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Pharmacogenetic Determinants of Valproic Acid Efficacy, Toxicity, and Serum Levels in Genetic Generalized Epilepsy

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ABSTRACT

Responses to antiseizure medications (ASM) vary widely from one individual to another, both in terms of benefit and adverse reactions. Genetic differences are believed to play a meaningful role in shaping these outcomes. Valproic acid (VPA), a commonly prescribed ASM, is influenced by numerous pharmacogenetic elements. Yet, unlike agents such as carbamazepine or phenytoin, the amount of evidence linking VPA to specific genetic variants remains limited. This study was therefore designed to investigate how selected pharmacogenetic markers relate to VPA effectiveness, tolerability, and serum levels in a uniform group of newly diagnosed patients with genetic generalized epilepsies (GGE). This prospective cohort project extracted demographic, clinical, and treatment information from the medical charts of individuals with GGE. Whole-exome sequencing was completed in partnership with Epi25. Variants connected to VPA response, biotransformation, or toxicity were gathered from PharmGKB. The subsequent analysis assessed whether these variants showed measurable associations with clinical outcomes during VPA therapy.

Among 166 enrolled participants, 60 (36.1%) did not respond adequately to treatment, whereas 106 (63.9%) showed successful outcomes. After adjusting for the VPA maintenance dose, carriers of the rs3892097 variant (CYP2D6) demonstrated a 2.5-fold higher probability of treatment failure compared with noncarriers ($p = 0.026$). The rs1057910 allele (CYP2C9*3) was linked to higher circulating VPA concentrations ($p = 0.034$). The rs1137101 variant (LEPR, a regulator of metabolic processes) was tied to an increased likelihood of weight gain (coefficient 3.430 [0.674; 6.186], $p = 0.015$) as well as more frequent hair loss (OR = 3.394 [1.157; 9.956], $p = 0.026$). In contrast, rs4480 in SOD2, which encodes a mitochondrial antioxidant enzyme, corresponded to a reduced rate of hair loss (OR = 0.276 [0.089; 0.858], $p = 0.026$). The results emphasize that inherited genetic variation contributes meaningfully to differences in VPA treatment outcomes and point toward the value of developing individualized therapeutic strategies aimed at improving efficacy and reducing adverse effects.

Keywords: Pharmacogenetics, VPA, Epilepsy, Efficacy, Toxicity, Concentrations

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Introduction

Epilepsy is a widespread neurological condition defined by repeated seizures occurring without provocation [1]. Although incidence peaks in infancy and older adulthood, the disorder can emerge at any point in life and presents with varied underlying causes and clinical patterns [2].

Approximately 70% of individuals beginning antiseizure therapy achieve seizure control, resulting in substantial improvements in daily function and long-term health [1, 2]. Despite this, responses to ASMs differ considerably across patients, including differences in effectiveness, drug-related adverse effects (ADEs), and measured drug levels—even when seizure type and treatment plan are similar [3]. Increasing evidence suggests that inherited genetic differences significantly shape these disparities [3]. This highlights the value of pharmacogenetics—the study of how genetic makeup influences drug response—in refining treatment strategies for epilepsy.

Valproic acid (VPA), a broad-spectrum ASM, is routinely used in managing genetic generalized epilepsies (GGE) [4–6]. Many genes have been proposed to affect how individuals respond to VPA based on its mechanisms and

pharmacokinetic behavior [7]. VPA increases GABAergic signaling partly by blocking ABAT and ALDH5A, enzymes responsible for GABA breakdown [8, 9]. It also acts on voltage-gated sodium, potassium, and calcium channels—including CACNA1C, CACNA1D, CACNA1N, CACNA1F, and the SCN family—to reduce excessive neuronal firing [9, 10]. VPA is extensively metabolized in the liver through UGT enzymes (UGT1A3, UGT1A4, UGT1A6, UGT1A8, UGT1A9, UGT1A10, UGT2B7, UGT2B15), generating valproate-glucuronide as its main urinary metabolite [11–13]. Additionally, ABCB1 and ABCC1 encode blood–brain-barrier efflux pumps capable of transporting VPA out of the brain, possibly lowering CNS concentrations and contributing to drug resistance [14, 15].

