Pharmacogenomics: Bridging the gap between science and practice
Kelly C. Lee, Joseph D. Ma, and Grace M. Kuo

Abstract

Objective: To educate pharmacists about principles and concepts in pharmacogenomics, clinical applications of pharmacogenomic information, and the social, ethical, and legal aspects of pharmacogenomics and to describe a Centers for Disease Control and Prevention (CDC)-supported pharmacogenomics education program for pharmacists and other health professionals.

Data sources: Primary literature from PubMed, recommendations from the Food and Drug Administration and Evaluation of Genomic Applications in Practice and Prevention Working Group, prescribing information, websites of government agencies and professional organizations, and relevant textbooks.

Study selection: Not applicable.

Data extraction: Not applicable.

Data synthesis: Principles and concepts of pharmacogenomic nomenclature, polymorphism types, and systematic approach to understanding polymorphisms were reviewed. Drug therapy for select therapeutic areas that highlight the applicability of pharmacogenomics are presented, including abacavir (Ziagen—GlaxoSmithKline), selective serotonin reuptake inhibitors, tamoxifen, and warfarin. Challenges of translating pharmacogenomics into clinical practice included ethical, social, legal, and economic issues. We have developed a pharmacogenomics education program to disseminate evidence-based pharmacogenomics information and provide a resource for health professionals, including pharmacists.

Conclusion: Pharmacists play a critical role in the education of patients and health professionals in the area of pharmacogenomics.

Keywords: Pharmacogenomics, pharmacogenetics, education, clinical intervention.

Pharmacy Today. 2009(Dec);15(12):36–48

Learning objectives
At the conclusion of this program, the pharmacist will be able to:
- Define common terminologies used to discuss pharmacogenomics.
- Discuss pharmacogenomic tests in select therapeutic areas based on varying levels of evidence for testing recommendations and clinical use.
- Summarize current recommendations for pharmacogenomic testing based on manufacturer and/or consensus group evidence-based recommendations.
- List resources for and barriers to integrating pharmacogenomics into practice.

ACPE Activity Type: Knowledge-Based
Medication response rates for treating many diseases (e.g., depression, schizophrenia, rheumatoid arthritis) are in the range of 30% to 60%, which is far from optimal. Recent reports indicate that reported adverse drug events have increased from 30,000 to 90,000 events from 1998 to 2005, whereas the number of prescriptions during the same time period increased approximately 27%. Genetics play a role in personalized medicine. Growing evidence suggests that certain drug therapies (e.g., abacavir [Ziagen—GlaxoSmithKline], warfarin), based on a patient’s genetics, may result not only in an improved treatment response but also in a clinically important reduction in adverse drug reactions. The scientific field that studies the role of genetics and the possible relationship to medication therapy is referred to as pharmacogenomics or pharmacogenetics. Professional organizations, regulatory agencies, and researchers provide various but similar definitions for these terms, which are used interchangeably. The American Association of Pharmaceutical Scientists defines pharmacogenetics as “the study of genetic causes of individual variations in drug response” (Table 1). Pharmacogenomics is defined as “the genome-wide analysis of genetic determinants of drug efficacy and toxicity.” Pharmacogenetics focuses on a single or a few genes in a chromosome, while pharmacogenomics encompasses a much larger perspective by examining genes in all chromosomes. Recent research and technologies have expanded into analyzing entire genomes in a quick and efficient manner. Consequently, for the purposes of this article, pharmacogenomics is the preferred term.

The implications of pharmacogenomics affect drug development issues such as drug safety, productivity, and personalized health care. An increasing number of drug labels approved by the Food and Drug Administration (FDA) contain pharmacogenomic information. Integration and use of biomarkers in drug development, regulation, and clinical practice will continue to increase. However, a gap appears to exist between health providers’ knowledge and the expectations of patients regarding pharmacogenomic testing. To meet the expectations and needs of patients, educational campaigns geared toward clinicians need to be developed to bridge the gap between the science of pharmacogenomics and clinical practice.

Objective
Pharmacogenomics is a rapidly developing field that has important implications in individualized treatment for patients. These scientific advances present opportunities and challenges for pharmacists. This article seeks to educate pharmacists about principles and concepts in pharmacogenomics, clinical applications of pharmacogenomic information, and social, ethical, and legal aspects of pharmacogenomics. The first section contains principles and concepts related to pharmacogenomics, while the next section discusses the clinical application of pharmacogenomics for select therapeutic areas. In addition, a Centers for Disease Control and Prevention (CDC)-supported pharmacogenomics education program for pharmacists and other health professionals is described.

