Dopamine Transporter Gene Moderates Response to Behavioral Parent Training in Children With ADHD: A Pilot Study

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There is great variability in the degree to which children with attention deficit/hyperactivity disorder (ADHD) improve through behavioral treatments. This study investigates the influence of the dopamine transporter gene (SCL6A3/DAT1) on outcome of behavioral parent training (BPT). Study subjects were a subsample (n = 50, for whom DAT1 genotypes were available) of a randomized controlled BPT effectiveness study (N = 94) comparing BPT plus ongoing routine clinical care (RCC) versus RCC alone in referred children (4–12 years old) with ADHD. Treatment outcome was based on parent-reported ADHD symptoms and behavioral problems. Presence of 2 versus no or 1 DAT1 10-repeat allele served as moderator variable. Time × Treatment × Genotype effect was analyzed with repeated-measures analysis of variance, controlling for baseline medication status. Results indicate that DAT1 moderated treatment response (p=.009). In children with no or 1 DAT1 10-repeat allele, superior treatment effects of BPT + RCC compared with RCC alone were present (p=.005), which was not the case in children with 2 DAT1 10-repeat alleles (p=.57). Our findings suggest that genetic differences in DAT1 in children with ADHD influence their susceptibility to a behavioral intervention directed at shaping their environment through their parents. The role of the dopamine system in motivation and learning and in the aberrant sensitivity to reinforcement in children with ADHD may explain this moderating effect, given that the management of contingencies is typically addressed in BPT.

Keywords: ADHD, parent training, dopamine transporter gene, moderator, randomized controlled trial

Although a number of studies have provided evidence for the efficacy of behavioral parent training (BPT) as a treatment for children with attention-deficit/hyperactivity disorder (ADHD; for an overview of the available evidence, see National Collaborating Centre for Mental Health, 2009), there remains great variability in the degree to which children with ADHD improve through behavioral treatments. Several factors associated with response to behavioral treatments in ADHD have been identified, including presence of comorbidity, parental depression, and socioeconomic variables (for an overview, see Hinshaw, 2007). However, virtually nothing is known about the possible association of genetic factors with response to psychosocial interventions. This is surprising, given the growing evidence of the role of gene–environment interactions in the etiology of ADHD and associated behavioral problems (Thapar, Harold, Rice, Langley, & O’Donovan, 2007; Thapar, Langley, Asherson, & Gill, 2007). BPT can be seen as an intervention that particularly focuses on the child’s environment. It may well be the case that genetic factors play a role in the child’s susceptibility to interventions that are directed at changing his or her environment in a positive way. A theoretical framework for this hypothesis is provided by Belsky’s differential susceptibility perspective (see Belsky & Pluess, 2009; Belsky, Bakermans-Kranenburg, & Van Ijzendoorn, 2007; Ellis, Boyce, Belsky, Bakermans-Kranenburg, & Van Ijzendoorn, 2009).
2011), which particularly stresses that some individuals are more susceptible than others to both negative and positive environmental influences, with specific moderating genes that seem to function as plasticity factors (for an overview, see Belsky & Pluess, 2009).

To our knowledge, only one study has examined the interaction between genetic factors and response to behavioral treatment (Bakermans-Kranenburg, Van IJzendoorn, Pijlman, Mesman, & Juffer, 2008). That study investigated the role of dopamine receptor D4 (DRD4) polymorphisms in explaining differential effects of a behavioral parent treatment in preschool children with externalizing behavior problems. Results indicated that children with the DRD4 seven-repeat allele showed larger intervention effects at 1-year follow-up than did children without the DRD4 seven-repeat allele. Treatment effects were particularly large in children with the DRD4 seven-repeat allele whose parents showed the largest increase in positive discipline.

