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Assessing the clinical impact of *CYP2C9* pharmacogenetic variation on phenytoin prescribing practice and patient response in an integrated health system

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Abstract

Objective: To assess the impact of *CYP2C9* variation on phenytoin patient response and clinician prescribing practice where genotype was unknown during treatment.

Methods: A retrospective analysis of Resource on Genetic Epidemiology Research on Adult Health and Aging cohort participants who filled a phenytoin prescription between 1996–2017. We used laboratory test results, medication dispensing records, and medical notes to identify associations of *CYP2C9* genotype with phenytoin blood concentration, neurologic side effects, and medication dispensing patterns reflecting clinician prescribing practice and patient response.

Results: Among 993 participants, we identified 69% extensive, 20% high-intermediate, 10% low-intermediate, and 2% poor metabolizers based on *CYP2C9* genotypes. Compared to extensive metabolizer genotype, low-intermediate/poor metabolizer genotype was associated with increased dose-adjusted phenytoin blood concentration (21.3 pg/mL, 95% confidence interval (CI): 13.6 – 29.0 pg/mL; $p < 0.01$) and increased risk of neurologic side effects (hazard ratio: 2.40, 95% CI: 1.24 – 4.64; $p < 0.01$). *CYP2C9* genotypes were associated with medication dispensing patterns indicating dose decrease, use of alternative anticonvulsants, and worse adherence, though these associations varied by treatment indication for phenytoin.

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Conclusion: *CYP2C9* variation was associated with clinically-meaningful differences in clinician prescribing practice and patient response, with potential implications for healthcare utilization and treatment efficacy.

Keywords

anticonvulsant; adherence; prescribing; seizure; Dilantin; precision medicine; electronic health record; pharmacogenetics

Introduction:

Phenytoin is the most commonly used anticonvulsant for preventing seizures among patients with chronic conditions and acute injury.[1] It has a narrow therapeutic index leading to half of patients experiencing minor toxicities. In addition, 10–20% suffer major adverse events, including neurological side effects (i.e., nystagmus, slurred speech, loss of balance) and Stevens-Johnson Syndrome/toxic epidermal necrolysis, a severe skin and mucosal reaction that can be fatal.[2–4] Genetic variation increases risk for these adverse events. For example, patients carrying the *Human Leukocyte Antigen (HLA)* allele *HLA-B*15:02* have at least fivefold odds of developing Stevens-Johnson Syndrome compared to patients without this allele.[5–7] Similarly, genetic variation that reduces the function of the hepatic enzyme cytochrome P450 2C9 (CYP2C9) increases phenytoin circulating blood concentrations, which raises risk of neurological side effects.[3, 4, 8–14]

For this reason, among patients with at least one copy of the *HLA-B*15:02* allele, the Clinical Pharmacogenetics Implementation Consortium (CPIC) recommends using an alternative anticonvulsant medication. Among those with one or two copies of *CYP2C9* alleles that are expected to reduce enzyme function, CPIC and the Royal Dutch Pharmacists Association - Pharmacogenetics Working Group (DPWG) recommends reducing the starting dose of phenytoin.[15, 16] The most well characterized reduced-function *CYP2C9* alleles include **2* (rs1799853) and **3* (rs1057910), which together are found in roughly one third of patients of European descent.[15] Other alleles that are found more commonly in patients of non-European descent likely also decrease CYP2C9 function, but these alleles and their consequences for diverse populations are poorly described.[15, 17–22]

While the associations of *CYP2C9*2* and *CYP2C9*3* with decreased phenytoin metabolism and increased phenytoin blood concentrations are well-established in clinical research settings, preemptive genetic testing is still uncommon in many health systems and evidence for an association of *CYP2C9* variation with the risk of phenytoin-induced neurological side effects in community care settings is lacking.[15] Thus, we investigated whether the associations of *CYP2C9* variation with known pharmacogenetic outcomes of phenytoin blood concentrations and risk for neurological side effects were present in a routine clinical care setting. We also examined how *CYP2C9* pharmacogenetic variation might affect clinician prescribing practice and patient response. Using data from a large, multi-ethnic patient population in which *CYP2C9* variation was unknown throughout treatment, we used medication dispensing patterns to evaluate clinician action in optimizing phenytoin dose or anticonvulsant choice, and investigated patient adherence to phenytoin treatment.

Methods:

IRB.

