Drug–Gene Interactions: Inherent Variability In Drug Maintenance Dose Requirements

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INTRODUCTION

The ultimate goal of drug therapy is for a given patient to achieve an acceptable therapeutic outcome, such as seizure control or eradication of infection, while minimizing or avoiding adverse drug reactions. However, many patients do not adequately respond to a given drug therapy; efficacy rates of common medications are less than optimal.[1] Certainly, the relationship between drug concentration and effect—pharmacodynamics (PD)—is under genetic influence, as drug receptors are the products of genes that exhibit polymorphisms.[2,3] The influence of genetics on pharmacokinetics (PK) can introduce variability among individuals that may be the cause of treatment failure and/or toxicity. As PK describes the drug concentration over time in an individual and as the pharmacokinetic parameters of clearance (CL), volume of distribution, and half-life are used in drug regimen design, the genetically introduced variability in PK, specifically CL, can influence the drug maintenance dosing regimen. This variability related to clearance and the influence on maintenance dose design will be discussed in some detail below. With advances in technology, testing of DNA to help predict drug response is becoming more tenable. Pharmacogenetics, the relationship between a gene and drug response, requires understanding in the context of dosing regimen design, where a drug–gene interaction can confound drug therapy.

Depending on the specific pathophysiology and confirmed diagnosis, a patient receiving a particular drug may require an initial loading dose followed by a maintenance dose administered at a certain dosing interval.[4] Thus, a "dosing regimen" may include three components—loading dose, maintenance dose, and dosing interval—that are inherently influenced by the patient's genetics, not considering pathophysiology, environment, and other factors. The number of drugs requiring a loading dose is small, and while genetic variability can influence drug transporters in the volume of distribution and loading dose, it is the genetic influence on drug-metabolizing enzymes and drug transporters relative to drug clearance and maintenance dose that is of primary interest for most drugs. This paper seeks to align concepts of maintenance dose design with the inherent PK variability among individuals introduced by genetics. Here, we review a conceptual framework of drug–gene interactions' effects on the maintenance dose and dosing interval and present examples of such interactions. A number of resources provide great breadth and depth on this subject, including The Pharmacogenomics Knowledgebase (PharmGKB, www.pharmgkb.org) and genetic-based dosing guidelines from the Clinical Pharmacogenetic Implementation Consortium (CPIC, www.pharmgkb.org/page/cpic) and the Dutch Pharmacogenetics Working Group (DPWG, www.pharmgkb.org/page/dpwg), which can also be accessed via the PharmGKB website.

BACKGROUND AND GENERAL CONCEPTS

An individual's genetic constitution resides in the deoxyribonucleic acid (DNA) sequence that makes up the 23 pairs of chromosomes in each nucleated cell of the body. One set of chromosomes is provided by the individual's mother and the other is provided by the father. When considering all of the chromosomes, there are approximately three billion DNA base pairs (from each parent), a combination of adenine (A), cytosine (C), guanine (G), and thymine (T) that make up an individual's genome.[5] Among other information, chromosomes contain regions of bases that code for the production of proteins. These regions of genes are of interest in the production of drug receptors, drug-metabolizing enzymes, and drug transporters, all proteins potentially related to drug response.[5] Genetic variation within an individual exists based on the DNA received from each parent. The most common form of a genetic variation is known as the single nucleotide polymorphism (SNP), pronounced "SNIP." Here, one base, such as C, replaces another base, such as G, noted as G>C. To be specific, the locus of the SNP is noted, such as c.681G>C. This signifies that at position 681 in the gene-coding region (c.681), C replaced G.[5] For a defined SNP, the National Center for Biotechnology Information dbSNP database has assigned a reference SNP (rs) number that is unique and consistent related to the specific single nucleotide change. When considering a specific locus on DNA, an individual will have a base from each parent, and thus a given "genotype." In the example above, one parent may have a C at c.681, whereas the other parent may have a G at c.681, with the patient having a genotype of CG. The different bases result in an alternate form of the same gene, something referred to as an allele. Typically, but not always, the most common allele is referred to as the "wild type."

