, paternal over-transmission of the HTR1B G861 allele to offspring with the inattentive ADHD subtype was observed in 12 multi-generational Centre d’Etude du Polymorphisme Humain (CEPH) pedigrees, comprising 229 families of ADHD probands (Smoller et al., 2006). A haplotype block encompassing the gene was associated with the inattentive ADHD subtype. In addition, three polymorphisms in this block were nominally associated with this subtype but did not remain significant after correction for multiple testing (Smoller et al., 2006). Meta-analysis from nine studies indicates a modest, yet significant, risk associated with the HTR1B 861 G allele on susceptibility to ADHD (fixed effects $P = 0.01$) with no heterogeneity in study-effect sizes (Gizer et al., 2009).

3.7. Dopamine beta hydroxylase (DBH)

Dopamine beta hydroxylase gene (DBH) polymorphisms have been shown to influence plasma dopamine beta hydroxylase (DBH) activity. Low plasma DBH levels are said to be associated with CD (Bowden et al., 1998). A significant association between an SNP in DBH (rs2519152) has been shown in 118 children with ADHD (Daly et al., 1999). Meta-analysis of six studies (Gizer et al., 2009) found a trend towards an association between the DBH A2 allele and ADHD ($P = 0.074$). However, there was significant heterogeneity between the study-effect sizes ($P = 0.004$). After correcting for this by removing one study from the analysis, heterogeneity was reduced ($P = 0.057$), increasing evidence for an association between the A2 allele and ADHD. Two functional mutations, rs1108580 and rs1611115 associated with plasma levels of DBH were both shown not to be associated with ADHD in a meta-analysis (Gizer et al., 2009).

3.8. The alpha2A and 1A adrenergic receptors (ADRA2A and ADRA1A)

Methylphenidate, prescribed to treat ADHD, increases catecholaminergic activity through increased activation of alpha2A adrenergic receptors (Andrews and Lavin, 2006). In a Brazilian sample of 92 ADHD patients and their biologic parents, the GG genotype of the $-1291C$-G SNP (rs180054) of the ADRA2A gene was associated with inattention and combined ADHD scores ($P = 0.02$, Roman et al., 2003). In a new sample of 128 Brazilian ADHD probands, the ADRA2A GG genotype was also associated with symptoms of inattention (Roman et al., 2006). However a meta-analysis of 11 studies could not confirm the ADRA2A association with ADHD (Gizer et al., 2009). A recent study has found allelic association between the ADRA1A gene and ADHD (Elaia et al., 2009a).

3.9. The dopamine D2 receptor (DRD2)

Early studies focused on a Taq1 polymorphism (rs1800497) 10 kb downstream of DRD2. This SNP causes a non-synonymous Glu to Lys change in an exon of neighbouring ANKK1 gene. SNP rs1800497 has been shown to affect DRD2 expression (Hirvonen et al., 2004), the levels of the dopamine metabolite homovanillic acid (Ponce et al., 2004) and has been implicated in alcohol dependence (Blum et al., 1991). The first study to report an association between the rs1800497 A2A2 genotype and the highest mean level of symptoms in children with ADHD did not find significant effect sizes of the SNP (Rowe et al., 1999). Results of six studies were reviewed in a meta-analysis, which showed a significant association between the A1 allele and childhood ADHD (fixed effects $P < 0.001$). There was only a trend towards an association with random effects analysis ($P = 0.110$), although this may be explained by the significant heterogeneity of effect sizes across studies (Gizer et al., 2009). This gene has been convincingly associated with alcoholism in many genetic association studies as well as in a linkage study (Cook et al., 1996). A recent genetic association report showed that DRD2 may be associated with dis-social psychopathic personality traits rather than alcoholism per se (Ponce et al., 2008).

