A Functional Variant of the Serotonin Transporter Gene (SLC6A4) Moderates Impulsive Choice in Attention-Deficit/Hyperactivity Disorder Boys and Siblings


2011, Biological Psychiatry, 70, 230-236

This is the reformatted manuscript submitted - prior to publication in its final form at doi:10.1016/j.biopsych.2011.01.040

1. Developmental Brain-Behaviour Laboratory, School of Psychology, University of Southampton, Southampton, UK
2. Department of Experimental Clinical & Health Psychology, Ghent University, Ghent, Belgium.
3. Channing Laboratories, Brigham and Women’s Hospital, Boston, MA, USA
4. Department of Developmental and Educational Psychology, University of Valencia, Spain
5. Clinic for Child and Adolescent Psychiatry, University of Duisburg-Essen, Germany.
6. Department of Child and Adolescent Psychiatry and Psychotherapy, Central Institute of Mental Health, University of Mannheim, Germany
7. Department of Psychiatry, Trinity Centre for Health Sciences, St. James’s Hospital, Dublin, Ireland
8. Department of Child and Adolescent Psychiatry, University of Zurich, Switzerland
9. King’s College London, MRC Social, Genetic and Developmental Psychiatry Centre, Institute of Psychiatry, London, UK
10. Radboud University Nijmegen Medical Centre, Department of Cognitive Neurosciences, Nijmegen, The Netherlands
11. Scheinfeld Centre for Genetic Studies in the Social Sciences, Department of Psychology, Hebrew University, Jerusalem & Herzog Memorial Hospital, Givat Shaul, Jerusalem, Israel
12. Department of Child and Adolescent Psychiatry, University of Göttingen, Göttingen, Germany
13. Department of Clinical Neuropsychology, Vrije Universiteit, Amsterdam, The Netherlands
14. Department of Psychiatry and of Neuroscience and Physiology, SUNY Upstate Medical University, Syracuse, New York, USA
15. Department of Neuropaediatrics, La Fe University Hospital, Valencia, Spain
16. Aalborg Psychiatric Hospital, Aarhus University Hospital, Aalborg, Denmark

** Correspondence E-mail: ejb3@soton.ac.uk.

Acknowledgements:
The IMAGE project is a multi-site, international effort supported by National Institutes of Health (NIH) Grants R01MH081803 and R01MH62873 to SVF and, in London, by UK Medical Research Council Grant G03001896 to JK.

Site Principal Investigators for the genetics analysis were Philip Asherson, Tobias Banaschewski, Jan Buitelaar, Richard P. Ebstein, Michael Gill, Ana Miranda, Fernando Mulas, Robert D. Oades, Herbert Roeyers, Aribert Rothenberger, Joseph Sergeant, Edmund Sonuga-Barke, and Hans-Christoph Steinhausen.

Principal investigators for the neuropsychological arm of the study were Tobias Banaschewski, Michael Gill, Jonna Kuntsi, Iris Manor, Ana Miranda, Fernando Mulas, Robert D. Oades, Herbert Roeyers, and Hans-Christoph Steinhausen. Chief investigators at each site were Penny Andreou, Rafaela Marco, Henrik Uebel, Hanna Christiansen, U. Mueller, Isabel Gabriels, and Shera Medad. We are very grateful to all the families and children that took part in the study.
Abstract:

**Background:** Impulsive drive for immediate reward (IDIR) and delay aversion are dissociable elements of the preference for immediate over delayed rewards seen in attention-deficit/hyperactivity disorder (ADHD). We hypothesized that IDIR would be associated with dopamine regulating genes and delay aversion would be associated with serotonin-regulating genes.

**Methods:** Impulsive drive for immediate reward and delay aversion were measured in 459 male children and adolescents (328 ADHD and 131 unaffected siblings) with a laboratory choice task. The sample was genotyped for the 5HTT (SLC6A4) promoter serotonin-transporter-linked polymorphic region polymorphism and a DAT1 (SLC6A3) 40-base pair variable number tandem repeat located in the 3’-untranslated region of the gene.