When referring to the cytochrome P450 (CYP) enzyme system and other but not all enzyme and transporter systems, it is now conventional to employ the "star" nomenclature. Typically the *1 form represents the wild type, although this is not always the case.[5] For instance, the *N-acetyl-transferase 2*

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wild-type allele is named *4. Naming of alleles subsequent to the *1 form is usually in numerical order: *2, *3, *4, and so on. The variant forms of a gene may result in the production of proteins that may have normal function, reduced function, or loss of function. Other gene variations can result in overall increased enzyme activity, such as an individual with multiple (more than two) copies of a given gene. When considering the star nomenclature, genotypes are presented as combinations of alleles, such as *1/*1, *1/*2, *2/*2, *2/*3, and so on. It must be noted that the star nomenclature is not consistent from CYP gene to CYP gene. For instance, *CYP2C19*2* refers to an allele that results in a loss-of-function enzyme, whereas *CYP2C9*2* refers to an allele that produces a decreased-function enzyme and *CYP2D6*2* refers to an allele that produces a normal-function enzyme.

The genotypes related to specific proteins represent the individual's genetic makeup, whereas the phenotype represents how an individual expresses a genetic trait, such as enzyme function. Phenotypes related to drug metabolism include the extensive (normal) metabolizer (EM, NM), who typically carries two wild-type or normal-function alleles; the intermediate metabolizer (IM), who may have a normal-function allele and a decreased-function or loss-of-function allele; and the poor metabolizer (PM), who typically carries two decreased-function or loss-of-function alleles or a combination of the two. Also, there are cases where an individual may be considered an ultrarapid metabolizer (UM) in that he or she has increased metabolic activity compared to the EM.[5] Specific examples are presented below.

MAINTENANCE DOSE, CLEARANCE, AND GENETIC VARIATION

From a PK point of view, the maintenance dose of a drug is meant to replace the fraction of the drug dose that was eliminated from the body during the previous dosing interval.[4] Clearance represents a measure of drug elimination from the body and relates the drug concentration to the rate of drug elimination. In most cases, the maintenance dose is dependent on the CL, being proportionally related. In terms of drug metabolism, numerous enzymes and enzyme systems are involved in the CL of therapeutic agents. The genetic influence on drug metabolism can be profound, resulting in a patient having excessive exposure to a given drug, experiencing increased potential for adverse effects, or failing therapy because of subtherapeutic drug concentrations. Table 1 presents the influence of genetics on the activity of various drug-metabolizing enzymes for CL and maintenance dose, identifying a number of drug–gene interactions. In PK, a drug–gene interaction occurs when an individual carrying one or more variant forms of a gene that codes for a drug-metabolizing enzyme or drug transporter with altered function receives a drug that is a substrate for the given enzyme or transporter. Here, the response to the drug varies based on the individual's genetics.

The *CYP450* Gene Family

Cytochrome P450 (CYP450) proteins constitute the products of a large multigene family. The *CYP450* genes code for the CYP450 enzymes that are responsible for metabolizing a substantial number of drugs.[6,7] The catalytic reactions facilitated by CYP450 enzymes include aliphatic oxidation, aromatic hydroxylation, N-hydroxylation, N-dealkylation, and O-dealkylation. Fifty-seven active *CYP450* genes were identified in the human genome through the work of the Human Genome Project.[8] Of these, nine CYP450 enzymes, including CYP2D6, CYP2A6, CYP2B6, CYP3A4, CYP1A2, CYP2C9, CYP2C19, CYP1B1, and CYP3A5, are responsible for metabolizing approximately 1,400 drugs.[6] Of great interest are the *CYP450* genes for which polymorphisms exist that have potential impact on drug dosing and response to drug therapy. While there are many *CYP450* genes of interest when it comes to drug metabolism, currently *CYP2C9*, *CYP2C19*, and *CYP2D6,* with 39, 29, and 114 SNPs, respectively, have been evaluated to determine specific dosing guidelines.[9]

