The Pharmacogenetics of Cytochrome P-450 and its Effect on Drug Metabolism

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ABSTRACT

Cytochrome P-450 (CYP-450) enzyme plays an essential role in the oxidation of most drugs, and thus it can affect the toxicity and efficacy of many medications. Factors that influence the function and presence of cytochrome have a key impact on the outcomes of therapy. More specifically, characteristics of cytochrome pharmacogenetics and procedures of cytochrome enzymes induction and inhibition can greatly influence the rate of drug biotransformation and the rate of elimination. So, an understanding of genetic variants which are associated with drug responses and illnesses could improve and enhance the outcome of treatment. Clinical data in genetic tests may be useful in order to develop methods for assessing genetic risks. Positively, genetic tests for monogenic sicknesses have already proven to be a useful test and any changes may trigger a new field of personalized drug. This review will look at the influence of pharmacogenetics in drug metabolism and the importance of using personal genetic data in achieving optimal therapy and in preventing any possible adverse effects.

Keywords: Pharmacogenetics, Biotransformation, CYP-450

1. INTRODUCTION

Pharmacogenetics or genetic variability is the study of how people respond to drug therapy. In humans, genes affect hair and eye color, gender, and also the likelihood of developing certain diseases. Genes can also amend the risk of developing different diseases. It is well known that African persons are more likely to develop sickle cell anemia than other races. Armenians, Turks, and Arabs are more able to tolerate Mediterranean fever than other ethnicities. More recently, research has been presented, showing that genes can allocate other traits of each individual, depending on the quantity of each enzyme formed in the liver, while the amount of those enzymes can help to predict how quickly a drug can be cleared from the body 1.

Present drugs face several challenges such as adverse drug effect, drug-drug interactions, and nonresponse to typical treatment. Adverse drug reactions (ADRs) could be a result of individual risk factors like drug interactions or body condition such as liver and renal function or alcohol consumption 2. However, genetic variability might also be a risk factor for adverse drug reactions 3. The variability in patient response to pharmacotherapy, in part, depends on recognised factors that are easily assessable, like age and gender. In addition to the previous factors, inherited variants in biotransformation enzymes might have a great impact on drug response. More specific studies identified particular genes which can determine how patients will respond to particular medicines. For instance, some genetic factors might define whether people will respond to codeine as a pain killer, or the method of response. The possible cause of genetic variation among individuals in drug response may be due to a single mutation of an allele. For example, a single nucleotide polymorphism (SNP) can influence the capacity of protein coding and can also affect the method of protein expression or regulation 4. However, more recently, researchers have become concerned about a varied range of structural variations in the human genome and the number of polymorphisms 5,6.

It is obvious that variability in genetic encoding drug biotransforming enzymes have a notable effect in the outcome of pharmacotherapy to a very wide range, and this is because of polymorphism of the CYP 450 enzymes which play a key role in this deference. Positively, genetic tests for monogenic sicknesses have been proven to be useful 7, thus understanding of CYP enzyme genetic variations may open a new field of personalized drugs.
The variability of populations can be categorised into three major phenotypes. These classes are ultra-rapid metabolizers (UM), extensive metabolizers (EM), and poor metabolizers (PM) 5.

The objective of this review is to review the effect of genetic polymorphisms of cytochrome P450 in drug biotransformation and the importance of using of personal genetic data in achieving optimal therapy, and in preventing any possible adverse effect.

1.1. Role of cytochrome enzymes in the oxidative biotransformation of drugs

Phase I hepatic oxidation, which represents the main method of elimination of most drugs, is mainly catalysed by the cytochrome enzymes (CYPs). The CYP-450 enzymes in family I and in family II are responsible for nearly 75% of phase I drug biotransformation 8. Cytochromes are members of a complicated multi-genie family. Every cytochrome enzyme is encoded by a specific gene and the amino acid sequence of the encoded polypeptide can specify the substrate for CYP [9]. Some oxidation metabolisms are catalysed by multiple cytochromes but other biotransformation reactions could be attributed to the activities of individual enzymes. Therefore, the clearance of certain drugs might be useful indicators for specific activity of cytochrome P-450 enzymes. According to Nelson et al 1996, approximately 40% of cytochrome enzymes are grouped within the same family at the amino acid sequence level, and about fifty five percent correlated to members of a subfamily. For instance, cytochrome P2D6 is a humanoid enzyme which belongs to subfamily D within family II 8.