Although VPA is widely used, the amount of pharmacogenetic research devoted to it remains noticeably smaller than that available for ASMs like carbamazepine or phenytoin, and the strength of the existing evidence is comparatively weak [16, 17]. One example is a multi-center investigation of 142 Caucasian adult patients, which examined whether the T1405 variant in the CPS1 gene contributes to VPA-related hyperammonemia; the study found that individuals with the T1405 allele had a higher likelihood of developing this complication [18]. Another analysis involving 52 pediatric patients demonstrated that dose adjustments based on CYP2C9 genotype helped reduce hyperammonemia cases [19]. A separate study examining 170 Chinese children assessed several ABCB1 SNPs and showed that homozygous rs1128503 carriers were more prone to persistent seizures, whereas homozygous rs3789243 carriers had fewer gastrointestinal adverse effects [20]. Even so, the role of ABCB1 variants in predicting ASM resistance remains unsettled, and clinical testing is not advised at this time [17]. In addition, a cohort of 212 Han Chinese patients treated with VPA identified links between weight gain and genetic variation in LEPR and ANKK1 [21]. All these findings require replication, as the collective evidence remains uncertain [17].

To help clarify these unresolved questions, we conducted a prospective cohort study assessing whether selected pharmacogenetic markers influence VPA efficacy, adverse effects, and serum levels in a uniform group of newly diagnosed GGE patients.

Materials and Methods

Approval for the study was obtained from the AUBMC Institutional Review Board (IRB) (IM.AB1.01). Informed consent for genetic testing was collected from all adult participants or legal guardians, and assent forms were signed by children when required.

Prospective enrollment of patients with newly diagnosed epilepsy

The individuals included in this pharmacogenetic sub-study were drawn from a larger longitudinal project initiated in 2010, focusing on newly identified epilepsy in adults and children. By August 2024, the broader cohort had reached 3010 enrolled subjects [22]. The study is coordinated through AUBMC in partnership with the Lebanese Chapter of the ILAE. Neurologists across Lebanon refer patients with suspected first-time seizures to AUBMC, where each undergoes a standardized diagnostic protocol. This evaluation includes a 3-hour sleep-deprived video-EEG, interpreted by two independent electroencephalographers, along with an epilepsy-protocol brain MRI, assessed by a neuroradiologist who is blinded to clinical history. The results are communicated back to the referring clinician, who determines whether to initiate ASM therapy and chooses the appropriate medication. Once started, the ASM dose is titrated to achieve seizure remission or until adverse effects become limiting. Follow-up occurs every 3 months during the first year and every 6 months thereafter, with additional visits depending on clinical need. Patients presenting with absence, myoclonic, or primary generalized tonic-clonic seizures, combined with generalized spike-wave discharges on EEG, were classified as having GGE [23].

Whole exome sequencing in collaboration with the Epi25 collaborative

In 2017, the AUBMC cohort joined the international Epi25 Collaboration, which aims to carry out Whole Exome Sequencing (WES) to better understand how genetic variation contributes to both common and rare forms of epilepsy and to determine the relative roles of de novo and inherited variants [6]. Sequencing is performed at the Broad Institute using the Illumina Infinium Global Screening Array, GSA-MD v1.0 (Illumina, San Diego, United States) [24–26].

Blood samples were collected in EDTA tubes, separated into aliquots, and stored at −80°C prior to DNA extraction with the Flexigene DNA isolation kit (Qiagen, CA, United States). Purified DNA was subsequently stored at −20°C.

By 2020, 1238 peripheral blood DNA samples had been transferred from AUBMC to the Broad Institute for whole-exome sequencing (WES). After completion of sequencing, the institute returned both BAM and VCF files, which were archived and processed by the institutional Bioinformatics core. The unaligned BAM (uBAM) files were mapped to the hg38 human reference genome using the Burrows–Wheeler Aligner (BWA) [27]. Once aligned, reads were reordered by genomic position and duplicate fragments were flagged through GATK's MarkDuplicates [28]. Base quality scores were then corrected using ApplyBQSR. Variant detection for single-nucleotide changes (SNVs) and INDELs was carried out using GATK HaplotypeCaller. Multi-sample GVCFs were integrated with GenomicsDBImport, and joint genotyping was subsequently executed with GenotypeGVCFs. Variant filtering and recalibration were performed using VariantFiltration and the applyVQSR procedure. The Broad Institute's pipeline generated the final VCF (format v4.2) using GATK v4.1.1.0. Variant annotation was performed with the Ensembl Variant Effect Predictor (VEP) v104 [29]. Only variants marked with a "PASS" label were retained for later analyses. Genotype and variant-level information were extracted from VCFs via BCFtools [30]. Departure from Hardy–Weinberg Equilibrium (HWE) was assessed via Chi-square testing.

