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ORIGINAL RESEARCH ARTICLE

The Effect of Genetic Polymorphisms of Cytochrome P450 CYP2C9, CYP2C19, and CYP2D6 on Drug-Resistant Epilepsy in Turkish Children

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Abstract

Background and Objective Despite the availability of several antiepileptic drugs, drug resistance remains one of the major challenges in epilepsy therapy. Genetic factors are known to play a significant role in the prognosis and treatment of epilepsy. The aim of this study was to determine the frequencies of alleles for *CYP2C9*, *CYP2C19*, and *CYP2D6* genes in Turkish children with epilepsy, and to investigate the relationship between the genetic polymorphism of these genes with multiple drug resistance in epilepsy patients.

Methods We genotyped 132 epileptic patients (60 drug resistant and 72 drug responsive) and 55 healthy controls for six single nucleotide polymorphisms (SNPs) in *CYP2C9*, *CYP2C19,* and *CYP2D6*. Genotype, allele, and haplotype frequencies were compared between groups.

Results The frequencies of *CYP2C9*3/*3* genotype and *CYP2C9*3* allele, and the haplotype CCGG (*CYP2C9*2* C>T, *CYP2C9*3* A>C, and *CYP2C19*2* G>A, *CYP2C19** G>A) were significantly higher in drug-resistant versus -responsive patients.

Conclusion Our results demonstrated the important role of the *CYP2C9*3* allelic variant in preventing epilepsy patients from developing drug resistance. These data suggest that *CYP2C9*, *CYP2C19*, and *CYP2D6* SNPs and haplotypes may affect the response to antiepileptic drugs.

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1 Introduction

Epilepsy is a neurological disorder that usually affects children under the age of 15 years or adults over the age of 65 years. It can be inherited, or it can result from a birth defect, labor trauma, head injury, brain tumor, or an infection in the brain. The cause of epilepsy can be determined in only 30 % of cases; in the other 70 %, the exact cause cannot be determined [1]. Despite significant therapeutic advances, almost every second an epileptic patient develops resistance to the current drug regimen [2]. The mechanisms underlying the resistance to antiepileptic drugs (AEDs) in some epilepsy patients are still elusive, but some potential mechanisms have been suggested [3]. Among them, genetic factors have a prominent place. The existence of a substantial heterogeneity of pharmacologic action of the same drug in specific individuals can be explained with high probability by differences in the patients' genotypes [4]. Genetic variations can potentially affect the individual responsiveness to the drug at several steps, including drug absorption, drug distribution, drug metabolism, drug elimination, and drug concentration at target sites [5].

Drug metabolism represents the prominent pathway, both in qualitative and quantitative elimination of drugs, including AEDs. Cytochrome P450 (CYP450) enzymes are major phase I drug-metabolizing enzymes, responsible for the metabolism of approximately 90 % of clinically prescribed drugs. Among the 18 identified human CYP450 families, CYP2 is the most diverse, associated primarily with phase I metabolism of a large number of foreign compounds [6].

The CYP2 family includes a variety of drug-metabolizing enzymes coded by polymorphic genes. The polymorphisms of three members from this gene family,

CYP2C9, *CYP2C19*, and *CYP2D6* have been studied extensively in a large number of populations and show significant heterogeneity in the frequency of different alleles/genotypes and the resulting metabolizer phenotypes [7].

CYP2D6, located at 22q13.1, mediates oxidative metabolism of a variety of clinical medicines, such as antidepressants, opioids, neuroleptics, and β-adrenergic-blocking agents [8]. *CYP2D6* is highly polymorphic, with over 100 known allelic variants. Some polymorphisms lead to a complete loss of *CYP2D6* function, while others reduce its activity. These polymorphisms seem to cause large inter-individual and ethnic differences in *CYP2D6* activity in vivo. It has been reported that 5–10 % of Caucasians have the *CYP2D6* poor metabolizer phenotype. The main variant alleles resulting in this phenotype in Caucasians are thought to be *CYP2D6*3* (frameshift) and *CYP2D6*4* (splicing defect) [9].

CYP2C9 and *CYP2C19* are part of the *CYP2C* gene cluster on chromosome 10q24 [10]. *CYP2C9* is responsible for the catalysis of the oxidation and metabolic clearance of up to 15–20 % of clinically important drugs, including phenytoin, warfarin, tolbutamide, losartan, and a large number of non-steroidal anti-inflammatory drugs [11, 12]. *CYP2C9* is highly polymorphic, with more than 35 variants, including functional variants of major pharmacogenetic importance. *CYP2C9*2* and *CYP2C9*3* are recognized as main *CYP2C9* variants in humans and are present in approximately 35 % of Caucasian individuals [13]. *CYP2C9*2* and *CYP2C9*3* have reduced catalytic activity compared with the wild type (*CYP2C9*1*) [14].

