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PSYCHIATRY AND PRECLINICAL PSYCHIATRIC STUDIES - REVIEW ARTICLE

# Application of pharmacogenetics in clinical practice: problems and solutions

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**Abstract**
This paper discusses difficulties of pharmacogenomic data integration into clinical practice. It emphasizes the need for developing simple and easy to use bioinformatics tools to help prescribers to rapidly access and use genetic data in clinical decision-making at the point of encounter.

**Keywords** Clinical informatics tools · Medication side effect estimation · Drug–drug interactions · Drug–gene interactions

## Introduction

How would you individualize medication therapy? You have a new patient whom you have diagnosed with a major depressive disorder. There are over 20 antidepressants available on the market, and you would like medical evidence-based guidance to help you decide which one to prescribe. One approach is often referred to as personalized or precision medicine. Its early uses can be traced back to 1900, when Austrian Karl Landsteiner identified three blood groups, and to 1907 when Reuben Ottenberg performed the first successful blood transfusion at Mount Sinai Hospital in New York based on Landsteiner's findings (Landsteiner 1900). In 1956, the genetic basis for the selective toxicity of fava beans ("favism") and primaquine was discovered to be a deficiency in glucose-6-phospho dehydrogenase or G6PD (Alving et al. 1956). In 1977, cytochrome P450 (CYP) enzyme CYP2D6 was identified as responsible for increased duration and intensity of an anti-hypertensive drug debrisoquine effect, marking the beginning of the modern era of genetics-based treatments (Mahgoub et al. 1977).

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Advances in pharmacogenomics have greatly increased our understanding of the relationship between genetic variations, and drug effectiveness or their side effects. Pharmacogenetic testing (PGx) to identify genetic mutations that predict patient responses to pharmacotherapy are emerging as a science-based method to select the optimal treatment regimen for individual patients. PGx-based testing services offering analysis of a patient's DNA have recently become commercially available. Since the adverse drug reaction-related mortality ranks the 5th in the overall US mortality statistics (Lazarou et al. 1998), the attractiveness of PGx in informing medication prescribing should be difficult to contest. Nevertheless, PGx implementation into clinical practice has been slow, and acceptance by prescribers has been wavering (Patel et al. 2014; Hess et al. 2015; Peterson et al. 2016). The question that inevitably arises is why? To address this question let's review the basic concepts of PGx.

## Overview

Recall from basic pharmacology that the relationship between a drug dose and its effectiveness can be separated into pharmacokinetic (dose → concentration) and pharmacodynamic (concentration → physiological function, pathological process, etc.) components (Katzung et al. 2009). Tissue proteins ultimately influence a patient's response to a drug; since proteins are encoded by genes and genes vary from person to person, drug responses will also vary. Genetic variants associated with the pharmacodynamic component are usually directly associated with a drug target. For example, carriers of a G

allele in the opiate mu-receptor 1 (*OPMR1*) in a single nucleotide polymorphism (referred to as SNP) known as 118A>G (rs1799971) have a greater sensitivity to pain and require two to four times more of an analgesic drug to achieve a comparable degree of analgesia as non-carriers (Ren et al. 2015).

Pharmacokinetic processes govern absorption, distribution, and elimination of drugs; the genetic aspects of the latter process being most intensely studied. Drug metabolism by cytochrome P450 (CYP) mixed function oxidase system accounts for the elimination of 60–80% of all commercial drugs. Because of a high prevalence of SNPs, deletions, duplications, and splicing defects in CYP genes, effectiveness of CYP enzymes varies greatly between individuals and ethnic groups. A computational analysis of genetic and functional differences of the 57 CYP450 genes performed using the Human Genome Diversity Project and HapMap (http://www.1000genomes.org/) data that included approximately 1694 individuals belonging to 62 human populations discovered a total of 449 SNPs distributed across the 57 CYP450 genes that could affect individual responses to drugs [Polimanti et al. 2012]. For example, *CYP2C19* (Cytochrome P450 Family 2 Subfamily C Member 19) gene is responsible for the metabolism of antidepressants such as citalopram, clomipramine and amitriptyline, proton pump inhibitors and antiplatelet drug clopidogrel (Mrazec 2010). A SNP rs4244285 responsible for a nucleotide change from G to A creates an aberrant splice site resulting in reduced CYP2C19 enzyme function found in carriers of altered alleles (e.g. *CYP2C19 *2*, see de Morais et al. 1994). A loss of function by one or both alleles results in a partially or completely metabolically inactive enzyme and slows the elimination of drugs predominantly metabolized by CYP2C19 (e.g. sertraline). Alternatively, the presence of *CYP2C19 *17* allele results in an accelerated drug metabolism and a potential loss of drug effectiveness.

