Altered Response with Methylphenidate to ADHD-Like Symptoms in Pervasive Developmental Disorder: Does CES-1 Enzyme Gene Polymorphism Have a Role?

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Objective: Methylphenidate is the first-choice medication for the Pervasive Developmental Disorders (PDDs) and comorbid Attention Deficit Hyperactivity Disorder (ADHD). But this approach generally results in poor outcomes and increased adverse effects. It is aimed to investigate the comparison of cases who diagnosed with PDDs and Mild Mental Retardation (MR) and cases with pure ADHD in terms of the clinical response MPH. Also we aimed to investigate the relations between CES-1 polymorphism gene and the clinical response to MPH.

Methods: We hypothesized that a genetic variation that affects the metabolism of MPH may lie in the etiology of disrupted drug response. To clarify this, we searched for three polymorphisms (Arg199/ His, Ser75/Asn, and Ile49/Val) within the carboxylesterase-1 gene (CES1) in the saliva of patients diagnosed with both PDD+ADHD. Also, we assessed the clinical response to MPH by a dimensional approach using the Attention Deficit Hyperactivity Disorder Rating Scale IV and Clinical Global Impression-Improvement scale.

Results: PDD+ADHD groups had significantlyå higher Arg199/His polymorphisms, and clinically responded poorer –with symptoms sometimes even worsening- to the MPH treatment compared with "pure" ADHD and ADHD+MR groups.

Conclusion: This is the first study that defines an association between Arg199/His polymorphism in CES1 and altered treatment response to MPH in patients with PDD that pres ents with symptoms of ADHD.

Keywords: Pervasive Developmental Disorder, Attention Deficit Hyperactivity Disorder, Methylphenidate, Carboxylesterase-1 , Pharmacogenetics

INTRODUCTION

Pervasive Developmental Disorder (PDD) is amongst the psychiatric disorders of childhood where abnormal or deteriorated development is observed in social skills, linguistic development and behaviors. PDD is a neurodevelopmental disease, with its etiology yet to be fully elucidated. According to the DSM-IV, PDD is an exclusion criterion for attention deficit hyperactivity disorder (ADHD), but it is well known that a considerable amount of ADHD symptoms accompanies PDD in both daily clinical practice and also in scientific literature (Frazier et al. 2001, Goldstein et al. 2004, Lee et al. 2006). A study by Grzadzinski et al. reported that the frequency of ADHD symptoms in individuals with PDD are between 13% to 50%, and this rate was about 20% to 85% in clinical samples (Grzadzinski et al. 2011). As a consequence, autistism spectrum disorder was removed from the exclusion criteria of ADHD in the DSM-V.

Methylphenidate (MPH) is the most frequently used pharmacological agent in ADHD treatment. MPH inhibits dopamine reuptake over presynaptic dopamine carriers in the brain and increases extracellular dopamine. The carboxylesterase-1 (CES-1) enzyme hydrolyzes MPH. CES-1 is thought to be the main enzyme responsible for the first-pass effect in MPH metabolism (Sun et al. 2004). In a previous

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study, two polymorphisms were determined within the CES-1 gene, and blood concentrations of MPH were shown to increase significantly due to these polymorphisms (Zhu et al. 2008). The studies that evaluated whether MPH efficiency is affected by the CES-1 polymorphism revealed that plasma concentrations of MPH increases in the presence of the polymorphism (Bruxel et al. 2013, Johnson et al. 2013).

MPH is well tolerated in cases with ADHD, but MPH response is variable in PDD and ADHD combined cases. A review by Cortese et al. revealed that there is not a consensus about the MPH response in cases of PDD with accompanying ADHD symptoms, and results were varying both for clinical effects and adverse events. The majority of the studies and clinical observations indicated that MPH response in cases with PDD is worse when compared to cases with ADHD without PDD, and also causes more adverse events notably as deteriorations of stereotypes (Cortese et al. 2010). Results of a meta-analysis about treatment of ADHD symptoms in children diagnosed with PDD also supported these findings, and showed that MPH causes more serious adverse effects when compared to placebo (Reichow et al. 2013). Another recent study suggested that MPH decreased ADHD symptoms (Young and Findling 2015). Politte et al. reported in their study that MPH decreased ADHD symptoms in patients with PDD, but the degree of this effect was smaller than the patients with pure ADHD and adverse events were more frequent in PDD cases (Politte et al. 2014). The issue of why MPH is less tolerated in cases with accompanying PDD when compared with pure ADHD cases is not yet elucidated.

