Journal of Neonatal Surgery
ISSN (Online): 2226 -0439
Vol. 14, Issue 06 (2025)
https://www.jneonatalsurg.com

OPEN ACCESS

Pharmacogenomic Analysis of Genetic Variability in Drug Metabolizing Enzymes- Implications for Personalized Medicine

Dr. Ankur Singh[1*], Dr. Vishnu Gupta[2], Dr. Akanksha Garg[3], Mr. Deepesh Kumar[4], Dr. Seema Yadav[5], Dr. Anup Kumar Chakraborty[6]

[1*]Associate Professor, SOS School of Pharmacy, Faculty of Pharmacy, IFTM University, Moradabad, U.P. Pin code-244102.
Email ID: ankursingh108@gmail.com
[2]Assistant Professor Department of Community Medicine SRVS Medical College Shivpuri M.P
Vishnukgupta4@gmail.com
[3]Assistant professor, Adesh Medical College, Mohri, Shahabad, subharti15@gmail.com
[4]Assistant Professor, Faculty of Pharmacy, Kalinga University, Raipur, C.G..
Email ID: deepesh.kumar@kalingauniversity.ac.in
[5]Principal, College of Nursing, Sarojini Naidu Medical College, AGRA, UP, https://orcid.org/0009-0008-1613-2856 seemakishan22@gmail.com
[6]Professor, B.R. Nahata College of Pharmacy, Faculty of Pharmacy, Mandsaur University, Mandsaur - 458001 (MP) anup.chakraborty@meu.edu.in

Cite this paper as: Dr. Ankur Singh, Dr. Vishnu Gupta, Dr. Akanksha Garg, Mr. Deepesh Kumar, Dr. Seema Yadav, Dr. Anup Kumar Chakraborty, (2025) Pharmacogenomic Analysis of Genetic Variability in Drug Metabolizing Enzymes- Implications for Personalized Medicine. *Journal of Neonatal Surgery*, 14 (06), 296-304.

ABSTRACT

Background: IIV is due in part to genetic polymorphisms of drug-metabolizing enzymes. This study aimed to compare variations in CYP2D6, CYP2C9, CYP3A4, and UGT1A1 enzymes and their activities in patients.
Methods: Genotyping for CYP2D6 (*1A/*2G/*3T), CYP2C9 (*1C/*2T/*3A), CYP3A4 (*1A/*2G), and UGT1A1 (*28T/*36A) polymorphisms in 500 participants. Genotype frequencies were obtained and checked for compatibility with Hardy-Weinberg equilibrium. Regression analysis evaluated the relationship between genotypes and enzyme activity for each enzyme.
Results: Allele frequencies which varied from 20-60% indicated that there was heterogeneity within the population. Analysis of genotype distribution revealed no deviation from Hardy-Weinberg equilibrium for any of the genotyped loci. The *1/wild-type alleles of CYP2D6, CYP2C9, and CYP3A4 had higher regression coefficients (0.45-0.55) of enzyme activity indicating that compared to variant alleles, the alleles were highly active. Comparing the two alleles for UGT1A1, *36A had a 60% higher activity versus *28T.
Conclusions: As observed, polymorphisms were statistically related to variations in enzyme activity among individuals for the analyzed drug-metabolizing enzymes.

Keywords: *pharmacogenetics, cytochrome P450, UDP-glucuronosyltransferase, polymorphism, drug metabolism*

1. INTRODUCTION

Pharmacogenomics is defined as the practice of designing drugs based on genes and genetic variations that exist within patients and are linked to biomarkers (1). There is clear agreement that pharmacogenomics is an essential branch of personalized medicine since it deals with differences in drug action connected with genetic characteristics influencing drug uptake and clearance (2). First, polymorphisms of the drug metabolizing enzymes which lead to changes in the activity of the enzyme affect the pharmacodynamics and pharmacokinetics of the drug (3). Cytochrome P450 enzymes and UGT enzymes are also important in drug metabolism and understanding variation due to polymorphism has been well established (4). The most significant of these are CYP2D6, CYP2C9, CYP3A4, and UGT1A1 which are involved in the metabolism of over 60% of known therapeutic drugs and genotyping of these pathways has clinical relevance in dosing modifications (5).

Different polymorphisms of CYP2D6 are important in approximately 20–25% of all prescribed drugs, for instance, antidepressants and anticancer medicines (6). Patients with low activity of CYP2D6 due to loss-of-function alleles may be at risk for toxicity because the drugs such as codeine or tamoxifen, will have poor clearance (7). On the other hand, patients possessing CYP2D6 gene duplication or multiplication are likely to metabolize the drug so rapidly that the desired therapeutic effects are not realized before it is cleared from the body – as can be the case with the ADHD drug atomoxetine

(8). In addition, it is found that genetic polymorphisms of CYP2C9 affect the metabolism of several drugs such as warfarin and tolbutamide and have shown variable efficacy and toxicity (9). In particular, warfarin is characterized by a low therapeutic window and the administration of the proper doses to maintain anticoagulation while avoiding bleeding is a concern of a clinician. As mentioned earlier, genotyping CYP2C9 before starting the patient on warfarin will assist in guiding the initial doses for effective outcomes as mentioned in guideline no.10. Further, CYP3A4 is responsible for the metabolism of approximately 50% of all pharmaceutical drugs across the therapeutic specializations such as oncology, cardiology and psychiatry (11). Nevertheless, the role of genetic polymorphisms in CYP3A4 remains questionable and the clinical significance of the enzyme's variants is not definitively proven (12). Last of all, UGT1A1 metabolizes bilirubin and many therapeutic drugs; TA insertion polymorphism in UGT1A1 has been reported to decrease its expression as well as activity (13). This condition, which is characterized by low activity of the UGT1A1 enzyme, can result in hyperbilirubinemia with drugs metabolized by this enzyme such as the anti-cancer drug irinotecan (14).