3.10. Tryptophan hydroxylase 1 (TPH1)

Tryptophan hydroxylase (TPH) catalyzes tryptophan in a rate-limiting step to synthesise 5-hydroxy-tryptophan (5-HT), the precursor for serotonin. A silent polymorphism in intron 7 (rs1800532) of tryptophan hydroxylase gene, TPH1, was identified and the rare 218A$\rightarrow$6526G haplotype was significantly under-transmitted to probands with ADHD ($P = 0.034$) in the Chinese Han population (Li et al., 2006b). This association was confirmed in a meta-analysis of four studies (Gizer et al., 2009).

3.11. Tryptophan hydroxylase 2 (TPH2)

A second isoform of TPH (Walther and Bader, 2003), coded for by the TPH2 gene, was found to be responsible for TPH expression in the brain, not TPH1. Excess transmission of two SNPs in the TPH2 gene in 225 children with ADHD in 103 families was found using the pedigree disequilibrium test (Walitza et al., 2005). A second study reported association with SNPs in intron 5 (Sheehan et al., 2005). Neither marker was significantly associated with ADHD in a meta-analysis (Gizer et al., 2009; Sheehan et al., 2005).
3.12. Serotonin 2A receptor (HTR2A)

Serotonin-2A receptor antagonists block increases in dopamine activity and hyperlocomotion following amphetamine treatment (O’Neill et al., 1999). However, meta-analysis of three SNPs (rs6311, rs6313 and rs6314), previously implicated in ADHD, showed the association was non-significant (Gizer et al., 2009).

3.13. Solute carrier 6A2 noradrenaline transporter (SLC6A2)

The noradrenaline transporter gene (SLC6A2) is highly expressed in the frontal cortex and regulates noradrenaline and dopamine reuptake. It is an initial site of action for therapeutic drugs such as amphetamines (Pacholczyk et al., 1991) that are used in the treatment of ADHD (Solanto, 1998). Neither a synonymous SNP (rs5569) in exon 9 of SLC6A2 nor a T to C SNP in intron 13 (rs2242447) were significantly associated with ADHD following meta-analysis of five studies (Gizer et al., 2009).


A functional 44 bp deletion/insertion polymorphism in the promoter region 5-hydroxytryptamine (5-HT, serotonin) transporter (5-HTT) gene, SLC6A4, has been associated with depression and anxiety (Acosta et al., 2004; Lotrich and Pollock, 2004; Munafo et al., 2006; Schinka et al., 2004). The long promoter variant is associated with more rapid reuptake of serotonin than the short allele (Lesch et al., 1996). Meta-analysis of ten TDT studies and nine case control and haplotype relative risk studies shows a significant association with the long allele and ADHD (fixed effects P = 0.004, random effects P = 0.010), with significant heterogeneity across studies (P = 0.00003, Gizer et al., 2009). Furthermore, a 17 bp VNTR in intron 2 and an SNP in the 3’ UTR (rs3813034) have been associated with ADHD. The 12-repeat allele of the VNTR has been shown to enhance transcription more than the 10 repeat allele (Fiskerstrand et al., 1999; MacKenzie and Quinn, 1999). SNP rs3813034 is a putative polyadenylation site that may affect mRNA stability (Battersby et al., 1999). Neither polymorphism was found to be significantly associated with ADHD when results from nine studies were tested in a meta-analysis (Gizer et al., 2009).

3.15. Synaptosomal-associated protein 25 isoform (SNARE protein SNAP25)

The synaptosomal-associated protein of 25 kDa gene (SNAP25) has been suggested as a genetic susceptibility factor in ADHD based on the mouse strain coloboma. Four polymorphisms in SNAP25 were significantly associated with ADHD in 186 Canadian families with 234 ADHD children (P = 0.039–0.005) (Feng et al., 2005a). However, these results were not replicated in an independent sample of 99 families with 102 ADHD children from southern California, possibly due to differences in selection criteria, ethnicity, medication response and other clinical characteristics of the samples (Feng et al., 2005a). Analysis of the DSM-IV subtypes in the Toronto sample indicated that the differential results were not attributable to ADHD subtype. Quantitative trait analyses of the dimensions of hyperactivity/impulsivity and inattention in the Toronto sample found that both behavioral traits were associated with SNAP25. Meta-analysis revealed that only one of the four SNPs (rs3746544) in SNAP25 to be significantly associated with ADHD across seven studies (Gizer et al., 2009).