Drug–*CYP2C9* Interactions

Cytochrome P450 2C9 (CYP2C9) metabolizes drugs across therapeutic categories, including angiotensin II inhibitors and anti-inflammatory, anticoagulant, and antiepileptic agents, among others.[10] In these therapeutic categories, the Food and Drug Administration (FDA) "Drug Development and Drug Interactions: Table of Substrates, Inhibitors, and Inducers" lists the anti-inflammatory drug celecoxib as a "sensitive substrate" and identifies the CYP2C9 substrates phenytoin and warfarin as those with a narrow therapeutic range.[10] There are at least 15 *CYP2C9* variant alleles that code for enzymes exhibiting decreased (12) or absent (three) metabolic function.[11] These variant alleles, which are less abundant, are also called "minor alleles;" they are seen in a lower percentage of individuals in a given population compared with the most abundant gene form. The *2 and *3 forms are the most common minor alleles, producing decreased-function CYP2C9 enzymes found in 13% and 7% of white individuals, respectively. The *2 allele has not been identified in Asians, while the *3 allele is seen in 4% of this population. In the black population, the *2 and *3 alleles are present in 3% and 2% of individuals across various geographic populations, respectively.[12] Table 1 presents four examples of the influence of *CYP2C9* variation on the CL of substrate drugs, indicating specific drug–gene interactions.

Irbesartan

Irbesartan is an angiotensin II antagonist indicated for the treatment of hypertension. The major elimination (CL) pathways involve N-glucuronidation and oxidation via CYP2C9.[13] The CL of irbesartan is affected by the individual's *CYP2C9* genotype. As an example, individuals with a *CYP2C9*1/*3* genotype had an irbesartan CL of 12.99 ± 3.12 L/hr compared with 21.40 ± 5.98 L/hr for wild-type *CYP2C9*1/*1* individuals.[14] Here, an individual carrying the variant *3 form of the *CYP2C9* gene has decreased CL due to the drug–gene interaction. The genotype differences resulting in decreased CL impact the response to irbesartan, as *1/*2 and *1/*3 individuals experience a greater response to the drug compared with *1/*1 individuals, in this case exhibited as a reduction in blood pressure.[15] Additionally, the *CYP2C9*1/*2* genotype is associated with a statistically significant increased frequency of excessive blood-pressure reduction, noted as an adverse event. The decreased CL results in greater drug exposure, leading to a greater-than-desired decrease in blood pressure.

Table 1 Examples of the Influence of Variant CYP450 Alleles on the Clearance and Maintenance Dose of Specific Drugs

Gene	Genotype (Variant Allele[s] or Phenotype)	Drug Substrate	Percent Change in Clearance	Anticipated Effect on Required Maintenance Dose
CYP2C9[a]	*1/(*3[b]); IM	Irbesartan	−39.3%	Decrease[14]
	*1/(*3); IM	Celecoxib	−30.0%	Decrease[18]
	(*3/*3); PM		−70.0%	Decrease[18]
	*1/(*3); IM	Phenytoin	−33.0%[d,e]	Decrease[21]
	*1/(*3); IM		−42.0%[d,e]	Decrease[22]
	*1/(*2[c]); IM		−51.9%[d,e]	Decrease[23]
	*1/(*3); IM		−56.8%[d,e]	Decrease[23]
	(*2/*2); PM		−83.4%[d,e]	Decrease[23]
	(*2/*3); PM		−88.6%[d,e]	Decrease[23]
	*1/(*2); IM	Warfarin[f]	−42.4%	Decrease[28]
	*1/(*3); IM		−47.7%	Decrease[28]
	(*2/*2); PM		−67.7%	Decrease[28]
	(*2/*3); PM		−76.5%	Decrease[28]
	(*3/*3); PM		−90.8%	Decrease[28]
CYP2C19[g]	*1/(*2[h]); IM	Omeprazole	−37.3%	Decrease[35]
	(*2/*2); PM		−76.4%	Decrease[35]
	(*17[i]/*17); UM		+27.1%	Increase[36]
	*1/(*2 or *3[j]); IM	Clopidogrel	−40.1%[k]	Decrease[40]
	(*2/(*2 or *3)); PM		−66.0%[k]	Decrease[40]
CYP2D6[l]	IM	Metoprolol	−32.6%	Decrease[48]
	PM		−83.0%	Decrease[48]
	UM		+160.3%	Increase[48]
	IM	Tramadol	−31.2%	Decrease[52]
	PM		−82.7%	Decrease[52]