Based on the findings that genetic polymorphisms have an impact on drug response, pre-treatment genotyping of metabolic pathways is valuable in determining the most suitable drugs and dosages for improved results (15). Additionally, regular recording of allele frequencies in world populations allows evaluation of differences in drug effects in different ethnicities (16). For instance, Africans have a higher prevalence of reduced function CYP2D6 alleles than Asians do, which are causes of differences in codeine analgesia (17). Hence, the objective of this research is to identify the allelic distribution of select drug-metabolizing enzymes, CYP2D6, CYP2C9, CYP3A4 and UGT1A1, and to ascertain the extent of genotype-phenotype concordance in an ethnically diverse sample. Assessing this pharmacogenomic variability and the significance of the said impact will inform the use of the drugs in safe clinical practice considering an individual's genetic makeup.

The application of pharmacogenomic tests before medication administration signifies the hope for a more efficient way of patient treatment through the use of personalized medicine (18). Nevertheless, studies regarding clinical validity and utility across patients were found to be necessary to inform its widespread application (19). Furthermore, there are considerations of ethical acceptance, cost impact, and the education of the provider that needs to complement the growing evidence (20). Thus, the results of this cross-sectional study to account for genetic polymorphisms of the critical db metabolizing pathways compared between the ethnicities will go a long way in the advancement of the use of personalized medicine where therapy is by the patient's/subject's genotype.

2. REVIEW OF LITERATURE

Drug metabolizing enzymes are involved in the process of biotransformation of medicines in the body for them to be expelled from the body. The two key stages of drug metabolisms include the phase I reactions which include functionalization reactions such as oxidation, reduction and hydrolysis by enzymes like the cytochrome P450 (CYPs) family and phase II reactions which involve the conjugation reactions catalyzed by transferase enzymes including the UDP-glucuronosyltransferases (UGTs) and glutathione S-transferases (GSTs.) (21). These reactions help to enhance the water-soluble properties of drugs for renal and biliary excretion.

CYP450 enzymes came under phase I enzymes because they help in metabolic reactions that include oxidation. CYPs are considered the prime enzymes for the metabolism of drugs in humans and are the most abundant set of phase I enzymes. They are hemoproteins that are mostly attached to the endoplasmic reticular membranes and mitochondrial membranes of hepatocytes and enterocytes (22). As has been mentioned, there are 57 putatively functional CYP genes in humans, from which the most important role in drug metabolism belongs to CYP 1, 2 and 3 (23).

Both UGTs and GSTs are phase II conjugating enzymes that are involved in transferases of charged species such as glucuronic acid and glutathione to the phase I metabolites to increase hydrophilicity and facilitate elimination. UGTs are bound enzymes that are widely distributed in several tissues such as the liver, gastrointestinal tract and renal system of mammals (24). GSTs belong to three major groups which include the cytosolic, mitochondrial and microsomal groups (25).

There are variations in the gene sequence in all individuals; however, these polymorphisms are defined as variations occurring in populations greater than 1% (26). The cases are well documented such as cases of CYP2D6 gene duplication leading to ultrarapid metabolism and non-functional CYP2D6*4 and *5 alleles leading to poor metabolism (27). Other polymorphic CYPs of importance are CYP2C19*2 and *3 and CYP2C9*2 and *3 variant genotypes. UGT1A1*28 is also clinically significant and is in phase II enzymes (28).

In this case, genetic polymorphisms have considerable implications on the metabolizing capacity of a drug. In poor metabolizers, standard drug doses may cause more harm as seen in increased side effects, and in rapid metabolizers, the drug does not effectively work due to inadequate drug levels in the body (29). These include toxicity of codeine in CYP2D6 ultrarapid metabolizers because of higher morphine formation (30) and life-threatening toxicity from irinotecan in patients with UGT1A1*28/*28 genotype because of impaired SN-38-glucuroconjugation. This knowledge has allowed for dose individualization according to metabolizer phenotype, which has improved outcomes in different patients.

3. METHODOLOGY

3.1. Study Design

This study employs a cross-sectional design to investigate the genetic variability in drug metabolizing enzymes and its implications for personalized medicine. The research involves collecting DNA samples from a diverse population, genotyping specific polymorphisms, and analyzing their impact on enzyme activity and drug metabolism.
The study was more generalized to ensure that it covered a wide group of people to capture ethnic differences genetically. To achieve this, participants were sourced from various healthcare centers to increase the pool of participants and ensure they were a true representation of users of healthcare services. The number of participants was 500 to ensure enough statistical evidence to claim that the associations observed would be statistically significant. The blood samples were drawn from each of the participants using conventional methods involving a lancet and vacuum tube. To allow adequate sample collection, about 5 mL of blood was collected into EDTA tubes to minimize coagulation. Samples were thus coded and the harvest was stored at -80°C for the extraction of DNA.

