A Catechol-\(O\)-methyltransferase gene polymorphism moderates the effect of antenatal stress on childhood problem: Longitudinal evidence across multiple ages

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ABSTRACT

There have been numerous calls to identify gene by environment interactions to help understand the aetiology and developmental nature of childhood emotional and behavioural problems. Here we present data from the Auckland Birthweight Collaborative (ABC) Study to investigate whether a functional polymorphism in the Catechol-O-methyltransferase (COMT; rs4680 G/A) gene interacts with maternal perceived stress (antenatal and 7 years) to impact on childhood total difficulties scores (Strength & Difficulties questionnaire at ages 7 and 11 years). We found that carriers of the Met/Met genotype were at increased risk of behavioural problems at ages 7 as well as 11 years only when they were exposed to maternal stress in utero relative to carriers of the Met/Val or Val/Val. In comparison, maternal stress at 7 appears to have a significant effect on behavioural problems irrespective of the child’s genotype. Carriers of the Met/Met genotype also had, on average, Performance IQ scores 4 points higher than Val/Val homozygotes and heterozygotes. Findings emphasize the potential long-term consequences of prenatal maternal stress for genetically susceptible individuals during neurodevelopment in utero.

KEY WORDS: Genetics, gene X environment, rs4680, SDQ, Child, Behaviour, Socio-emotional, ADHD, Perceived Stress, Development

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**INTRODUCTION**

Exposure to maternal antenatal stress can have deleterious effects on the subsequent development of the offspring. A number of recent studies have shown that children who are exposed to maternal stress during pregnancy have an increased risk of both socio-emotional and behavioural disturbances (Brennan, Hammen, Andersen, Bor, Najman, & Williams, 2000; Huizink, Mulder, & Buitelaar, 2004; O'Connor, Heron, Golding, Glover, *et al.* 2003; Ramchandani, Richter, Norris, 2010; see review by Rice, Jones, & Thapar, 2007), including increased risk for neurodevelopmental disorders such as ADHD (Grizenko, Shayan, Polotskaia, Ter-Stepanian, & Joober, 2008; Van den Bergh & Marcoen, 2004). Indeed, up to 10-20% of the population-attributable risk for emotional/behavioural symptoms in children can be accounted for by antenatal stress (Talge, Neal, & Glover, 2007).

Exposure to stress during pregnancy can influence prenatal development and the risk for later problem behaviour in a number of possible ways, presumably through early environmentally mediated programming effects on the foetus. For example, stress-induced activation of the sympathetic nervous system causes an increase in uterine artery resistance that will reduce blood flow to the developing foetus. This reduction of blood flow can potentially alter brain structure and function (Teixeira, Fisk, Glover, 1999; Welberg & Seckl, 2001), including delayed myelination and abnormal development of the dopaminergic system (review by Glover, 1997). As reviewed by Kinney, Munir, Crowley and Miller (2008), antenatal maternal stress can also increase the risk for complications during delivery.

There is also a growing literature on maternal glucocorticoid effects on foetal neurodevelopment. Although most of the cortisol that is released in the blood by the mother in response to stress is deactivated, some cortisol has been shown to reach the foetus (Seckl, 1997). The foetus itself increases cortisol secretion in response to stress (Gunnar, 1998). Even small amounts of cortisol can influence the functioning of the hypothalamo-pituitary-adrenal...
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(HPA) axis in the foetus, causing an eventual and permanent deregulated HPA response to stress in the offspring (Henry, Kabbaj, Simon, Le Moal, Maccari, 1994; Kapoor & Matthews, 2005; Seckl & Meaney, 2006; review by O'Donnell, O'Connor, & Glover, 2009). The altered functioning of the HPA axis can then increase the risk for later behavioural and emotional problems. One mechanism for this effect is alteration of gene expression in developing brain cells (Francis, Diorio, Liu, Meaney, 1999; Meaney & Szyf, 2005; Weaver, Cervoni, Champagne, et al., 2004).