CYP2C19 acts on 5–10 % of drugs in current clinical uses, including antidepressants, barbiturates, proton pump inhibitors, antimalarial, and antitumor drugs [15]. At least 28 variant alleles for *CYP2C19* have been identified, the most extensively described of which are *CYP2C19*2* and *CYP2C19*3* (http://www.cypalleles.ki.se/). Both *CYP2C19*2*, which causes a 40-nucleotide deletion and a frameshift, and *CYP2C19*3*, which leads to a premature stop codon, result in the production of a truncated protein without enzymatic activity [16]. The frequency of the *CYP2C19*2* allele appears in 10–13 % of Caucasians, while the frequency of the *CYP2C19*3* allele varies from 5–9 % [17]. This is the first study in Turkish children investigating an association between polymorphisms of CYP450 enzymes and drug resistance to AEDs.

The variations in *CYP2D6*, *CYP2C9*, and *CYP2C19* genes could influence inter-individual variation in AED metabolism that could be responsible for the drug-responsive or drug-resistant phenotype. Whether the polymorphisms of these genes are associated with AED resistance is still not clear; for this reason, we investigated the effect of *CYP2D6*, *CYP2C9,* and *CYP2C19* genetic variants on multiple drug resistance in Turkish children with epilepsy.

2 Materials and Methods

2.1 Subjects

A total of 132 epileptic patients treated with AEDs were included in this study. The inclusion criteria comprised: (i) diagnosis of epilepsy (defined by the occurrence of two or more unprovoked seizures) confirmed by an electroencephalogram examination by an expert, (ii) known clinical response to the AED administered. According to therapeutic efficacy, these patients were divided into two groups: a drug-responsive subgroup and a drug-resistant subgroup. The main criterion for drug resistance was the occurrence of at least four seizures over a period of 1 year with three appropriate AEDs, such as carbamazepine + valproate + levetiracetam, or carbamazepine + valproate + phenytoin, according to the epileptic seizure type at maximum tolerated doses. Patients in the drug-responsive subgroup responded to at least one AED. Of the 132 patients, 60 were classified as drug resistant and 72 were drug responsive. Demographic details of the patients are given in Table 1.

Exclusion criteria included severe adverse drug reactions, poor compliance with AEDs, history of pseudo-seizures, alcohol or drug abuse, or any other malignant diseases such as brain tumor, secondary metastasis, hepatic failure, or renal failure.

The protocol for this study was approved by the Regional Ethics Committee and was carried out in accordance with The Code of Ethics of the World Medical Association (Declaration of Helsinki) for experiments involving humans. An informed consent was signed by each participant or responsible adult.

The control group consisted of 55 individuals with no previous history of epileptic seizures or other neurological diseases in a similar age group as the patients.

2.2 DNA Extraction and Genotyping

Blood was collected in EDTA-containing tubes, and DNA was extracted from the leukocytes using a high-pure template preparation kit (Roche diagnostics, GmbH, Mannheim, Germany). Genotyping of *CYP2C9* (*2 and *3 alleles), *CYP2C19* (*2 and *3 alleles), and *CYP2D6* (*3 and *4 alleles) was performed with melting curve analysis (LightCycler system) of polymerase chain reaction (PCR) amplified products using LightMix kits (TibMolbiol, Berlin, Germany) on LightCycler 1.5 capillary PCR machine (Roche diagnostics, GmbH, Mannheim, Germany). For each single nucleotide polymorphism (SNP), the primers flanking the SNP and the oligonucleotide probes were designed and validated by TIB-MOLBIOL, Berlin, Germany (LightMix to detect human *CYP2C9*2* and

Table 1 Demographic and clinical characteristics of epilepsy patients and healthy controls

Parameters	Drug resistant (n = 60)	Drug responsive (n = 72)	Controls (n = 55)
Gender			
Male	35 (58)	44 (61)	31 (56)
Female	25 (42)	28 (39)	24 (44)
Age, years (mean ± SD)	8.8 ± 4.0	9.6 ± 3.9	8.6 ± 3.9
Age, years (range)	1–15	2–14	1–15
Seizure type			
Partial	21 (33)	21 (29)	–
Generalized	39 (67)	51 (71)	–
Etiology			
Idiopathic	27 (45)	35 (49)	–
Symptomatic	25 (42)	23 (32)	–
Cryptogenic	8 (13)	14 (19)	–