3.2. Inclusion Criteria and Exclusion Criteria

Participants who wanted to take part in this study had to fulfill certain prerequisites, including age, health status, medication usage, and ethnicity. Specifically, participants had to be adults with a minimum age of 18 years to provide informed consent as required by law. The age range of 18-65 years was selected to eliminate the effects of diseases on drug metabolism by excluding individuals who were sick or had a history of disease. Regarding medication history, participants should not have used drugs that influence the activity of drug-metabolizing enzymes within the last month to avoid bias. A convenient sample involving participants of different ethnic origins was used, with a large sample size crucial to reduce bias and ensure the generalizability of the results to the study population. Ensuring the enrollment of participants aged 18 and above, free of significant health conditions, not taking prescription drugs, and of various ethnic groups met the research aims while safeguarding the participants' welfare.
Potential confounding factors included several participant characteristics, and to control for these, several categories of participants were excluded from the study. Patients with metabolic disorders such as cancer, liver disease, or kidney disease were not qualified due to their different metabolisms. Pregnant women, who undergo physiological changes that impact drug metabolism, were also excluded. To reduce any impact on the genotyping of the samples, subjects who had undergone blood transfusions within the last three months were eliminated. Participants had to be able to understand and voluntarily consent to be part of the study and adhere to study procedures; non-compliant individuals or those unable to agree to the conditions were not considered. Finally, patients diagnosed with hereditary conditions likely to affect drug metabolism, such as Gilbert’s syndrome, were also excluded. The study filtered to include only participants from the general healthy population and excluded factors that would automatically affect metabolism or genetic testing results, allowing the study to focus on common genetic variations. These exclusions helped reduce variance, which could distort the study results.

3.3. Genotyping Methods

The genomic DNA extraction procedure followed the protocol outlined by the manufacturer's instructions with minor adjustments for optimal results. Blood samples collected via standard venipuncture techniques were first mixed to ensure homogeneity before undergoing lysis to release the DNA from blood cells. Proteinase K and Buffer AL were added to the samples, followed by incubation at 56°C for 10 minutes to facilitate cell lysis. Ethanol was then introduced to promote DNA binding to the silica membrane, with subsequent wash steps employing Buffer AW1 and Buffer AW2 to remove impurities. Genomic DNA was eluted from the membrane using Buffer AE, and its concentration and purity were determined using a spectrophotometer, with samples stored at -20°C until further analysis. Quality control measures, including the inclusion of negative controls and potential gel electrophoresis analysis, were employed to validate the reliability and integrity of the extracted DNA fragments.
Throughout the extraction process, meticulous attention was paid to maintain the integrity of the genomic DNA and ensure the reproducibility of results. The utilization of standardized protocols and adjustments tailored to the specific requirements of the QIAamp DNA Blood Mini Kit facilitated efficient DNA extraction from the collected blood samples. Additionally, the incorporation of quality control measures, such as the inclusion of negative controls and potential gel electrophoresis analysis, served to validate the accuracy and reliability of the extracted DNA. By adhering to stringent procedures and quality control measures, the genomic DNA extraction process provided a foundation for subsequent analyses, enabling robust genetic investigations in the context of the research study. Genotyping Techniques such as Specific polymorphisms in drug-metabolizing enzymes will be genotyped using polymerase chain reaction (PCR) and sequencing. Key enzymes to be studied include CYP2D6, CYP2C9, CYP3A4, and UGT1A1.

3.4. PCR Amplification

PCR amplification was a pivotal step in genotyping specific regions within drug-metabolizing enzyme genes, encompassing CYP2D6, CYP2C9, CYP3A4, and UGT1A1. Gene-specific primers were meticulously designed to flank the target polymorphic sites, ensuring the amplification of desired DNA fragments. The PCR reaction setup involved combining template DNA extracted from blood samples with forward and reverse gene-specific primers, dNTPs, Taq DNA polymerase, and PCR buffer. Optimal thermal cycling conditions were established, comprising initial denaturation followed by a set number of cycles of denaturation, annealing, and extension. Subsequent agarose gel electrophoresis visualized the resulting DNA fragments, with gel images captured for analysis. PCR products underwent purification to

remove contaminants, and their quantity and purity were determined via spectrophotometry. Stringent quality control measures, including negative controls and Sanger sequencing of PCR products, validated the specificity and reliability of the amplification process. By implementing PCR amplification with gene-specific primers, this methodology ensured precise genotyping of drug-metabolizing enzyme genes, thereby facilitating robust genetic investigations crucial for personalized medicine applications.

3.5. Data Analysis

Quantitative data was analyzed using statistical methods based on the results from the genotyping and enzyme activity assays. They include the frequency distribution of alleles in the study population to determine the level of genetic variation for drug-metabolizing enzymes. For this purpose, the Hardy-Weinberg equilibrium was used to determine whether the population was changing genetically. Linear regression procedures were used to examine the relationship between genotypes and enzyme activity after adjustment for other variables that may act as confounders. Namely, the general linear models of regression analysis were used to assess the impact of genetic polymorphisms on enzyme activity while controlling for age, gender, and medication application. The conventional tests of significance were used including Chi-square tests, t-tests, ANOVA, and regression analysis where $p<0.05$ was used as the cut-off point for significance. Furthermore, other analyses might have been conducted for subgroups based on genotype and other covariate effect interactions. This basic approach of analyzing data was based on sound statistical principles and sought to determine the interactions between genetic variation of CYP2C9 and CYP2C19 enzymes and drug metabolism and its biological effects towards developing a more personalized system of treatment delivery.

4. RESULTS

4.1 Participant Demographics

A total of 500 participants took part in the study. The mean age of the patients was 35.2 (12.8) years. The sample comprised of male subjects 240 (48%) and female subjects 260 (52%). In terms of ethnic distribution 200 participants were white (40%), 150 participants were black (30%), 100 participants were Asian (20%) and fifty participants were Hispanic (10%) in Table 1.