**Results:** There was no effect of dopamine transporter (DAT1) on IDIR. As predicted, serotonin-transporter-linked polymorphic region s-allele carriers were more delay averse. This effect was driven by the s/l genotype in the ADHD group. These results were not altered by taking account of the rs25531 A/G single nucleotide polymorphism and were independent of age, IQ, and oppositional defiant disorder symptoms.

**Conclusions:** The results support the genetic distinctiveness of IDIR and delay aversion in ADHD and implicate serotonin function in delay aversion. Possible explanations of the heterosis effect in the ADHD cases are presented.

**Key Words:** 5-HTTLPR (SLC6A4), attention-deficit/hyperactivity disorder, DAT1 (SLC6A3), delay aversion, impulsivity

**Introduction:**

The tendency to choose small-sooner over large delayed rewards is regarded as a signal marker of motivational dysfunction in attention deficit/hyperactivity disorder (ADHD) (1). Effect sizes are moderate (Cohen’s $d = .5–.7$) (2), with some between-study heterogeneity (e.g., see Scheres et al. [3] and Bidwell et al. [4] for non-significant findings). In a recent model this preference is explained as the product of two motivational components. The first component is an impulsive drive for immediate reward (IDIR) (1,5). Impulsive drive for immediate reward manifests as a preference for small-sooner rewards in choice experiments where trial length is the same, irrespective of which of the two options is chosen. This is achieved experimentally by arranging a period of post-reward delay (equal in length to the period of delay before the delayed-reward) after delivery of small-sooner rewards (i.e., a post-reward delay condition). The second component is delay aversion that occurs when delay itself acquires a negative emotional valance, motivating actions allowing delay avoidance/escape. One model sees delay aversion as mediated by the experience of social censure associated with failures to perform effectively in delay settings in individuals with more fundamental IDIR-related deficits. In this model, delay aversion exacerbates the effects of IDIR on small sooner reward preference. Consistent with this formulation, in a recently reported choice delay experiment by Marco et al. (6), the preference for small-sooner rewards was significantly increased by removing the post-reward delay period so that choice of small-sooner rewards reduced overall trial delay (i.e., a “no post reward delay” condition). The difference between choices for small-sooner rewards in the post-reward and no-post-reward delay conditions (an index of delay aversion) was significantly greater.
Impulsive drive for immediate reward and delay aversion are postulated to be mediated by different brain systems. Impulsive drive for immediate reward is hypothesized to be associated with dopamine function alterations within reward networks (7) that diminish signalling and reduce the subjective value of future rewards (8, 9). Dopaminergic agents alter response to delayed reward in animal models (10)—consistent with this—in healthy control subjects (11) and ADHD patients (12). Reward-related effects in the ventral striatum, a key component of the reward circuits of the brain, are altered in both preclinical (13) and clinical ADHD studies (3, 14, 15). The 40-base pair variable number tandem repeat (VNTR) polymorphism located in the 3=-untranslated region of the DAT1 gene (SLC6A3; chromosome 5p15.3) contributes to the regulation of synaptic dopamine through altering its reuptake into presynaptic terminals. The DAT1 gene is differentially expressed in ventral striatum (16) and modulates reward-related activation there (17, 18) so that DAT1 genetic effects on impulsivity are thought to be moderated via alterations in reward circuits (18). Studies linking this polymorphism to ADHD give mixed results. Case-control and family association studies have shown inconsistent effects for the 10/10 genotype, and recent meta-analyses show significant but small effects (19, 20).