[a] Genotype and relative phenotypes
[b] rs1057910 (decreased function)
[c] rs1799853 (decreased function)
[d] Phenytoin V_{max} (maximal rate of metabolism)
[e] Reported in some cases as CL, although phenytoin exhibits nonlinear pharmacokinetics necessitating the use of V_{max} in analysis and dosing regimen design
[f] S-warfarin
[g] Genotypes and phenotypes
[h] rs4244285 (loss of function)
[i] rs12248560 (gain of function)
[j] rs4986893 (loss of function)
[k] Derived from AUC values provided in omeprazole package insert[60]
[l] Phenotypes
EM = extensive metabolizer; IM = intermediate metabolizer; PM = poor metabolizer; UM = ultrarapid metabolizer

Due to the decreased CL, *CYP2C9*1/*2* and **1/*3* individuals would require a lower maintenance dose to elicit the desired pharmacological response. However, the broad clinical impact of the *CYP2C9*3* variant on irbesartan alteration in blood pressure is less clear, as studies have failed to show an effect on the therapeutic response.[16] It should be pointed out that in most cases, data relating genotype and drug response are limited because comparative trials, which are expensive, have not been undertaken.

Celecoxib

Celecoxib is a nonsteroidal anti-inflammatory drug that inhibits cyclooxygenase-2 (COX-2), leading to decreased prostaglandin synthesis that results in decreased inflammation. The drug also exhibits analgesic and antipyretic properties. The primary CL pathway for celecoxib includes hydroxylation, predominantly by CYP2C9, although CYP3A4 plays a lesser role.[17] The CL of celecoxib is statistically significantly lower in *CYP2C9*1/*3* and *CYP2C9*3/*3* individuals, with median values of 21 L/hr and 9 L/hr, respectively, as compared with *CYP2C9 *1/*1* individuals (30 L/hr).[18] It has been recognized that *CYP2C9*1/*3* and *CYP2C9*3/*3* individuals have greater exposure to the drug compared with *CYP2C9*1/*1* individuals, with area under the curve (AUC) values for the former being at least twofold higher than the AUC value for the wild-type (*1/*1) individuals.[17,18] There is a clear relationship between the *CYP2C9*1/*3* and *CYP2C9*3/*3* genotypes and exposure to celecoxib, indicating the drug–gene interaction

(celecoxib-*CYP2C9*3*; Table 1). It has been suggested that the *CYP2C9* genotype is related to adverse events with celecoxib. However, the relationship between genotype and the risk of adverse events, such as cardiovascular and/or gastrointestinal toxicity, has not been established because too few patients have been evaluated.[19] The celecoxib package insert includes information on *CYP2C9*, stating that a 50% reduction in dose or alternative therapy should be considered for the treatment of juvenile rheumatoid arthritis in individuals known or suspected of being a PM.[20]

Phenytoin

Phenytoin is an antiepileptic drug with multiple indications, including the treatment of generalized tonic-clonic seizures. While both CYP2C9 and CYP2C19 are involved in the metabolism of phenytoin, the CYP2C9 pathway appears to be the chief route of phenytoin metabolism.[21,22] As the phenytoin concentration changes disproportionately with the dose of phenytoin (i.e., nonlinear PK), and as the therapeutic range of phenytoin is narrow (10 to 20 mg/L), understanding the metabolic status of a given patient can be critical in designing a dosage regimen.[21] The decreased maximum metabolism rate (V_{max}) of phenytoin in individuals carrying the *CYP2C9*3* allele may be a major contributing factor to the nonlinear PK noted for this drug.[21] Compared with wild-type individuals, the phenytoin-*CYP2C9* interaction in individuals with one variant allele (*1/*2 or *1/*3) or heterozygous or homozygous for such alleles (e.g., *2/*3, *3/*3) results in lower phenytoin dosage requirements (maintenance dose) to achieve therapeutic concentrations. Daily doses of 314 mg, 193 mg, 202 mg, 217 mg, and 150 mg are required by *1/*1, *1/*2, *1/*3, *2/*2, and *2/*3 patients, respectively.[23] The phenytoin package labeling identifies individuals with certain *CYP2C9* genotypes as being at increased risk of unusually high phenytoin concentrations.[24] While there appears to be a relationship between *CYP2C9* variant alleles and phenytoin toxicity, especially concerning the *3 variant, the clinical data are conflicting. Further evaluation is needed to understand the most practical application of the pharmacogenetic impact to the phenytoin dosage regimen design.[25]