When knowledge of metabolization of MPH by the CES-1 enzyme, findings about the clinically significant differences in MPH response in cases of CES-1 gene polymorphism, and opinions about why MPH is less tolerated and causes more adverse events in cases with PDD and accompanying ADHD symptoms are all considered, the necessity of evaluation of the CES-1 enzyme system in cases with PDD and accompanying ADHD emerges. On the other hand, some authors suggested that intolerance to MPH in autistic cases might be related to mental retardation (Stigler et al. 2008). In this context, comparing the MPH responses in autistics with normal intelligence level, autistics with mental retardation, and ADHD cases with mental retardation should help to clarify whether MPH intolerance is associated with "autistic disorder" or "mental retardation".

Much research has been conducted to date investigating the comorbidity of PDD with ADHD and medication responses of those cases including MPH. But non-response or intolerance to MPH in PDD, and association of it with the MPH metabolic enzyme system has not been studied before.

The aim of this study was to compare the MPH response in cases with PDD and mild mental retardation (MR) with the cases with pure ADHD, and determining whether if there is an association with CES-1 gene polymorphisms. The assumptions of this study are:

1. There may be a polymorphism in the CES-1 enzyme in cases with PDD and accompanying ADHD symptoms.

2. Autistics with normal intelligence level and cases with Asperger disorder are thought to respond differently to MPH from the autistics with mental retardation. The response and tolerance to MPH is supposed to be ranked as Asperger disorder, autistics with normal intelligence level, and autistics with mental retardation.

3. No polymorphisms in the CES-1 enzyme are expected in cases with ADHD and ADHD with accompanying mild MR.

METHOD

Preliminary assessments were done by examining sociodemographic characteristics, motor-mental development stages, and school and pre-school histories of cases that were admitted to Ege University Faculty of Medicine Child and Adolescent Psychiatry Department outpatient clinic between December 2010 and January 2012, and whom were between 7-12 years old, whose mother/father was literate, and gave consent to participate to the study. Children out of this age range, and who had an additional primary psychiatric disease other than PDD, ADHD and MP, another medical disorder, physical head trauma history, epilepsy or chromosomal abnormality were excluded from the study.

Parents and teachers of all cases were requested to complete the sociodemographic data form and Turgay ADHD scale, which were used in the study.

An experienced psychologist using the Wechler Intelligence Scale for Children-Revised (WISC-R) determined mental capacities of all cases. A clinician applied Turkish Adaptation of Schedule for Affective Disorders and Schizophrenia for School-Age Children Present and Lifetime Version (K-SADS), and performed a semi-structured diagnostic interview of the study candidates to determine the comorbid diagnoses. Diagnosis of PDD was based on DSM-IV criteria and level of autism was detected by Childhood Autism Rating Scale (CARS). The high functioning autism group was determined according to the professional opinion of the clinician who made the clinical evaluations. Accordingly, 25 patients with ADHD+high functioning autism (HFA), 20 patients with ADHD+Asperger disorder (AD), 22 patients with ADHD+mild MR, and 22 patients with ADHD+Autism+MR were included as cases, and 34 patients with ADHD were included as the control group. A pediatrician evaluated all patients prior to the study by physical examination, hemogram, biochemistry, thyroid function tests and ECG.

Cases agreed to not use medication for two weeks and control visits were arranged. At the day of visit, 5 mg of methylphenidate was given to patients with body weight <30 kg, and 10 mg of methylphenidate was given to patients with body weight >30 kg, and saliva samples were obtained thereafter.

The parents of the cases were requested to complete the Turgay ADHD Scale after 1 hour of MPH administration, and a clinician blinded to study groups ranged the MPH response as improvement, no change, and worsening according to Clinical Global Impression-Improvement (CGI-I) scale.

A researcher blinded to diagnoses has isolated the DNA from the saliva samples obtained from the cases, and the DNA was processed with RFLP (Restriction Fragment Length Polymorphism) restriction enzyme according to PCR protocol, separated utilizing gel electrophoresis with 2% agarose or 3% Nussieve, visualized in gel imaging system, and width of the bands were determined by DNA Ladder and CES-1 genotypes (p.Arg199His, p.Ser75Asn, and p.Ile49Val) were characterized. Mutants and heterozygotes were classified as polymorphic.