Nested sample for the current study

The study included individuals whose samples had undergone sequencing by 2020, carried a diagnosis of GGE, had been initiated on valproic acid (VPA) monotherapy, and had adequate clinical documentation. Chart review encompassed data from the initial consultation through the final follow-up, including sex, body weight (kg), height (cm), age at diagnosis, electroclinical syndrome (ECS), VPA maintenance dose (mg/kg), measured VPA serum levels (mg/L) with corresponding dosages, adherence information, duration of observation, and reasons for discontinuation when applicable. Trough VPA concentrations were obtained from samples collected either 8–12 hours after the previous dose or immediately prior to the next dose. Serum VPA measurements were performed using a chemiluminescent microparticle immunoassay (CMIA) on the Abbott Architect system (Abbott Park, Illinois, United States), with a detection limit of 2 mg/L. At each encounter or phone assessment, adherence was evaluated by questioning the patient or caregiver regarding proper administration of the antiseizure medication (ASM). Treatment response was determined by measuring the period of seizure remission. Patients lacking follow-up consistency or demonstrating non-adherence were excluded.

Selection of genetic variants

Candidate variants associated with VPA-related genotype–phenotype relationships were identified through a search performed on 14 December 2023 using the PharmGKB curated "clinical annotations" and "variant annotations" resources [31]. The screening retained only statistically significant SNPs ($p < 0.05$) and discarded entries classified as evidence level 4. This process yielded 31 variants across 25 genes, grouped into four phenotypic domains: dosage, metabolism/pharmacokinetics, efficacy, and toxicity. Additional details—including functional category, nucleotide substitution type, gnomAD allele frequencies [32], and population-specific frequency data with emphasis on European cohorts—were retrieved from dbSNP [33].

Statistical analysis

Analyses were carried out using SPSS (Version 25.0, Armonk, NY: IBM Corp.) to investigate possible relationships between genotypes and VPA effectiveness, adverse effects, and adjusted drug concentration (ADC), which accounts for both dose and body weight [34]. Continuous variables are reported as mean ± standard deviation (SD), whereas categorical variables are presented as counts and percentages.

A patient was considered to have a successful therapeutic outcome if at least one year of seizure freedom was achieved on stable VPA monotherapy. Individuals who did not reach one year of remission, required an additional ASM, switched therapy after starting VPA, or discontinued treatment because of adverse events were categorized as treatment failures. Toxicities attributed to VPA included weight gain (defined as the change between pre-treatment and final on-treatment weight), alopecia, gastrointestinal symptoms (nausea/vomiting), and tremor.

ADC values were obtained by normalizing each serum VPA concentration (μg/mL) to the product of body weight (kg) and daily VPA dose (mg/day), yielding units of [(μg/mL/day)/(mg/kg)]. Body weight used for normalization

corresponded to the measurement taken at the time of VPA level collection or within a 6-month window; if unavailable, the average of the closest preceding and subsequent weights was used. For patients with several concentration measurements across different time points, the mean ADC was calculated.

Initially, demographic factors and genetic markers were explored one by one to determine how they related to treatment outcome, adverse effects, and ADC values. Categorical parameters were evaluated using the chi-square test, while continuous measures—including age at first assessment, duration of monitoring, maintenance VPA dose, serum levels, and weight variation—were processed with the Mann–Whitney U test, since the Shapiro–Wilk test indicated non-normal distributions. Any remaining continuous variables were assessed with the Student t-test.

A subsequent multivariable stage was introduced to correct for factors that may distort the associations. Binary logistic regression, expressed with ORs and 95% CIs, was used for variables representing treatment success or toxicity (both coded as yes/no). Linear regression, reporting β values and 95% CIs, was applied to the ADC and weight-gain analyses. The following elements were examined as potential confounders: sex, age at diagnosis, total follow-up length, and VPA dose normalized to body weight (mg/kg/day). Only those confounders reaching significance ($p < 0.05$) in the univariate step were retained in the adjusted models.

Differences in VPA maintenance dosing were taken into account when modelling treatment outcome and hair-loss risk. Variations in sex distribution were controlled for in the nausea/vomiting and hair-loss analyses. The tremor and adjusted-concentration models were corrected for age at the first diagnosis. Because growth can influence weight trajectories in children, both age at the initial visit and follow-up duration were included in the weight-gain model, regardless of univariate significance. Age at diagnosis corresponded to the patient's age at the first clinical encounter. For VPA dose selection: the stable dose was used for responders, the highest given dose for patients with inadequate control, and the withdrawal dose for those who stopped due to intolerance. All genetic associations were evaluated under a recessive model, contrasting wild-type homozygotes against combined carriers and mutant homozygotes.