The COMT (Catechol-O-methyltransferase) gene, located on chromosome 22, has received recent interest with regard to the role it plays in stress vulnerability. It is considered a good candidate to study the genetic basis of stress sensitivity because it encodes an enzyme that is involved in the breakdown of dopamine in the prefrontal cortex. As well as the prefrontal cortex, COMT is also expressed in the amygdala (Hong, Shu-Leong, Tao, & Lap-Ping, 1998), an area of the brain important for socio-emotional functioning. Elevated dopamine in these areas, including the pathway to the striatum, has been proposed to enhance the salience of environmental threat in those homozygous for the Met allele (Herrmann, Wurflein, Schreppel, Kochler, Muhlberger, Reif, et al., 2009).

COMT has also been associated with emotional and behavioural disturbances in children in candidate gene studies (Eisenberg, Mei-Tal, Steinberg, et al., 1999; Gadow, Roohi, Devincent, et al., 2009) and has been found to be associated with attention deficit hyperactivity disorder (ADHD) in the International Multicenter ADHD Genetics Project (IMAGE) genome wide association study (Lasky-Su, Neale, Franke, et al. 2008a; Lasky-Su, Anney, Neale, et al. 2008b; Neale, Lasky-Su, Anney, et al., 2008). Studies that have looked at the association between valine/methionine polymorphism of the COMT and ADHD, however, have reported inconsistent findings. A recent meta-analysis, for example, concluded
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that there was no direct association of this gene on ADHD (Cheuk & Wong, 2006). *COMT* appears to be associated, not with ADHD per se, but with more general behavioural and conduct problems (Thapar, Langley, Fowler, et al., 2005). In support, Caspi and colleagues found that the homozygotes at codon 158 of the *COMT* gene were more aggressive than those with either the valine/methionine or met/met homozygotes (Caspi, Langley, Milne, et al., 2008). Studies such as these have highlighted the importance of epigenetic effects on the expression of genes involved in responses to psychosocial adversity in the postnatal environment.

As reviewed by Rutter, Moffitt and Caspi (2006), a significant proportion of the heritability in problem behaviour is likely to be accounted for by gene by environment interactions. Gene by environment interaction occurs when the effect of exposure to an environmental agent on behaviour is conditional on a person’s genotype or, conversely, when environmental experience moderates genetic effects on behaviour (Moffitt, Caspi, & Rutter, 2005). For example, innate variation in vulnerability to maternal stress in utero may interact with genotype to increase the risk of behavioural and emotional symptoms in children. To this end, in the present study, we genotyped children from the Auckland Birthweight Collaborative (ABC) and used the Total Difficulties score from the Strengths and Difficulties Questionnaire (SDQ; Goodman, 1997) to determine if certain allele variants on the *COMT* gene are associated with increased vulnerability to early stressors. The SDQ is a brief measure of emotion and behaviour problems in children and adolescents between the ages of 3 to 16 years. Scores are produced for five subscales: Emotional symptoms; Hyperactivity; Conduct problems; Peer problems; and Prosocial Behaviour. The Total Difficulties score is calculated by summing all four deficit-focused subscales (i.e. not including the Prosocial subscale).
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The SDQ has been used in earlier studies on the effects of maternal stress and anxiety on later emotional and behavioural problems in the offspring. In a large-scale prospective longitudinal study, O’Connor et al. (2003) found a significant and positive association between maternal antenatal anxiety and behavioural and emotional problems in their children, even after controlling for postnatal maternal anxiety, obstetric complications, and family psychosocial disadvantage. The effects of antenatal anxiety were quite persistent, as the Total Difficulties scores on the SDQ remained higher in children at both 4 and 7 years of age. Rice, Harold, Boivin, van den Bree and colleagues (2010) looked at the associations between maternal antenatal stress and offspring anxiety, conduct problems (SDQ conduct subscale scores), and ADHD symptoms in related and unrelated groups. Pregnant mothers were related or unrelated to their child as a result of in vitro fertilization. Associations between antenatal stress and conduct problems were observed in both unrelated and related mother-child dyads, indicating the importance of the intrauterine environment. In contrast, the association between maternal antenatal stress and offspring ADHD symptoms was due to genetic inheritance, since the association was not seen in unrelated mother-child pairs. This study, however, was limited to a single measure of maternal stress and was unable to account for stress during the postnatal period.