Data are presented as n (%) unless otherwise indicated

*CYP2C9*3* [catalog no: 3 266 982]; LightMix to detect human *CYP2C19*2* and *CYP2C19*3* [catalog no: 3 267 008]; LightMix to detect human *CYP2D6*3* and *CYP2D6*4* [catalog no: 40 0180 16]). PCR/restriction fragment-length polymorphism (RFLP) was used to confirm the mutations [18–20].

2.3 Statistical Analysis

Mean and standard deviations (SD) are presented in the case of continuous variables. Chi-squared analysis (χ^2 tests) and Fisher's exact test were used to compare the gender distribution and to test the association between the genotypes and alleles in relation to the cases and controls, as well as to test for deviation of genotype distribution from Hardy–Weinberg equilibrium (HWE). The odds ratio (OR) and their 95 % confidence intervals (CI) were calculated to estimate the strength of the association between polymorphism genotype alleles of patients and controls. Differences between the means of the two continuous variables were evaluated with the Student's t test. Analysis was performed with SPSS 11.5 statistical software. Linkage disequilibrium and haplotypes were analyzed using SHEsis software [21]. A value of $P < 0.05$ was considered statistically significant.

3 Results

Demographic details of the patients and controls are shown in Table 1. There were no statistically significant differences in age ($P = 0.31$) and gender ($P = 0.34$) between patients and controls. The mean age of the patients was 9.2 ± 3.9 years, with no statistical difference between the drug-resistant (8.8 ± 4.0 years) and drug-responsive (9.6 ± 3.9) patients ($P = 0.29$). There were also no statistically significant differences with respect to the genders between the drug-resistant and drug-responsive patients ($P = 0.59$). There were no statistically significant differences in terms of age ($P = 0.70$ for drug-resistant; $P = 0.16$ for drug-responsive) and gender ($P = 0.54$ for drug resistant; $P = 0.33$ for drug-responsive) between drug-resistant or drug-responsive patients and the control group. The seizure types were generalized and partial in 68.2 and 31.8 % of the patients, respectively; and did not differ between the drug-resistant and drug-responsive patients (OR = 1.30, 95 % CI 0.59–2.90, $P = 0.60$). There were also no statistically significant differences regarding the etiology between the drug-resistant and drug-responsive patients ($\chi^2 = 1.67$, $df = 2$, $P = 0.43$).

The observed genotype frequency distributions did not show a significant deviation from the HWE. In our study population, none of the individuals were homozygous for any of the *CYP2C9*2* or *CYP2C19*3* or *CYP2D6*3* polymorphisms. The genotype and allele frequencies of *CYP2C9*2*, *CYP2C9*3*, *CYP2C19*2*, *CYP2C19*3*, and *CYPD6*3* did not differ significantly in patients compared with controls (Table 2). Only the *CYPD6*4* polymorphism showed a significant difference between patients and controls for **1/*4* genotype (OR = 2.39, 95 % CI 1.10–5.19, $P = 0.03$).

The polymorphisms of *CYP2C9* (except *CYP2C9*3*), *CYP2C19*, and *CYP2D6* did not differ significantly in the drug-resistant versus the drug-responsive patients (Table 3). *CYP2C9**3/*3 genotype and *CYP2C9**3 allele were both associated with the drug-resistant phenotype in epilepsy patients ($P = 0.04$ and $P = 0.02$, respectively).

Six haplotypes for *CYP2C9*2* and **3*, and *CYP2C19*2* and **3* gene polymorphism and two haplotypes for *CYP2D6*3* and **4* gene polymorphism were observed in drug-responsive and drug-resistant patients. These haplotype distributions were compared for any difference between both groups (Table 4). For *CYP2C9* and *CYP2C19* haplotypes, the frequency of CCGG (*CYP2C9*2* C>T, **3*

Table 2 *CYP2C9*, *CYP2C19*, and *CYP2D6* genotype and allele frequencies in patients and controls