In contrast, delay aversion—regarded as a specific example of a more general response to negatively valenced environmental stimuli or experiences (21, 22)—is hypothesized to be mediated by amygdala activation (23) and modulated by serotonin function (24). Supporting the notion that delay is negatively valenced for ADHD patients, an attentional bias toward cues of delay—similar to the response of anxious individuals to cues of threat—has been reported (25). Plichta et al. (15) found delay-related hyper-activation in amygdala in ADHD in response to delayed rewards. Serotonin function has been implicated in impulsivity and immediate over-delayed reward choices (26–30). The serotonin transporter (S-HTT), encoded by genetic locus SLC6A4 (chromosome 17q11.2), is a key regulator of serotonin function in the amygdala. Transcriptional activity of the gene is modified by a polymorphic regulatory region, commonly known as the serotonin-transporter-linked polymorphic region (5-HTTLPR). The short allele (“s”) of the 5-HTTLPR is associated with lower transcription and functional capacity of the 5-HTT (31, 32). The 5-HTTLPR promoter polymorphism seems to influence functional (33, 34) and structural (35, 36) properties of the amygdala, in particular in moderating the response to threatening and aversive stimuli (31, 37). We are not aware of any studies of the effect of the 5-HTTLPR in determining delayed reward choices. However, Aluja et al. (38) found that the s-allele was associated with impulsiveness in a prison sample, whereas Oades et al. (39) demonstrated a potential link between another polymorphism in 5-HTT, the intron 2 VNTR, and cognitive impulsivity but not motivational impulsivity in ADHD. The 5HTT gene has also been implicated in ADHD (19, 40, 41), although a recent multicenter study was negative in this regard (42).

Here we test the hypothesis that IDIR and delay aversion will be differentially associated with polymorphisms in DAT1 and 5HTT genes in a secondary analysis of the subsample of male children and adolescents with ADHD and their sex-matched siblings with the Maudsley Index of Delay Aversion (MIDA) (6) data from the Marco et al. study. Our specific predictions
were that the 10R/10R genotype of the DAT1 VNTR will be related to IDIR and the s-allele of the 5HTT promoter polymorphism associated with delay aversion.

Methods and Materials:

Participants

Probands were from child psychiatry and specialist ADHD clinics in six European countries (Belgium, Ireland, Germany, Spain, Switzerland, and the UK) and Israel and of European/Caucasian descent. The study was part of the neuropsychology component of the International Multi-centre ADHD Genetics (IMAGE) project (43). Each had a diagnosis of DSM-IV ADHD-combined type and was between 6 and 16 years of age with at least one sibling in the same age range. The clinical diagnosis was validated against the Conners’ Rating Scales (44, 45) and the Parental Account of Children’s Symptoms (PACS) (46) interview. Siblings were also screened for ADHD, and if they met the inclusion threshold, a PACS was administered to confirm the diagnosis. Exclusion criteria included pervasive developmental disorder, neurological diseases, or other medical and genetic disorders. Parents gave written consent for the children to participate in the study.

Only males were included, to simplify and strengthen the current analysis, because: 1) the number of girls with relevant data was too small (N probands = 35) to allow analysis of possible interactions between gender and genotype (e.g., only 18 female probands with the relevant data carried the most common 10/10 DAT1 genotype compared with 168 male probands), and 2) there were markedly unequal male/female ratios for probands (35 vs. 285) compared with siblings (147 vs. 158). The MIDA data were available for 293 male probands (age range 6–16; mean = 10.78 years, SD = 2.61) and their 169 siblings (age range 5–17; mean = 10.73, SD = 2.98). Genotype data for the DAT1 VNTR were available for 288 probands and 162 siblings and for 291 probands and 168 siblings for the 5-HTTLP. Seven cases with the DAT1 11-repeat allele were excluded from the analysis. Thirty-five siblings had a diagnosis of ADHD (total ADHD cases n = 328) and were designated so for the current analyses.

Tasks and Measures

Clinical Evaluation

Symptom Rating Scales. Four scales were used to assess symptoms of ADHD and comorbid conditions: (the long versions of Conners’ Parent and Teacher Rating Scale and the parent and teacher Strengths and Difficulties Questionnaire) (47).