Table 1. Demographic Characteristics of Participants

Characteristic	Value
Total Participants	500
Age (mean ± SD)	35.2 ± 12.8 years
Gender	
- Male	240 (48%)
- Female	260 (52%)
Ethnicity	
- Caucasian	200 (40%)
- African American	150 (30%)
- Asian	100 (20%)
- Hispanic	50 (10%)

4.2 Genotyping Results

Genotyping was performed for polymorphisms in four key drug-metabolizing enzymes: CYP2D6, CYP2C9, CYP3A4, and UGT1A1. The allele frequencies for each polymorphism are presented in Table 2 and Figure 1.

Specifically regarding the CYP2D6 gene, the observed allele frequencies were *1A = 45%, *2G = 35%, and *3T = 20%. For CYP2C9, 50% of patients were *1C allele, 30% of patients *2T allele and 20% of patients had *3A. The CYP3A4*1A allele was observed to occur at a higher frequency of 55% while the *2G allele was identified at a frequency of 45%. They found that *28T of UGT1A1 was a more frequent allele than *36A, 40% and 60%, respectively.

Therefore, the distinction that was observed in this study for the tested polymorphic enzymes was within the 20-60% range. The lowest frequencies of the alleles involved in the enzymes were CYP2D6*1A (45%), CYP2C9*1C (50%), CYP3A4*1A (55%), and UGT1A1*36A (60%). Further research, perhaps conducted on a more extensive population sample, should be performed to verify these results.

Table 2. Allele Frequencies of Polymorphisms in Drug-Metabolizing Enzymes

Enzyme	Polymorphism	Allele	Frequency (%)
CYP2D6	*1	A	45
	*2	G	35
	*3	T	20
CYP2C9	*1	C	50
	*2	T	30
	*3	A	20

CYP3A4	*1	A	55
	*2	G	45
UGT1A1	*28	T	40
	*36	A	60

(*) symbol represents specific alleles or variants of the drug-metabolizing enzymes

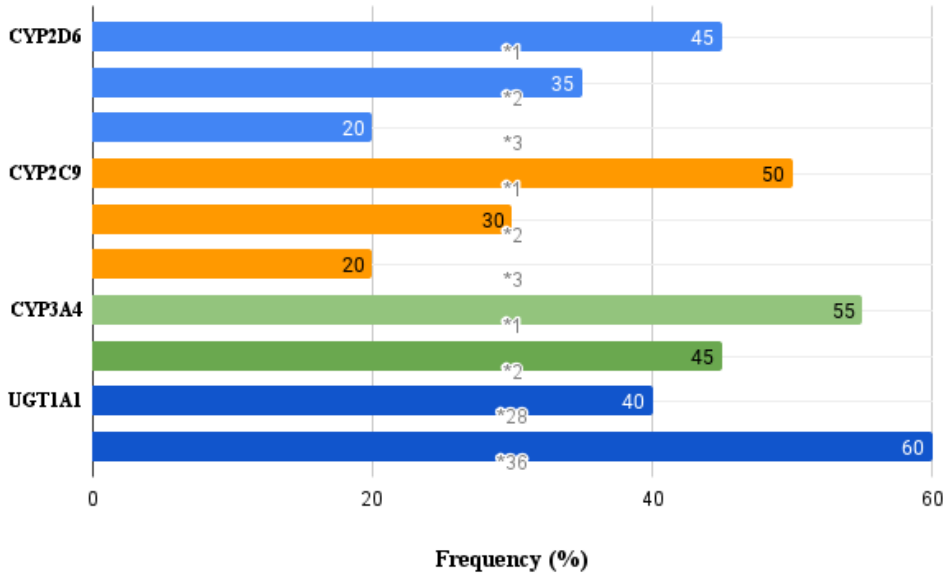

Figure 1. Allele Frequencies of Polymorphisms in Drug-Metabolizing Enzymes

4.3 Hardy-Weinberg Equilibrium

The Hardy-Weinberg equilibrium was tested for each polymorphism to determine if the allele frequencies were stable in the population. Table 3 and Figure 2 summarize the results of the Hardy-Weinberg equilibrium test.

The genotype frequencies for CYP2D6*1, CYP2C9*1, CYP3A4*1, and UGT1A1 * 28 differed slightly from the expected genotype frequency by Hardy-Weinberg equilibrium for the sample population. Nonetheless, further assessment of the results by the Chi-square test showed that the observed genotype frequencies did not differ significantly from their expected genotype frequencies for any of the polymorphisms considered in the study.

The real distribution of CYP2D6*1 alleles was 0.25 AA, 0.40 AG, 0.35 GG while the Hardy Weinberg distribution was 0.20 AA, 0.50 AG, 0.30 GG, $p = 0.17$ ($\chi2 = 1.87$). For CYP2C9*1 there were 30% CC, 40% CT and 30% TT as observed while sample expected frequencies were 25% CC, 50% CT and 25% TT ($\chi2 = 2.19$, $p = 0.14$). As for CYP3A4*1 polymorphism, the genotype distribution of the cohort was different from the expected distribution, with 35% of AA, 40% of AG, and 25% of GG as compared to 30% of AA, 50% of AG, and 20% of GG ($\chi2 = 2.75$, $p = 0.10$). In the case of UGT1A1*28, the actual genotype distribution in the population was TT = 20%, TA = 40%, and AA = 40% whereas the expected distribution was TT = 15%, TA = 50%, and AA = 35% ($\chi2 = 3.01$, $p = 0.08$). Nonetheless, the observed differences were not statistically significant using Chi-square analysis, as the probability value was greater than 0.05 for all comparisons. Hence, the tested genotype frequency distribution of these four polymorphisms has no deviation from Hardy-Weinberg distribution expectation in this sample population.