Another example of genetic variation effects on drug pharmacokinetics is prodrugs such as codeine. Codeine is a week analgesic until it is metabolized by CYP2D6 enzyme yielding morphine, a potent analgesic. Mutations in *CYP2D6* gene impairing CYP2D6 enzyme-mediated metabolic activation of codeine result in low concentration of morphine and, consequently, poor analgesia, leaving the patient in pain even at high doses of codeine (Brousseau et al. 2007). On the other hand, duplication of *CYP2D6* gene can result in a rapid increase in morphine concentration and life-threatening toxicity due to accelerated metabolic activation of codeine.

## Metabolizer types

Most PGx services report test results in terms of extensive, intermediate, poor or ultrarapid metabolizer phenotypes (EM, IM, PM and UM). These terms refer to metabolic phenotypes defined by allelic combinations. The *extensive metabolizer* or EM phenotype usually consists of two functional alleles each encoding a fully functional CYP enzyme. The *intermediate metabolizer* or IM phenotype arises from one functional and one non-functional allele, and in most cases show normal or slightly reduced CYP enzyme activity. *Poor metabolizers* or PMs are individuals who have two non-functional alleles. If prescribed a medication that is metabolized by the affected enzyme, PMs will likely have a higher than expected drug plasma concentration often resulting in medication side effects but will show a lack of efficacy with prodrugs (e.g. codeine or tramadol). *Ultrarapid metabolizers* or UMs possess either a duplication of a CYP gene or carry a gene variant that increases the rate of drug metabolism by the encoded enzyme. If such an individual is taking a medication that is metabolized by that enzyme, they may have a reduced drug concentration in the plasma, which could make the drug ineffective at regular doses. Toxic effects can occur with prodrugs such as codeine (Gammal et al. 2016) UMs are more common in some populations than in others: for instance, nearly 30% of individuals in Saudi Arabia or Ethiopia are CYP2D6 UMs but only 1–6% of Europeans and about 1% of East Asians possess this phenotype (Teh and Bertilsson 2012; McGraw and Waller 2012).

Taken together, four phenotypes have been identified in association with CYP genetic polymorphisms that can impact drug metabolic rates and, consequently, have an impact on adverse drug reactions (ADRs) as well as drug efficacy. The US Food and Drug Administration (FDA) has published a "Table of Pharmacogenomic Biomarkers in Drug Labeling" that includes *CYP2D6* and *CYP2C19* genotype-specific dosing recommendations for several popular psychotropic drugs when these drugs are being prescribed to PMs and UMs (Baskys 2017).

## CYPs and drug–drug interactions

In addition to genetic polymorphisms, there are other factors influencing the expression and functions of CYPs. They include physiological states or processes such as age, sex, hormones, pregnancy, environment as well as pathological states such as cancer, inflammation and cholestasis. Epigenetic mechanisms such as DNA methylation and histone modifications also regulate CYP gene expression. These factors together with drug–drug interactions (DDIs) contribute to the clinical manifestations of ADRs that arise as a consequence of CYP-catalyzed reactions. CYP induction or inhibition is another major mechanism that underlies DDIs (Samer et al. 2013; Manikandan and Nagini 2018). For a detailed analysis of the molecular mechanisms underlying the induction or inhibition of specific CYPs, as well as epigenetic regulation of CYP expression, the reader is referred