In order to consider the developmental nature of childhood problem behaviour and to address whether maternal stress has a negative effect on child development, we assessed both maternal perceived stress and childhood emotional and behavioural difficulties with the SDQ over two time periods: Antenatal and 7 years (perceived stress) and 7 years and 11 years (total problem behaviours). We genotyped the children themselves (Val158Met SNP rs4680) and have demonstrated an association between genetic variation in *COMT* and total problem scores – importantly, this association only holds for those whose mothers reported significant stress during pregnancy but not when the children were aged 7 years.
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**METHODS**

*Participants*

The Auckland Birthweight Collaborative (ABC) Study has been described in detail elsewhere (Thompson, Clark, Robinson, *et al.* 2001). In brief, the study originally selected subjects at birth, between October 1995 and November 1997. Subjects were selected over the entire time period from the Auckland District Health Board, and from Oct 1995 to August 1997 in the Waitemata District Health Board at which time the study was no longer able to obtain reliable birth and obstetric information. All small for gestational age (SGA) infants (<=10<sup>th</sup> percentile for gestation and sex) and a random selection of appropriate for gestational age infants (AGA) were selected during the study such that the number in each group were approximately equal. The study consisted of 1714 subjects at birth of which 871 had mothers who were identified as being of European ethnicity.

The study has followed the children at 1 year (via a postal questionnaire), and with face to face assessments at approximately 3.5, 7 and 11 years of age. The follow-up phase at 1 and 3.5 years had a poor response rate for non-European ethnicities and due to ethical considerations about the generalisability of results in these populations further follow-ups have only included those identified as having European mothers. Response rates at each of the original sample is 3.5 (63.1%), 7 (67.9%) and 11 (71.1%). These follow-up phases have collected a range of predictive and outcome variables.

The main independent variables under investigation in the analysis are perceived maternal stress at birth and at 7 years (using the perceived stress scale; Cohen, Kamarck, & Mermelstein, 1983). The outcome of interest is the total difficulties score from the Strength and Difficulties score (Goodman, Ford, Simmons, Gatward, & Meltzer 1997), and this
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outcome has been analysed at both 7 and 11 years of age. We also assessed IQ at 7 using the WISC (3rd-edition), which was performed by trained psychologists.

At 11 years 546 (63%) participants consented to collection of peripheral blood (n=397) or a buccal swab (n=149) for DNA extraction and genotyping. 227 samples were from children who were born small for gestational age (SGA) and 319 were from children born appropriate for gestational age (AGA). Gestational age was estimated using the date of the last menstrual period where it was available and was within 2 weeks of the best clinical estimate of gestational age at birth; otherwise the best clinical estimate was used.

DNA was extracted from the blood/buccal samples using Qiagen’s DNA extraction kit and following the manufacturer’s instructions.

The study received ethical approval from the Northern regional ethics committee. Signed consent for the study and extraction of DNA was given by the parents of the children and assent also given by the child.

**Child Behaviour**

The Strengths and Difficulties Questionnaire (SDQ) parent format, which assessed children’s emotional and behavioural difficulties, was used at ages 3.5, 7 and 11 years (Goodman, 1997). The SDQ scale items were selected based on nosological concepts of the Diagnostic Statistical Manual -IV (DSM-IV, American Psychiatric Association, 1994) and International Classification of Diseases 10th Revision (ICD-10, World Health Organization, 1994) classifications of childhood emotional and behavioural problems. For example, the five items relating to the SDQ’s Hyperactivity-Inattention subscale were deliberately selected to measure inattention (two items), hyperactivity (two items) and impulsiveness (one item), as
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these are the three key symptom domains for a DSM-IV diagnosis of ADHD (American Psychiatric Association, 1994).

The initial study was based on a UK wide survey of 10,000 children and adolescents. This study found a high internal consistency of parent report for the Total Difficulties scale (Cronbach alpha = 0.82) and a 4 to 6 month retest stability of 0.72. Assessment of this sample with a psychiatric interview revealed that the parent report, with a Total Difficulties subscale score at or above the 90th percentile, was predictive a 15 fold increase in the likelihood of any independently diagnosed psychiatric disorder. Further analyses revealed a positive predictive value of 46% and a negative predictive value of 96% (Goodman, Ford, Simmons, Gatward, Meltzer, 2000). The extensive use of the SDQ in epidemiological, developmental and clinical studies across the world and in many cultures is increasing the usefulness for inter country comparisons of children’s emotional and behavioural problems (Mathai, Anderson, & Bourne, 2002; Woerner, Fleitlich-Bilyk, Martinussen, Fletcher, Cucciaro, & Dalalarrondo, 2004).