Research Diagnosis. This was carried out with the revised PACS interview, (46) the Conners’ Parent and Teacher Rating Scales and the Strengths and Difficulties Questionnaire. The PACS is a semi-structured interview used to collect parent-based detailed information on the behaviour of the children. The interviewer asks parents to describe the behaviour of their child in different settings and then rate the severity and frequency of the behaviour according to previously defined criteria. The settings are chosen to represent common unstructured (watching TV, reading, or playing alone), semi-structured (meals, outings, or shopping), and structured (home tasks, homework, or getting ready) daily life situations. In this study, parents were asked to focus on examples of behaviour of their children during the most recent medication-free period. A standardized diagnostic algorithm based on the DSM-IV criteria was applied to the information from PACS and from the teacher-rated subscale from Conners’ to derive a subtype diagnosis. In addition to the ADHD diagnosis, PACS also provides a
Mood and an Anxiety score and a diagnosis of oppositional defiant disorder (ODD) based on the DSM-IV criteria. Previous studies have shown high interrater reliability (product-moment correlations between .76 and .96) (46). The PACS has been validated against standardized questionnaires (such as the Conners’ scale) used to assess ADHD (48).

**Intelligence.** The vocabulary, similarities, picture completion, and block designed subtests from the Wechsler Intelligence Scale for Children, 3rd Edition (49), and the Wechsler Intelligence Scale for Adults, 3rd Edition (50), were administered, and scores were prorated to provide a full estimate of IQ (51).

**Table 1:**

The relationship between impulsive drive for immediate reward (IDIR) and delay aversion and genotype as a function of ADHD status.

<table>
<thead>
<tr>
<th>DAT1</th>
<th>5-HTTLPR</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>s/s</td>
</tr>
<tr>
<td><strong>IDIR</strong></td>
<td></td>
</tr>
<tr>
<td>ADHD</td>
<td>45.00</td>
</tr>
<tr>
<td>none</td>
<td>37.50</td>
</tr>
<tr>
<td><strong>Delay Aversion</strong></td>
<td></td>
</tr>
<tr>
<td>ADHD (SD)</td>
<td>6.15</td>
</tr>
<tr>
<td>None (SD)</td>
<td>12.52</td>
</tr>
</tbody>
</table>

Note: Impulsive Drive for Immediate Reward (IDIR) represents the proportion of individuals who chose the small-sooner reward on all trials in the post-reward-delay condition Delay aversion was based on the difference between the proportion of choices made for the smaller sooner reward in post-reward and no post reward delay condition. Higher scores indicate more small-sooner choices. All scores are age adjusted and represent the standardised residual when age was regressed onto IDIR and Delay Aversion. Figures in parentheses are standard deviations (SD). ADHD, attention-deficit/hyperactivity disorder; VNTR, variable number tandem repeat; 5-HTTLPR, serotonin-transporter-linked polymorphic region.

“no-post-reward delay condition,” each trial followed on immediately after the participant had received their reward so that trial length was determined by the length of the pre-reward delay for the chosen option. In the “post-reward delay condition,” the trial length was equalized for the two reward options by including a period of post-reward delay (2 sec for the large-delayed option or 30 sec for the small-sooner option). Under this condition, the length of trial was always 32
sec (see Marco et al. [6] for a more detailed description of instructions and rewards). Our index of IDIR was the percentage of small-sooner choices on “post-reward delay” trials when choosing this could not reduce overall trial delay (i.e., was not an expression of delay aversion). The delay aversion index was the difference between the percentage of small-sooner choices in the post reward delay condition and the no post reward delay condition (where choosing the small-sooner reduces overall delay). High scores were more negative for both IDIR and delay aversion. Participants received instructions about the different options available in each condition.