to several excellent reviews on the topic (e.g. He et al. 2015; Manikandan and Nagini 2018). From a prescriber perspective, CYP enzyme induction and inhibition should be always considered in conjunction with the pharmacogenetic data. This can be illustrated by a phenomenon of *phenoconversion*, which occurs when a person with an IM phenotype is prescribed a medication inhibiting the altered CYP enzyme and becomes a poor metabolizer as a result. Clinically, this may present as a sudden appearance of side effects associated with a drug that a patient has been taking for some time without problems. Significance of phenoconversion is reflected in regulatory agency (such as FDA) warnings and in manufacturers' package inserts. Thus, the FDA warns that fluoxetine (other examples are quinidine, paroxetine) inhibits the activity of CYP2D6 and may make individuals with a typical CYP2D6 metabolic activity resemble poor metabolizers. Prevalence of variant alleles responsible for the IM phenotype can be disproportionally high in some geographic areas and low in others. For example, 50–70% of East Asians are CYP2C19 IMs (Teh and Bertilsson 2012; McGraw and Waller 2012) and could be prone to developing ADRs to CYP2C19 substrate drugs (e.g. voriconazole) when a CYP2C19 inhibitor (e.g. fluvoxamine) is added to their regimen.

## Ordering and interpretation of PGx test results

Knowledge of a person's CYP polymorphisms is essential for the development of personalized drug therapy. The polymorphisms of CYP genes can be identified by examining a person's phenotype or genotype. Phenotyping involves administration of a probe drug (e.g. debrisoquine, dextrametorphan) followed by a subsequent measurement of drug/metabolite concentration ratio in blood, urine or saliva. The advantage of CYP phenotyping is that it offers clinically relevant information reflected by genetic, environmental, and endogenous factors (Manikandan and Nagini 2018). Genotyping provides an accurate individual DNA sequence and information on loss-of-function or gain-of-function mutations coding for specific enzymes involved in metabolism of pharmacological agents. There are numerous methods of CYP genotyping ranging from PCR to high-throughput next generation sequencing (NGS). Comparison of these methods is beyond the scope of this review and interested readers are directed to several recently published excellent reviews that cover this topic (e.g. Samer et al. 2013; Manikandan and Nagini 2018). One important disadvantage of genotyping (which is the foundation of PGx) is the inability to measure the influence of the environmental factors on the CYP enzymes.

When should one order a pharmacogenetic test? The answer is rather simple: if it has not yet been done before, always order PGx when starting a new medication. There are numerous reasons for this. Thus, health, patient safety, financial and consumer satisfaction benefits of PGx have already been well documented in adult and pediatric populations (Snyder et al. 2014; Ferreri et al. 2014; Plumpton et al. 2016; Gammal et al. 2016; Aka et al. 2017; Moretti et al. 2018), and there are ongoing projects focusing on the implementation of this technology into clinical practice (Blagec et al. 2018). The Ubiquitous-PGx or U-PGx project is an effort of seven European countries to integrate PGx into routine clinical care, which is often hampered by insufficient or fragmented infrastructures. This project aims to set up and implement a unique multimodal, multilingual clinical decision support intervention system consisting of digital-, paper-, and mobile-based tools deployed across implementation sites in the participating countries (Blagec et al. 2018). Further, unlike traditional laboratory tests, PGx test results will remain valid for life and can be applied not only to drugs that the patient is currently taking but also to those that may be taken in the distant future. While it may be argued that *preemptive* genetic tests should be done only in certain circumstances, so far it has not been possible to *apriori* define such circumstances (Hinderer et al. 2017). Moreover, recent studies (Cheung et al. 2016; Ji et al. 2016) found that on average there are at least 1–2 actionable variant CYP genes per general psychiatry clinic patient (Fig. 1). These findings suggest that without the knowledge of genetic variants impacting drug metabolism there is a significant likelihood that undesirable drug–gene interactions will be encountered by most prescribers in most patients. Finally, compliance with drug labeling that clearly spell out dose adjustment requirements based on genotype along with fully informing the patient of all available options are undeniably prudent approaches.