Principles and concepts
Molecular biology principles
An in-depth review of molecular biology and genetics is beyond the scope of this article. A detailed review is available elsewhere. Briefly, a chromosome is the structural component of DNA that resides in the cell nucleus. Humans possess a total of 46 chromosomes (23 pairs). Each chromosome contains a single DNA molecule, DNA is the double-helix molecule, and segments or regions of DNA are known as genes. Genes contain noncoding and coding nucleotide sequences needed for messenger ribonucleic acid (mRNA) transcription. A nucleotide is a structural unit of DNA containing a sugar moiety, a phosphate group, and a base. Four nucleotide bases exist in DNA. The two pyrimidine bases are thymine (T) and cytosine (C), and the two purine bases are adenine (A) and guanine (G). Complementary base pair formation occurs when an A pairs with a T or when a G pairs with a C. The arrangement of base pairs provides the DNA sequence or template by which amino acids are determined, resulting in protein synthesis.

At a Glance
Synopsis: Pharmacists can play a critical role in educating patients and health professionals regarding pharmacogenomics—a rapidly developing field that has important implications in individualized patient treatment. The current work addresses drug therapy for select therapeutic areas (e.g., abacavir, selective serotonin reuptake inhibitors, tamoxifen, warfarin) that highlight the applicability of pharmacogenomics. Ethical, social, legal, and economic challenges of translating pharmacogenomics into clinical practice are also discussed. Pharmacists are encouraged to remain current on pharmacogenomic information in order to interpret the literature and testing recommendations, make clinical decisions, and provide counseling for patients.

Analysis: A gap appears to exist between health providers’ knowledge and the expectations of patients regarding pharmacogenomic testing. PharmGenEd is a 3-year program (2008–2011) designed to provide pharmacists, physicians, students, and other health professionals access to evidence-based pharmacogenomic information, increase their knowledge of pharmacogenomic testing for clinical application, influence their attitudes to overcome barriers to discussing this topic with their patients, and improve their skills in using practice tools. The program is currently reaching out to health professionals through collaboration with health care organizations, schools/colleges, pharmacogenomic experts, and clinicians willing to be trained and to train others.

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**Polymorphisms: nomenclature and concepts**

Variation in the DNA sequence present in more than 1% of the population is defined as a polymorphism (Table 1). Several polymorphism types exist. Variation in a single nucleotide base is known as a single nucleotide polymorphism (SNP; pronounced “snip”). Other polymorphisms include large segments of the DNA sequence that consist of a specific gene or numerous genes. Examples include a gene deletion polymorphism and a gene copy number variant polymorphism. Polymorphisms may affect gene expression, protein function, and activity. However, alterations in gene expression can also occur in the absence of DNA sequence variation (e.g., epigenetics).

An understanding of SNP nomenclature is warranted before discussing the clinical application of pharmacogenomics. Numerous methods are used to describe SNPs because a universally accepted SNP nomenclature method does not exist. This results in confusion among pharmacists and health professionals. A SNP can be described in a numeric and alphabetic, star, genotype), the focus is on polymorphisms of a single gene. In certain instances, the functional protein activities of the P450 (CYP) gene: CYP2C19, CYP2C19*1, CYP2C19*2, and CYP2C19*3 are examples of three allele subtypes. The first few letters and numbers identify the protein (e.g., CYP2C19). The star (*) and subsequent number (e.g., 1, 2, 3) identify the allele subtype. Identifying subtypes is relevant to the functional effect for each allele. The CYP2C19*1 allele results in normal (or wild-type) enzyme activity in individuals. In contrast, both the CYP2C19*2 and CYP2C19*3 alleles result in no enzyme activity.

An individual’s genotype identifies both alleles for a specific gene. The CYP2C19 example can be further detailed. If both alleles in an individual are CYP2C19*2, then the respective genotype would be homozygous and identified as CYP2C19*2/*2. Conversely, if an individual has one CYP2C19*2 allele and one CYP2C19*3 allele, then the respective genotype would be heterozygous and identified as CYP2C19*2/*3. Based on the enzyme activities of the CYP2C19 allele subtypes, one can deduce which genotypes will have normal (CYP2C19*1/*1), reduced (CYP2C19*1/*2, CYP2C19*1/*3), or no (CYP2C19*2/*2, CYP2C19*2/*3, CYP2C19*3/*3) enzyme activity.