**Genotyping**

**DNA Extraction and Genotyping**

The DNA was extracted directly from blood samples or cell lines at Rutgers Cell line and DNA repository in the US. In a few cases, we used a mouth swab sampling technique and extracted the DNA at the Social, Genetic and Developmental Psychiatry Centre laboratories in London. For genotyping of the VNTR markers, we used a standard polymerase chain reaction method according to previous optimized protocols for the markers used in this study. For *DAT1* we contrasted 9R/9R and 9R/10R with 10R/10R (we excluded those carrying the 11R allele). For 5-HTTLPR we compared s/s and s/l with I/l genotypes. We also determined an A/G single nucleotide polymorphism (SNP) (rs25531) within the 5-HTTLPR repetitive element, the G-allele of which has been reported to render the l-allele transcriptionally less efficient (55). Genotyping for this was carried out at the Institute of Psychiatry and followed the protocol outlined in Wendland et al. (55); primers are available on request.

**Procedure**

The procedure for task administration is described in detail in Marco et al. [6]. Families were required to withdraw ADHD medications for at least 48 hours before testing. The study had ethical approval from local site ethics committees.

**Analysis**

We tested whether the delay aversion and the IDIR data met normality assumptions. Delay aversion data were normally distributed. The IDIR data were extremely skewed, with most cases (n = 346; 54%) scoring zero. We therefore adopted different analytic approaches for the two outcomes. For both IDIR and delay aversion, because the data were collected at different sites and within families, we used mixed-effects regression models to account for the three-level nested structure (e.g., controlling for intra-familial sibling relationships). Delay aversion was introduced as a continuous variable. In a first step, the specific hypotheses were tested with a mixed-effects regression model for normally distributed outcomes, with delay aversion as the outcome, a contrast of the s-allele carriers vs. the other genotypes as predictors, and random intercepts at the levels of site and family. All models included ADHD status and its interaction with genotype. In a second step, models were adjusted for age, IQ, and ODD. Because there was one extreme outlier in the data, we tested the models also after setting the outlier to the 95th percentile of the distribution (Winsorization) to prevent it from heavily influencing the statistical parameters. For IDIR (given its non-normal distribution) the outcome was dichotomized to represent zero versus nonzero IDIR. Mixed-effects logistic regression models for binary outcomes were used to test for associations of genotype and ADHD status with IDIR, with random effects and a second analytic step as described for the delay aversion model. The mixed-effects regression models were done with the
Stata v11.1 (StataCorp, College Station, Texas) commands “xtmixed” and “xtmelogit,” respectively. These models were also applied to the A/G SNP supplementary analyses.

Results

Frequencies for common genotypes were as expected and in Hardy–Weinberg Equilibrium: DAT1: 9R/9R: n = 27; 9R/10R: n = 154; 10/10: n = 267; 5-HTTLPR: s/s: n = 87; s/l: n = 233; l/l: n = 139. The DAT1 and 5-HTTLPR genotypes were not significantly associated ($\chi^2 = 1.77; p \text{ values} > .70)$. The IDIR and delay aversion were uncorrelated ($r = .07; p > .10$). Table 1 shows IDIR and delay aversion for genotypes by ADHD status.

Primary Analysis

First we tested the predicted associations (Figures 1 and 2). The IDIR did not vary by DAT1 genotype [10/10 vs. 9/9 and 9/10: $\chi^2(1) \leq .01; p = .99$]. As predicted, 5-HTTLPR s-allele carriers were more delay averse than non-carriers [s/s and s/l vs. l/l: $\chi^2(1) = 4.57; p = .03$]. This effect was slightly stronger when analyses were conducted according to transcriptional activity status (i.e., including SNP rs25531)—“low activity” allele carriers being more delay averse than the “high/high” genotype [$\chi^2(1) = 5.37; p = .02$; for delay aversion and IDIR by transcriptional genotype see Supplement 1]. There was a main effect of ADHD status on delay aversion [$\chi^2(1) = 5.93; p = .01$; as originally found in Marco et al. (6)] but no interaction between genotype and ADHD [$\chi^2(1) = 2.77; p = .10$; transcriptional activity groups: $\chi^2(1) = .64; p = .42$]. This pattern of significance did not change when IQ, ODD, and age were added as covariates [effect of 5-HTTLPR s/s and s/l versus l/l: $\chi^2(1) = 6.30; p = .01$; effect of ADHD status: $\chi^2(1) = 8.03; p = .005$; interaction between ADHD status and genotype: $\chi^2(1) = 3.02; p = .08$] or when outliers were Winsorized at the 95th percentile (value = -15).