1.2. The Pharmacogenetics of CYP-450 and drug-drug interactions

During last years, the field of cytochrome P450 pharmacogenetics has been grown. Most of cytochrome P-450 enzymes and their difference have been recognized. There is a notable variation in cytochrome P-450 enzymes’ expression among individuals. Individuals which are poor metabolizers, have inactive allele substrates for the encoded CYPs, and they eliminate drugs at a very slow rate 10. While the CYP alleles encode enzymes for extensive metabolizers (EM), these are completely functional and thus can eliminate medications at a normal rate. Furthermore, ultra-rapid metabolizers (UM) can eliminate substrates at a very rapid rate because they have high level enzyme expression and multiple copies of the gene 5. The differences in cytochrome P-450 allelic variation are important factors which cause variation in drug response and safety among individuals as well as causing variation among different ethnic populations. For example, the Asian race usually shows a lower prevalence of poor metabolizer and UM than in Caucasian people 11. Although there is a lower prevalence of the active cytochrome P2D6 *1 allele in Asian people, the encoded enzymes are more active in drug oxidation than the cytochrome P2D6*10 genes 12, 13. These problems may be associated with psychopharmacological factors. Fortunately, modern microarray-based systems for CYP2D6 genotyping have been created and might be accepted for the personalizing of therapy with a number of particular drugs 14.

Cytochrome enzymes accommodate many drug substrates, and consequently the possibility for inhibition interactions is high. In addition, the ingredients of cigarette smoke and certain environmental chemicals could also activate cytochrome gene transcription in human cells.

The inhibitory drug interactions caused by cytochrome enzymes may possibly encompass irreversible or reversible inhibitions. In the case of irreversible interactions, new enzymes must be synthesized to recover the activity of cytochrome but, in the case of reversible interactions, the cytochrome function is quickly re-established after the inhibitory agent is cleared by hepatic metabolism 4. For example, most patients with schizophrenia are treated with combinations of psychoactive agents for long periods. Therefore, the potential risks of adverse events from polypharmacy are highly significant within those patients. The primary inducible cytochrome enzymes which have been shown to have a role in the biotransformation of most medications are CYP1A2, CYP2C9, CYP2C19 and CYP3A4 15.

2. DISCUSSION

2.1. Effect of cytochrome polymorphism on biotransformation of anti-platelet drug clopidogrel

Clopidogrel inhibits platelet aggregation. It needs to be metabolised into an active metabolite by cytochrome enzymes in order to give the desirable antipatelet effect. Many studies in clinical trials showed that clopidogrel is effectively able to prevent thrombotic events in patients with atherosclerosis and cardiovascular disease 16, 17. The pharmacological response to clopidogrel is widely diverse from person to person. For instance, around a quarter of patients treated with standard clopidogrel doses do not respond properly and show low in-vivo inhibition of ADP-induced platelet aggregation 17. This poor response to clopidogrel causes an elevated risk of recurrent ischemic events 18. The mechanism explaining clopidogrel resistance is still unclear.