Results and Discussion

Final sample

From the 1238 individuals whose DNA underwent sequencing, clinical files were available for 218 who matched the eligibility requirements. Fifty-two were removed for inadequate documentation, loss to follow-up, or poor medication adherence. This yielded 166 participants for the treatment-efficacy analysis. Ages spanned 6 months to 40 years (mean = 12.85 ± 7.28), and baseline body weight ranged from 7.5 to 116 kg (mean = 47.32 ± 22.26, N = 163).

For toxicity outcomes, 4 individuals lacked sufficient descriptions of hair loss and nausea/vomiting, leaving 162 participants (N = 162) for those endpoints. The tremor analysis excluded 3 individuals, leaving 163 subjects (N = 163).

Weight-change calculations were possible for 163 of the 166 patients (N = 163). ADC computations required serum VPA measurements, which were available for 150 participants (N = 150).

Genotyping results

Among the 31 initially selected variants, three were absent entirely (all participants were wild-type), one had no wild-type representation, six showed insufficient sequencing depth, and four did not meet Hardy–Weinberg expectations. Thus, 17 variants were retained for analyses, with allele frequencies similar to European reference populations.

Five variants (rs2279020, rs9332120, rs3892097, rs7438284, rs2269577) across GABRA1, CYP2C9, CYP2D6, UGT2B7, and XBP1 were used in the treatment-outcome models. Four variants (rs1137101, rs1800497, rs4880, rs3087374) in LEPR, ANKK1, SOD2, and POLG were studied for toxicity outcomes. Eight variants (rs6759892, rs1105879, rs1105880, rs7592281, rs2070959, rs1057910, rs1799853, rs7668258) from UGT1A, CYP2C9, and UGT2B7 were included in the ADC-related analysis, following the PharmGKB phenotype regroupings.

Association with VPA efficacy

In the final cohort of 166, 60 (36.1%) were categorized as treatment failures, whereas 106 (63.9%) met the predefined criteria for success **(Table 1)**. The non-responder group required a significantly larger mean maintenance dose of 19.01 ± 9.03 mg/kg/day, compared to 15.88 ± 6.7 mg/kg/day used in responders ($p = 0.036$).

This pattern is expected, as dose escalation is more common in individuals who do not achieve seizure control promptly.

No other demographic or genetic variable demonstrated a statistically meaningful link to treatment outcome.

Table 1. Demographic and genetic correlates of 12-month treatment status.

Characteristic	Total Cohort (N = 166)	Treatment Failure at 12 Months (N = 60)	Treatment Success at 12 Months (N = 106)	p-value
Age at first visit (years), mean ± SD	12.85 ± 7.28	13.15 ± 6.65	12.68 ± 7.64	0.443
Valproic acid maintenance dose (mg/kg/day), mean ± SD	17.01 ± 7.75	19.01 ± 9.03	15.88 ± 6.70	0.036
Duration of follow-up (years), mean ± SD	6.58 ± 2.61	6.90 ± 2.66	6.40 ± 2.58	0.245
Female sex, n (%)	76 (45.8%)	31 (51.7%)	45 (42.5%)	0.252
Male sex, n (%)	90 (54.2%)	29 (48.3%)	61 (57.5%)	—
rs2279020 (ABCB1)				
Wild-type, n (%)	36 (21.7%)	16 (26.7%)	20 (18.9%)	0.241
Variant carrier (heterozygous + homozygous), n (%)	130 (78.3%)	44 (73.3%)	86 (81.1%)	—
rs9332120 (ABCB1)				
Wild-type, n (%)	107 (64.5%)	34 (56.7%)	73 (68.9%)	0.115
Variant carrier (heterozygous + homozygous), n (%)	59 (35.5%)	26 (43.3%)	33 (31.1%)	—
rs3892097 (CYP2D6)				
Wild-type, n (%)	135 (81.3%)	44 (73.3%)	91 (85.9%)	0.047
Variant carrier (heterozygous + homozygous), n (%)	31 (18.7%)	16 (26.7%)	15 (14.2%)	—
rs7438284 (ABCB1)				
Wild-type, n (%)	55 (33.1%)	18 (30.0%)	37 (34.9%)	0.519
Variant carrier (heterozygous + homozygous), n (%)	111 (66.9%)	42 (70.0%)	69 (65.1%)	—
rs2269577 (XPC)				
Wild-type, n (%)	69 (41.6%)	22 (36.7%)	47 (44.3%)	0.335
Variant carrier (heterozygous + homozygous), n (%)	97 (58.4%)	38 (63.3%)	59 (55.7%)	—

Continuous variables were assessed with the Mann–Whitney U test; categorical variables with the chi-square test. Means are expressed as ± SD; categorical values as N (%). Statistically significant p-values (< 0.05) are shown in bold.