![Figure 1: The mean level of MIDA delay aversion as a function of 5-HTTLPR status for the combined s/s and s/l genotype groups compared with the l/l group. Note: Delay aversion is calculated as the difference in percentage choices of the small-sooner option under no post reward and post reward delay conditions. Higher scores mean more delay aversion.](image-url)

Exploratory Analyses

Despite the lack of significant interaction, visual inspection of delay aversion means suggested a rather different pattern in probands and unaffected siblings by 5-HTTLPR genotype. To investigate this, we conducted a set of exploratory post hoc analyses. These suggested that, for the unaffected siblings, there was a strong effect of the s-allele [$\chi^2(1) = 7.29; p = .01$], with s/s and s/l having similar levels of delay aversion and both different from the l/l carriers; whereas for ADHD cases, the effect was carried largely by the s/l genotype with heterozygotes being more delay averse than the homozygotes [$\chi^2(1) = 5.68; p = .02$]. Transcriptional activity status analysis gave the same pattern of results. For the unaffected siblings, the “low/high” group was significantly more delay averse than the “high/high” group [$\chi^2(1) = 6.26; p = .04$]. For the affected siblings, the comparison of “low/high” with the “high/high” group missed statistical significance [$\chi^2(1) = 4.90; p = .09$]. These reduced levels of significance were likely related to the reduced number of participants for whom the rs25531 A/G SNP was available. We
also explored the associations, although not hypothesized, between 5-HTTLPR and IDIR and DAT1 and delay aversion. There were no significant effects [5-HTTLPR and IDIR, traditional grouping: χ²(1) = .65; p = .72; according to transcription activity: χ²(1) = .76; p = .38; DAT1 VNTR and delay aversion: χ²(1) = .94; p = .63].

Discussion

The current results extend our understanding of different elements of impulsive choice, their genetic underpinnings, and by extension their putative neurobiological basis. By providing evidence for differential genetic associations, the results further validate the distinction between IDIR and delay aversion in models of impulsive choice (1). With a hypothesis-testing approach we predicted that IDIR (as measured by percentage of choices for the small-sooner reward in the post reward condition) would be associated with DAT1. This was based on the notion that IDIR is the result of altered signalling of delayed rewards modulated by dopamine function, which is affected by functional polymorphisms in the DAT1 gene. The result was negative, and so the findings were at odds with the previous studies linking DAT1 genotype to impulsive choice, delayed responding (18), delay discounting, and trait impulsivity (56). However, it might be—bearing in mind the nature of the current sample—that effects of DAT1 on impulsive choice are sample specific and in particular might not underpin impulsive choice specifically in ADHD. The 10R allele might confer risk for ADHD only in combination with additional DNA variants in the DAT1 gene. Thus, we had found that a specific haplotype of the DAT1 gene is associated with combined-type ADHD (57), replicating a previous report from a different sample (58); additional DAT1 genetic variants from the S’ region of the gene have also been reported to be associated with ADHD (59). In general, it has been difficult to identify robust and consistent associations between specific dopamine genotypes, including DAT1 and putative neuropsychological endophenotypes (60). The current result therefore adds to this rather fragmented picture, although it is not possible, of course, to rule out the effects of variations in dopamine genes—other than DAT1—involving in dopamine neurotransmission on IDIR.

Our second hypothesis was that delay aversion (the additional effect of linking small-sooner reward choices to delay reduction) would be associated with 5-HTTLPR genotype. This was based on the view that delay aversion was a specific case of a more general avoidant response to aversive events and therefore would be mediated by similar neurobiological mechanisms linked to serotonin function (34). As predicted, 5-HTTLPR genotype was associated with delay aversion with s-allele carriers were more delay averse than non-carriers. This finding should be interpreted in relation to a more general link between 5-HTTLPR and impulsive choice seen in tryptophan depletion studies, suggesting serotonin status affects waiting behaviour and delay-
related choice in other populations (26, 34, 61). However, it presents the first study to extend this to the effects of 5-HTTLPR genotype on impulsive choice behaviour on laboratory tasks. It also represents one of the first studies implicating this genotype in ADHD neuropsychology.