The Kaiser Permanente Northern California (KPNC) Internal Review Board approved this study.

Cohort.

We conducted a retrospective cohort study in the Resource for Genetic Epidemiology Research on Adult Health and Aging (GERA) cohort, which includes 110,266 participants ≥18 years old, who were members of the KPNC healthcare delivery system, donated a DNA sample, completed a health survey including self-identifying race/ethnicity, and agreed to share their electronic health record (EHR), as described elsewhere.[23, 24] KPNC is an integrated healthcare delivery system that serves >4 million people in northern California who are generally representative of the regional population with respect to race/ethnicity and socioeconomic status.

Using outpatient medication dispensing records, we identified members of the GERA cohort who filled at least one prescription for phenytoin as an outpatient at any point between January 1, 1996 and September 1, 2017. We extracted all instances of phenytoin dispensing in an outpatient setting, including date, days supply, and daily dose, and all dispensing records for alternative anticonvulsants, including phenobarbital, carbamazepine, clonazepam, diazepam, divalproex, ethosuximide, gabapentin, felbamate, lacosamide, lamotrigine, levetiracetam, lorazepam, oxcarbazepine, topiramate, pregabalin, primidone, valproic acid, and zonisamide. Using data from the KPNC EHR, we determined age at first phenytoin dispensing, year of first dispensing, gender, dates of health insurance coverage within KPNC, and death date, if applicable. We excluded participants who died or left KPNC membership within 30 days of initiating phenytoin therapy. All data were deidentified prior to analysis.

Genotyping:

DNA collection and genotyping are described elsewhere.[24,23] *HLA* typing was previously performed for all members of the GERA cohort using SNP2HLA.[25] To identify variation in the *CYP2C9* gene, we compiled a list of genomic reference numbers (rs numbers) for known variants using the Pharmacogene Variation Consortium (PharmVar) reference database (pharmvar.org).[26] Within the phenytoin cohort, only variants rs1799853 (**2*), rs1057910 (**3*), rs28371686 (**5*), rs7900194 (**8*), rs28371685 (**11*), and rs9332239 (**12*) were present. We tested for all variants for Hardy-Weinberg Equilibrium using an exact test. Supplemental Table 1 illustrates the assignment of each of these variants in the star nomenclature and their expected effect on CYP2C9 activity.

According to the PharmVar database, the **2* and **3* alleles definitively decrease CYP2C9 enzymatic function while the other variants may decrease function.[26] Because of the low frequency of the *5, *8, *11 and *12 alleles and the likelihood that not all can be clustered in the same phenotypic category, we excluded participants with these alleles from analysis. The **3* allele has a greater than twofold deleterious effect on enzyme activity compared to

the **2* allele.[18, 27] Therefore, we assigned each participant an expected CYP2C9 metabolic activity as "Extensive Metabolizer" (neither **2* nor **3* identified), "High-Intermediate Metabolizer" (one **2* variant identified), "Low-Intermediate Metabolizer" (one **3* variant or two **2* variants identified), or "Poor Metabolizer" (two **3* or one **2* plus one **3* variants identified).

Clinical phenotyping:

Dose-adjusted phenytoin blood concentrations.—We identified all results of total (free and bound) circulating phenytoin blood level laboratory tests (μg/mL), which were measured in serum using a chemiluminescent microparticle immunoassay. We calculated the dose-adjusted blood level for each patient as the first phenytoin blood level laboratory test result (μg/mL) that occurred within the first 100 days of treatment, divided by the first daily phenytoin dose (mg).

Chart reviews of clinical text notes to assess neurological side effects.—We reviewed electronic health record clinical notes for all participants with a first phenytoin dispensing date after January 1, 2005, which is when electronic notes became embedded in the EHR, so that we could capture the early patient symptoms and clinician assessment of phenytoin treatment in these notes. We recorded instances of clinical notes that mentioned a side effect with phenytoin within 100 days of first phenytoin dispensing, and classified these side effects using the IBM Micromedex system[28] into neurological (i.e., nystagmus, slurred speech, dizziness, somnolence) and other (i.e., gum hyperplasia, nausea, rash). Because phenytoin side effects are characterized by central nervous system dysfunction, we included only the neurological side effects in analysis. We did not adjudicate these clinical impressions for their plausibility that the symptom was phenytoin-induced given that these were historical records. Using clinical notes, we also noted the indication for phenytoin treatment, and classified these reasons into seizure (i.e., all types of epilepsy or chronic seizure) or other (e.g., neuropathy and prophylaxis for occurrence of seizures following trauma or meningioma resection). Chart reviews were conducted while blinded to genotype.