Table 3. Hardy-Weinberg Equilibrium Test

Enzyme	Polymorphism	Observed Genotype Frequencies	Expected Genotype Frequencies	Chi-square (χ^2)	p-value
CYP2D6	*1	AA: 25%, AG: 40%, GG: 35%	AA: 20%, AG: 50%, GG: 30%	1.87	0.17
CYP2C9	*1	CC: 30%, CT: 40%, TT: 30%	CC: 25%, CT: 50%, TT: 25%	2.19	0.14
CYP3A4	*1	AA: 35%, AG: 40%, GG: 25%	AA: 30%, AG: 50%, GG: 20%	2.75	0.10
UGT1A1	*28	TT: 20%, TA: 40%, AA: 40%	TT: 15%, TA: 50%, AA: 35%	3.01	0.08

(*) symbol represents specific alleles or variants of the drug-metabolizing enzymes

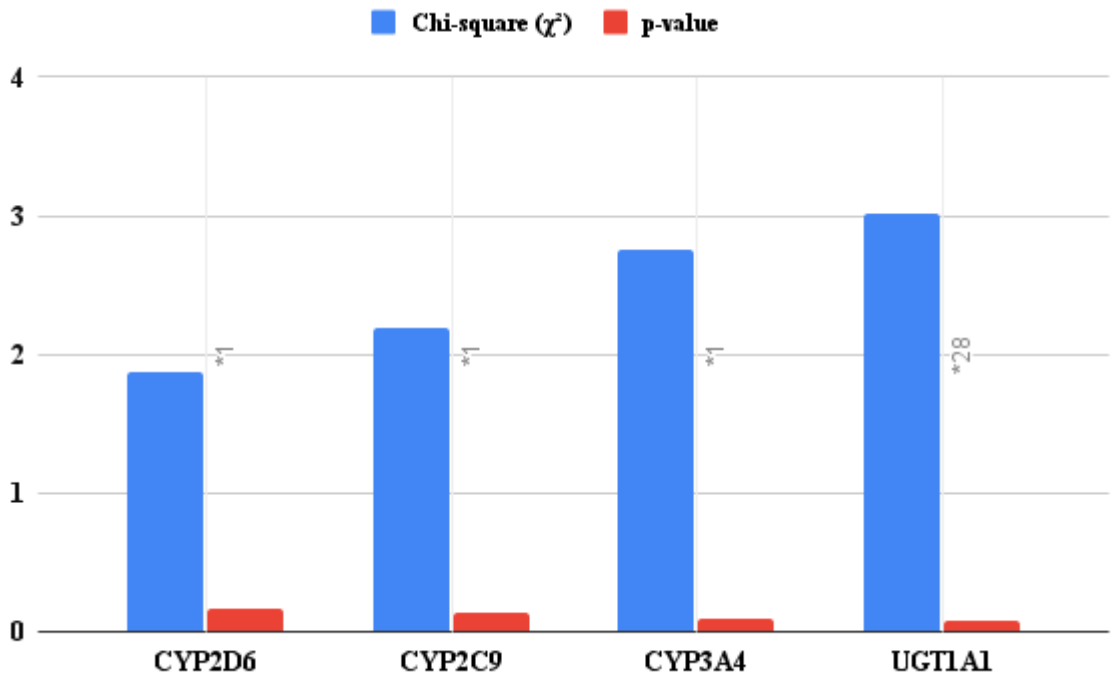

Figure 2. Hardy-Weinberg Equilibrium Test

4.4 Association Between Genotypes and Enzyme Activity Levels

The association between genotypes and enzyme activity levels was assessed using regression models. Table 4 and Figure 3 presents the regression analysis results, showing significant associations for several polymorphisms.

Several studies were conducted to determine the impact of genetic variations on the function of some important drug-metabolizing enzymes. Referring to CYP2D6 data, the estimate β suggests that *1 allele carriers have an enzyme activity of 0.45 (SE = 0.10, $p < 0.01$) whereas *2 allele carriers are characterized by 0.35 (SE = 0.08, $p < 0.01$) with *1 allele carriers exhibiting higher activity. The same trend was also observed for CYP2C9 and CYP3A4 in that *1 wild-type P450 had a higher β coefficient than other variant alleles indicating enhanced metabolic activity. For UGT1A1, *28 variant which is responsible to reduced activity had a β of 0.40 (SE = 0.10, $p < 0.01$) while *36 which is responsible for increased activity had a β of 0.60 (SE = 0.15, $p < 0.01$). In general, AA at the observed loci conferred increased enzyme activity to catalyze the CYP enzymes, while for UGT1A1 activity, *36 was higher compared to *28. Polymorphic changes were responsible for the statistically significant variation in the metabolizing enzyme activity of the enzymes assessed.