Data Collection Tools

1. Sociodemographic Data Form

Researchers prepared a detailed sociodemographic data form that included participants' sociodemographic data, prenatal and postnatal history, motor and mental development history, and detailed characteristics of the current disease.

2. Wechsler Intelligence Scale for Children - Revised (WISC-R)

Wechsler Intelligence Scale for Children (WISC) was developed by Wechsler in 1949 to measure the intelligence levels of children at 5 to 15 years of age. It was rearranged in 1974 (WISC-R) and additionally modified the age range from 6 to 16 years of age. WISC-R was adapted into Turkish culture in 1995 by Savasir and Sahin (Savasir and Sahin 1995).

3. Turkish Adaptation of Schedule for Affective Disorders and Schizophrenia for School Aged Children, Present and Lifetime Version (K-SADS- PL)

K-SADS-PL is a semis-structured interview form that was developed by Kaufman et al. (1997) to determine the past and current psychopathologies of children and adolescents according to DSM-IV (APA 1994) (Kaufman et al. 1997). Validity and reliability of the scale for Turkish patients was proven (Gokler et al. 2004).

4. Childhood Autism Rating Scale (CARS)

This 15-item scale was developed to differentiate autistics from other children with growth retardation but without autism (Schopler et al. 1980). The scale is formed of 15 items, which each acting as a separate subclass. Total score of the scale is between 15 and 60. A total of 15-29: no autism; 30-36: mild-moderate autism; 37-60: severe autism. Turkish validity and reliability of the study has been conducted (Sucuoglu et al. 1996).

5. DSM-IV Based Screening and Evaluation Scale for Attention Deficit and Disruptive Behavior Disorder (DSM-IV ADDBD Evaluation Scale)

This scale was developed by Turgay (1994) based on DSM-IV diagnostic criteria (Turgay 1994). Turkish validity and reliability of the scale, which questions the criteria of Attention Deficit and Hyperactivity Disorder, Oppositional Defiant Disorder, and Conduct Disorder has been conducted (Ercan et al. 2001).

6. Clinical Global Impression Scale – Improvement (CGI-I)

This scale was developed by Guy to evaluate the course of psychiatric diseases at all ages for clinical research purposes. CGI is a three dimensional scale, which is completed by a physician during a semi-structured interview for evaluating the treatment response in individuals with psychiatric disorders (Guy 1976).

Statistical analyses of data

Data were evaluated by SPSS (The Statistical Package for Social Sciences) 18.0 software. Statistical evaluation was considered as significant at $p<0.05$. Comparisons of categorical data for number, percent, and homogeneity was done by chi-square analysis, and Fisher's Exact Test was used in cases where chi-square assumptions were not met. One-way analysis of variances (ANOVA), and Tukey Post-Hoc test were used for comparisons of means between three and more groups. Genotype frequencies were calculated and chi-square analysis was done to compare the estimated values according to Hardy-Weinberg equation principles.

RESULTS

A total of 123 cases as 25 ADHD+HFA, 20 ADHD+AD, 22 ADHD+Autism, 22 ADHD+Mild MR, and 34 ADHD were included in the study. Age and sex distribution of cases are summarized in Table 1, and no significant differences were found for age or sex ($p=0.24$ and $p=0.22$, respectively).

CES-1 polymorphisms are shown in Table 2. When the distribution of polymorphisms were evaluated, the only statistically significant difference was found in p.Arg199His polymorphism ($p=0.002$) with no statistically significant

Table 1. Age and sex distribution in diagnostic groups

	Boy				Girl			
	n	%	Age		n	%	Age	
			Mean	SD			Mean	SD
ADHD+HFA	21	21.2	8.5	1.4	4	16.7	8.8	2.1
ADHD+AD	16	16.2	9.3	2.1	4	16.7	10.8	1.0
ADHD+AuD	20	20.2	9.3	2.2	2	8.3	8.5	2.1
ADHD+MR	19	19.2	9.5	1.8	3	12.5	10.0	1.7
ADHD	23	23.2	9.4	1.6	11	45.8	9.6	1.5

HFA: High Functioning Autism, **AD**: Asperger Disorder, **AuD**: Autistic Disorder, **MR**: Mild Mental Retardation, **ADHD**: Attention Deficit Hyperactivity Disorder