Pharmacogenetic outcomes derived from medication dispensing records.—In response to high phenytoin blood concentrations or report of side effects, clinicians frequently reduce the daily dose of phenytoin or prescribe an alternative anticonvulsant medication. Patients who experience side effects from high blood concentrations of phenytoin may try to reduce symptoms themselves by taking the medication less often (i.e.,poor adherence), as has been shown previously in other chronic conditions. [29–32] We investigated these features of clinician prescribing practice and patient response with the following objective and quantifiable measures based on medication dispensing patterns, also summarized in Supplemental Table 2. The analytical flow, including which participants were included in each step of the analysis is illustrated in Supplemental Figure 1.

Dose decrease.: Among participants with more than one phenytoin prescription fill, we determined whether phenytoin dose decreased from first phenytoin daily dose to last phenytoin daily dose within the first year of treatment, adjusting for whether that patient took concomitant anticonvulsants at any point during that year.

Switching to alternative anticonvulsant.: We identified participants who filled a prescription for any alternative anticonvulsant in the first 100 days after the first dispensing of phenytoin and who did not subsequently fill another phenytoin prescription. We excluded participants who had filled a prescription for an alternative anticonvulsant in the 180 days prior to first phenytoin dispensing, as any ongoing fills of these alternative anticonvulsants might not reflect clinical issues with phenytoin.

Poor patient adherence.: Among participants who filled more than one prescription, we identified participants as having poor adherence in the first year of treatment if they had a gap of >30 days at any point between the end of supply and the date of a new prescription dispensing record.

Statistical analysis:

We performed all data processing and analysis in R programming language (version 3.5). We used multivariate linear regression to model dose-adjusted blood concentrations and multivariable logistic regression to model binary outcomes (dose reduction, switching anticonvulsants, and poor medication adherence). We performed a Cox proportional hazard model to determine the association between genotype and neurological side effect, censoring participants when, if ever, they filled a prescription for an alternative anticonvulsant. All models were adjusted for age by decade at first phenytoin dispensing, sex, race/ethnicity, and whether the patient was anticonvulsant-naïve based on the prior 6 months. All models include *CYP2C9* metabolizer genotype as categorical variables, with extensive metabolizers as reference. We combined the low-intermediate and poor metabolizer subgroups due to low sample size in the poor metabolizer group (n = 16 in full cohort; n = 4 in seizure subset). The significance threshold was $p \leq 0.05$ for all analyses. Clinical indication for phenytoin may affect clinician and patient behavior. Therefore, we also conducted analyses in a subset of patients with a confirmed indication for seizures (seizure subset) under the assumption that these patients would receive long-term anticonvulsant treatment. Supplemental Table 3 presents results from sensitivity analyses regarding non-normal distribution of dose-adjusted blood concentrations and potential selection biases arising from our inability to account for ongoing phenytoin treatment initiated prior to electronic records being available.

Results:

Overall cohort summary

We identified 1070 participants in the GERA cohort who had filled a phenytoin prescription as a KPNC member and excluded 65 for death or loss of KPNC membership within 30 days of first phenytoin dispensing. Our data consisted of 22,122 instances of phenytoin dispensing and 10,245 total phenytoin blood laboratory test results. Mean age at first dose was 62 (+/− 15) years, median number of phenytoin fill records was 4 (interquartile range (IQR) 1 – 29) per participant, and median number of phenytoin blood laboratory tests was 4 (IQR 2 – 12) per participant. The median starting dose for all participants was 300 mg/day (IQR: 300 – 300).

CYP2C9 and HLA-B genotyping results

Based on the 6 *CYP2C9* single nucleotide polymorphism locations, the **2* allele was most common, with a frequency of 12.0%. The **3* was found with a frequency of 5.1%. Together, the **5, *8, *11,* and **12* alleles were found in 12 of the remaining participants, at a frequency of 0.6%. The *1 allele (no variant identified) was found at 82.2%. All variants were in Hardy-Weinberg Equilibrium. Table 1 presents the frequency of *CYP2C9* allele combinations in each of the metabolizer groups. Only four individuals had an *HLA-B*15:02* allele. Because of this small number, we did not study associations between the *HLA-B*15:02* allele and risk of Stevens-Johnson Syndrome/toxic epidermal necrolysis in this cohort.