In contrast to the scarcity of studies on genetic susceptibility to behavioral interventions stands the growing interest in identifying genetic polymorphisms associated with stimulant medication response (i.e., pharmacogenetic studies). The studies in that area have largely focused on dopamine-related genes, on the basis of the notion that stimulant medications affect dopamine release or reuptake through blockade of the dopamine transporter, thus leading to increased synaptic dopamine (Thapar, O’Donovan, & Owen, 2005; Volkow & Swanson, 2003; Volkow, Fowler, Wang, & Swanson, 2004). With respect to the genes that have been examined in association with methylphenidate response (for an overview, see McGough, 2005), the most evidence is available for the dopamine transporter gene (DAT1 40bp variable number of tandem repeats). However, until now, findings have been equivocal (McGough, 2005), with some studies pointing to an association of homozygosity of the 10-repeat allele (Roman, Rohde, & Hutz, 2004; Winsberg & Comings, 1999) and others of homozygosity of the nine-repeat allele (Joober et al., 2007; Stein et al., 2005) and poor response to methylphenidate.

It could be hypothesized that DAT1 is also associated with response to BPT. The dopamine transporter has a crucial role in regulating the extracellular midbrain dopamine level in the ventral tegmental area and substantia nigra, which modulate a wide variety of functions, including attention, reward, learning, and operant conditioning (Ettenberg, 1989; Le Moal, 1995; Schultz, 1997, 2001, 2006; Waelti, Dickinson, & Schultz, 2001). Particularly operant conditioning is the central mechanism through which BPT programs appear to work (Berkowitz & Graziano, 1972). Reinforcement is one of the main interventions in BPT, whereas several findings support the suggestion that children with ADHD are aberrantly sensitive to reinforcement (for a review, see Luman, Oosterlaan, & Sergeant, 2005); for example, children with ADHD prefer immediate over delayed rewards (Rapport, Tucker, DuPaul, Merlo, & Stoner, 1986; Sonuga-Barke, Taylor, Semb, & Smith, 1992; Tripp & Alsop, 2001) and need more frequent and intensive reinforcement to increase their task performance and motivation level (Haenlein & Caul, 1987). These reward contingencies are typically addressed in BPT.

Our aim in the present pilot study was to examine whether DAT1 polymorphisms are associated with BPT treatment response in children with ADHD. We conducted our study as a post hoc, secondary, pilot analysis within a previously conducted randomized controlled treatment outcome study (Van den Hoofdakker et al., 2007). In that study, our primary goal was to investigate the effectiveness of a 12-session BPT group program targeting both behavioral problems and ADHD symptoms for parents of referred children with ADHD versus ongoing routine clinical care (RCC). We found that a treatment program that incorporated both BPT and RCC (BPT + RCC) was superior in reducing behavioral (oppositional) problems compared with treatment with ongoing RCC alone, whereas ADHD symptoms decreased over time regardless of treatment group. Children’s medication status at baseline did not affect outcome. Also, treatment allocation did not appear to affect medication status posttreatment, other than that children allocated to RCC alone were more likely to receive polypharmaceutical treatment.

Method

Participants

The 50 children who participated in the present study came from a larger sample of 94 referred children with ADHD whose parents had participated in a randomized controlled treatment study (Van den Hoofdakker et al., 2007), which examined the effectiveness of BPT + RCC compared with RCC alone.

About two years after finishing the treatment study, we contacted families to ask them to participate in the current study, that is, to provide children’s DNA. We tried to approach all 94 families, by mail and, in cases of nonresponse, by telephone. This resulted in 59 families who gave informed consent (in line with Dutch medical-ethical regulations) to collect DNA and 35 families who did not participate. These 35 families included 17 families who refused to participate and 18 families who did not respond at all or who initially agreed to participate but did not return DNA.

DNA was of insufficient quality for DAT1 genotyping in nine of the 59 children, resulting in 50 children for whom DAT1 genotypings were available.

Child and family characteristics of the children participating in the current genetic study are given in Table 1. No statistically significant differences in age, IQ, sex, baseline medication status, comorbidity, ethnicity, and parental education level (examined through univariate analyses of variance [ANOVAs] in the case of continuous variables and chi-square tests in the case of dichotomous variables) were present between participants and nonparticipants in the genetic study.