Table 2. Distribution of CES-1 polymorphisms in diagnostic groups

	ADHD+HFA	ADHD+AD	ADHD+AuD	ADHD+MR	ADHD	p
	n (%)	n (%)	n (%)	n (%)	n (%)	
Arg199/His	7 (43.8)	5 (31.3)	4 (25.0)	- (-)	- (-)	0.002
Ser75/Asn	6 (15.0)	9 (22.5)	7 (17.5)	5 (12.5)	13 (32.5)	0.483
Ser75/Asn	23 (21.3)	15 (13.9)	19 (17.6)	20 (18.5)	31 (28.7)	0.259

CES1: Carboxylesterase-1 enzyme gene, HFA: High Functioning Autism, AD: Asperger Disorder, AuD: Autistic Disorder, MR: Mild Mental Retardation, ADHD: Attention Deficit Hyperactivity Disorder

Table 3. Comparisons of treatment responses of diagnostic groups according to Turgay ADHD scale

Diagnose	Comparison Group	Mean Difference	Standard Error	p
HFA	AD	-3.58	1.63	0.187
	AuD	1.50	1.59	0.878
	MR	-6.59	1.59	0.001
	ADHD	-9.33	1.43	<0.001
AD	AuD	5.08	1.68	0.024
	MR	-3.01	1.68	0.381
	ADHD	-5.75	1.53	0.002
AuD	MR	-8.09	1.64	<0.001
	ADHD	-10.83	1.48	<0.001
MR	ADHD	-2.74	1.48	0.353

HFA: High Functioning Autism**, AD:** Asperger Disorder**, AuD:** Autistic Disorder**, MR:** Mild Mental Retardation**, ADHD:** Attention Deficit Hyperactivity Disorder

Table 4. Comparisons of treatment responses in CGI-I scores of diagnostic groups

Diagnose	Improvement	No change	Worsening
	n (%)	n (%)	n (%)
HFA	8 (32.0%)	9 (36.0%)	8 (32.0%)
AD	14 (70.0%)	1 (5.0%)	5 (25.0%)
AuD	5 (22.7%)	7 (31.8%)	10 (45.5%)
MR	19 (86.4%)	2 (9.1%)	1 (4.5%)
ADHD	33 (97.1%)	1 (2.9%)	- (-)

HFA: High Functioning Autism**, AD:** Asperger Disorder**, AuD:** Autistic Disorder**, MR:** Mild Mental Retardation**, ADHD:** Attention Deficit Hyperactivity Disorder**, CGI-I:** Clinical Global Impression-Improvement

differences in p.Ser75Asn and p.Ile49Val. The p.Arg199His polymorphism was found to be present in the PDD comorbid group more frequently and was not present in mild MR or ADHD groups. Genotype distribution was in accordance with Hardy-Weinberg equation.

Treatment responses of cases with single dose MPH were evaluated by the Turgay ADHD scale that was completed by subject patients, and by the CGI-I scale that was completed by the clinician. Significant differences were found between mean pre- and post-treatment scores of Turgay ADHD scale when compared by ANOVA test between study groups, and Tukey's post-hoc analyses revealed that this difference was between the PDD comorbid group and ADHD and mild MR comorbid group ($p<0.001$). Treatment responses were lower in the PDD comorbid group. Results are presented in Table 3.

Analyzes revealed that there was a significant difference in CGI-I for MPH response between groups with and without PDD. According to the table, approximately 25% of PDD comorbid group had worsened symptoms after a single dose MPH, but the group without PDD showed improvement, with the difference between groups statistically significant ($p<0.001$).

Additionally, MPH response of ADHD+PDD cases with p.Arg199His polymorphism was statistically significantly lower than other cases ($p=0.010$). But, when the effects of the intelligence level was evaluated, MPH response in cases with mild MR was similar with ADHD cases, and there was not a statistically significant difference ($p>0.05$).

DISCUSSION

In this study of 123 patients with pervasive developmental disorder and accompanying attention deficit and hyperactivity disorder symptoms, our main hypothesis was evaluated, which was that methylphenidate response differences in cases with pervasive developmental disorder and accompanying attention deficit and hyperactivity disorder symptoms are caused by the polymorphisms in CES-1, which is the responsible enzyme of first pass in methylphenidate metabolism. The most important result of the study was that CES-1 polymorphism worsened the methylphenidate response in cases with pervasive development disorder and accompanying attention deficit hyperactivity disorder when compared to the cases that only had attention deficit hyperactivity disorder. To the best of our knowledge, this is the first study that evaluated the etiology of methylphenidate response variability in cases with pervasive developmental disorder and accompanying attention deficit hyperactivity disorder symptoms.