One significant difficulty associated with PGx is a relatively large number of data points for a prescriber to consider. This brings us to the question posed at the beginning of this article: which antidepressant to prescribe? While PGx-guided selection of an antidepressant is a science-based answer to this question, it is not a simple answer. Given a large number of potential genotypes [449 SNPs distributed across the 57 CYP450 genes that could affect individual responses to drugs (Polimanti et al. 2012)], at least four phenotypes and drug actions on CYP enzymes (substrates, inducers and inhibitors), and their coding genes, the task of PGx-guided prescribing may be difficult if not impossible for a human mind. While most PGx companies do provide detailed genotyping reports, reading these reports could be unacceptably time consuming. For example, Ferreri et al. (2014) found that a clinical pharmacist on average spends 76.6 min with a

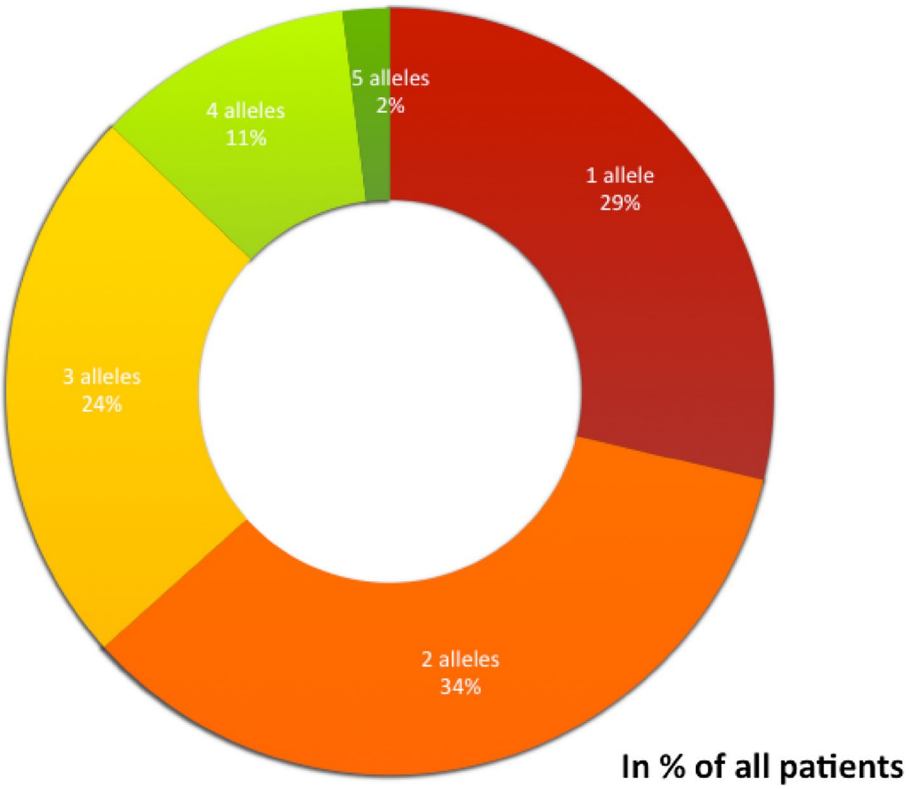


**Fig. 1** Frequencies of actionable CYP gene variants in a general psychiatry clinic patient population. More than 50% of the study samples were found to have at least one actionable CYP gene allele in six pharmacogenes (*CYP2B6, CYP2C9, CYP2D6, CYP2C19, CYP3A4 and CYP3A5*). On average, there were 2.22 variant alleles within the six pharmacogenes per subject (Adapted from Cheung et al. 2016 with permission)

patient communicating PGx test results. Further, static PGx reports typically do not account for the phenoconversion phenomenon. To address these problems and facilitate PGx acceptance by prescribers, user-friendly bioinformatics tools are urgently needed. There are several user-friendly static databases that allow to explore drug–drug or drug–genotype interactions. Examples include EuroPharmagenics drug–gene–disease interaction database by EuroEspes SA, (http://www.europharmagenics.com), Stanford University managed PharmGKB database (Whirl-Carrillo et al. 2012) and SuperCYP, a comprehensive database on Cytochrome P450 enzymes that includes a tool for analysis of CYP–drug interactions (Preissner et al. 2012). An innovative interactive tool that allows drug–gene and drug–drug interaction visualization and the Dynamic Medication Selection™ algorithm is available for download from http://www.medpicker.com.

In summary, strengths of PGx and other molecular techniques are in their accuracy and a potential to improve outcomes, patient safety and experiences. Their drawbacks are large quantities of data that cannot be swiftly integrated into the clinical decision process without resorting to bioinformatics tools. The successful implementation of PGx in clinical practice depends on the development of user-friendly bioinformatics tools that can process and present genetic and drug interaction data to clinicians at the point of service.

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