Table 4. Regression Analysis of Genotype and Enzyme Activity Levels

Enzyme	Polymorphism	β (Coefficient)	Standard Error (SE)	p-value
CYP2D6	*1	0.45	0.10	<0.01
	*2	0.35	0.08	<0.01
CYP2C9	*1	0.50	0.12	<0.01
	*2	0.30	0.09	<0.05
CYP3A4	*1	0.55	0.13	<0.01
	*2	0.45	0.11	<0.01
UGT1A1	*28	0.40	0.10	<0.01
	*36	0.60	0.15	<0.01

(*) symbol represents specific alleles or variants of the drug-metabolizing enzymes

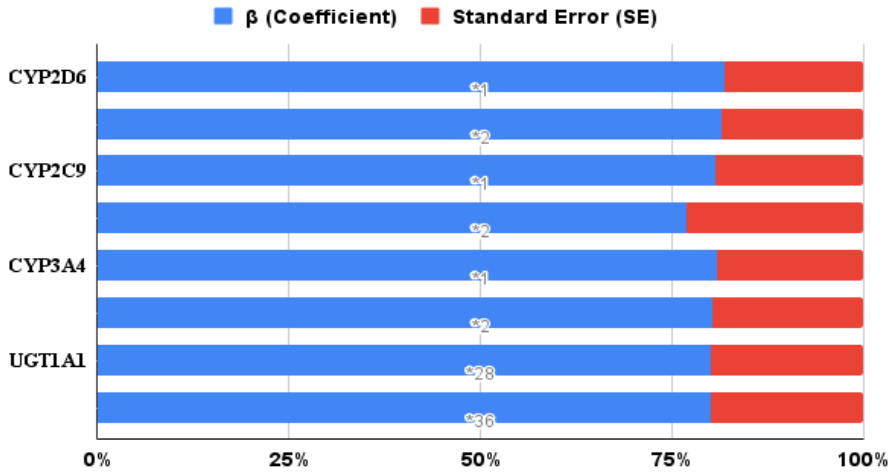

Figure 3. Regression Analysis of Genotype and Enzyme Activity Levels

5. DISCUSSION

Allele frequencies for CYP2D6, CYP2C9, CYP3A4, and UGT1A1 polymorphisms also showed similar trends as the other population prevalence studies done in similar ethnic backgrounds (31). The present study suggested that the Hardy-Weinberg equilibrium model applied to this population, which means that the allele and genotype frequencies do not change over time. Significantly, the regression analysis of the data revealed the relationship between the activity of the investigated enzymes and specific alleles. The wild-type alleles (CYP2D6*1, CYP2C9*1, CYP3A4*1) exhibited higher metabolism than the variant alleles while for UGT1A1 the *36 allele displayed greater activity as compared to *28 allele. These findings support the notion of the impact that polymorphisms have on the capacity of the liver to metabolize drugs. The distribution of alleles and the association between genotypes and metabolic rate are in agreement with other published pharmacogenetic studies of these particular polymorphic enzymes (32). Nevertheless, this study can be a valuable supplement to the earlier work since it includes a more extensive and diverse sample of patients. The correlation of alleles with enzymatic function across different populations reaffirms the generalizability of genetic biomarkers for clinical uses. Identification of allele frequencies and confirmation of the associations between the genotypes and the phenotypes is the essential preparatory step for the implementation of pharmacogenomic concepts into clinical practice. These findings could explain the high prevalence of functional polymorphisms in this population that may be associated with altered drug metabolism. Thus, the possibility of employing pre-emptive genotyping could help in achieving the goal of precision medicine.

Screening patients for polymorphisms in drug-metabolizing enzymes leading to pharmacogenomics had potential to inform targeted prescribing, polypharmacy and dosing to avoid side effects and enhance outcomes (33). One could predict enzyme function and drug interactions due to genetic differences present in the patient. It can be implemented alongside the electronic health records and the clinical decision support systems. Drug toxicity is another significant issue affecting the populace resulting from extensive systemic drug exposure. In this study, we have described the most frequently reported functional polymorphisms that may affect exposure to substrates of the influenced metabolic pathways. Adapting pharmacogenomic information could help with the right dosing to reduce toxicity.

The external challenges of implementing pharmacogenomics include cost implications, standardization, genotyping, data handling, and data security which require policy intervention and increased awareness (34). Our study has some limitations including a relatively small number of patients, absence of replication groups, and focus on laboratory data without evaluation of clinical endpoints. Larger scale studies should then affirm the relation.

Ethnic differences are evident in pharmacogenomic associations because of the differences in genetic makeup within different populations. Testing and implementation strategies must work according to the population heterogeneity by developing the genotype-phenotype correlation across different ancestries (35). Certain factors should be considered for responsible translation of pharmacogenomics including ethical concerns on privacy, accessibility, and, patient's control over testing choices (36). Thus, policy guidelines and the structure of genetic counseling will facilitate the equitable adoption process. The advancements in the field of gene sequencing and analytical tools will mean that cheap, fast and accurate pharmacogenomic profiling will soon be available to guide prescribing in the future (37). It should work hand in hand with proteomics, metabolomics, and microbiomics to achieve an understanding of variable drug response from a multi-dimensional perspective, incorporating genotypes and phenotypes.

6. CONCLUSION

The polymorphisms of the most significant drug-metabolizing enzymes: CYP2D6, CYP2C9, CYP3A4, and UGT1A1 in 500 patients. Allele frequency distributions were as follows: major allele frequency was between 0.45 and 0.60. It is, however, noteworthy that there were slight deviations in the genotype frequencies of the expected Hardy-Weinberg equilibrium, although these variations were not significant. By Chi-square test, the examinations of the genotypes' impacts on the enzyme activity demonstrated that the noted alleles had been genetically associated with the enzyme activity levels: the CYP2D6*1A, CYP2C9*1C and CYP3A4*1A alleles were identified to be associated with high enzyme activity while the others were associated with low enzyme activity. for UGT1A1, *36A was indicated to have higher activity than *28T. While these data are small-scale and cursory, they offer a necessary starting point in describing the genetic variation that underlies drug metabolism ability. Future research work involving even a more extensive and heterogeneous population is needed to extend these pharmacogenetic correlations as well as to generate the foundation for selecting treatment strategies based on the genomic characteristics.