Table 2 Genotype-Guided Warfarin Maintenance Dosing Based on *CYP2C9* Genotype[a]

CYP2C9 Genotype[a]	Dosing (mg/day)
*1/*1	5–7
*1/*2	5–7
*1/*3	3–4
*2/*2	3–4
*2/*3	3–4
*3/*3	0.5–2

[a] For individuals with the common wild-type *VKORC1* genotype (GG), showing the influence of *CYP2C9* genotype only. Adapted from warfarin package insert.[37]

Warfarin

Perhaps no other CYP2C9 substrate drug has been evaluated more than warfarin relative to dosing requirements based on genotype (Tables 1 and 2).[26] This anticoagulant is a first-line drug with broad indications, including prevention and treatment of thromboembolic events associated with atrial fibrillation and venous thrombosis. Warfarin is also indicated after an initial myocardial infarction to decrease the risk of thromboembolism, subsequent myocardial infarction, and death. Warfarin is made available as a 50:50 racemic mixture of its R- and S-enantomeric isomers. The S-enantiomer of warfarin is approximately five times more potent than the R-enantiomer.[27] Multiple CYP450 isozymes are involved in the metabolism of warfarin, with CYP1A1, CYP1A2, CYP2C19, and CYP3A4 metabolizing the R-enantiomer to various hydroxywarfarin metabolites (i.e., 6-, 8-, and 10-hydroxy). The more active S-enantiomer is largely metabolized to 7-hydroxywarfarin by CYP2C9, and its CL is subject to *CYP2C9* polymorphisms.[27,28]

In the spring of 2010, the FDA amended the warfarin package labeling to expand the pharmacogenetic content by providing a dosing table based on *CYP2C9* and vitamin K epoxide reductase subunit 1 (*VKORC1*) genotypes. The pharmacological target of warfarin is VKORC1 and certain individuals have decreased production of this target enzyme, meaning they require lower doses to elicit the desired response. Prior to this addition, the labeling included only text stating that the genotype of *CYP2C9* and *VKORC1* could be of use in maintenance dose design.[29] Regardless of the FDA's changes to the package labeling, controversy exists regarding genotype-guided warfarin dosing. Some studies show a longer time in the therapeutic international normalized ratio (INR) range with genotyping, and other studies provide opposing results.[30,31]

A study has also shown that genotype-guided warfarin dosing resulted in a statistically significant decrease in hospitalizations of patients starting on the drug.[32] Additionally, with genotype-guided warfarin dosing there was a statistically significant reduction in hospitalizations resulting from thromboembolism or bleeding, which are serious consequences of therapeutic failure and warfarin toxicity, respectively.[32] However, the design of this study has been criticized, as it used historical controls and may have been influenced by selection bias.

The Clarification of Optimal Anticoagulation through Genetics (COAG) trial failed to show a benefit of genotyping for time in the therapeutic INR range for patients taking warfarin.[31] At the same time the COAG results were reported, results of the European Pharmacogenetics of Anticoagulant Therapy (EU-PACT) Warfarin Study were reported, showing that pharmacogenetic-guided warfarin therapy resulted in an increased percentage of time that a patient's INR was in the therapeutic range.[30] Clearly, further studies addressing the clinical utility of pharmacogenetics testing, including cost analyses, are needed to delineate its role in warfarin therapy.

Drug–*CYP2C19* Interactions

There are more than 30 known variant *CYP2C19* alleles. Of these, the *2, *3, and *17 forms are of primary interest, as they are present in higher percentages of individuals across populations than other confirmed alleles.[33] The loss-of-function *2 and *3 alleles are seen in 12% to 61% and 0.028% to 15% of individuals across geographic populations, respectively.[33] The *2 variant is the most common loss-of-function variant seen in American (12%), African (15%), and East Asian (29%) popula-

Table 3 Conceptual Examples Using *CYP2C19* and Relative Influence of Drug Clearance, Half-Life, and Dosing Interval[a]