With the previously described SNP nomenclature (e.g., numeric and alphabetic, star, genotype), the focus is on polymorphisms of a single gene. In certain instances, the functional effect of a protein may result from more than a single gene. A haplotype is a combination of alleles that are closely linked in a single chromosome and are inherited together.

**Table 1. Definitions of commonly used terms**

<table>
<thead>
<tr>
<th>Term</th>
<th>Definition</th>
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<tbody>
<tr>
<td>Allele</td>
<td>The original or the variant form of a gene at a particular location on a chromosome</td>
</tr>
<tr>
<td>Extensive metabolizer</td>
<td>Phenotype and/or genotype of an individual who possesses a high (or extensive) rate of metabolism</td>
</tr>
<tr>
<td>Genotype</td>
<td>The clinical presentation of an individual with a particular genotype</td>
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<tr>
<td>Nucleotide</td>
<td>A structural unit of DNA containing a sugar moiety, a phosphate group, and a base</td>
</tr>
<tr>
<td>Pharmacogenetics</td>
<td>The study of genetic causes of individual variations in drug response</td>
</tr>
<tr>
<td>Pharmacogenomics</td>
<td>The genome-wide analysis of genetic determinants of drug efficacy and toxicity</td>
</tr>
<tr>
<td>Phenotype</td>
<td>The clinical presentation of an individual with a particular genotype</td>
</tr>
<tr>
<td>Polymorphism</td>
<td>Variation in the DNA sequence present in more than 1% of the population</td>
</tr>
<tr>
<td>Poor metabolizer</td>
<td>Phenotype and/or genotype of an individual who possesses a low (or poor) rate of metabolism</td>
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<tr>
<td>Single nucleotide polymorphism (SNP)</td>
<td>Pronounced “snip”; a type of polymorphism for which variation occurs in a single nucleotide base</td>
</tr>
<tr>
<td>Ultra-rapid metabolizer</td>
<td>Phenotype and/or genotype of an individual who possesses a very high (or ultra-rapid) rate of metabolism</td>
</tr>
<tr>
<td>Validity</td>
<td>Extent to which a test actually measures what it claims to measure</td>
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**Systematic approach to understanding polymorphisms**

A systematic approach to understanding polymorphisms is shown in Table 2. First, the polymorphism type and the protein that is affected by the polymorphism are identified. Proteins can include, but are not limited to, enzymes (e.g., CYP),
drug transporters (e.g., P-gp), and receptors. In some cases, the functional effect of a polymorphism can be increased protein activity, decreased protein activity, or no effect. Second, the populations that are affected by the polymorphism are considered because individual and population variation exists. For example, a specific CYP2C19 SNP is present in a higher frequency in Asians than in whites. Third, the relationship between the polymorphism and a drug for an individual is analyzed. A polymorphism may affect drug dosing, efficacy, toxicity, pharmacokinetics, or pharmacodynamics. Alternatively, a polymorphism may have no effect on drug efficacy or toxicity. Fourth, the relationship between the polymorphism and a disease for an individual is investigated. A polymorphism may influence disease status, prognosis, or susceptibility. Additionally, a polymorphism can be used as a screening or diagnostic test for certain diseases.

### Polymorphism types

#### Nonsynonymous SNP

A nonsynonymous SNP refers to when a nucleotide change occurs and the resultant amino acid has changed (Figure 1). Thiopurine methyltransferase (TPMT) is an enzyme that metabolizes thiopurine drugs (e.g., azathiopurine, 6-mercaptopurine). The TPMT*3A polymorphism is an example of a non-synonymous SNP for which the nucleotide guanine (G) is substituted for an adenine (A), resulting in an amino acid change from alanine (Ala) to threonine (Thr). Another nonsynonymous SNP is present elsewhere on the DNA, where the nucleotide adenine (A) is substituted for a guanine (G), resulting in an amino acid change from tyrosine (Tyr) to cysteine (Cys). The functional effect of the TPMT*3A polymorphism includes both nonsynonymous SNPs and results in decreased TPMT enzyme activity. This is clinically important because an increased risk of thiopurine toxicity (e.g., myelosuppression) is associated with decreased TPMT enzyme activity.