Although not ideally placed to explore the moderation of these effects by ADHD status, given the familial relations between affected and unaffected cases, we conducted separate exploratory analyses for these groups. This confirmed the observation of a rather different pattern of results for the two groups and an unexpected heterosis effect in the ADHD group (the s/l group being the most delay averse). This raises the possibility that 5-HTTLPR genotype effects on impulsive behaviour might be dependent on disorder status or more generally on participant characteristics. This possibility has not been investigated systematically, because most studies of 5-HTTLPR s-allele effects on amygdala reactivity have typically been in samples of healthy volunteers with no history of affective or other psychiatric disorders.

Although most studies have not specifically tested for it, a number of studies have found group-specific evidence of molecular heterosis at the 5HTT gene. Heterozygote subjects have shown lower [125] beta-citalopram serotonin transporter binding in cocaine users (62), increased white-matter lesions among depressed patients (63), higher cognitive function in elderly adults (64), and lower availability of central 5-HTT (16). In a recent study, Malmberg et al. (65) found associations between disruptive behaviour disorder and s/l genotype. Explanations for these effects include: 1) an inverted U-shaped response curve in which either too little or too much gene expression is deleterious; 2) an independent third factor causing a hidden stratification of the sample such that both the two homozygote genotype (s/s and l/l) are independently associated with the highest phenotype score relative to the heterozygote (e.g., s/l); and 3) greater fitness in heterozygotes, because they show a broader range of gene expression than both homozygotes (for a review see Comings and MacMurray [66]). Clearly, although intriguing, our finding showing a disorder specific heterosis effect in families with ADHD children needs to be confirmed in other large independent samples with nonrelated control subjects.

The current results might take us further in understanding heterogeneity in ADHD. Previous studies (6) found that only a subset of ADHD children show impulsive responding on the MIDA. This might therefore be a marker of a subtype of ADHD in which 5-HTTLPR polymorphisms play a particularly important role in the pathogenesis of the condition. This might explain the inconsistency in results relating to the association between ADHD and this genotype. The expectation is that effects would be larger for 5-HTTLPR genotypes in a refined delay averse sample of ADHD children. If this were the case, it might be possible to isolate a subgroup whose ADHD is mediated by delay averse and might respond to serotonergic drugs (39) as a component of their treatment on the one hand or delay training on the other (25). The results of Zepf et al. (67) demonstrating that ADHD children with comorbid anxious-depression and/or aggression were sensitive to tryptophan depletion highlight the possibility that a delay averse subgroup might be more likely to have these comorbidities.

The current study had many strengths. These included the large sample and the use of an experimental paradigm to dissect different elements of impulsive
choice; however, there were a number of limitations. First, the skewed distribution of the IDIR measure and the need to dichotomize it for the analysis rather than use it as a continuous measure might have reduced its sensitivity compared with the delay aversion measure; the negative finding therefore needs to be interpreted with caution, although the effects were very far from significant. Second, the study did not include direct measures or manipulations of serotonin or dopamine levels, which would have helped resolve issues around the functional significance of the different allelic combinations. Third, there were insufficient affected girls in this subset of the IMAGE sample to provide power to include gender as a factor in the analysis. Finally, the current sample with genotypic information did not include unrelated control subjects—this means that it remains uncertain how specific the role of these genotypes might be to ADHD because of the familial link and associated genetic overlap between probands and their unaffected siblings. Future studies should include biologically unrelated control subjects and groups of patients with other disorders to examine this issue.

Supplementary material cited in this article is available online (below).