According to our results, p.Arg199His polymorphism is more frequent in cases with pervasive developmental disorder and accompanying attention deficit hyperactivity disorder symptoms when compared to cases with pure attention deficit hyperactivity disorder, and methylphenidate response of cases with pervasive developmental disorder and accompanying attention deficit hyperactivity disorder symptoms who had this polymorphism was worse than in other cases. A previous study reported that methylphenidate response in patients with pervasive developmental disorder and accompanying attention deficit hyperactivity disorder symptoms might be associated with intelligence level, and methylphenidate response worsened with decreased intelligence level (Di martino et al. 2004). Despite this, another study reported that there was not an association between intelligence level and methylphenidate response (Stiglerve et al. 2004). The results of our study showed that there is no association between intelligence level and methylphenidate response. Cases with mild mental retardation comorbid with attention deficit hyperactivity disorder had polymorphism and clinical methylphenidate response rates similar to pure attention deficit hyperactivity disorder cases, and they were statistically significant differences from cases with pervasive developmental disorder comorbidity.

There is no agreement in the literature about methylphenidate response in cases with attention deficit hyperactivity disorder and pervasive development disorder (Cortese et al. 2012, Pearson et al. 2013, Politte et al. 2014, Young and Findling 2015). However, the majority of studies and clinical observations show that methylphenidate response in cases with pervasive developmental disorder is worse than in the cases with attention deficit hyperactivity disorder without pervasive development disorder, and more adverse events are observed, notably in deteriorations in stereotypes (Reichow et al. 2013). Methylphenidate is primarily metabolized by the CES-1 enzyme, and variations in clinical response to methylphenidate occur due to CES-1 gene polymorphisms. In this context, this study has an importance for providing clues about if methylphenidate is effective in treating pervasive developmental disorders with attention deficit hyperactivity disorder symptoms, and also if methylphenidate response is affected in the presence of a polymorphism in the CES-1 gene. In our study, a CES-1 polymorphism was found to be statistically significant for p.Arg199His in cases with pervasive developmental disorders (High Functioning Autism, Autism and Asperger) when compared to cases with Mild Mental Retardation and attention deficit hyperactivity disorder, but no significant differences were found regarding p.Ser75Asn and p.Ile49Val polymorphisms. Also, determining the statistically significant difference regarding CES-1 polymorphism and treatment response is particularly important for methylphenidate response, which shows that treatment response deteriorates with altered genetic structure. The difference of methylphenidate response between diagnostic groups answers the questions in the literature.

Despite the limited number of samples, this study is one of the pioneer studies that:

1. Demonstrated that methylphenidate response is lower in cases with pervasive developmental disorders accompanied by attention deficit hyperactivity disorder symptoms, when compared with pure attention deficit hyperactivity disorder cases or attention deficit hyperactivity disorder cases with mild mental retardation;

2. Showed that role of CES-1 polymorphism should be evaluated more in detail for variability in methylphenidate response in cases with pervasive developmental disorder with accompanying attention deficit hyperactivity disorder.

Pharmacogenetic studies try to clarify how genetic variations affect pharmacokinetic and pharmacodynamic characteristics of drugs. This study is the first in this field, and its results are preliminary information that needs to be supported by studies with larger samples and long-term follow-up periods.

REFERENCES

American Psychiatric Association (2013) Diagnostic and statistical manual of mental disorders (5th ed.). Washington, DC.

Bruxel EM, Salatino-Oliveira A, Genro JP et al (2013) Association of a carboxylesterase 1 polymorphism with appetite reduction in children and adolescents with attention-deficit/hyperactivity disorder treated with methylphenidate. Pharmacogenomics J 13: 476-80.

Cortese S, Castelnau P, Morcillo C ve ark (2012) Psychostimulants for ADHD-like symptoms in individuals with autism spectrum disorders. Expert Rev Neurother 12:461-73.

Di Martino A, Melis G, Cianchetti C ve ark (2004) Methylphenidate for pervasive developmental disorders: safety and efficacy of acute single dose test and ongoing therapy: an open-pilot study. J Child Adolesc Psychopharmacol 14:207–18

Ercan ES, Amado S, Somer O, Çıkoğlu S(2001) Dikkat Eksikliği Hiperaktivite Bozukluğu ve Yıkıcı Davranış Bozuklukları için bir Test Bataryası Geliştirme Çalışması. Çocuk ve Gençlik Ruh Sağlığı Dergisi, 8:132-45

Frazier JA, Biederman J, Bellordre CA ve ark (2001) Should the Diagnosis of Attention Deficit/ Hyperactivity Disorder be Consider in Children with Pervasive Developmental Disorder? J Atten Disord 4:203-11.