Carriers of rs3892097 (CYP2D6) made up 26.67% of the group with treatment failure, whereas only 14.15% of the successful-treatment group carried this variant (p = 0.047). When the analysis was adjusted for the maintenance dose of VPA, the resulting odds ratio was 0.389 [0.169; 0.894] (p = 0.026). This corresponds to an estimated 2.5-fold higher likelihood of possessing the polymorphism among individuals who did not respond to therapy. Thus, rs3892097 (CYP2D6) appears to impede the therapeutic efficacy of VPA monotherapy. No other genetic markers displayed statistically meaningful relationships with treatment outcomes **(Figure 1)**.

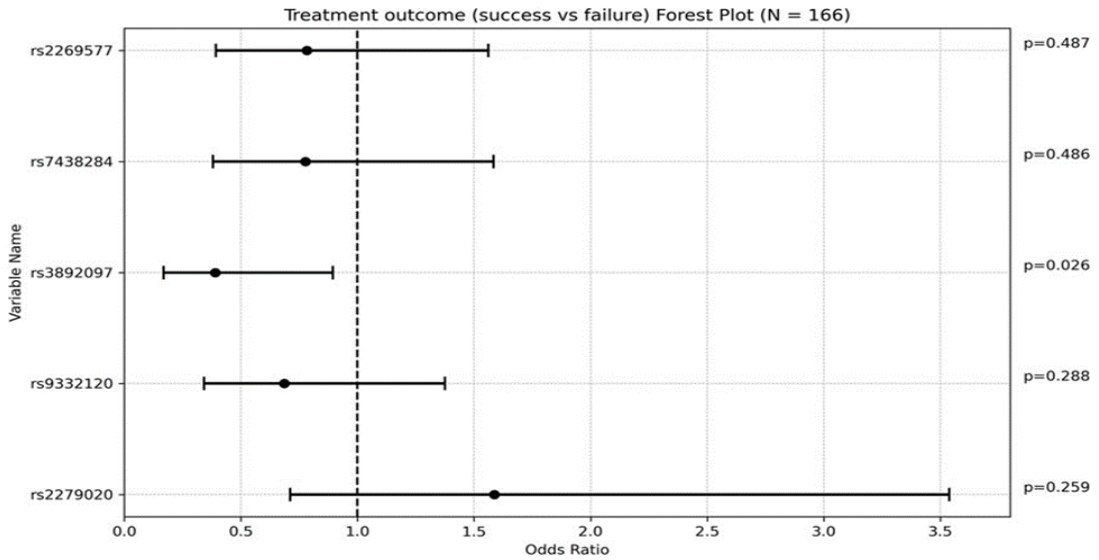

Figure 1. Multivariate odds ratios (95% CI) comparing successful versus failed treatment for each SNP.

Association with VPA toxicity

Within the study cohort, spontaneous reports of adverse events during VPA monotherapy included nausea/vomiting (4.93%), tremor (12.26%), and hair loss (11.72%).

All individuals who reported nausea or vomiting were female, while females constituted 42.86% of those without these symptoms (p = 0.002). After controlling for potential confounders, no SNPs demonstrated a significant relationship with nausea or vomiting.

Similarly, none of the genetic variants examined showed significant associations with tremor.

A single SNP, rs1137101 (LEPR), was linked to greater weight gain when analyses were adjusted for maintenance VPA dose, diagnostic age, and follow-up duration. Its multivariate regression coefficient was 3.430 [0.674; 6.186], p = 0.015 **(Figure 2)**.