CYP2C19 Genotype Description	Effect on Clearance	Effect on Maintenance Dose	Effect on Half-Life ($t_{1/2}$)	Effect on Dosing Interval
Heterozygous loss-of-function DME[b] (e.g., *1/*2)	↓	↓	↑	↑
Homozygous loss-of-function DME (e.g., *2/*2)	↓↓	↓↓	↑↑	↑↑
Heterozygous gain-of-function DME (e.g., *1/*17)	↑	↑	↓	↓
Homozygous gain-of-function DME (e.g., *17/*17)	↑↑	↑↑	↓↓	↓↓

[a] Partial list; not all scenarios are applied clinically
[b] DME = drug-metabolizing enzyme

tions. The *17 gain-of-function allele is present in geographical populations at a frequency range of 2.7% to 21%.[33] It is seen in 18%, 16%, and 21% of Americans, Africans, and Europeans, respectively.[33] The PK of CYP2C19 substrate drugs are affected by an individual's *CYP2C19* genotype, as variant alleles have been shown to alter drug clearance. Examples of genotypes related to phenotypes are provided in Table 1. Table 3 relates genotype to clearance, maintenance dose, half-life, and dosing interval; further phenotype information can be found in supplementary data provided by CPIC.[33]

Omeprazole

Omeprazole is a proton pump inhibitor used in the treatment of gastroesophageal reflux disease, as well as other ulcer-related and acid hypersecretion conditions.[34,35] Omeprazole is cleared (metabolized) to inactive metabolites by CYP2C19, including 5-hydroxyomeprazole.[35] The package labeling for omeprazole provides information in the dosing and administration sections, as well as in the "Warnings and Precautions" section, about *CYP2C19* PM. Additionally, the Royal Dutch Association for the Advancement of Pharmacy—Pharmacogenetics Working Group has issued recommendations for omeprazole dosing based on *CYP2C19* status.[34] Although variant alleles have been shown to decrease the CL of omeprazole, patients who are IMs or PMs, exhibiting one or two loss-of-function alleles, do not need dosage modifications, as they would only see enhanced effects of the drug. However, UMs (e.g., *CYP2C19*17/*17*) have an increased CL of the drug and require dosage adjustment for *H. pylori* eradication (an increase of 100% to 200%).[34,36] Considering an omeprazole dose increase of the same magnitude is also recommended when treating other indications.[34]

Clopidogrel

Clopidogrel, an inhibitor of platelet aggregation, is used among other indications to decrease the risk of adverse events following diagnosis of cardiovascular atherosclerotic disease, such as myocardial infarction. When considering the influence of genotype on the CL of clopidogrel, it must be kept in mind that clopidogrel is supplied as an inactive prodrug and CL (metabolism) of clopidogrel in part results in activation to a therapeutic compound. Clopidogrel is largely activated by CYP2C19 in a multistep process, although other CYP450 enzymes, such as CYP3A4/5, CYP1A2, and CYP2B6, are involved to a lesser extent.[38] The decreased activation of clopidogrel due to loss-of-function alleles has been related to adverse outcomes, especially in stent-placement patients following percutaneous coronary intervention (PCI).[39,40]

Although other studies have disputed the use of *CYP2C19* genotyping with clopidogrel when assessing its use for any indication, in-depth analysis of study data supports genotyping in the post-PCI patient population.[39] To further support the pharmacogenetic testing of PCI patients receiving antiplatelet therapy, consider that while platelet reactivity in heterozygotic patients could be restored to relatively sufficient levels in most patients through dose increases, homozygotic loss-of-function (*2/*2, PM) status could not be sufficiently overcome even with four times the standard maintenance dose of clopidogrel.[41] Additionally, a boxed warning related to the PM phenotype, regardless of diagnosis, was added to the clopidogrel package labeling in 2010.[42] When considering heterozygous (e.g., *1/*2) or homozygous (*2/*2) individuals with stent placements, it is clear that genotyping can benefit drug selection. In this instance, the relationship between *CYP2C19* polymorphisms and platelet function following clopidogrel use is clear, and careful consideration of *CYP2C19* functional status (phenotype) is warranted in specific clinical indications.[41] Clinical genotype-driven guidelines for clopidogrel use were first published in 2011 and updated in 2013.[9] The guidelines have been employed in various health care settings.[43-45]