Clopidogrel is given as an in-active prodrug and its activation is controlled by numerous CYP-450 enzymes in the liver and involving CYP2C19. Variants in cytochrome P2C19 may lead to a reduction in clopidogrel efficacy due to loss of enzymatic function 7, 18. There were two clinical relevant studies which assessed therapeutic outcomes of clopidogrel. The first trial was conducted by Mega et al for optimizing platelet inhibition with Prasugrel-thrombolysis in myocardial infarction (MI) patients (TRITON-TIMI38). In this study, individuals who are carrying the reduced function CYP2C19 allele were treated with clopidogrel. Post clopidogrel treatment, those patients had considerably low levels of the active metabolite of clopidogrel and they were at a higher rate of thrombosis (major cardiovascular adverse events) compared with non-carriers 6. The second study is a French study. In this trial, patients who had two loss of function alleles and who were undergoing percutaneous coronary intervention for acute MI had a three-fold higher risk of MI, stroke, and even death 15. Furthermore, another
genetic sub-study of platelet inhibition and patient outcomes (PLATO) confirmed that any individual patients who were carrying loss of function variant of CYP2C19 belong to a higher rate of vascular event 30. As a result and response to these results, clopidogrel is warned with a black box by the Food and Drug Administration (FDA). However, there is a debate about this result. Two studies investigated the ability of clopidogrel to reduce cardiovascular events compared with a placebo combined with aspirin, and found no difference 21.

2.2 Effect of cytochrome enzymes on biotransformation of antipsychotic drug Clozapine

Clozapine is an important anti-psychotic medication because it can work properly with many subjects who are insensitive to typical antipsychotic drugs. However, its agranulocytosis and cardiotoxicity limit its use and the occurrence of toxicity normally lead to the end of remedy 22. The metabolism of clozapine yields a range of toxic effects in human body cells and organs. Peroxidase enzymes oxidises clozapine to the active metabolite (nitrenium), which has been linked to toxic mechanism 4. Although classic anti-psychotic drugs are less toxic, they do not show the same ability as clozapine to treat psychosis. For individuals suffering from drug-induced neutropenia, it is still recommended to continue treatment with clozapine 23.

Clozapine undergoes oxidation metabolism to yield different metabolites. The major metabolites are N-demethyl- (norclozapine) and N-oxide metabolites and the minor metabolites are mono- and dihydroxylated products 24. Norclozapine is the pharmacologically active compound.

![Genomic organization of the human CYP1A2 gene](image)

**Figure 1:** Genomic organization of the human CYP1A2 gene shows the seven exons and locations of SNPs which give rise to allelic variants 4.

On the other hand, evidence for the involvement of other cytochrome p-450 enzymes in the clozapine oxidation have been proven by in-vitro studies. Cytochrome P2C19 and CYP3A4 have been identified to catalyse N-demethylation of clozapine, CYP3A4 and the monooxygenase reacts to mediate clozapine N-oxidation 31. Some in-vitro experiments with microsomal fractions and cDNA expressed cytochrome enzymes have also presented CYP2D6 to have a role in the oxidation of clozapine 4. Moreover, there are also some occasional reports of adverse events in schizophrenic individuals treated with clozapine and risperidone at the same time 35, and these studies have stated that the plasma concentrations of clozapine were increased by co-administration of risperidone 35. This has been explained in terms of cytochrome P2D6 interactions, although the value of evidence is in contradiction of a significant role for this enzyme in clozapine metabolism. 36. A possible cause is that patients, who are suffering from a deficiency of CYP1A2 might use cytochrome P2D6 for clozapine clearance and such individual patients may be more susceptible to interactions from co-administered drugs rather than others 4.

2.3 Effect of cytochrome polymorphous on biotransformation of anticoagulant drug Warfarin

Warfarin is the most widely used oral anticoagulant agent in the world. In America, about thirty million patients were treated with this medication in 2004 37. The determination of the correct dose of warfarin is challenging because it relies on the variety of factor X among subjects, and therefore the consequences of taking the wrong dose could be lethal.

Studies have found that demographic variables, clinical factors, and variation of CYP450 in two genes, family II, polypeptide 9 (CYP2C9) and vitamin K epoxide reductase complex, subunit 1 (VKORC1) are the main contributors to the variability of response to warfarin dosed among individual patients 38. Genetic polymorphism between individual patients plays a vital role in determining the initial dose of warfarin that should be given when oral anti-
coagulation medication is started. However, there are no practical methods that have been evaluated to use genetic information in a varied and large population. In 2007, the FDA added pharmacogenetic information to the warfarin product but did not recommend a particular technique for using genetic information to establish the required dose for each patient. A trial performed by (WPC 2009) found that, by using the validation cohort, the pharmacogenetic algorithm can accurately identify the required dose of warfarin. In that study, the pharmacogenetic algorithm successfully identified larger proportions of individuals who required less than 22 mg of warfarin per week and of those who required more than 49 mg per week to obtain the desired international normalized ratio (INR). Moreover, that study concludes that the use of a pharmacogenetic algorithm for assessing the suitable initial dose of warfarin can produce recommendations which are significantly closer to the required therapeutic dose than those derived from a fixed-dose approach 39.