Genotype	Patients	Controls	*P* value	OR (95 % CI)
*CYP2C9*2*				
*1/*1	103 (78)	43 (78)	Reference	
*1/*2	29 (22)	12 (22)	0.98	1.01 (0.47–2.16)
*2/*2	–	–		
*1 allele frequency	0.89	0.89		
*2 allele frequency	0.11	0.11	1.00	1.00 (0.06–16.24)
*CYP2C9*3*				
*1/*1	101 (77)	48 (87)	Reference	
*1/*3	23 (17)	6 (11)	0.31	0.55 (0.18–1.54)
*3/*3	8 (6)	1 (2)	0.17	
*1 allele frequency	0.88	0.94		
*3 allele frequency	0.12	0.06	0.14	2.14 (0.77–5.94)
*CYP2C19*2*				
*1/*1	98 (75)	38 (69)	Reference	
*1/*2	31 (23)	14 (25)	0.68	0.86 (0.41–1.79)
*2/*2	3 (2)	3 (6)	0.36	0.39 (0.08–2.01)
*1 allele frequency	0.86	0.82		
*2 allele frequency	0.14	0.18	0.44	0.74 (0.35–1.59)
*CYP2C19*3*				
*1/*1	128 (97)	55 (100)	Reference	
*1/*3	4 (3)	–	0.32	
*3/*3	–	–		
*1 allele frequency	0.98	1.00		
*3 allele frequency	0.02	0.00	0.50	
*CYP2D6*3*				
*1/*1	131 (99)	53 (96)	Reference	
*1/*3	1 (1)	2 (4)	0.22	0.21 (0.02–2.36)
*3/*3	–	–		
*1 allele frequency	0.99	0.98		
*3 allele frequency	0.01	0.02	1.00	0.50 (0.04–5.55)
*CYP2D6*4*				
*1/*1	83 (63)	44 (80)	Reference	
*1/*4	45 (34)	10 (18)	0.03	2.39 (1.10–5.19)
*4/*4	4 (3)	1 (2)	0.66	2.12 (0.23–19.56)
*1 allele frequency	0.80	0.89		
*4 allele frequency	0.20	0.11	0.08	2.02 (0.91–4.48)

Data are presented as n (%) unless otherwise indicated

CI confidence interval, *CYP* cytochrome P450, *OR* odds ratio

A>C and *CYP2C19*2* G>A, *3 G>A) haplotype in resistant patients was significantly higher than those in responsive patients (14.3.0 vs. 6.8 %, $P < 0.04$); however, no significant differences for *CYP2D6* haplotypes were observed among responsive and resistant patients.

4 Discussion

Despite extensive research efforts on AED resistance, we still know little about its mechanism. Studies are still needed to identify genetic and biomarkers to identify epileptic patients at risk of developing drug resistance. Genetic variability has been found to contribute towards both the susceptibility of occurrence of the diseases as well as to the variability in therapeutic response [22]. According to Grover et al. [23], genetics can account for 20–95 % of clinical variability in drug disposition and effects. SNPs have been reported to have a major impact on the pharmacokinetic as well as the pharmacodynamic profile of AEDs [24].

The polymorphisms in the CYP450 family are known to affect biotransformation of many drugs, resulting in inter-individual variability in drug response. The polymorphic

Table 3 *CYP2C9*, *CYP2C19*, and *CYP2D6* genotype and allele frequencies in drug-responsive versus drug-resistant patients with epilepsy

Genotype	Drug responsive	Drug resistant	*P* value	OR (95 % CI)
*CYP2C9*2*				
*1/*1	45 (75)	58 (81)	Reference	
*1/*2	15 (25)	14 (19)	0.58	0.72 (0.29–1.79)
*2/*2	–	–		
*1 allele frequency	0.87	0.90	Reference	
*2 allele frequency	0.13	0.10	0.66	0.74 (0.28–1.93)
*CYP2C9*3*				
*1/*1	51 (86)	50 (69)	Reference	
*1/*3	8 (12)	15 (21)	0.26	1.91 (0.68–5.46)
*3/*3	1 (2)	7 (10)	0.04	
*1 allele frequency	0.92	0.80		
*3 allele frequency	0.08	0.20	0.02	2.87 (1.12–7.56)
*CYP2C19*2*				
*1/*1	43 (72)	55 (77)	Reference	
*1/*2	15 (25)	16 (22)	0.82	0.83 (0.34–2.02)
*2/*2	2 (3)	1 (1)	0.42	
*1 allele frequency	0.84	0.88	Reference	
*2 allele frequency	0.16	0.12	0.54	0.72 (0.30–1.72)
*CYP2C19*3*				
*1/*1	58 (97)	70 (97)	Reference	
*1/*3	2 (3)	2 (3)	0.62	
*3/*3	–	–		
*1 allele frequency	0.98	0.99	Reference	
*3 allele frequency	0.02	0.01	0.50	
*CYP2D6*3*				
*1/*1	59 (98)	72 (100)	Reference	
*1/*3	1 (2)	0 (0)	0.45	
*3/*3	–	–		
*1 allele frequency	0.99	1.00	Reference	
*3 allele frequency	0.01	0.00	0.50	
*CYP2D6*4*				
*1/*1	36 (60)	47 (65)	Reference	
*1/*4	22 (37)	23 (32)	0.68	0.80 (0.36–1.77)
*4/*4	2 (3)	2 (3)	0.59	
*1 allele frequency	0.78	0.80	Reference	
*4 allele frequency	0.22	0.20	0.86	0.88 (0.42–1.84)