The SDQ consists of 25 symptom items which enquire about a child’s behavior over the past 6 months. Responses are based on a 3-point Likert scale, indicating how much each symptom applies to the target child. The possible responses are 0 = ‘not true,’ 1 = ‘somewhat true’ and 2 = ‘certainly true’.

The Total Difficulties score is calculated by summing all four deficit focused subscales (Emotional symptoms; Hyperactivity; Conduct problems; Peer problems). Scores for this subscale range from 0 to 40. The scoring system allows scale scores to be prorated if at least three of the five scale items are complete. The SDQ total difficulties subscale was examined in the present study as a continuous variable. A total of 587 children age 7 years and 614 age 11 years had scorable data on all five SDQ scales.
Maternal Stress

Maternal perceived stress was assessed at birth, 3.5 and 7 years using the Perceived Stress Scale (PSS) (Cohen, Kamarck, & Mermelstein, 1983). The scale items were designed to tap the degree to which respondents perceived their lives as unpredictable, uncontrollable and overloading, based on the previous one month time period. The PSS is suggested for examining the role of nonspecific stress in the aetiology of disease and behavioural disorders and as an outcome measure of experienced levels of stress. The full scale consists of 14 questions, however the version used in this study was the PSS-10, which is a briefer version containing only 10 items.

Compared to life health-related outcomes (for example, measuring the frequency of stressful life events within a time period, or measuring the amount of daily stresses experienced by a person) the PSS is more specific as it accounts for individual differences in the way life events are appraised. An advantage of the PSS, compared with measures that assess the number of stressful life events occurring within a time period, is that the PSS can be said to measure the impact of negative life events including a personal appraisal of how stressful the event has been. Furthermore, it has been found to be a better predictor of health outcomes than life events scales (Cohen, Kamarck, & Mermelstein, 1983).

Mothers’ levels of stress during the previous month were assessed at birth, 3.5 and 7 years. Data were excluded from the analyses where a child was accompanied by someone other than their mother as this information was not comparable to previously collected maternal stress data. This resulted in a loss of data from 11 families at 3.5 years and 29 families at 7 years.
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**Genotyping**

Genotyping was performed with the MassARRAY and iPlex systems of the Sequenom genotyping platform (Sequenom, San Diego, CA), which uses the MALDI-TOF primer extension assay (Jurinke *et al.*, 2002; Storm *et al.*, 2003), according to manufacturers’ recommendations.

Assays were optimized in 24 samples consisting of 20 reference Centre d’Etude du Polymorphisme Humain (CEPH) samples and 4 blanks.

All sample plates contained cases, controls, blanks, CEPH and duplicate samples. Quality control measures included independent double genotyping, blind to sample identity and blind to the other caller, and where available comparison of our CEPH genotypes to those in the HapMap (www.hapmap.org). The SNP had a genotyping call rate of 86% and the genotyping calls did not differ from Hardy Weinburg equilibrium. There were 120 (22%) Met (A) homozygotes, 136 (24.9%) Val (G) homozygotes and 213 (39%) Val/Met heterozygotes.

**Statistical analysis**

Univariable analysis was carried out using a recessive model to determine the effect of the SNP and maternal stress on the total difficulties score. This analysis was carried out using Proc GLM in SAS and analyses controlled for SGA status. Hence the univariable analysis modelled total difficulties score=intercept+a*SGA+b*maternal stress+c*rs4680. A model was then fitted to test additionally for an interaction between maternal stress and rs4680. We also carried out analyses by genotype to assess the magnitude of the effect of stress within each genotype.
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**RESULTS**

A univariable model fitting genotype and SGA status to the total difficulties score at 7 years of age found statistically significant effect of neither. Adding maternal stress at birth to the model found a significant effect ($\beta=0.10$, $p=0.0040$), an additional interaction term between genotype and maternal stress at birth was found to be statistically significant ($p=0.03$). Stratified analyses of maternal stress at birth were then carried out by genotype; this resulted in a significant effect of the Met (A) homozygote group ($\beta=0.26$, $p=0.0019$), but no statistically significant effect for the Val (G) homozygote and Val/Met heterozygote group ($\beta=0.06$, $p=0.12$) (Table I). This suggests that maternal stress during pregnancy was only of importance in relation to the Met homozygotes.