2.4 Effect of Cytochrome polymorphisms on biotransformation of cholesterol-lowering medication statins

In 1998, the percentage of statin drugs dispensed in the United States pharmacies increased from about fifty three percent in 1991 to approximately eighty six percent 40. More recently, data from the United States indicate that statin consumption rose from 12.5 million patients in 2000 to twenty four million in 2003 41. In 2005, atorvastatin and simvastatin were described as the first and second most prescribed medications worldwide, respectively 42. Long-term clinical trials conducted by Verre et al 2003 including statins highlighted that there is a significant reduction in the potential risk of major coronary events by more than thirty percent in primary prevention and by nearly twenty six percent in secondary prevention experiments 43. Furthermore, similar studies have shown similar results with respect to the prevention of cardiovascular disease (CVD) 44. However, there are serious ADRs associated with statin therapy. The most commonly reported statin-induced adverse drug reactions are elevations in liver enzymes and elevations in plasma creatine kinase (CK) levels 45. A study involving eighteen randomized trials showed about 1017 adverse reactions in patients with statins compared with about eight hundred in the placebo, indicating that statin therapy can significantly increase the risk of any adverse event by nearly 40% 46.

Statins are either delivered in inactive form (prodrug) such as simvastatin and lovastatin or in the active form, as with atorvastatin and rosuvastatin. The inactive forms of statins are activated to the corresponding active forms in the intestine via non-specific hydrolysis by carboxysterase enzymes. The primary pathway of statin biotransformation is attributed to microsomal cytochrome P-450 enzymes 44. It has been verified that the cytochrome P3A4 is the major pathway of statin drug biotransformation 47 and the acidic metabolites which are produced by cytochrome P450 metabolism are responsible for the cholesterol-lowering effect 44. CYP2D6 and CYP2C9 are also encompassed in the biotransformation of the statins but at a lower level than CYP3A4 47.

Polymorphisms affecting particular CYP-450 enzymes can affect the kinetic character of statin metabolites and resulting efficacy 48. The activity of the cytochrome P3A4 gene may vary between individuals, consequently affecting the activity and the efficacy of statin pharmacotherapy. A study conducted by Kajjami et al 2006 found that the expression of SNP variant (A-290G variant) in CYP3A4 can cause an increase in low density lipoprotein (LDL) level with 12.4% compared with the wild type allele. On the other hand, in the same study, another SNP variant (M445T variant) was found to be associated with a low level of LDL before and after atorvastatin treatment, enhancing the efficacy of atorvastatin treatment among 11.2–17.6% of patients against non-carriers 49. Thus, these variants may either result in a fall in the metabolism of atorvastatin, or enhanced atorvastatin efficacy.

CONCLUSION

It is clear that drug response and adverse reactions can be affected by polymorphic nature of the cytochrome P-450 to a great extent. The understanding of the mechanisms by which genetic factors and co-administered medicines can affect the therapeutic efficacy and safety of any medications can help achieve optimal therapy with lower ADRs. In order to optimize drug therapy, new pharmacogenetic approaches and clinical phenotyping methods in individual patients need to be considered. It is also crucial to consider the feasible influence of cytochrome P-450 genotype and phenotype as factors that can affect the prevalence of pharmacokinetic interactions in diverse ethnic groups. It is expected in the following years to involve individual genetic data in patient management and social implications 50, 51, and, such genetic testing could be allocated straight to the patient 7.

The application of pharmacogenetics in medicine to individualized drug treatment appears to be clinically and economically valuable. So, genetic variation that can influence the pharmacokinetic aspects of any medications requires further research.

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