Associations of reduced-function CYP2C9 with dose-adjusted phenytoin blood concentration.

Table 2 presents participant characteristics overall and by predicted CYP2C9 metabolizer subgroup. Compared to CYP2C9 extensive metabolizers, high-intermediate metabolizers had an 8.6 pg/mL increase in mean dose-adjusted phenytoin blood concentrations (95% confidence interval (CI): 2.3 – 14.8 pg/mL; $p < 0.01$), and low-intermediate/poor metabolizers had a 21.3 pg/mL increase (95% CI: 13.6 – 29.0 pg/mL; $p < 0.01$) (Table 3). Age and race/ethnicity were not associated with dose-adjusted blood concentration, but male participants had significantly lower dose-adjusted blood levels compared to females (8.6 pg/mL, 95% CI: 3.8 – 13.6; $p < 0.01$). Based on therapeutic monitoring test results among participants who had a starting dose of 300 mg/day (n = 363), we observed a greater increase in phenytoin blood concentration with **1/*3* genotype (mean concentration 16.2 μg/mL +/− 7.7 μg/mL) than with the **1/*2* genotype (mean concentration 12.7 μg/mL +/− 7.1 μg/mL) when compared to the **1/*1* genotype (mean concentration 8.8 μg/mL +/− 5.2 μg/mL) (Supplemental Figure 2).

Associations of reduced-function CYP2C9 with neurological side effects

We reviewed the EHR clinical notes for all of the 382 (39%) participants who filled a first phenytoin prescription in 2005 or later (Table 4). Of these, over half (n = 232, 60%) received phenytoin for active seizure treatment, including epilepsy. Other indications included prophylaxis for brain neoplasm or head trauma (n = 95, 25%) and neuropathy (n = 25, 7%). *CYP2C9* genotype was not associated with seizure indication (chi-squared test $p = 0.97$).

Among participants with a first phenytoin dispensing record in 2005 and later, we identified 72 (19%) who had an EHR clinical note indicating a neurological side effect within 100 days of first phenytoin dispensing, including 13 (34%) of participants expected to be low-intermediate or poor CYP2C9 metabolizers (Table 4). Compared to extensive metabolizers, high-intermediate metabolizers did not have increased risk of neurological side effects in the full cohort (hazard ratio (HR) 1.06; 95% CI: 0.53 – 2.11; p = 086), or in the seizure subset (HR 1.31; 95% CI: 0.65 – 2.65; $p = 0.46$). Low-intermediate/poor metabolizers had significantly increased risk of a neurological side effect in the full cohort (HR 2.40; 95% CI: 1.24 – 4.64; $p < 0.01$) and increased risk, though not significantly, in the seizure subset (HR 2.09; 95% CI: 0.95 – 4.60; $p = 0.07$). Males were less likely to have neurological side effects

in the full cohort (HR 0.51; 95% CI: 0.31 – 0.86; p = 0.01), but age, race/ethnicity, and being anticonvulsant-naïve were not associated with neurological side effects.

Associations of reduced-function CYP2C9 genotype with clinician prescribing practice and patient response.

Compared to among extensive metabolizers, the mean starting dose did not vary for high-intermediate metabolizers (p = 0.76) or low-intermediate/poor metabolizers (p = 0.71) after adjusting for age, sex, and race/ethnicity. Only sex was associated with a significantly different starting dose, with males receiving 25.1 mg/day more than females (p = 0.01). However, low-intermediate/poor *CYP2C9* genotype was associated with greater odds of having a lower dose by the end of the first year of treatment in the full cohort (OR 1.11; 95% CI: 1.02 – 1.22; p = 0.02), though this association was not significant in the smaller seizure subset (low-intermediate/poor OR 1.13; 95% CI: 0.90 – 1.43; p = 0.30) (Table 5). In the full cohort, the average change in dose after a year was 49.2 mg/day greater in the low-intermediate/poor metabolizers compared to extensive metabolizers (95% CI: 20.4 – 78.0 mg/day; p < 0.01).