Data are presented as n (%) unless otherwise indicated

CI confidence interval, *CYP* cytochrome P450, *OR* odds ratio

alleles lead to altered activity of these isoenzymes causing absent, decreased, or increased metabolism. *CYP2D6*, *CYP2C9*, and *CYP2C19* polymorphisms account for the most frequent variations in phase I metabolism of drugs. In general, AED monotherapy has been found to be effective in ~60–70 % of newly diagnosed epileptic patients, while up to ~50 % of patients who fail initial AED treatment can be managed by a switch to an alternative AED [25]. Epileptic patients who do not respond to monotherapy are typically administered a combination of AEDs to optimize seizure control. No single combination is perfect for everyone. Sometimes a series of combinations must be tried before finding what is best for the individual patient. Population-based studies of drug utilization demonstrate that 19–24 % of patients with epilepsy use polytherapy with AEDs [26]. Factors influencing the choice of the additional AED may include potential interactions and the mechanisms of action of the two medications.

The mainstay of epilepsy treatment uses more than 20 AEDs with several different mechanisms of action [27]. The commonly prescribed AEDs are carbamazepine, phenytoin, valproic acid, phenobarbital, gabapentin, topiramate, and tiagabine. The drug-resistant patients in our study were using a combination of AEDs, such as

Table 4 Haplotype frequencies of *CYP2C9*2* and **3*, *CYP2C19*2* and **3*, and *CYP2D6*3* and **4* gene polymorphism in drug-resistant and drug-responsive patients

Haplotypes	Drug responsive (%)	Drug resistant (%)	*P* value
CYP2C9 and *CYP2C19*			
CAGG	0.647	0.640	0.90
CAGA	0.132	0.094	0.33
CCGG	0.068	0.143	0.04
TAGG	0.103	0.088	0.68
TAGA	0.018	0.010	0.60
CCGA	0.010	0.017	0.59
CYP2D6			
GA	0.803	0.788	0.76
AA	0.189	0.212	0.64

Data are presented as percentages unless otherwise indicated

CYP cytochrome P450

carbamazepine + valproate + levetiracetam, or carbamazepine + valproate + phenytoin. Phenytoin (diphenylhydantoin) and valproic acid (valproate) are mainly metabolized by CYP2C9 and CYP2C19. The differences in phenytoin dose response can partially be explained by genetic polymorphisms in CYP2C9 and 2C19 enzymes, but a great deal of individual variability remains unexplained. Apart from these enzymes, involvement of many other CYP enzymes in phenytoin metabolism, including CYP2D6, has already been reported [28]. On the other hand, levetiracetam does not act as an inducer or inhibitor of the hepatic CYP450 system or other enzymes. Carbamazepine is the major enzyme-inducing AED that stimulates the rate of metabolism of most co-administered AEDs, including valproic acid and, to some extent, levetiracetam. Consequently, the interaction between drugs might exist but the direction of interactions is the same.

This study investigated six SNPs in *CYP2C9*, *CYP2C19*, and *CYP2D6* genes in 132 responsive or resistant epilepsy patients and found that only allele '*3' of the *CYP2C9* gene is associated with AED resistance. This association is attributable to the very low enzymatic activity of the *CYP2C9*3* allele (93 % decreased enzyme activity) compared with the *CYP2C9*2* allele (29 % decreased enzyme activity) [16]. Two *CYP2C9* alleles that produce a phenotype of poor metabolism occur in 11 and 8 % of Whites but only 3 and 0.8 % of Blacks [29]. The substitution encoded by *CYP2C9*2* is not in any of the substrate recognition sites, and several studies have shown that it has less impact on enzyme activity than does *CYP2C9*3* [30]. The metabolism of many CYP2C9 substrates was reported to be significantly decreased in individuals with the *CYP2C9* allele 3 [31, 32]. Genetic variants that lead to poor metabolizer phenotypes of *CYP2C19* are relatively common in Asian groups [29].