#### Synonymous SNP

A synonymous SNP refers to when a nucleotide change occurs but the resultant amino acid is unchanged from the original or reference amino acid (Figure 1). Abcb1 3435 C>T is an example of a synonymous SNP. The nucleotide cytosine (C) is substituted for a thymine (T); however, the resultant amino acid glycine (Gly) remains unchanged. Evidence suggests that the ABCB1 3435 C>T SNP decreases P-gp expression and function, possibly via a mechanism of decreased mRNA stability. Efavirenz is a drug that is a P-gp substrate. In a case–control study of human immunodeficiency virus (HIV)-infected patients, a decreased risk of efavirenz-related hepatotoxicity was associated with the ABCB1 3435 C>T SNP. Conversely, for cyclosporine (a P-gp substrate), patients with the ABCB1 3435 C>T SNP have not shown a significant difference in cyclosporine phar-
macokinetics or toxicity risk. One potential explanation for the discrepancy between efavirenz and cyclosporine is that a "silent" polymorphism alters the interaction sites for P-gp substrates. Recent studies have confirmed that P-gp recognizes multiple substrates via partially overlapping but distinct binding sites.

**Premature stop codon SNP**

A premature stop codon SNP refers to when a nucleotide change occurs, the resultant amino acid is no longer coded, and protein synthesis is terminated (Figure 1). The CYP2C19*3 allele is an example of a premature stop codon SNP, for which the nucleotide guanine (G) is substituted for an adenine (A). The change results in termination of protein synthesis. The functional effect of the CYP2C19*3 allele is no CYP2C19 enzyme activity. The proton pump inhibitors (PPIs) omeprazole and lansoprazole are drugs that are affected by the CYP2C19 genotype. In one study, omeprazole area under the concenzyme activity. The changes in uridine diphosphate (UDP) nucleotide repeats. The functional effect of the polymorphism whereby an extra dinucleotide sequence of thymine (T) and cytosine (C) results in the deletion of the entire CYP2D6 gene and a complete loss of CYP2D6 enzyme activity. The polymorphism results in the deletion of the entire CYP2D6 gene and a complete loss of CYP2D6 enzyme activity. The polymorphism observed with CYP2D6, whereby extra copies of the CYP2D6*2 allele confer an ultra-rapid metabolizer phenotype. The selective serotonin reuptake inhibitors (SSRIs), as well as tamoxifen, are drugs for which CYP2D6 polymorphisms may affect drug dosing, efficacy, and toxicity.

**Variable number tandem repeat polymorphism**

A variable number tandem repeat polymorphism occurs when there is an insertion of repeat nucleotides into the DNA sequence. The functional effect may be an increase, a decrease, or no effect on protein activity. Uridine diphosphate (UDP) glucuronosyl transferase (UGT)1A1 is an enzyme in which the UGT1A1*1 allele includes six dinucleotide repeats of thymine (T) and adenine (A) and results in normal enzyme activity. The UGT1A1*28 allele is an example of a tandem repeat polymorphism whereby an extra dinucleotide sequence of thymine (T) and adenine (A) occurs. This results in a total of seven dinucleotide repeats. The functional effect of the UGT1A1*28 allele is decreased UGT1A1 expression and enzyme activity. The risk of irinotecan toxicity has been associated with the UGT1A1*28 variable number tandem repeat polymorphism.

**Gene deletion polymorphism**

In gene deletion, several thousand nucleotides of a specific gene are deleted. More than 50 variant allele subtypes exist for the CYP2D6 enzyme subfamily. The CYP2D6*5 allele is an example of a gene deletion polymorphism. The polymorphism results in the deletion of the entire CYP2D6 gene and a complete loss of CYP2D6 enzyme activity.

**Gene copy number variant polymorphism**

A gene copy number variant refers to the duplication of several thousand nucleotides comprising a specific gene. In essence, an extra copy of the exact same gene is now present along the DNA. In some cases, 2 to 13 extra copies of the same gene can be present. A gene copy number variant polymorphism is also observed with CYP2D6, whereby extra copies of the CYP2D6*2 allele confer an ultra-rapid metabolizer phenotype. The selective serotonin reuptake inhibitors (SSRIs), as well as tamoxifen, are drugs for which CYP2D6 polymorphisms may affect drug dosing, efficacy, and toxicity.