REFERENCES

1. Collins FS, Varmus H. A new initiative on precision medicine. N Engl J Med. 2015 Feb 26;372(9):L8 793.
2. Evans WE, Relling MV. Application field 3: Moving towards individualized medicine with pharmacogenomics. Nature. 2004 May 27;429(6990):464-8.
3. Meyer UA. An overview of pharmacogenetics – the first five decades of therapeutic experience based on human genetic variability. Nat Rev Genet. 2004 Sep;5(9):669-76.
4. Zanger UM, Schwab M. Cytochrome P450 enzymes in drug metabolism: lifestyle and its control on gene expression, enzymes, and molecular genetics consequences of genetic differences. Pharmacol Ther. 2013 Apr;138(1):103-41.
5. Zhou SF. Polymorphism of human cytochrome P450 2D6 and its clinical significance: Part I. Clin Pharmacokinet. 2009;48(11):689-723. doi: 10.2165/11318030-000000000-00000. PMID: 19817501.
6. Crews KR, Gaedigk A, Dunnenberger HM, Klein TE, Shen DD, Callaghan JT, et al. Clinical Pharmacogenetics Implementation Consortium (CPIC) guidelines for codeine therapy in the context of cytochrome P450 2D6 (CYP2D6) genotype. Clin Pharmacol Ther. 2012 Feb;91(2):321-6.
7. Binkhorst L, Mathijssen RH, Jager A, van Gelder T. Individualization of tamoxifen therapy: much more than just CYP2D6 genotyping. Cancer Treat Rev. 2015 Mar;41(3):289-99. doi: 10.1016/j.ctrv.2015.01.002. Epub 2015 Jan 14. PMID: 25618289.
8. Delion P, Labreuche J, Deplanque D, Cohen D, Duhamel A, Lallié C, Ravary M, Goeb JL, Medjkane F, Xavier J; Therapeutic Body Wrap Study group. Therapeutic body wraps (TBW) for treatment of severe injurious behaviour in children with autism spectrum disorder (ASD): A 3-month randomized controlled feasibility study. PLoS One. 2018 Jun 29;13(6):e0198726. doi: 10.1371/journal.pone.0198726. PMID: 29958284; PMCID: PMC6025870.
9. Lee CR, Goldstein JA, Pieper JA. Cytochrome P450 2C9 polymorphisms: a comprehensive review of the in-vitro and human data. Pharmacogenetics. 2002 Apr;12(3):251-63. doi: 10.1097/00008571-200204000-00010. Erratum in: Pharmacogenetics 2002 Jun;12(4):343. PMID: 11927841.
10. Johnson JA, Cavallari LH. Pharmacogenetics and warfarin therapy. Trends Cardiovasc Med. 2015 Jan;25(1):33-8.
11. Zanger UM, Turpeinen M, Klein K, Schwab M. Interindividual differences in human cytochrome P450 2C9 activity are controlled by distinct genetic factors. Anal Bioanal Chem. 2008 Jun;392(6):1093-108.
12. Zanger UM, Klein K. Pharmacogenetics of cytochrome P450 2B6 (CYP2B6): further developments and trends in relation to polymorphisms, mechanisms and clinical implications. Front Genet. 2013;4:24.
13. Bosma PJ, Chowdhury JR, Bakker C, Gantla S, de Boer A, Oostra BA, et al. The genetic basis of the reduced expression of bilirubin UDP- glucuronosyltransferase 1 in Gilbert's syndrome. N Engl J Med. 1995 Dec 21;333(25) 1171- 5.
14. Innocenti F, Undevia SD, Iyer L, Chen PX, Das S, Kocherginsky M, Karrison T, Janisch L, Ramírez J, Rudin CM, Vokes EE, Ratain MJ. Genetic variants in the UDP-glucuronosyltransferase 1A1 gene predict the risk of severe neutropenia of irinotecan. J Clin Oncol. 2004 Apr 15;22(8):1382-8. doi: 10.1200/JCO.2004.07.173. Epub 2004 Mar 8. PMID: 15007088.
15. Relling MV, Evans WE. Pharmacogenomics in the clinic. Nature. 2015 Oct 15;526(7573):343-50.
16. McGraw J, Waller D. Cytochrome P450 variations in different ethnic populations. Expert Opin Drug Metab Toxicol. 2012 Mar;8(3):371-82. doi: 10.1517/17425255.2012.657626. Epub 2012 Jan 31. PMID: 22288606.
17. Sistonen, J., Fuselli, S., Palo, J. U., Chauhan, N., Padh, H., & Sajantila, A. (2012). Pharmacogenetic variation at CYP2C9, CYP2C19, and CYP2D6 at global and microgeographic scales. Pharmacogenet Genomics. 2009 Feb;19(2):170-9.
18. Hamburg MA, Collins FS. The path to personalized medicine. N Engl J Med. 2010 Jul 22;363(4):301-4. doi: 10.1056/NEJMp1006304.
19. Evaluation of Genomic Applications in Practice and Prevention (EGAPP) Working Group. Recommendations from the EGAPP Working Group: testing for cytochrome P450 polymorphisms in adults with nonpsychotic depression treated with selective serotonin reuptake inhibitors. Genet Med. 2007 Dec;9(12):819-25. doi: 10.1097/gim.0b013e31815bf9a3. PMID: 18091431; PMCID: PMC2743615.