Procedure of the Treatment Study

To be eligible for the treatment study, children had to meet Diagnostic and Statistical Manual of Mental Disorders (4th ed.; DSM–IV; American Psychiatric Association, 1994) criteria for ADHD, as assessed by the Dutch parent version of the Diagnostic Interview Schedule for Children—IV (Shaffer, Fisher, Lucas, Dulcan, & Schwab-Stone, 2000), with or without comorbid disorders. The treatment study involved either parents who preferred behavioral treatment over medication as a first line of ADHD treatment or parents of children who still had behavioral problems despite medication treatment, that is, if medication was (partially) ineffective or had to be stopped because of side effects. Other inclusion criteria for the treatment study were an IQ above 80 and age
between 4 and 12 years. Parents who had already received intensive BPT the year before or had problems that required immediate intervention (e.g., crisis in the family) were excluded.

On referral, families received routine intake assessments and were subsequently offered RCC by a child psychiatrist or a supervised trainee. If parents still reported behavioral problems after this first phase of RCC, BPT was offered and the parents were asked to participate in the treatment study. After written informed consent was provided by the parents, subjects were randomly assigned to either BPT plus ongoing RCC \( (n = 11005) \) or RCC alone \( (n = 11005) \). Parents in the latter condition were placed on a waiting list for BPT. A detailed description of recruitment strategies, patient selection, and demographic variables of that sample is available in Van den Hoofdakker et al. (2007).

### Treatments

BPT was delivered by experienced psychologists in twelve 2-hr group sessions spread over 5 months, with six children’s parents per group. The program included information about the nature of ADHD; cognitive restructuring of parental cognitions; and behavioral management techniques such as structuring the environment, setting rules, giving commands, anticipating misbehaviors, reinforcing positive behaviors, ignoring, using punishment, and implementing token systems. Reinforcement of positive behaviors through praise and rewards played a major role in the BPT program. This parental skill was introduced in Session 3 and rehearsed in all following sessions. Parents were trained to praise and reward prosocial behaviors immediately, that is, without delay, and with high frequency, intensity, and consistency.

Problem behaviors that were addressed in the training were individualized for each child, as were most of the homework assignments. More details about the BPT treatment, therapists, and treatment integrity are available in Van den Hoofdakker et al. (2007). A more detailed description of the BPT manual can be obtained from Barbara J. van den Hoofdakker.

### Collection of DNA

Buccal smears were collected using cervical brushes, stored in buffer containing proteinase K and sodium dodecylsulfate, and sent to the laboratory by mail. Samples were stored at 4 °C until the DNA was isolated. DNA was isolated using salt extraction followed by isopropanol precipitation.

### Genotyping

\( DAT1 \) variable number of tandem repeats were analyzed using polymerase chain reaction (PCR) after fragment analysis. PCR was
performed under standard reaction conditions using a 6-carboxyfluorescein (FAM)-labeled forward primer (FAM 5’-TGT GGT GTA GGG AAC GCC CTG AG-3’) and a reverse primer (5’-CTT CCT GGA GGT CAC GGC TCA AGG-3’; Kirley et al., 2002). We performed 35 cycles of denaturation (95 °C, 30 s), annealing (65 °C, 30 s), and extension (72 °C, 1 min), followed by a final extension step of 10 min at 72 °C. The resulting PCR products were separated using capillary electrophoresis (ABI 3130 analyzer; Applied Biosystems, Nieuwerkerk aan den IJssel, the Netherlands), and fragment sizes were estimated using ABI Prism GeneMapper (2002) software. Fragments of 360, 400, 440, 480, and 520 base pairs correspond to seven, eight, nine, 10, and 11 repeats, respectively. Our sample consisted of two DAT1 9/9-, 21 DAT1 9/10-, and 27 DAT1 10/10-repeat allele individuals. This genotype distribution was consistent with Hardy–Weinberg equilibrium (the expected respective numbers of individuals was 27.4, 18.8, and three, respectively; the 9-repeat allele frequency of 0.25 was χ² = 0.68, p = .41). Children were subdivided into two groups: DAT1 10-repeat allele homozygote subjects (n = 27) versus subjects with one or no 10-repeat allele (n = 23). This division was based on a previous study showing that presence of two DAT1 alleles is associated with ADHD symptoms and response inhibition (Cornish et al., 2005).