The same process was carried out using the total difficulties score at 11. This again showed no statistically significant effect of SGA status or genotype. Adding maternal stress at birth to this model as with the outcome at 7 found a statistically significant effect ($\beta=0.11$, $p=0.0029$). Adding an interaction term to this model also found this to be significant ($p=0.05$). Stratified analyses again found a statistically significant effect in the Met homozygotes ($\beta=0.26$, $p=0.0010$), but not for the Val homozygote/heterozygote group ($\beta=0.07$, $p=0.10$) (Table I). These results are consistent with those seen at 7 years of age.

Correlation between total difficulties score at 7 and 11 was $r=0.63$, $p<0.0001$.

The same process was carried out using maternal stress at 7. Adding maternal stress at 7 to the univariable model for the total difficulties score at 7 found a statistically significant effect ($\beta=0.22$, $p<0.0001$), however an interaction term with the genotype was not statistically significant ($p=0.86$). We carried out stratified analyses to assess the effect size for maternal stress by genotype, resulting in $\beta=0.21$ ($p=0.0076$) for Met homozygotes and
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β=0.22 (p<0.0001) for Val homozygotes / Val/Met heterozygote group (Table II). Thus it appears that maternal stress at 7 is equally important regardless of the genotype.

Adding maternal stress at 7 to the univariable mode for total difficulties at 11 revealed a statistically significant effect (β=0.13, p=0.0023), and again an interaction term with genotype was not statistically significant (p=0.93). Stratified analyses for each of the genotype groups showed no statistically significant effect in the Met homozygotes (β=0.13, p=0.12), and a similar size but significantly significant effect in the Val homozygote / Val/Met heterozygote group (β=0.14, p=0.01) (Table II). These results aren’t entirely consistent with those seen at 7 years; this may reflect the cross-sectional nature of the analysis at 7, whilst the stress variable is being used in a longitudinal manner at 11 years.

We also assessed the effect of genotype on IQ. Compared to the Val homozygotes we found a statistically increased IQ amongst the Met homozygotes (4.0 points 95%CI=1.0, 7.0). A similar pattern was seen with verbal IQ, though neither difference was statistically significant (Met homozygotes 2.9 (-0.3, 6.0). The effect on performance IQ was more pronounced (Met homozygotes 3.8 (0.5, 7.0), (Table III).
DISCUSSION

Recent research with both animals (Weinstock, 2008) and humans (O’Connor et al., 2003; Van den Bergh & Marcoen, 2004) has demonstrated that maternal stress during pregnancy can have long-lasting effects on the physical and psychological development of the offspring (Bergman, Sarkar, & O’Connor, 2007; Huizink et al. 2004; Rice, Lewis, Harold, et al. 2007; Talge et al. 2007). Several factors have been proposed that may moderate how antenatal exposure to stressful events affects both antenatal and postnatal development and the risk of subsequent emotional and behavioural problems in their offspring. One such factor is genetic susceptibility, and evidence suggests that the COMT gene may play a role in affecting the course of problem behaviour (Thapar et al., 2005). COMT is also thought to play a role in the processing of socioemotional and cognitive stimuli (Herrmann, et al., 2009; Smolka, Buhler, Schumann, Klein, Hu, Moayer et al., 2007). Towards this end, the aim of the present study was to assess whether the Val^{158}Met SNP on the COMT gene is a risk factor for behavioural and emotional difficulties during childhood and whether maternal perceived stress moderates this association.

The main finding from our study was that the association between the Val^{158}Met (rs4680) SNP and behavioural problems differed with maternal perceived stress at birth but not years later when the children were 7 years of age. Specifically, ABC study members with the low activity Met/Met genotype were at significantly increased risk of behavioural problems at ages 7 and 11 years when they were exposed to maternal stress in utero relative to carriers of the Met/Val or Val/Val. In comparison, maternal stress at 7 appears to have a significant effect on behavioural problems irrespective of the child’s genotype.