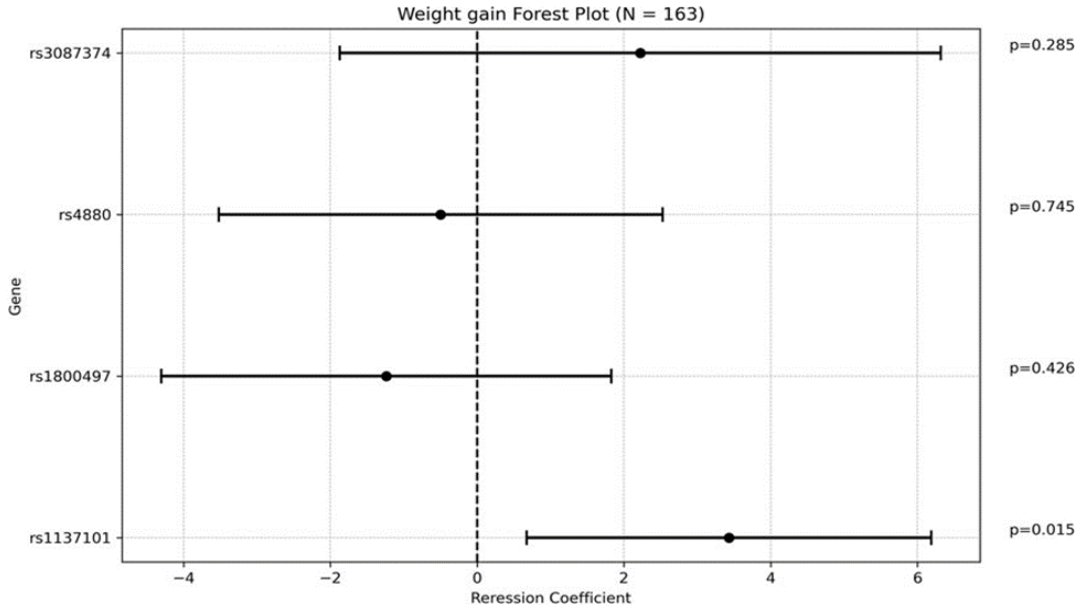

Figure 2. Multivariate regression coefficients (95% CI) describing weight gain for each SNP.

Regarding hair loss, individuals reporting this effect were maintained on a lower mean VPA dose than those without the symptom (13.38 ± 4.00 mg/kg/d vs 17.38 ± 8.04 mg/kg/d, p = 0.001). Because the number of affected participants was small, the relevance of this result is limited.

A pronounced sex imbalance was observed: 78.95% of the hair-loss group were female, compared with 41.26% in the unaffected group (p = 0.002). No additional confounders showed significant associations.

The rs1137101 (LEPR) variant was present in 68.42% of patients with hair loss but only 40.56% of those without (p = 0.021). Adjustment for sex and VPA dose produced an OR of 3.394 [1.157; 9.956], suggesting approximately a three-fold elevation in risk.

In contrast, rs4880 (SOD2) appeared more often among individuals without hair loss (73.43%) compared to those who experienced it (52.63%, p = 0.061). In the adjusted model, the OR decreased to 0.276 [0.089; 0.858], indicating a possible protective influence. No other SNPs were associated with this adverse effect **(Figure 3)**.

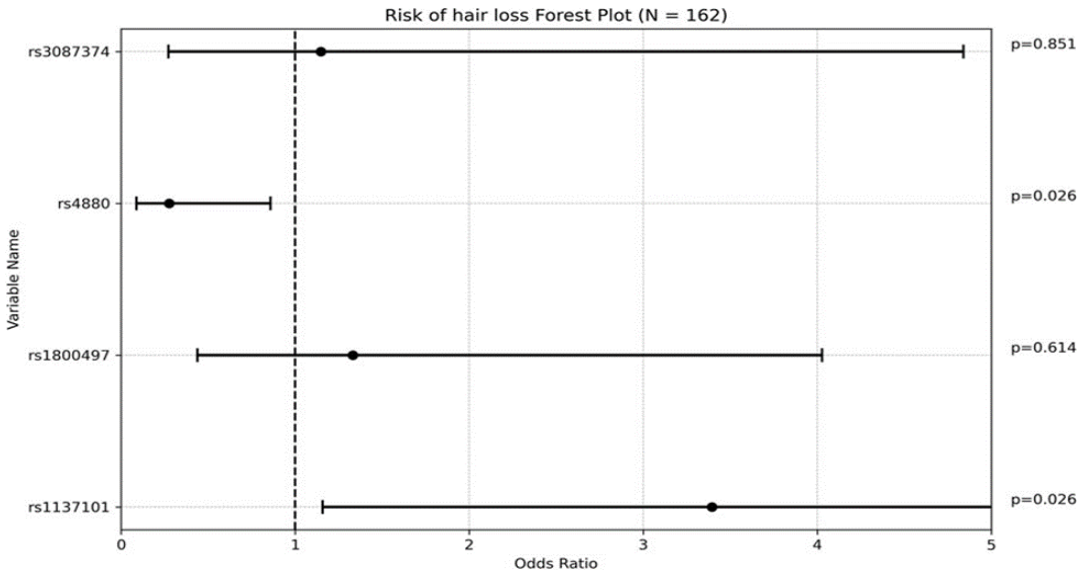

Figure 3. Multivariate odds ratios (95% CI) for the risk of hair loss across SNPs.