Outcome Measures

For moderator analyses, a composite may be the preferred outcome measure in that it more validly captures treatment outcome in a single measure, compared with a range of separate measures (Conners et al., 2001), especially considering the fact that a distinction between ADHD target symptoms and behavioral problems is not explicitly made within the BPT treatment. Therefore, as our primary outcome measure, we used a composite score constructed from rating scales that cover the primary targets of the BPT program: behavioral problems and ADHD symptoms.

The composite score was derived from three parent-based outcome measures, that is, the ADHD index subscale of the Conners’ Parent Rating Scale—Revised: Short Form (CPRS–R:S; Conners, 2001), the Externalizing scale of the Child Behavior Checklist (CBCL; Achenbach, 1991), and occurrence and severity of five individually chosen target problems. The target problems were assessed with short telephone interviews (for details, see Van den Hoofdakker et al., 2007). Examples of target problems include being noncompliant, arguing or discussing, displaying aggression against others, being easily and often angry, and finishing tasks not playing on his or her own, and crying easily. Respective Pearson correlation coefficients between the three measures were .12 (ADHD index and individual target problems), .34 (CBCL Externalizing and individual target problems), and .48 (CBCL Externalizing and ADHD index). These intercorrelations are medium to small, indicating some relation but also some specific variance. Following the procedure described in Rosenthal and Rosnow (1991), we computed the composite score by averaging the z scores of each of the three instruments over the two assessments at Time 1 (T1) and Time 2 (T2), thus giving equal weight to the three outcome measures.

The CPRS–R:S and CBCL were filled in by the primary caregiver, in all cases the mothers; the individual target problems were evaluated through 1–2 min daily telephone calls with one of the parents on 10 consecutive school days. In these phone calls, one of the parents was asked by a trained psychologist, in a neutral way, if any of the target behaviors had occurred during the past 24 hr. If the answer was yes, he or she was asked to rate the severity of each observed target behavior on a 5-point scale ranging from 1 = not severe to 5 = exceptionally severe.

Given that some studies into the efficacy of BPT for children with ADHD demonstrated improvement in ADHD symptoms (e.g., Anastopoulos, Shelton, DuPaul, & Guevremont, 1993; Sonuga-Barke et al., 2002), whereas others showed effects on behavioral problems (e.g., Bor, Sanders, & Markie-Dadds, 2002; Pisterman et al., 1992), we also investigated the role of DAT1 genotype specifically on improvement of behavior problems versus ADHD symptoms. For this aim, we used the ADHD index of the CPRS–R:S, the CBCL Externalizing scale, and the assessment of the five individually chosen target problems as separate, secondary outcome measures.

We assessed all measures at two time points: T1 and T2, directly before the start of BPT or continued RCC (T1), and again directly after the BPT or ongoing RCC (T2), approximately 20 weeks after T1.

Statistical Analyses

The subjects were analyzed on an intention-to-treat basis. For two subjects, composite measure data at T2 were partially missing. We used the last observation carried forward technique to replace missing data at T2.

We assessed the statistical significance of the effects of time, of Time × Treatment Group, and of Time × Treatment Group × DAT1 Genotype (i.e., presence of two vs. no or one DAT1 10-repeat allele) by examining the interaction Fs, using repeated measures (mixed) ANOVAs. The composite measure was entered as the dependent variable, with time points T1 and T2, whereas treatment group and DAT1 genotype were subsequently included as between-subjects factors. Medication status (i.e., presence or absence of baseline psychotropic medication) was included as a covariate to control for possible confounding effects of medication. Finally, the analyses of the effects of Time × Treatment Group × DAT1 Genotype were repeated with the ADHD index of the CPRS–R:S, the CBCL Externalizing scale, and the assessment of five individually chosen target problems as outcome measures.