Compared to extensive metabolizers, high-intermediate *CYP2C9* metabolizers were at increased odds of switching to an alternative anticonvulsant within 100 days after first phenytoin dispensing in the seizure subset (high-intermediate OR 1.22; 95% CI: 1.05 – 1.43; p = 0.01), but not among low-intermediate/poor metabolizers in the seizure cohort (OR 1.10; 95% CI: 0.88 – 1.37; p = 0.41) or in the cohort overall (high-intermediate OR 1.06; 95% CI: 0.99 – 1.13; p=0.08; low-intermediate/poor OR 1.06; 95% CI: 0.98 – 1.14; p = 0.13) (Table 5). The most common alternative anticonvulsants replacing phenytoin were levetiracetam (n = 40; 23%), carbamazepine (n = 39; 22%), gabapentin (n = 22; 13%), lorazepam (n = 21; 12%), and divalproex (n = 13; 7%).

Similarly, among participants who filled more than one prescription, low-intermediate/poor metabolizers had worse adherence compared to extensive metabolizers in the first year within the full cohort (OR for poor adherence: 1.95; 95% CI: 1.12 – 3.39; p =0.01) but not in the seizure subset (OR 0.96; 95% CI: 0.18 – 5.01; p = 0.96) (Table 5).

Discussion:

This study is the largest analysis to-date of *CYP2C9* pharmacogenetic associations with phenytoin response. It is novel for three major reasons: 1) it differentiates the relative effects of the *CYP2C9*2* and *CYP2C9*3* alleles on phenytoin metabolism, which have been debated in clinical research; 2) it reveals a concrete association between *CYP2C9* genotype and risk for neurological side effects; and 3) it is the first analysis, to our knowledge, to evaluate pharmacogenetic associations with clinician prescribing practice and patient response using objective medication dispensing measures. Using previously-studied and innovative measures of pharmacogenetic outcomes, we comprehensively describe the clinically-meaningful associations of *CYP2C9* pharmacogenetic variation with phenytoin treatment in population-based care.

In a cohort of nearly 1000 patients, we confirmed the well-known association between reduced-function *CYP2C9* genotypes and increased dose-adjusted phenytoin blood concentrations. These clinical laboratory results confirm *in vitro* findings[18, 27] that the decrease in metabolism due to a **3* allele is more severe than that for a **2* allele. CPIC and DPWG have the difficult task of assigning alleles into functional categories tenable for dosing recommendations, and data such as these illustrate the importance of carefully assessing the different effects of alleles on CYP2C9 function when designing effective decision-support tools for phenytoin and other CYP2C9 substrates.

The risk of neurologic side effects was similar among expected extensive and high-intermediate metabolizers, but increased dramatically among expected low-intermediate/poor metabolizers, suggesting that these patients with the lowest CYP2C9 function may be particularly vulnerable to neurological side effects. This analysis is complicated by the difficulty of assessing side effects objectively in the EHR – identifying them relies on patients reporting their symptoms to healthcare providers, on those providers documenting symptoms in clinical notes, and on researchers manually reviewing those text notes to extract them. Because of this subjectivity, we sought more objective measures of patient experience.

We hypothesized that we could identify associations of *CYP2C9* genotypes with patterns of clinician prescribing practice and patient response evident in objective measures of medication dispensing records. Interestingly, reduced-function *CYP2C9* genotypes were associated with increased odds of switching to an alternative anticonvulsant only in the seizure subset and not in the full cohort. Conversely, low-intermediate/poor *CYP2C9* genotype was associated with dose reduction and poor adherence only in the full cohort and not in the seizure subset. Indication for phenytoin did not vary by *CYP2C9* genotype. Thus the indication for phenytoin likely affects clinician and patient response to side effects and dosing challenges.

Compared to patients taking phenytoin for neuropathy or seizure prophylaxis, patients with active and ongoing seizures require long-term medication use for seizure prevention, and the consequence of sub-therapeutic blood concentrations from too low of a dose or poor adherence – a breakthrough seizure – is severe. Therefore, clinicians may be more likely to seek alternative, more tolerable anticonvulsants for patients experiencing difficulties on phenytoin rather than to decrease their phenytoin dose. Patients with active seizures may be more likely to adhere to treatment despite minor side effects, having already experienced seizures and having greater perceived consequences of being noncompliant.