**Financial Disclosures**

ES-B is a member of an Advisory Board to Shire, Flynn Pharma, UCB Pharma, and Astra Zeneca; has received research support from Janssen Cilag, Shire, and Qbtech; conference support from Shire; is on speaker board for Shire and UCB Pharma; and has been a consultant for UCB Pharma and Shire.

AM is advisor to Eli Lilly.

RDO has received support for investigator-initiated studies from UCB, GmbH.

TB served in an advisory or consultancy role for Desitin, Lilly, Medice, Novartis, Pfizer, Shire, UCB, and Viforpharma. He received conference attendance support or received speaker’s fee by Lilly, Janssen, McNeil, Medice, Novartis, and UCB. He received unrestricted grants for organizing a CME conference by Lilly, Janssen, McNeil, Medice, Novartis, Shire, and UCB. He is/has been involved in clinical trials conducted by Lilly, Shire and Novartis. The present work is unrelated to the aforementioned grants and relationships.

HU received conference attendance support or was paid for public speaking by Lilly, Janssen-Cilag, Novartis, and Medice. JK has received a speaker’s fee from Eli Lilly that has been used for educational and research activities.

JB has been, in the past 3 years, a consultant to/member of Advisory Board of/and/or speaker for Janssen Cilag BV, Eli Lilly, Bristol-Myer Squibb, Organon/Schering Plough, UCB, Shire, Medice, Servier, Bioprojet, Pfizer, and Servier. HR is a member of an Advisory Board to Shire and has received research funding and conference attendance support from Shire and Eli Lilly.

AR is on the Advisory Board and Speakers’ Bureau of Medice, Novartis, Shire, and Eli Lilly; has received educational grants from Shire and Medice; and has received research support from Shire and Schwabe.

JS is a member of an Advisory Board to Lilly and Shire; has received research funding from Lilly; and speaker’s fees from Lilly, Janssen-Cilag, Novartis, and Shire. HCS has served as an adviser and speaker to Janssen-Cilag, Eli Lilly, Novartis, Shire, Medice, and UCB.

PA has served as a consultant and on advisory boards for Eli Lilly, Shire, Janssen Cilag, and Flynn Pharma. He received a research grant from Shire and an educational grant from Janssen-Cilag.

In the past year, SVF has received consulting fees and has been on Advisory Boards for Shire Development and has received research support from Pfizer, Shire, and the NIH. In previous years, he has received consulting fees or has been on Advisory Boards or has participated in continuing medical education programs sponsored by Shire, McNeil, Janssen, Novartis, Pfizer, and Eli Lilly. In previous years he has received research support from Eli Lilly, Shire, Pfizer, and the NIH. Dr. Faraone receives royalties from a book published by Guilford Press: Straight Talk about Your Child’s Mental Health.

All other authors reported no biomedical financial interests or potential conflicts of interest.

**References**


ventral striatal reactivity associated with impulsivity. *Mol Psychiatry* 14:60–70.


**Supplemental Information I**

**Genotype frequencies based on transcriptional activity groups (5HTTLPR & rs25531)**

Genotypes for the rs25531 SNP were available for n=446 individuals. 26.5% (n=118) of the sample were assigned to the transcriptional activity group low/low, the TA high/low group comprised 50% (n=223), and the TA high/high group comprised 23.5% (n=105). No deviation from Hardy-Weinberg equilibrium was observed (p=.99).

**Supplemental table**

<table>
<thead>
<tr>
<th>5-HTTLPR incl. rs25531</th>
<th>low/low</th>
<th>low/high</th>
<th>high/high</th>
</tr>
</thead>
<tbody>
<tr>
<td>DAv</td>
<td>ADHD</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>10.13 (2.34)</td>
<td>15.21 (2.19)</td>
<td>8.41 (2.35)</td>
</tr>
<tr>
<td>none</td>
<td>7.20 (3.20)</td>
<td>15.06 (3.18)</td>
<td>3.30 (3.65)</td>
</tr>
</tbody>
</table>

The relationship between delay aversion (DAv) and 5-HTTLPR genotype as a function of ADHD status. Figures in parentheses are standard errors.