**Clinical applications**

In this section, we will highlight four pharmacotherapy topics (abacavir, SSRIs, tamoxifen, and warfarin) and discuss the following: (1) polymorphism (e.g., gene/allele) of interest, (2) functional effect of the polymorphism, (3) population variation (when available), (4) available genetic tests, (5) literature supporting or refuting the clinical relevance of pharmacogenomic testing, and (6) testing recommendations. The specific therapeutic areas included in the current work were selected based on their varying levels of evidence for testing recommendations and clinical use. The level of evidence was based on the existence of formal recommendations that either support or refute the role of pharmacogenomic testing for that drug or drug class. Clinical use was based on the frequency of use of the pharmacogenomic test in the clinical setting. For example, with abacavir, both the level of evidence of pharmacogenomic testing and the drug’s clinical use are high. In contrast, with warfarin, the level of evidence for pharmacogenomic testing is moderate, while the clinical use is low at this time. A case presentation will precede each therapeutic area discussion.

**Abacavir**

H.S. is a 29-year-old white man with a 3-month history of acquired immunodeficiency syndrome (AIDS) who presents with fever, gastrointestinal upset, and a maculopapular rash on his forearms and trunk of his body for the past 3 days. He has no known drug allergies, and his last viral load was 50,000 copies and CD4 count 100/mm³. His current antiretroviral regimen is abacavir, zidovudine, and efavirenz. He is prescribed sulfamethoxazole-trimethoprim for Pneumocystis carinii pneumonia prophylaxis. What is the cause of H.S.’s maculopapular rash? How would you evaluate this clinical scenario? Would pharmacogenomic testing be helpful in formulating your clinical decision?

Abacavir is a nonnucleoside reverse transcriptase inhibitor for treating HIV infection and AIDS and has been extensively used in highly active antiretroviral treatment regimens. Abacavir is associated with a treatment-limiting hypersensitivity reaction (HSR) that may include fever, skin rash, gastrointestinal symptoms, constitutional symptoms (e.g., malaise, fatigue), and respiratory symptoms. The rash can lead to more severe forms of dermatologic reactions such as Stevens-Johnson syndrome, systemic lupus erythematosus, or toxic epidermal necrolysis. Other symptoms of HSR include lactic acidosis, severe hepatomegaly, and death. The incidence of abacavir-induced HSR is 2% to 8%, and the reaction usually occurs within the first 6 weeks of initiating the drug. If a patient develops HSR caused by abacavir, the clinician and patient must decide whether to continue the drug. Oftentimes, establishing the causality of HSR, as depicted in the featured case, is difficult, and...
inadvertently discontinuing the antiretroviral agent may have devastating consequences for the patient’s disease treatment.

**Polymorphism of interest, functional effect, and population variation**

A known polymorphism of the human leukocyte antigen (HLA) gene is associated with an increased sensitivity or likelihood of developing an abacavir-induced HSR. The HLA-B*5701 allele is prevalent in 5% to 8% of whites with no appreciable difference in minority populations. Systemic reactions to abacavir are mediated through drug-specific activation of cytokine-producing CD8+ T cells.

**Available pharmacogenomic tests**

Patients may be tested for the HLA-B*5701 allele with a blood test or buccal swab. The genetic sequences coding for the HLAB*5701 are probed and reported as positive if the allele is present or negative if the allele is absent.

**Summary of clinically relevant literature**

At least three studies have demonstrated the use of HLA-B*5701 allele screening. PREDICT-I (Prospective Randomized Evaluation of DNA Screening in a Clinical Trial) showed that HLA-B*5701 screening could accurately predict patients who might be at risk for HSR due to abacavir. When abacavir was avoided in patients who screened positive for HLA-B*5701, the incidence of clinically suspected abacavir-induced HSR was 3.4% compared with 7.8% in patients who were not screened (P < 0.001). SHAPE (Study of Hypersensitivity to Abacavir and Pharmacogenetic Evaluation) demonstrated a similar trend in whites and blacks. Of 99 patients who had positive skin patch tests to confirm HSR, 47 carried the HLA-B*5701 allele. Finally, in the North American multicenter prospective study ARIES (Atazanavir Ritonavir Induction/Simplification with Epzicom), 41 (5.7%) of 725 patients were positive for the HLA-B*5701 allele, including 7.2% in whites, 2.8% in blacks, and 5.6% in other groups. Among patients who were HLA-B*5701 negative, none had positive skin patch tests at 30 weeks.

Patients with the HLA-B*5701 allele appear to have increased risk of developing HSR from abacavir. By identifying patients at risk, clinicians may offer an alternative antiretroviral agent that should be initiated in patients who have the HLA-B*5701 allele.