Conversely, patients who do not need long-term treatment for seizures may be less tolerant of even mild side effects, and clinicians may lower the phenytoin dose in these patients, less concerned with breakthrough seizures. Side effects, even those deemed too minor to report to clinicians, are known to decrease medication adherence, as patients are more likely to self-titrate by reducing or missing doses in order to reduce side effect symptoms.[33–36] The effects of side effects on decreased medication adherence have been observed in patients with chronic conditions, such as hypertension[31, 32], diabetes[30], and mental health disorders[29]. Furthermore, although nearly all participants began treatment with 300 mg/day,

the lower the predicted function of CYP2C9, the more likely participants were to have a lower dose after one year. This dose titration process may reflect continued and perhaps increased utilization of healthcare resources compared to patients who are immediately within therapeutic range. These results suggest that pharmacogenetic variation may impact both therapeutic efficacy and healthcare utilization by increasing risk for mild side effects.

Limitations of this study include an inability to account for all factors known to affect phenytoin pharmacokinetics, including participant weight, comorbidities, and concomitant use of CYP2C9 inducers or inhibitors. Gender was a significant predictor of dose-adjusted blood concentrations, perhaps due to weight differences. We were unable to ensure capturing phenytoin steady state in the first laboratory test result or to account for the nonlinear kinetics of phenytoin. Furthermore, the **5, *8, *11,* and **12* alleles were too rare for us to study in this cohort, but are important to characterize to improve equity in pharmacogenetic knowledge because they are more prevalent in non-European populations. Due to using electronically available data from a single health system, we were unable to account for medical care or medications received outside of this health system or prior to the start of the study period, which is when pharmacy and laboratory records became electronic. Participants who were already on phenytoin therapy when joining the health plan or at the start of the study period may be more likely to have responded well to phenytoin, as we would not detect participants who initiated phenytoin outside of this time, but stopped due to adverse events. While we were able to identify neurological side effects through manually reviewing clinical notes, we were not able to differentiate their severity, adjudicate their plausibility, or link them with specific phenytoin blood concentrations. Future studies linking medication dispensing patterns to side effects, breakthrough seizures, and healthcare utilization and cost metrics may help clarify these broad consequences of pharmacogenetic variation in routine phenytoin treatment.

In summary, our results suggest that the impact of varied CYP2C9 activity on clinician prescribing practice and patient compliance may depend on the indication for treatment and the relative importance of risk of adverse side effects and therapeutic failure. These results provide evidence for clinically-meaningful effects of *CYP2C9* genotypes beyond their well-studied associations with altered phenytoin blood concentrations and risk of neurological side effects, suggesting that preemptive pharmacogenetic testing to improve treatment decisions may have broad benefits for healthcare delivery systems.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Table 1:

Transformation of *2 and *3 star alleles into genotypes and expected effect of CYP2C9 metabolic activity.

CYP2C9 Metabolizer Group	Star allele	Number with genotype, n (%) (total = 993)
Extensive	*1/*1	680 (68%)
High-Intermediate	*1/*2	196 (20%)
Low-Intermediate	*1/*3 *2/*2	85 (9%) 15 (2%)
Poor	*2/*3 *3/*3	16 (2%)

Table 2:

Cohort demographics and therapeutic blood monitoring test results according to *CYP2C9* metabolizer genotype. Metabolizer status was based on*1, *2, and *3 alleles. Individuals with *5, *8, *11 or *12 alleles were excluded from analysis.

		Expected CYP2C9 metabolic activity				
		Total	**Extensive**	**High-Intermediate**	**Low-Intermediate**	**Poor**
Total		993	680	196	101	16
Gender, n (%)						
	Female	521 (52%)	346 (51%)	113 (58%)	54 (53%)	8 (50%)
	Male	472 (48%)	334 (49%)	83 (42%)	47 (47%)	8 (50%)
Age at first fill						
	<40	75 (8%)	53 (8%)	12 (6%)	10 (9%)	
	40–60	353 (36%)	252 (37%)	63 (32%)	38 (34%)	
	61–80	449 (55%)	286 (42%)	104 (53%)	59 (50%)	
	81+	116 (12%)	89 (13%)	17 (9%)	10 (9%)	
Race/ethnicity, n (%)						
	Asian	38 (4%)	33 (5%)	< 10	< 10	< 10
	Black	45 (5%)	40 (6%)	< 10	< 10	< 10
	White, Hispanic	75 (8%)	57 (8%)	12 (6%)	< 10	< 10
	White, non-Hispanic	827 (82%)	545 (80%)	176 (90%)	91 (90%)	14 (88%)
Therapeutic blood monitoring						
Mean first blood test result µg/mL (sd)		10.1 (6.4)	8.8 (5.4)	12.1 (7.1)	15.0 (7.7)	

May not add to 100% due to rounding; cells with low counts are masked for participant privacy.