To our knowledge, no studies have examined the relationship between CYP450 polymorphisms and drug-resistant epilepsy in Turkish children. Our findings support previous observations that identified CYP2C9 as a major drug metabolizer for commonly prescribed AEDs [33]. Earlier studies have also observed a strong association between *CYP2C9* allelic variants (*CYP2C9*2* and *CYP2C9*3*) and phenytoin maintenance dose requirements [34, 35]. Van der Weide et al. [34] found that *CYP2C9* allelic variants affect phenytoin maintenance dose requirements. For patients carrying at least one mutant *CYP2C9* allele, the mean phenytoin dose required to achieve a therapeutic serum concentration was about 37 % lower than the mean dose required by wild-type individuals. Another similar study revealed that severe phenytoin intoxication occurred in a patient subsequently genotyped as homozygous for *CYP2C9*3* and heterozygous for the *CYP2C19*2* allele [35]. CYP2C9 is also the predominant catalyst of valproic acid metabolism [36].

Our haplotype study revealed that the CCGG (*CYP2C9*2* C>T, *3 A>C and *CYP2C19*2* G>A, *3 G>A) haplotype was associated with AED resistance. It has been shown that patients with variant alleles of the *CYP2C9* and *CYP2C19* genotypes are at increased risk of drug resistance, although the overall contribution of CYP2C19 toward metabolism of some AEDs was less important than CYP2C9 [37, 38].

Drug resistance is a complex phenotype resulting from contribution of numerous genes. In addition to drug-metabolizing enzymes, expression of multidrug transporters also influences phenytoin and carbamazepine disposition and may account for inter-individual pharmacokinetic variability [39]. Kerb et al. [40] performed a combined analysis of variable alleles of *CYP2C9*, *CYP2C19,* and *ABCB1* gene and revealed that the number of mutant *CYP2C9* alleles is a major determinant.

The *CYP2C9*2* and **3* alleles were reported to be the most common variants in various Caucasian populations with allelic frequencies of 0.08–0.14 and 0.04–0.16, respectively [24]. In the control group of our study, the allele frequencies for *CYP2C9*2* and *CYP2C9*3* were found to be 11 and 6 %, respectively. Our findings show that the frequencies of *CYP2C9*2* and **3* are within the range defined for other Caucasian populations. The observed *CYP2C9*3* allele frequency in our epileptic patients was higher than in controls, but this difference was not statistically significant. In a previous study, Babaoglu et al. [41] detected similar allele frequencies of *CYP2C9*2* and **3* variants in a Turkish population (10 and 8.8 %, respectively).

*CYP2C19*2* and *CYP2C19*3* variant alleles are the most characterized alleles of the *CYP2C19* gene [22]. *CYP2C19*2* is the most common allele among Caucasians: the frequency of 18 % found in our control group was close to values reported in studies of different Caucasian populations [42, 43]. The *CYP2C19*3* variant allele was not detected in our control group, which is consistent with findings of previous research suggesting the *CYP2C19*3* variant is infrequent in Caucasian populations (0–2 %) [44]. Aynacioglu et al. [45] detected similar allele frequencies of *CYP2C19*2* and **3* variants in a Turkish population (12 and 0.4 %, respectively).

The most common *CYP2D6* variants in Caucasians are variants **3* and **4,* resulting in a decreased enzyme activity and leading to poor metabolizer phenotypes [46]. In the control group in our study, we found a *CYP2D6*3* allele frequency of 2 % and a *CYP2D6*4* allele frequency of 11 %. These results were similar to the studies of Aynacioglu et al. [45], with 404 individuals (*CYP2D6*4* allele frequency was 0.11) and of Aydin-Sayitoglu et al. [47] with 140 individuals (*CYP2D6*3* and **4* alleles were 0.025 and 0.139, respectively).

5 Conclusion

This is the first study investigating an association between CYP450 polymorphisms and drug-resistant epilepsy in Turkish children. Our results confirm the importance of the *CYP2C9*3* variant in conferring a multiple drug-resistant phenotype against AEDs. Large-scale efforts are needed to unravel the genetic determinants of AED response.

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