Association with VPA concentrations

A mild positive association was detected between age at diagnosis and ADC, characterized by a regression value of 0.048 [0.011; 0.084], p = 0.0104.

Carriers of rs1057910 (CYP2C9*3) demonstrated higher ADC levels, with a mean of 4.9 ± 1.75 [(µg/mL/day)/(mg/kg)], compared to 4.15 ± 1.61 in homozygous wild-type subjects (p = 0.028). In multivariable analysis, the coefficient was 0.722 [0.053; 1.391], p = 0.034.

The rs7668258 (UGT2B7) variant reached significance only in the unadjusted evaluation. All other SNPs were not statistically linked to ADC **(Figure 4)**.

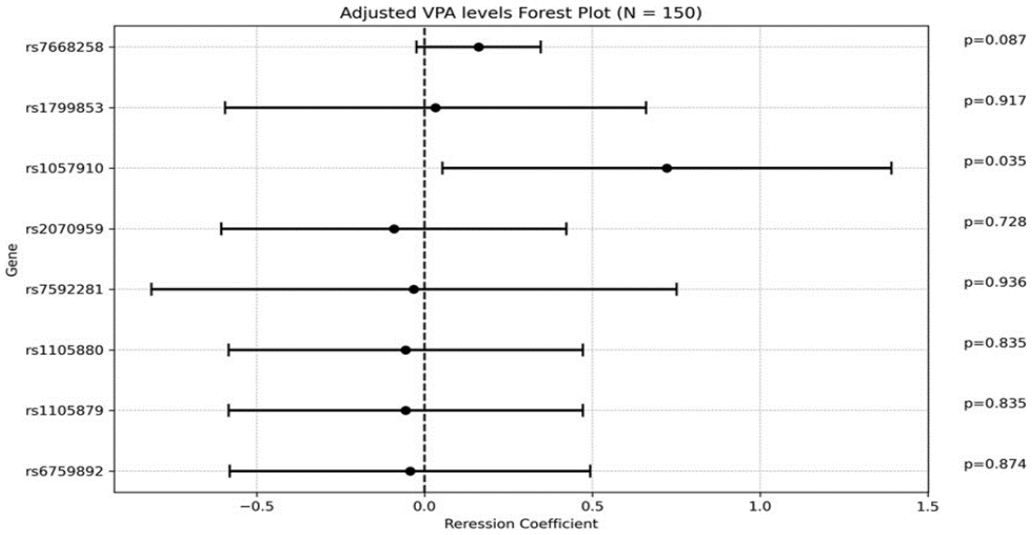

Figure 4. Forest plot displaying the multivariable regression estimates (95% CI) for ADC across the examined SNPs.

In this investigation, we detected a meaningful link between VPA treatment outcomes and the rs3892097 polymorphism in CYP2D6. We also observed that corrected VPA serum levels were influenced by the rs1057910 mutation (CYP2C9*3). In addition, several VPA-related adverse reactions appeared to depend on genetic background. Specifically, weight gain showed an association with rs1137101 (LEPR), while hair thinning corresponded to both rs1137101 (LEPR) and rs4480 (SOD2).

Our analysis demonstrated that the rs3892097 allele in CYP2D6 corresponded to a higher likelihood of therapeutic failure. CYP2D6 (also known as debrisoquine hydroxylase) contributes to the biotransformation of multiple neuropsychiatric agents [35–37]. Variants within this enzyme are known to markedly alter metabolic capacity, giving rise to established phenotypes such as ultrarapid, extensive, intermediate, and poor metabolizers, often requiring individualized dose modifications [38, 39]. Although evidence on how rs3892097 interacts with VPA is limited, prior work aligns with our results; a single study connected rs3892097 to drug-resistant epilepsy in children [40]. Our findings support this observation, and a plausible rationale is diminished VPA oxidation and accumulation of toxic intermediates linked to this genotype [41].

In contrast, the rs1057910 SNP of CYP2C9 (or CYP2C9*3) correlated with considerably elevated serum levels of VPA compared with wild-type individuals. CYP2C9 participates in metabolizing a broad range of therapeutic agents [42] and is the principal enzyme mediating VPA hydroxylation and desaturation [43]. A prior meta-analysis also concluded that heterozygotes for CYP2C9*3 maintain higher VPA concentrations relative to homozygous wild-type subjects [44]. Moreover, an earlier report documented an inverse association between VPA concentrations and seizure occurrence [45].