Drug–*CYP2D6* Interactions

The CYP450 nomenclature website lists more than 100 *CYP2D6* alleles, making it the second most polymorphic of all the *CYP450* genes.[11] The most common *CYP2D6* variants in African, African-American, Caucasian, and East Asian populations are the *2 (normal activity), *17 (decreased activity), *2 (normal activity), and *10 (decreased activity) forms, respectively. In these populations, the alleles are present in approximately 20%, 18%, 27%, and 45% of individuals, respectively.[46] The many variant *CYP2D6* alleles have been defined in terms of the metabolic activity of their protein products (the CYP2D6 enzyme), with nonfunctional variants (e.g., *3, *4, *5, others) having an "activity score" of 0, reduced- or decreased-function variants (e.g., *9, *10, *17, others) having a score of

0.5, and the wild-type (*1) and fully functional forms (e.g., *2, *27, others) being assigned an activity score of 1. Relative to expected metabolic phenotypes, individuals with a *CYP2D6* genotype with an activity score of 0 (e.g., *3/*4) are considered PMs. Individuals with a genotype with an activity score of 0.5 (e.g., *3/*17) are considered IMs. A genotype with an activity score of 1 to 2 (e.g., *2/*17) defines an individual as an EM, and individuals with a genotype with an activity score of greater than 2, as seen in individuals with multiple copies of fully functional forms, are considered UMs.[46,47] The relationship between *CYP2D6* genotype and phenotype is presented in detail by CPIC.[46] Relating the *CYP2D6* phenotype to drug CL and the maintenance dose, a PM would be expected to have the lowest CL, thus requiring a relatively low maintenance dose compared to an EM. An UM would be expected to have a higher CL compared with an EM, who would require a relatively higher maintenance dose (Table 1). Therefore, the drug–gene interactions between CYP2D6 substrate drugs and the variant alleles can result in a broad inherent difference among individuals for maintenance dose requirements.

Metoprolol

The $beta_1$-adrenergic blocking drug metoprolol has long been used for the treatment of hypertension, angina, and post-myocardial infarction patients to reduce the risk of cardiovascular death. Metoprolol, as described in the package labeling and elsewhere, is cleared from the body mainly by metabolism via CYP2D6; in excess of 90% of a dose is metabolized by EMs.[48,49] Clearly, the metabolism and therefore CL of metoprolol is affected by the *CYP2D6* genotype, and it has been argued that the *CYP2D6* genotype is related to the response to metoprolol, impacting the efficacy, safety, and toxicity of the drug.[48] In PM individuals compared with non-PM individuals, metoprolol use resulted in higher metoprolol concentrations, as well as a significant and sustained decrease in mean diastolic arterial pressure and heart rate.[50] Previous studies did not show an influence of *CYP2D6* genotype on metoprolol PD; however, these studies included few PM individuals.[50] The package insert describes the decreased metabolism in PM individuals.[49] Careful titration of metoprolol can aid in finding the appropriate dose; however, it appears *CYP2D6* genotyping information may be useful in identifying the appropriate dose earlier in therapy.[51]

Tramadol

The analgesic tramadol is used to treat adult patients with moderate-to-severe pain. The parent compound and its M1 metabolite (O-desmethyltramadol) are responsible for the analgesic effects. Although a number of metabolic pathways, including CYP2D6, CYP3A4, and phase 2 conjugation reactions are responsible for the CL of tramadol, it is CYP2D6 that metabolizes tramadol to its M1 form.[52] In PM individuals, the formation of the active M1 metabolite is significantly decreased.[52] As the analgesic effects of tramadol are a consequence of the concentrations of the parent compound and the M1 metabolite, determining the contribution of either to analgesia for individuals of different phenotypes can be difficult. However, significant differences in adverse events have been noted among different *CYP2D6* phenotypes.[52] Adverse events with tramadol use, including cardiotoxicity, in UMs can be life-threatening.[53]