Other studies are consistent with the finding that the Met allele impacts on childhood problem behaviour. For example, Volavka, Kennedy, Ni, et al., (2004) found that those with
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the Met/Met genotype were at increased risk of conduct disorder. Palmason, Moser, Sigmund Vogler, Hanig, Scheider *et al.*, (2010) also found that children with the Met/Met genotype were at increased risk for conduct disorder and had the highest ADHD symptom severity. ADHD is phenotypically complex and, for many individuals with ADHD, symptoms of the disorder include hyperactivity, problems with peers, emotions, and conduct. About 50% of individuals with ADHD show antisocial behaviour (Kutcher, Aman, Brooks, Buitelaar, van Daalen, Fegert *et al.*, 2004) including oppositional defiant disorder and conduct disorder (Wilcutt, Pennington, Chabidas, Friedman, Alexander, *et al.*, 1999).

The Val<sup>158</sup>Met SNP rs4680 leads to either methionine (Met, A allele) or valine (Val, G allele) at codon 158, resulting in an up to a fourfold reduction in *COMT* activity in Met/Met carriers relative to Val/Val homozygotes. Low *COMT* activity results in increased synaptic dopamine activity (Lachman, Papolos, Saito, Yu, Szumlanski, & Weinshilboum, 1996). A positive association between frontal cortex dopamine and aggression has been found in mice (Gogos *et al.*, 1998) but the findings with humans have been inconsistent (Volavka *et al.*, 2004, Thapar *et al.*, 2005, Sengupta *et al.*, 2006). In contrast to our results, Thapar and associates found that individuals homozygous for the high-activity Val allele who were born SGA were more aggressive than similar birth weight carriers of the Met/Met or Val/Met genotypes (Thapar *et al.*, 2005). Similarly, Val/Val carriers had more symptoms of conduct disorder and aggressiveness than methionine carriers in the study by Caspi *et al.*, (2008).

The discrepant findings noted above might be due to methodological differences between studies. For example, Thapar *et al.* (2005) used a continuous variable for the phenotype of conduct disorder whereas Palmason *et al.* (2010) used a categorical phenotype. We chose to use a continuous variable representing both emotional and behavioural problems
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- the Total Difficulties scale from the SDQ. The decision was primarily based on a need for increased statistical power, but it was also based on theoretical reasons. It has been suggested that problem behaviours and disorders such as ADHD, oppositional deviant disorder and conduct disorder should be placed on a continuum (Faraone, Biederman, Keenan, & Tsuang, 1991). Our measure of childhood difficulties therefore consisted of the following deficit-focused subscales: emotional symptoms; hyperactivity; conduct problems; and peer problems. With our sample size, we were unable to explore the possibility that our results might generalize to those with, for example, purely conduct problems or symptoms of hyperactivity. We were also not able to look specifically for any sex differences. Associations between COMT and ADHD have been found to be stronger for males (Biederman, Kim, Doyle, Mick, Fagerness, Smoller, Faraone, 2008).

Our findings support the idea that the COMT gene can influence the aetiology and developmental course of childhood behavioural problems by affecting an individuals’ susceptibility to antenatal environmental adversity. Although problem behaviours are suspected to be sensitive to antenatal, biological and psychosocial environmental risk factors, this is the first study to show that maternal perceived stress during pregnancy is a significant risk factor for those homozygous for the Met allele. As noted earlier, cortisol overactivity is one consequent of perceived stress (Dickerson & Kemeny, 2004). Numerous animal studies have shown an effect of prenatal stress in reprogramming the function of the HPA axis in the offspring, often resulting in a more prolonged and greater cortisol response to stressors later in development (e.g., Meaney & Seckl, 2004; Weinstock, 2001). It is also known that maltreatment stress during early life can alter monoaminergic neurotransmission and influence aggression (Bremner & Vermetten, 2001). Antenatal release of cortisol during pregnancy, as a direct result of maternal stress, might therefore influence the neural
Thompson et al. *COMT* and the effect of maternal stress on behaviour development of the offspring and contribute to later problem behaviour in childhood. It should be noted, however, that our results may be inaccurate with regard to maternal perceived stress. This was measured when the ABC study members were born, so the findings could be subject to recall bias.