We used an alpha level of .05 to indicate statistical significance. In the case of a significant interaction effect between treatment and DAT1 genotype, we conducted a repeated-measures ANOVA in the genetic subgroups separately to further analyze treatment effects and to investigate the direction of the interaction effect. Pre- and posttreatment effect sizes (Cohen’s d) in the four subgroups—2 (genotypes) × 2 (treatment groups)—as well as effect sizes regarding the difference between the effects of the RCC + BPT over the RCC treatment in the two genotype groups were calculated by dividing the difference in the mean (difference) scores at T1 and T2 by the pooled standard deviation (Rosnow & Rosenthal, 1996).

Results

The results of the repeated-measures ANOVA examining effects of time, of Time × Treatment Group, and of Time ×
TREATMENT GROUP × DAT1 GENOTYPE ON OUR PRIMARY OUTCOME MEASURE ARE PRESENTED IN TABLE 2. THE FINDINGS IN TABLE 2 INDICATE THAT EFFECTS OF TIME AND TIME × TREATMENT GROUP WERE STATistically significant, which is in line with our earlier findings (Van den Hoofdakker et al., 2007, 2010). In addition, a statistically significant interaction effect between time, treatment group, and DAT1 genotype was present.

Results of the repeated-measures ANOVA in the children carrying two DAT1 10-repeat alleles, F(1, 25) = 0.336, p = .568, indicate no significant outcome difference between the two treatment arms. Pre- and posttreatment effect sizes for the different genotypes and treatment groups are given in Table 2, and findings are illustrated in Figure 1A.

In contrast, results of the repeated-measures ANOVA in the subgroup of children carrying one or no DAT1 10-repeat allele, F(1, 21) = 9.722, p = .005, indicate a significant outcome difference between the two treatment arms, in favor of the BPT + RCC group. Pre- and posttreatment effect sizes for the different genotypes and treatment groups are given in Table 2; in Figure 1B, an illustration of the findings is presented. Effect sizes regarding the difference between the effects of the RCC + BPT over the RCC treatment were 0.21 for the DAT1 10-repeat homozygotes and 1.25 for the children with no or one DAT1 10-repeat allele.

The results of the analyses on the role of DAT1 genotype specifically on improvement of behavior problems versus ADHD symptoms were statistically significant for the five individually chosen target problems, F(1, 48) = 6.494, p = .014, and approached significance for the CBCL-Externalizing scale, F(1, 48) = 3.637, p = .063, and—to a lesser extent—for the ADHD index of the CPRS–R:S, F(1, 48) = 2.770, p = .103.

There is great variability in the degree in which children with ADHD improve through pharmacological and behavioral treatments. In contrast to the growing interest in the interaction between medication response and genetic factors in children with ADHD, the area of possible associations between behavioral treatment response and genes remains understudied, despite the large potential future impact on clinical decision making of insights into these interactions. Our aim in the present pilot study was to investigate the interaction between children’s DAT1 genotype (presence of two vs. no or one DAT1 10-repeat allele) and response to BPT in children with ADHD.

We found that DAT1 moderated treatment response: In children with two DAT1 10-repeat alleles, no differences between BPT + RCC and RCC alone were apparent, as opposed to the results with the children with no or one DAT1 10-repeat allele. Only in the latter group was BPT + RCC more effective than RCC alone. These findings suggest that genetic differences in DAT1 in children with ADHD play a role in their susceptibility to a behavioral intervention directed at shaping and enriching their environment through their parents. Thus, DAT1 polymorphisms may be a candidate plasticity factor of children with ADHD, determining susceptibility to both negative and positive environmental influences, in line with the differential-susceptibility perspective conceptualized by Belsky (Belsky & Pluess, 2009; Belsky, Bakermans-Kranenburg, & van Ijzendoorn, 2007; Boyce & Ellis, 2005; Ellis et al., 2011), although in the current study we did not investigate...
the effects of negative environmental factors in relation to DAT1 genotype.