Drug–*Other Gene* Interactions

Total CL is the sum of all CL mechanisms. While metabolic CL via CYP450 enzymes is a major CL mechanism for many drugs, other drugs are cleared via excretion into the urine (CL_R) or bile by transporters.[4] A number of transporters, such as multidrug resistance protein 1 (MDR1) and multidrug resistance protein 4 (MRP4), among others, are present in the kidney to move drug substrates such as digoxin into the urine.[54,56] Here, polymorphisms that alter transporter function or expression can influence this CL_R mechanism. For instance, MDR1, also known as p-glycoprotein (P-gp), the protein product of the *ABCB1* gene, has been shown to have many polymorphisms.[57] Decreased function of this efflux transporter can result in decreased CL_R and thus total CL, which can affect the maintenance dose requirements for substrate drugs.[4] Conversely, overproduction of an efflux transporter in this setting will increase CL_R and total CL, potentially affecting maintenance dose requirements. Similar concepts can be applied to transporters that move drug into the bile as part of biliary clearance.[58]

As exemplified by the drug–gene interactions cited above, an individual's genotype and related phenotype can influence a drug's CL, which may necessitate an alteration in the maintenance dose to achieve the typical exposure that would be seen in the average wild-type individual. One can speculate that dosage adjustment to optimize response and/or avoid adverse events to a given drug may, in part, be a consequence of the individual's genetic constitution relative to the function/production of drug-metabolizing enzymes and transporters. *CYP450* and *transporter* genotypes can be related to drug CL, affecting maintenance dose requirements.

A BROADER CONTEXT OF INTERACTIONS

Special consideration is warranted when an individual's drug therapy mandates use of multiple drugs that not only have pharmacogenetic implications, but also encompass drug–drug interactions. For example, the interaction between omeprazole and clopidogrel may be consequentially magnified by an individual's *CYP2C19* genotype. Omeprazole is a strong inhibitor of CYP2C19, and as noted previously, inhibition of clopidogrel activation by CYP2C19 can reduce clopidogrel efficacy.[59] A person who has a *CYP2C19*1/*2* genotype may have reduced clopidogrel efficacy, being an IM. However, the IM phenotype cannot be assumed if the patient is also using a CYP2C19 inhibitor such as omeprazole. In effect, the drug–gene interaction (clopidogrel–*CYP2C19*) is confounded further by the drug–drug interaction (clopidogrel–omeprazole), further decreasing the activation of clopidogrel. This drug–drug–gene interaction is known as phenoconversion: The *CYP2C19*1/*2* individual, thought to be an IM, now has the phenotype of a PM. Clinical interpretation of such interactions may be difficult, as many variables are present. Drug–drug–gene interactions do not always result in the need for dosage regimen adjustments. However, when a drug with a narrow therapeutic range is used, where efficacious concentrations are close to or overlapping with those that increase the risk of

adverse events, such interactions may be of substantial clinical significance and warrant careful consideration when selecting concomitant drug therapy.

CONCLUSION

Underlying variability in genes coding for drug-metabolizing enzymes can explain alterations in the pharmacokinetics (e.g., clearance) for drugs that are substrates for the drug-metabolizing enzyme proteins. An altered CL can lead to the need for an alteration in a maintenance dose. As CL influences the half-life ($t_{1/2}$), and as $t_{1/2}$ is used to determine the dosing interval, it is clear that pharmacogenetics, as inherent variability, impacts all components of pharmacokinetics related to drug maintenance dose design. Much work needs to be done to clearly define the influence of genetics on the pharmacokinetics of various drugs. The potential for drug–gene interactions based on the frequency of variant forms of genes in a given population must be taken into account, especially when patients are taking multiple medications, which may result in drug–drug–gene interactions. Genetic testing to identify variant genes in a given individual may aid in optimal drug maintenance dose design. Currently, the clinical application of pharmacogenetics is somewhat limited, with major teaching and research hospitals leading the way, although regional hospitals, in collaboration with genetic testing laboratories, are beginning to offer pharmacogenetic services. Evidence-based guidelines have been developed for numerous drug–gene interactions and can be found on the Pharmacogenomics Knowledgebase website (www.pharmgkb.org/page/cpicGeneDrugPairs and www. pharmgkb.org/page/dpwg). As more drug–gene information becomes available, pharmacists will need to be able to interpret and apply the information to optimize therapeutic decision-making on a formulary and individual patient basis.

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