Table 3:

Linear regression models of dose-adjusted phenytoin blood concentrations (pg/mL) with and without considering genetic variation in *CYP2C9*.

Dose-adjusted blood concentrations reflect the first phenytoin blood concentration test result within 100 days of starting treatment, divided by the first phenytoin daily dose.

	Base model (R^2: 0.01)		Genetic model (R^2: 0.07)	
Covariate	**Beta (standard error)**	**p-value**	**Beta (standard error)**	**p-value**
Age (by decade)	−0.1 (0.8)	0.85	−0.3 (0.8)	0.73
Male sex	−8.8 (2.6)	< 0.01	−8.6 (2.5)	< 0.01
Race/ethnicity (ref = White, non-Hispanic)				
Asian	4.7 (6.4)	0.46	5.7 (6.3)	0.37
Black	4.5 (5.8)	0.43	7.1 (5.7)	0.22
White, Hispanic	1.0 (4.6)	0.82	1.2 (4.5)	0.80
CYP2C9 genotype (ref = extensive metabolizer)				
High-Intermediate			8.6 (3.2)	< 0.01
Low-intermediate/Poor			21.3 (3.9)	< 0.01

ref: reference

Table 4:

Neurological side effects within the first 100 days of treatment, as reported in the clinical notes of participants who began phenytoin treatment after 2005, and presented by expected CYP2C9 function based on genotype and by confirmed indication for seizure. Hazard ratio presents the risk of neurological side effect for high-intermediate and low-intermediate/poor *CYP2C9* genotype compared to the risk for extensive metabolizer genotype, based on cox proportional hazards regression adjusting for age, sex, race/ethnicity, and whether the participant was anticonvulsant-naïve. Participants were censored when, if ever, they filled a prescription for an alternative anticonvulsant medication.

		Expected CYP2C9 metabolic activity		
	Total	**Extensive**	**High-Intermediate**	**Low-Intermediate / Poor**
Included in Chart Review, n	382	264	77	41
Neurological side effect, n (%)	72 (19%)	46 (17%)	13 (17%)	13 (32%)
HR compared to extensive metabolizers (95% CI); p-value			1.06 (0.53 – 2.11); p = 0.86	2.40 (1.24 – 4.64); p < 0.01
Validated seizure indication, n	232	158	51	23
Neurological side effect among participants with seizure indication, n (%)	59 (25%)	35 (22%)	13 (25%)	9 (39%)
HR compared to extensive metabolizers (95% CI); p-value			1.31 (0.65 – 2.65); p = 0.46	2.09 (0.95 – 4.60); p = 0.07

HR: Hazard ratio; CI: confidence interval

Table 5:

Odds of clinician prescribing practice and patient response outcomes among high-intermediate and low-intermediate/poor *CYP2C9* genotypes in the full cohort (n = 993) and the seizure subset (n = 232) compared to odds in extensive metabolizers. All analyses are adjusted for age, sex, and race/ethnicity. Dose reduction is also adjusted for being anticonvulsant-naïve and for dual anticonvulsant therapy. Adherence is adjusted for being anticonvulsant-naïve.

	Full cohort			Seizure cohort		
Prescribing outcome	**n**	**OR (95% CI)**	**p-value**	**n**	**OR (95% CI)**	**p-value**
Dose reduction	732			164		
High-intermediate	125	1.04 (0.97 – 1.12)	0.24	32	1.12 (0.95 – 1.31)	0.17
Low-intermediate/poor	68	1.11 (1.02 – 1.22)	0.02	12	1.13 (0.90 – 1.43)	0.30
Switch anticonvulsant	821			159		
High-intermediate	145	1.06 (0.99 – 1.13)	0.08	36	1.22 (1.05 – 1.43)	0.01
Low-intermediate/poor	90	1.06 (0.98 – 1.14)	0.13	15	1.10 (0.88 – 1.37)	0.41
Poor medication adherence	732			164		
High-intermediate	125	1.18 (0.73 – 1.89)	0.50	32	1.43 (0.52 – 3.94)	0.49
Low-intermediate/poor	68	1.95 (1.12 – 3.39)	0.01	12	0.96 (0.18 – 5.01)	0.96

OR: odds ratio; CI: confidence interva