The role of the dopamine system in motivation and learning (Ettenberg, 1989; Schultz, 1997, 2001, 2006; Waelti et al., 2001) and in the aberrant sensitivity to reinforcement in children with ADHD (for a review, see Luman et al., 2005) may explain the moderating role of a dopamine-related genetic polymorphism in response to BPT. Children with ADHD often have motivation problems when reinforcement is not readily available (Luman, Oosterlaan, & Sergeant, 2008), and continuous and immediate reinforcement appears to be effective (Haenlein & Caul, 1987). Furthermore, children with ADHD may be sensitive to the frequency but unaware of the magnitude of punishments (Luman, Oosterlaan, Knol, & Sergeant, 2008). The management of contingencies is typically addressed in BPT, in which parents learn how to praise (immediately and with high intensity) and how to punish. It is possible that one genetic subgroup of children with ADHD (i.e., children with no or one DAT1 10-repeat allele) is more sensitive to this parental approach, whereas the other subgroup (i.e., homozygote DAT1 10-repeat children) may be less susceptible to changes in their environment. More evidence for genetically based differences in sensitivity to family environment has recently been demonstrated by Sonuga-Barke et al. (2009), who found that sensitivity to the effects of positive maternal expressed emotion was moderated by dopamine and serotonin transporter genotypes. In this study, a reduced sensitivity to positive maternal expressed emotion was seen in homozygote DAT1 10-repeat children, which is clearly in line with the findings in our study.

Inherent to the character of our pilot study is the low number of participants. Clearly, future studies with larger sample sizes are warranted, either through newly designed studies or by collecting DNA on previously conducted treatment outcome studies, first to replicate our findings and second to throw more light on possible pathways of the differential treatment response in children with different polymorphisms of DAT1. How parenting behaviors interact with the suggested genetically determined differences in sensitivity to environmental differences of the child and why it appears that DAT1 moderates response to BPT should be investigated and currently remains unclear. Studies with larger sample sizes would also allow researchers to examine other genetic subgroups (e.g., DAT1 9-repeat allele homozygote children). Furthermore, the possible involvement of other dopamine-related genes (including DRD4 and DRD2) deserves further investigation (see Bakermans-Kranenburg & Van IJzendoorn, 2011). Moreover, future studies should include multiple informants, including teachers, rather than the parents alone. Finally, larger scale studies could more thoroughly investigate the effect of genotype on different outcome measures; the current findings suggest somewhat more specific effects of DAT1 genotype on improvement of behavior problems than of ADHD symptoms.

Some factors regarding the representativeness of our sample in this study need to be considered. First, we included children whose parents still reported behavioral problems after a first phase of RCC. Second, even if we investigated BPT treatment as an adjunct to possible pharmacotherapy, many children may not have been properly medicated. Although it would certainly be worthwhile for researchers conducting future studies to investigate the effectiveness of BPT after an optimal medication regime, the design of the present study may have had more ecological validity. Finally, although representative of our region and mental health clinic but in contrast to other ADHD treatment study samples (e.g., MTA Cooperative Group, 1999), our study included mainly participants of White origin and from two-parent families.

In conclusion, this is the first study that examined the interaction between DAT1 and response to BPT, showing that DAT1 polymorphisms moderate treatment response. This finding is intriguing, given the growing awareness that ADHD is caused by a complex interplay between genetic and environmental risk factors (Biederman & Faraone, 2005; Thapar, Harold, et al., 2007; Thapar, Langley, et al., 2007) and given current evidence that dopamine
transporter genotypes may moderate sensitivity to family environment in children with ADHD (Sonuga-Barke et al., 2009). BPT aims to improve parenting behavior, thus addressing an important aspect of the child’s environment.

References


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