**Testing recommendations**

The manufacturer of abacavir and the Department of Health and Human Services guideline both recommend that before initiating abacavir, the patient should be screened for the HLA-B*5701 allele. The association between HLA-B*5701 and HSR has been included as a black box warning in the prescribing information for abacavir as of July 2008. Screening should be conducted in patients who are naive to the drug and in those with unknown HLA-B*5701 status who have previously tolerated abacavir and will be restarting therapy. Patients who have negative HLA-B*5701 status may still be at risk of developing HSR; however, the risk is much less than that for patients who are positive for the HLA-B*5701 allele. In all patients, regardless of allele status, abacavir should be discontinued immediately if HSR is suspected and cannot be ruled out; these patients should not be rechallenged with abacavir in the future. Of important note, patients need to be monitored after the drug is discontinued.

**SSRIs**

J.M. is a 45-year-old Asian man who was diagnosed with major depressive disorder approximately 8 months ago and has been taking paroxetine. He reports that he has had considerable nausea and diarrhea from the medication and difficulty remaining adherent to the medication. His doctor recommended lowering the paroxetine dose from 20 to 10 mg/day to reduce the adverse effects. However, J.M. still complains of adverse effects and notes that his depression symptoms have not improved. He now presents to the pharmacy and asks the pharmacist about other antidepressant medications. What is the potential etiology of J.M.’s adverse effects and inadequate response to paroxetine? Could pharmacogenomic testing assist in evaluating J.M.’s drug therapy?

SSRIs are the mainstay treatment for depressive and anxiety disorders. The benefits of SSRIs include therapeutic effectiveness, a wide therapeutic index, good tolerability, and less toxicity in overdose situations. Six medications (fluoxetine, paroxetine, sertraline, fluvoxamine, citalopram, and escitalopram) are currently available in the SSRI class; however, five of them (excluding fluvoxamine) are used as first-line drug therapy for major depression. They share the same mechanisms of action but differ in severity of adverse effects and drug interactions. Individual SSRIs are metabolized by different CYP enzymes; for example, fluoxetine is metabolized by CYP2C9, CYP2C19, and CYP2D6, while paroxetine is metabolized in part by CYP2D6. In addition, each SSRI is capable of inhibiting other CYP enzymes; paroxetine is a CYP2D6 inhibitor while fluvoxamine is a CYP1A2 inhibitor.

**Polymorphism of interest, functional effect, and population variation**

A patient’s CYP enzyme activity may determine their ability to metabolize an SSRI. Patients who are poor metabolizers of CYP2D6 generally possess two copies of inactive CYP2D6 alleles, which confers no CYP2D6 enzyme activity. Patients may experience higher plasma concentrations of a drug such as paroxetine and may be at increased risk of experiencing an adverse drug reaction. Conversely, patients with more than two copies of active CYP2D6 alleles via a gene copy number variant polymorphism are considered ultra-rapid metabolizers and have the ability to metabolize drugs to a greater extent. This results in a lower concentration of the drug that undergoes CYP2D6 metabolism and potentially a decreased clinical effect from the drug. Multiple CYP isoenzymes and allelic variations affect SSRI metabolism. For example, CYP2D6 has at least five different alleles that differ in prevalence in whites, Asians, and blacks. Other CYP enzymes and alleles may also vary in different populations; consequently, predicting efficacy or toxicity for a drug
that is metabolized by numerous CYPs can be difficult.

**Available pharmacogenomic tests**
The AmpliChip CYP450 test from Roche Diagnostics is the first FDA-approved test for analyzing polymorphisms for **CYP2D6** and **CYP2C19**. The test may reveal whether the patient is a poor or ultra-rapid metabolizer. The test result would potentially allow the health provider to make appropriate adjustments in doses if necessary for various drug classes, including beta blockers, antipsychotics, opiates, anticonvulsants, and PPIs. Although the test is available commercially, its ability to predict clinical efficacy or toxicity is uncertain.

**Summary of clinically relevant literature**
Despite the potential impact of CYP enzyme activity on drug metabolism, evidence supporting the use of pharmacogenomic testing to predict clinical response or choice of drug is lacking. Single-dose SSRI pharmacokinetic studies show that CYP genotype and phenotype status correlates with circulating SSRI plasma concentrations. Despite this correlation, no evidence currently exists suggesting that CYP genetic testing influences SSRI choice or dose. Also, pharmacodynamic studies have not shown a correlation between metabolizer status and clinical response in adult patients treated with SSRIs. Evidence is conflicting regarding an individual’s CYP genotype and relationship to SSRI adverse effects.