3.16. Central nicotinic cholinergic A4 receptor (CHRNA4)

Nicotine agonists reduce ADHD symptom severity (Levin et al., 2001). Association between CHRNA4 markers and ADHD has been described, although an SNP found in exon 5 was not associated with ADHD in 70 children (Kent et al., 2001). A non-significant trend towards an association with two additional SNPs in exon 5 (rs2273506 and rs6090384) and ADHD has been reported (Gizer et al., 2009).

3.17. Catechol-O-methyl transferase (COMT)

Early-onset anti-social behaviour accompanied by ADHD is a clinically severe variant of anti-social behaviour with a poor outcome (Thapar et al., 2005). In 240 British children with ADHD or hyperkinetic disorder, the COMT val158met variation (rs4680) was associated with increased symptoms of CD and a significant gene–environment interaction between the COMT polymorphism and birth weight was confirmed (Thapar et al., 2005). A meta-analysis of 16 studies found no significant association between the COMT val158met polymorphism (Gizer et al., 2009).

3.18. Monoamine oxidase A (MAOA)

Monoamine oxidase A and B catalyze the oxidative deamination of naturally occurring monoamines, dopamine, noradrenaline, and serotonin. Furthermore, MAO inhibitors have been shown to improve the symptoms of ADHD (Zametkin et al., 1985). Impulsive and aggressive behaviour have shown to be caused by a stop codon mutation in MAOA in a large Dutch family (Brunner et al., 1993). Several polymorphisms near or in MAOA have been reported to be associated with ADHD (Jiang et al., 2006; Payton et al., 2001), while a functional 30 bp VNTR comprising 2, 3, 3.5, 4, or 5 copies, 1.2 kb upstream of MAOA, is associated with impulsivity and aggressive behaviour (Manuck et al., 2000). Meta-analysis of six studies indicates a non-significant association between ADHD and the high-activity MAOA alleles (Gizer et al., 2009).

4. Genome-wide association studies (GWAS)

A recent review of the first few attempts at genome-wide association studies (GWAS) of ADHD (Franke et al., 2009) reported that few studies had been completed and that sample sizes were still too small to unravel heterogeneity. Unlike previous genetic association studies of ADHD highlighting genes involved in monoaminergic neurotransmission the studies completed with genome-wide methods provide some evidence for the involvement of genes in cell division, cell adhesion especially via the cadherin gene, CDH13, and also the integrin system. Genes implicated in neuronal migration and plasticity; along with gene transcription, cell polarity and extracellular matrix regulation as well as cytoskeletal remodelling processes have shown some degree of allelic association with ADHD (Franke et al., 2009).

The first GWAS in ADHD was set up by the Genetic Association Information Network (GAIN) (Manolio et al., 2007), which collected 958 Caucasian case-parent trios as part of the International Multicentre ADHD genetics (IMAGE) study in children (Brookes et al., 2006a; Kutnsi et al., 2006). A TDT analysis of a categorically defined ADHD phenotype identified a highly significant imbalance in the number of major and minor allele over-transmissions. After applying a correction factor the research identified 25 SNPs in near genes with a putative role in ADHD, including nucleobindin 1, cannabinoid receptor 1 (CN1R), potassium channel interacting protein 1 isoform 2 and protein 4 isoform 3 (KCNI3 and KCNIP4), cytoskeletal-organizer doublecortin and CaM kinase-like 1.
5. Copy number variants