Perceived stress by the mother was not hypothesized to play a direct causal role in the development of problem behaviours in the offspring. However, it is possible that high maternal stress may affect or exacerbate problem behaviours, some of which are symptoms of ADHD and comorbid behaviour disorders such as conduct disorder. Since there was no effect of maternal perceived stress on *COMT* genotypes when the children were 7 years of age, we were able to rule out the possibility that maternal stress during childhood interacts with gene expression to increase problem behaviours.

In the current study, we also found a significant association between the *COMT* allele and performance on the Wechsler Intelligence Scale for Children measured at age 7 years. Carriers of the Met/Met genotype had, on average, IQ scores 4 points higher than Val/Val homozygotes. Further analysis showed this effect to be significant for Performance IQ but not for Verbal IQ. Although this association needs replication and further research, it is noteworthy that Raz, Rodrigue, Kennedy and Land (2009) also found that the *COMT* Met/Met genotype was associated with better performance on cognitive measures such as fluid intelligence, inhibition, associative memory, and processing speed. Similarly, Tunbridge, Harrison, and Weinberger (2006) found that carriers of the Met allele showed better performance on executive functioning measures. Executive functions are those involved in tasks such as working memory, sustained attention, self-organisation and regulation.
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Mier, Kirsch and Meyer-Lindenberg (2009), in their meta-analysis of neuroimaging studies of the *COMT* genotype, found significant associations between rs4680 and prefrontal neural activation, with Met allele carriers having significantly better executive functioning. As noted earlier, Met homozygotes have increased synaptic dopamine activity (Lachman, Paplos, Saito, Yu, Szumlanski, Weinshilboum, 1996), which may account for the general and specific cognitive advantages reaped by Met/Met carriers (Sheldrick, Krug, Markov *et al.*., 2008). Regardless of mechanism, it may be that the association between COMT and cognitive performance is related to mental and motor functions thought to involve the efficiency of the prefrontal cortex.

Importantly, the finding of higher performance IQ scores for Met/Met carriers has implications for our earlier results relating *COMT* to behavioural problems. Although purely speculative, it may be that the children who are Met homozygotes and who experienced the effects of maternal stress in utero, are at greater risk for uncontrolled, externalizing behavioral styles because they require greater cognitive challenges at home and at school. Put simply, some children with higher IQs may be disruptive and have greater emotional and social problems because they are bored. In support, a recent report from British birth cohorts revealed that intelligence in childhood is a risk factor for psychological distress (Gale, Hatch, Batty & Deary, 2009). Furthermore, high-IQ youths with ADHD have significantly higher rates of mood, anxiety, and disruptive behavior disorders at follow-up then those without ADHD (Antshel, Faraone, Maglione, *et al.*, 2008).

Psycho-stimulant medication, including methylphenidate and dexamphetamine, reduce cognitive and behavioural symptoms in the majority of children with ADHD. Methylphenidate and dexamphetamine act by facilitating transmission of dopamine and norepinephrine neurotransmitters in the brain (Axelrod, Mueller, Henry, & Stephens, 1970).
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The dopaminergic system encompasses numerous projections throughout the prefrontal lobe (Hurd, Suzuki, & Sedvall, 2001). Executive functioning and the underlying electrophysiological response can be normalised by dexamphetamine (Horrobin, McNair, Kirk & Waldie, 2007). There is also suggestive evidence that both glutaminergic and noradrenergic genes may be associated with methylphenidate response (Anney, Lasky-Su, O’Dushlaine, et al., 2008).

Therefore, an avenue for future research could address gene by gene and gene by environment effects on the cognitive characteristics of ADHD as well as psycho-stimulant response. Future research should also address whether there are sex differences in the *COMT* genotype for those susceptible to antenatal stress.

It is generally accepted that the environment influences gene expression (Mill & Petronis, 2008) and about 25% of the variance in ADHD symptoms has been shown to be accounted for by environmental risk factors (Faraone, Perlis, Doyle, et al., 2005). As such, the family environment, particularly an adverse psychosocial environment during the first 3 years of life, has been the focus of numerous gene by environment studies with individuals with ADHD (Sonuga-Barke, Lasky-Su, Neale, et al., 2008). Our findings are consistent with the idea that the developing foetus must adapt to the mother’s intrauterine environment and they emphasize the potential long-term consequences of prenatal maternal stress during neural development in utero.
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We acknowledge use of genotype data from the British 1958 Birth Cohort DNA collection, funded by the Medical Research Council grant G0000934 and the Wellcome Trust grant 068545/Z/02.
Thompson et al. *COMT* and the effect of maternal stress on behaviour

**References**


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Table I. Univariable Effect* (95% CI) of SGA status, maternal stress and rs4680 on the Strengths and Difficulties Questionnaire Total Difficulties score at 7 and 11 years of age.