**Testing recommendations**
The Evaluation of Genomic Applications in Practice and Prevention (EGAPP) Working Group found that there was insufficient evidence to support recommending for or against testing in nonpsychotic depression before initiating treatment with SSRIs. The EGAPP Working Group stated that additional studies are needed to examine individual drugs and not SSRIs as a class. In the prescribing label for SSRIs, information is provided regarding the influence of SSRIs on other CYP substrates and inhibitors. No specific recommendations exist for testing CYP enzyme activity in patients initiating treatment with SSRIs or during therapy.

**Tamoxifen**
H.M. is a 40-year-old white premenopausal female diagnosed with estrogen receptor–positive (ER+) breast cancer. She has been treated with chemotherapy and has been prescribed tamoxifen. After 12 months of treatment, H.M.’s disease has relapsed; another mass near the original tumor has been detected. The oncologist orders a pharmacogenomic test for H.M. The test result reveals that H.M. is a poor metabolizer of CYP2D6. What is the implication of this pharmacogenomic testing result? How does this result play a role in evaluating this clinical scenario?

Tamoxifen is a selective estrogen receptor modulator that is FDA approved for adjuvant treatment of breast cancer, treatment of metastatic breast cancer and ductal cancer in situ, and reduction of breast cancer incidence in high-risk women. Tamoxifen decreases breast cancer mortality by 30% in pre- and postmenopausal women with ER+ cancer when taken for 5 years after surgery.

**Polymorphism of interest, functional effect, and population variation**
Tamoxifen is a prodrug that requires biotransformation by CYP2D6 and -3A4 to produce two active metabolites, 4-hydroxy N-desmethyl-tamoxifen (or endoxifen) and 4-hydroxy-tamoxifen (Figure 2). 4-hydroxy-tamoxifen was considered the primary active metabolite because it has higher affinity to the estrogen receptor and is 30 to 100 times more potent than tamoxifen in suppressing estrogen-dependent cell proliferation. Recent evidence suggests that endoxifen, a metabolite that is formed from N-desmethyl-tamoxifen via CYP2D6, may be equipotent to 4-hydroxy-tamoxifen and present in concentrations 10 times higher than 4-hydroxy-tamoxifen. Evidence suggests that decreased endoxifen concentrations result in disease recurrence; however, less evidence supports endoxifen concentrations affecting adverse effects.

A patient’s CYP2D6 genotype is an important determinant for tamoxifen response and disease recurrence. The CYP2D6*4/*4 genotype results in no CYP2D6 enzyme activity. This results in a poor metabolizer phenotype with an inability to metabolize tamoxifen, thereby resulting in decreased endoxifen concentrations. Patients with the CYP2D6*4/*4 genotype have significantly decreased endoxifen concentrations compared with patients who have at least one CYP2D6 functional allele. Approximately 13% to 21% of whites carry the CYP2D6*4 allele compared with less than 2% prevalence in Asians and blacks. Additional polymorphisms for CYP2D6 exist, such as *3, *4, *6, *9, *10, *17, and *41, with varying frequencies.

**Available pharmacogenomic tests**
Blood tests are available to assess CYP enzyme polymorphisms. Based on the metabolism of tamoxifen, the test should determine various CYP2D6 allele subtypes, including *3, *4, *6, *9, *10, *17, and *41. As new alleles are identified, tests for additional allelic variations of CYP enzymes should be considered.

**Summary of clinically relevant literature**
Although studies have been conducted with tamoxifen and CYP2D6*4/*4 genotypes, results are conflicting in the relationship between genotype and overall survival, disease recurrence, and disease-free survival. In a retrospective study of postmenopausal patients with surgically resected ER+ breast cancer receiving adjuvant tamoxifen, women with the CYP2D6*4/*4 genotype had a shorter relapse-free survival time and worse disease-free survival time compared with women with one or no CYP2D6*4 alleles (log rank = 0.03, \( P = 0.017 \)). Another study analyzed several CYP2D6 allele subtypes; patients with CYP2D6*3, *4, *10, or *41 alleles had more breast cancer recurrences, shorter relapse-free periods, and worse disease-free survival rates than patients with CYP2D6 functional alleles. With regard to overall survival, no overall association with CYP2D6 genotypes was observed.