Copy number variants (CNVs) arising through non-allelic homologous recombination play an important role in the aetiology of psychiatric disorders such as autism, bipolar disorder, schizophrenia (Cichon et al., 2009) and ADHD. A girl with ADHD was shown to have a de novo 600 kb deletion on a maternal copy of chromosome 16p11.2, encompassing CORO1A (coronin-1A, essential for T cell release from the thymus), flanked by 146 kb segmental duplications (Shiov et al., 2009). Deletion as well as duplication at the 16q11.2 locus has also been associated with autism spectrum disorder and neurodevelopmental disorders (Ghebranious et al., 2007). One recent study has identified 222 structural variants in 335 ADHD patients and their parents, that were not detected in 2026 unrelated healthy controls (Elia et al., 2009b). No excess deletions or insertions were found in ADHD patients, although inherited rare CNVs encompassed genes that have been identified in structural variation studies carried out on neuropsychiatric and neurological disorders. These genes include A2B1P, APOL4, CHL1, CHN2, CNTNAP2, CPLX2, CNTND2, DPP6, GRM5, GRM7, NKAIN2, PARK2, RTN4, SEPP1, SP1N/V1 and TACR3. Some of these such as GRM7, DPP6 and TACR3 have been nominally associated with bipolar disorder in a recent GWAS (Ferreira et al., 2008). The deletion in the glutamate receptor gene (GRM5) was identified in an affected parent and three affected offspring, who displayed problems with spatial orientation (Elia et al., 2009b), a behavioural trait in the GRM5 knockout mouse (Lu et al., 1997). The CNV in GRM7 was present in an ADHD proband who presented with an anxiety disorder. Additional genes disrupted by CNVs in ADHD that are also thought to be involved in autism include AUTS2, and IMMP2L. Furthermore, other ADHD CNV-associated genes are implicated in learning, behaviour, synaptic transmission and neuronal development. These include the ADHD CNV disrupted gene ATM, which has been associated with ataxia-telangiectasia and neurodegeneration, while BLMH and PDCD10 are associated with Alzheimer’s disease and cerebral cavernous malformations, respectively. Four separate deletions were found in the protein tyrosine phosphatase gene (PTPRD), a gene thought to play a role in restless leg syndrome, which is a common symptom in ADHD (Elia et al., 2009b). It is of note that a CNV in the tachykinin receptor 3 (TACR3) gene was found to be associated with ADHD.

6. Discussion

Clinical genetic studies of ADHD demonstrate a rich variety of comorbidities and patterns of familial recurrence. The earlier linkage studies which showed heterogeneity of linkage showed a quite pleasing concordance with the later genetic association studies implicating single genes such as those on 5p and 17q. The evidence for association between DAT1 on 5p and ADHD is very strong and it has become a test bed for the molecular genetic approach to psychiatric disorders. Sequencing and gene expression studies should soon find aetiological base pair changes actually responsible for the underlying molecular pathology.

Once it is accepted that there are no common high frequency genetic markers associated with ADHD and that there is extensive heterogeneity it is possible to interpret the more recent genome-wide association literature. The earlier candidate gene association studies required much less stringent statistical correction for multiple testing and one can argue that they were successful. The multiple testing problem with microarray data means that it is premature to draw any strong statistical conclusions from the current GWAS data. In any case the extent GWAS are underpowered and none of the results would have been expected to show genome-wide significance levels of association with ADHD.
(Franke et al., 2009). Genes with dominant effects associated with ADHD subtype phenotypes have been proposed (Acosta et al., 2004). A plausible outcome is likely to be a mixture of dominant and recessive major genes with more complex, perhaps polygenic, transmission patterns. Genome-wide association studies based on clinical subtypes and endophenotypes of ADHD are needed in much larger samples. Inherited CNVs could plausibly play an increasingly important role in the diagnosis and treatment of ADHD after they have been further investigated. The field of ADHD genetics has moved into a highly productive phase and new treatment and preventive strategies are already being sought with the genetic information obtained.

References


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