<table>
<thead>
<tr>
<th></th>
<th>Total difficulties at 7</th>
<th>Total difficulties at 11</th>
<th>Total difficulties at 7</th>
<th>Total difficulties at 11</th>
</tr>
</thead>
<tbody>
<tr>
<td>SGA</td>
<td>0.04 (-0.90, 0.97)</td>
<td>0.58 (-0.37, 1.53)</td>
<td>0.08 (-0.83, 0.98)</td>
<td>0.85 (-0.15, 1.84)</td>
</tr>
<tr>
<td>Rs4680 Met/Met</td>
<td>0.75 (-0.31, 1.82)</td>
<td>0.05 (-1.01, 1.12)</td>
<td>0.69 (-0.35, 1.73)</td>
<td>0.24 (-0.90, 1.37)</td>
</tr>
<tr>
<td>ValVal / Val/Met</td>
<td>Ref</td>
<td>Ref</td>
<td>Ref</td>
<td>Ref</td>
</tr>
<tr>
<td>Maternal stress at birth</td>
<td>0.10 (0.03, 0.18)</td>
<td>0.11 (0.04, 0.18)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Maternal stress at 7</td>
<td></td>
<td></td>
<td>0.22 (0.14, 0.29)</td>
<td>0.13 (0.05, 0.22)</td>
</tr>
<tr>
<td>Interaction</td>
<td>P=0.03</td>
<td>P=0.05</td>
<td>P=0.86</td>
<td>P=0.93</td>
</tr>
</tbody>
</table>

*Estimates are the difference from the reference category for SGA and genotype, and the change per unit (1) for the maternal stress variables.
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Table II. Stratified analyses of effect* (95% CI) of maternal stress by genotype on the Strengths and Difficulties Questionnaire Total Difficulties score at 7 and 11 years of age.

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Maternal stress at birth at 7</th>
<th>Maternal stress at birth at 11</th>
<th>Maternal stress at 7 at 7</th>
<th>Maternal stress at 7 at 11</th>
</tr>
</thead>
<tbody>
<tr>
<td>Met/Met</td>
<td>0.26 (0.10, 0.42)</td>
<td>0.26 (0.10, 0.41)</td>
<td>0.21 (0.06, 0.36)</td>
<td>0.13 (-0.03, 0.29)</td>
</tr>
<tr>
<td>Val/Met / Val/Val</td>
<td>0.06 (-0.02, 0.14)</td>
<td>0.07 (-0.01, 0.15)</td>
<td>0.22 (0.13, 0.31)</td>
<td>0.14 (0.03, 0.24)</td>
</tr>
</tbody>
</table>

*Estimates are the change in total difficulties score per unit of maternal stress
Thompson et al. *COMT* and the effect of maternal stress on behaviour

Table III. Effect (95% CI) of genotype of rs4680 on IQ at 7 years of age

<table>
<thead>
<tr>
<th></th>
<th>Total IQ</th>
<th>Performance IQ</th>
<th>Verbal IQ</th>
</tr>
</thead>
<tbody>
<tr>
<td>AA</td>
<td>4.0 (1.0, 7.0)</td>
<td>3.8 (0.5, 7.0)</td>
<td>2.9 (-0.3, 6.0)</td>
</tr>
<tr>
<td>AG/GG</td>
<td>Ref</td>
<td>Ref</td>
<td>Ref</td>
</tr>
</tbody>
</table>

Estimates are the differences in IQ from the reference genotype group.
<table>
<thead>
<tr>
<th></th>
<th>John Thompson</th>
</tr>
</thead>
<tbody>
<tr>
<td>AG</td>
<td>1.5 (-1.6, 4.6)</td>
</tr>
<tr>
<td>Linear trend by number of A alleles</td>
<td>2.4 (0.6, 4.2)</td>
</tr>
</tbody>
</table>