Hair shedding is a frequent adverse outcome of VPA therapy, with an incidence between 3.5% and 12% [46]. Hypothesized mechanisms include reductions in biotin, vitamin D, and trace elements such as zinc, iron, copper, and magnesium [46–50]. Another explanation is VPA-triggered telogen effluvium, potentially associated with its aromatase-inhibiting properties, which may provoke hyperandrogenism and promote male-pattern baldness [50]. In our cohort, the rs1137101 (LEPR) polymorphism showed a strong relationship with elevated hair loss risk—a previously unreported observation. Because the leptin receptor contributes to initiating the anagen growth cycle in hair follicles [51], a dysfunctional receptor due to rs1137101 could hinder this transition and heighten susceptibility to VPA-related alopecia.

Additionally, our data indicated that the rs4480 variant of SOD2 corresponded to a reduced frequency of hair loss, implying a protective influence. SOD2 encodes the mitochondrial superoxide dismutase enzyme, which transforms superoxide generated during oxidative phosphorylation into hydrogen peroxide (H_2O_2) and oxygen (O_2) [52]. The rs4480 substitution (Val16Ala) yields the Ala form, which is imported into the mitochondrial matrix more efficiently than the Val form, promoting enhanced enzymatic activity [53]. Earlier studies linked this Ala variant to a lower risk of liver toxicity when compared with Val/Val genotypes [54]. Our work provides the first evidence connecting rs4480 to VPA-associated hair loss.

Supporting biological plausibility, oxidative stress has been implicated in hair follicle injury and alopecia [55, 56]. VPA has been reported to elevate oxidative stress markers in pediatric subjects [57]. Thus, improved SOD2 function may counterbalance VPA-related oxidative effects and lessen hair loss severity. Moreover, VPA-induced depletion of minerals such as zinc and copper—both cofactors for SOD1 [58]—could intensify oxidative damage. Therefore, the heightened SOD2 efficiency conferred by the rs4480 allele might offset these detrimental pathways.

Our results also indicate that individuals carrying the rs1137101 allele in LEPR faced a heightened likelihood of gaining weight during VPA monotherapy. This polymorphism lies within the gene encoding the leptin receptor, a central component in regulating energy usage and metabolic processes. Through this receptor, leptin communicates information about energy reserves to the central nervous system, thereby shaping appetite control and energy expenditure. Alterations in LEPR, including rs1137101, may impair leptin signaling and contribute to leptin resistance, in which responsiveness to leptin becomes diminished. Such impairment can disturb metabolic pathways tied to glucose regulation and lipid handling, ultimately promoting weight gain. Variants in LEPR have been linked to obesity and type II diabetes mellitus [59–61]. Consistent with our observations, a previous investigation involving 212 VPA-treated epilepsy patients also showed a positive association between weight gain and rs1137101 [62].

Our study is subject to several important constraints. First, the modest sample size may have produced broader confidence intervals. Although the design was prospective, we relied on information collected from clinical records, which can introduce inaccuracies or bias. Moreover, certain outcomes—especially adverse effects—were based on subjective reporting and may be influenced by recall limitations. Another drawback is the absence of a

population pharmacokinetic model to estimate VPA clearance in a cohort spanning different ages and body sizes [21]. Such an approach could have yielded more precise estimates and stronger genotype–phenotype correlations. Despite these challenges, the study offers meaningful advantages. It is one of the earliest to assess the relevance of rs1057910 (CYP2C9*3), rs1137101 (LEPR), rs4480 (SOD2), and rs3892097 (CYP2D6) in relation to VPA responsiveness and side-effect profiles. Furthermore, the use of multivariable modeling that adjusted for factors such as maintenance VPA dosage increases the reliability of our findings.

Conclusion

In summary, our work highlights the substantial impact of genetic variability on both therapeutic success and adverse reactions during VPA therapy for epilepsy. We found that rs3892097 (CYP2D6) showed a robust association with treatment non-response, consistent with earlier evidence pointing to its metabolic consequences. We also observed that individuals harboring rs1057910 exhibited higher adjusted VPA serum levels. Additionally, correlations between rs1137101 (LEPR) and rs4480 (SOD2) with weight gain and hair loss emphasize the relevance of genotype-guided treatment strategies. These results suggest that integrating genetic information into clinical decision-making may enhance outcomes and reduce toxicity. Further research is warranted to clarify the biological mechanisms underlying these variants and to confirm our findings in larger, prospective populations.

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Ethics Statement: None

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