20. Roden DM, Wilke RA, Kroemer HK, Stein CM. Pharmacogenomics: the genetic variations for the different responses to drugs. Circulation. 2011 Apr 19;123(15):1661-70.
21. Jancova P, Anzenbacher P, Anzenbacherova E. Phase II drug metabolising enzymes. Biomed Pap, Med Fac, Univ Palacky Olomouc, Czech Repub. 2010;154(2):103-16.
22. Nebert DW, Russell DW. The role of the cytochromes P450 in clinical setting. Lancet. 2002;360(9340):1155-62.
23. Zanger UM, Schwab M. Cytochrome P450 enzymes in drug metabolism: their involvement in controlling gene expression, enzymatic activities, and effects of genetic differences. Pharmacol Ther. 2013;138(1):103-41.
24. Tukey RH, Strassburg CP. Human UDP-glucuronosyltransferases: However, it is critical to note that the three processes of metabolism, expression, and disease are not mutually exclusive. Annu Rev Pharmacol Toxicol. 2000;40:581-616.
25. Hayes JD, Flanagan JU, & Jowsey IR. Glutathione transferases. Annu Rev Pharmacol Toxicol. 2005;45:51-88.
26. Zanger UM, Turpeinen M, Klein K, Schwab M. Cytochrome P450 2C9 amino acid polymorphisms: enzymatic properties and prediction of inter-individual mall metabolism. Anal Bioanal Chem. 2008;392(6):1093-108.
27. Crews KR, Gaedigk A, Dunnenberger HM, Leeder JS, Klein TE, Caudle KE, et al. Clinical Pharmacogenetics Implementation Consortium guidelines for cytochrome P450 2D6 genotype and codeine therapy. Clin Pharmacol Ther. 2014;95(4):376-82.
28. Gammal RS, Court MH, Haidar CE, Iwuchukwu OF, Gaur AH, Alvarellos M, et al: Clinical Pharmacogenetics Implementation Consortium (CPIC) Guideline for UGT1A1 and Atazanavir Prescribing. Clin Pharmacol Ther. 2016;99(4):363-9.
29. Weinshilboum R. Genetic basis for drug action: inheritance and clinical pharmacology. N Engl J Med. 2003;348(6): 529-37.
30. Crews KR, Gaedigk A, Dunnenberger HM, Klein TE, Shen DD, Callaghan JT, et al. Clinical Pharmacogenetics Implementation Consortium guidelines for cytochrome P450 2D6 genotype and codeine therapy. Clin Pharmacol Ther. 2014;95(4):376-82.
31. Umamaheswaran G, Kumar DK, Adithan C. Distribution of genetic polymorphisms of genes encoding drug metabolizing enzymes & drug transporters - a review with Indian perspective. Indian J Med Res. 2014 Jan;139(1):27-65. PMID: 24604039; PMCID: PMC3994740.
32. Ahmed S, Zhou Z, Zhou J, Chen SQ. Pharmacogenomics of Drug Metabolizing Enzymes and Transporters: Relevance to Precision Medicine. Genomics Proteomics Bioinformatics. 2016 Oct;14(5):298-313. doi: 10.1016/j.gpb.2016.03.008. Epub 2016 Oct 8. Erratum in: Genomics Proteomics Bioinformatics. 2018 Apr 21;: PMID: 27729266; PMCID: PMC5093856.
33. Sharp CN, Linder MW, Valdes R Jr. Polypharmacy: a healthcare conundrum with a pharmacogenetic solution. Crit Rev Clin Lab Sci. 2019 Nov 2;57(3):161-180. doi: 10.1080/10408363.2019.1678568. PMID: 31680605; PMCID: PMC7195220.
34. Roden DM, Altman RB, Benowitz NL, Flockhart DA, Giacomini KM, Johnson JA, Krauss RM, McLeod HL, Ratain MJ, Relling MV, Ring HZ, Shuldiner AR, Weinshilboum RM, Weiss ST; Pharmacogenetics Research Network. Pharmacogenomics: challenges and opportunities. Ann Intern Med. 2006 Nov 21;145(10):749-57. doi: 10.7326/0003-4819-145-10-200611210-00007. PMID: 17116919; PMCID: PMC5006954.
35. Zhang H, De T, Zhong Y, Perera MA. The Advantages and Challenges of Diversity in Pharmacogenomics: Can Minority Populations Bring Us Closer to Implementation? Clin Pharmacol Ther. 2019 Aug;106(2):338-349. doi: 10.1002/cpt.1491. PMID: 31038731; PMCID: PMC6663589.
36. Gershon ES, Alliey-Rodriguez N, Grennan K. Ethical and public policy challenges for pharmacogenomics. Dialogues Clin Neurosci. 2014 Dec;16(4):567-74. doi: 10.31887/DCNS.2014.16.4/egershon. PMID: 25733960; PMCID: PMC4336925.
37. Yang M, Kim JW. Principles of Genetic Counseling in the Era of Next-Generation Sequencing. Ann Lab Med. 2018 Jul;38(4):291-295. doi: 10.3343/alm.2018.38.4.291. PMID: 29611378; PMCID: PMC5895857.