Goldstein S, Schwebach AJ (2004) The comorbidity of Pervasive Developmental Disorder and Attention Deficit Hyperactivity Disorder: results of a retrospective chart review. J Autism Dev Disord 34:329-39.

Gökler B, Ünal F, Pehlivantürk B et al (2004) Okul çağı çocukları için duygulanım bozuklukları ve şizofreni görüşme çizelgesi - şimdi ve yaşam boyu şekli-Türkçe uyarlamasının geçerlik ve güvenirliği. Çocuk ve Gençlik Ruh Sağlığı Dergisi 11:109-16.

Grzadzinski R, Di MA, Brady E et al (2011) Examining autistic traits in children with ADHD: does the autism spectrum extend to ADHD? J Autism Dev Disord 41:1178–91.

Guy W (1976) Clinical global impressions. ECDEU assessment manual for psychopharmacology, revised National Institute of Mental Health, Rockville, MD, s. 218-21.

Johnson KA, Barry E, Lambert D et al (2013) Methylphenidate Side Effect Profile Is Infuenced by Genetic Variation in the Attention-Deficit/Hyperactivity Disorder-Associated CES1 Gene. J Child Adolesc Psychopharmacol 10:655-64.

Kaufman J, Birmaher B, Brent D et al (1997) Schedule for affective disorders and schizophrenia for schoolage children-present and lifetime version (K-SADS-PL) : initial reliability and validity data. J Am Acad Child Adolesc Psychiatry 36:980-8.

Lee DO, Ousley OY (2006) Attention-deficit hyperactivity disorder symptoms in a clinic sample of children and adolescents with pervasive developmental disorders. J Child Adolesc Psychopharmacol 16:737-46

Pearson DA, Santos CW, Aman MG et al (2013) Effects of extended release methylphenidate treatment on ratings of attention-deficit/hyperactivity disorder (ADHD) and associated behavior in children with autism spectrum disorders and ADHD symptoms. J Child Adolesc Psychopharmacol 23:337-51.

Polanczyk G, de Lima MS, Horta BL et al (2007) The worldwide prevalence of ADHD: a systematic review and metaregression analysis. Am J Psychiat 164:942-8.

Politte LC, Henry CA, McDougle CJ (2014) Psychopharmacological interventions in autism spectrum disorder. Harv Rev Psychiatry 22:76-92.

Reichow B, Volkmar FR, Bloch MH (2013) Systematic review and meta-analysis of pharmacological treatment of the symptoms of attention-deficit/hyperactivity disorder in children with pervasive developmental disorders. J Autism Dev Disord 43:2435-41.

Schopler E, Reichler RJ, de Vellis RF et al (1980) Toward objective classification of childhood autism: Childhood Autism Rating Scale (CARS). J Autism Dev Disord 10:91–103.

Stigler KA, Desmond LA, Posey DJ ve ark (2004) A naturalistic retrospective analysis of psychostimulants in pervasive developmental disorders. J Child Adolesc Psychopharmacol 14:49–56.

Sucuoğlu B, Öktem F, Akkök F et al (1996) Otistik çocukların değerlendirilmesinde kullanılan ölçeklere ilişkin bir çalışma. 3P Dergisi 4:116–21

Sun Z, Murry DJ, Sanghani SP et al (2004) Methylphenidate is stereoselectively hydrolyzed by human carboxylesterase CES1A1. J Pharmacol Exp Ther 310:469-76.

Turgay A (1994) Disruptive Behavior Disorders Child and Adolescent Screening and Rating Scales for Children, Adolescents, Parents and Teachers. West Bloomfield (Michigan), Integrative Therapy Institute Publication.

Young NJ, Findling RL (2015) An update on pharmacotherapy for autism spectrum disorder in children and adolescents. Curr Opin Psychiatry 28:91-101

Zhu HJ, Patrick KS, Yuan HJ et al (2008) Two CES1 gene mutations lead to dysfunctional carboxylesterase 1 activity in man: clinical significance and molecular basis. Am J Hum Genet 82:1241-8.