Data associating tamoxifen adverse effects with specific
CYP2D6 genotypes are conflicting. The incidence of hot flashes with tamoxifen, depending on the genotype, showed opposite trends in two studies. Of note, patients may appear to be poor metabolizers of CYP2D6 when they are given concomitant drugs that inhibit the same enzyme. Drugs that are CYP2D6 inhibitors, such as fluoxetine and paroxetine, can mimic a poor metabolizer phenotype and can result in decreased endoxifen concentrations. If the addition of an antidepressant is necessary, potent inhibitors of CYP2D6 (e.g., paroxetine, fluoxetine, bupropion, duloxetine [Cymbalta—Eli Lilly]) should be avoided. Mild inhibitors such as sertraline, citalopram, escitalopram, and venlafaxine are preferred.

**Testing recommendations**

Currently, the manufacturer of tamoxifen provides pharmacogenomic information and the potential effects on disease status with no specific recommendation for testing. The FDA advisory committee recommended a label update to reflect the increased risk of worsening disease outcomes in patients with a poor metabolizer of CYP2D6 status. However, there was insufficient evidence to recommend for or against use of tumor gene expression profiles to improve outcomes in women with breast cancer. As of the last available recommendation from October 2006, no formal consensus on genetic testing for tamoxifen existed.

**Warfarin**

S.K. is a 55-year-old white woman with a recent history of atrial fibrillation who requires long-term anticoagulation therapy with warfarin. She and her daughter are quite concerned about the potential for bleeding and ask the pharmacist about the use of genetic testing to avoid this adverse event. Could pharmacogenomic testing predict whether S.K. will have a serious bleeding event from warfarin? Is testing indicated at this time for S.K.?

Warfarin is an oral anticoagulant drug that is widely used to prevent and treat thromboembolic disease in patients with deep-vein thrombosis, pulmonary embolism, mechanical heart valves, and atrial fibrillation. Warfarin is a racemic mixture of two active isomers (R-isomer and S-isomer); the S-isomer is three to five times more potent than the R-isomer. S-warfarin is metabolized by CYP2C9 and inhibits vitamin K epoxide reductase complex subunit 1 (VKORC1). This results in decreased formation of vitamin K–dependent clotting factors and provides the therapeutic effect of anticoagulation. Although warfarin is an effective anticoagulant, it is associated with a substantial risk of major and sometimes fatal bleeding and carries a black box warning in the prescribing information. Patients taking warfarin must be closely monitored for signs and symptoms of bleeding; their international normalized ratios (INRs) must be kept within normal ranges to decrease the risk of bleeding.

**Polymorphism of interest, functional effect, and population variation**

Polymorphisms in either CYP2C9 or VKORC1 may influence either the metabolism of warfarin or the formation of active clotting factors. The functional effect of the CYP2C9*1 allele is normal (or wild-type) CYP2C9 activity. Patients who have the CYP2C9*2 or CYP2C9*3 allele have decreased CYP2C9 enzyme activity by 50% and 90%, respectively, resulting in impaired metabolism of S-warfarin. Decreased CYP2C9 enzyme activity results in increased S-warfarin concentrations, which predisposes the patient to an increased risk of bleeding. Although other alleles exist for CYP2C9 (e.g., *5, *6, *8, *11), these alleles are present in smaller frequencies compared with the *2 and *3 alleles but may still have the potential to impair the metabolism of S-warfarin.
The VKORC1 1173 G>T and VKORC1 -1639 G>A polymorphisms, on the other hand, may be responsible for the pharmacodynamic mechanism for warfarin resistance in certain patients. Ten different SNPs for VKORC1 have been shown to determine nine different haplotypes. Certain haplotypes have been found to be independently associated with higher or lower warfarin dose requirements. Haplotype A has been associated with requiring a lower warfarin dose, whereas haplotype B has been associated with requiring a higher warfarin dose. Considerable difference exists in allele frequencies for CYP2C9 and VKORC1 in ethnic populations. The allele frequencies of CYP2C9*2 and CYP2C9*3 among whites are 8% to 20% and 3% to 15%, respectively. CYP2C9*2 is present in low frequencies (2–4%) in Asians and in blacks. The frequency of CYP2C9*3 is also low (1–4%) in Asians and blacks. For VKORC1 haplotypes, the frequency for type A is 37% for whites, 14% for blacks, and 89% for Asians. For type B, the frequency is 58% for whites, 49% for blacks, and 10% for Asians. VKORC1 -1639 G>A is present in 82% to 83% of Asians compared with 14% of whites. For VKORC1 1173 C>T, Asians have the highest frequency (89%) compared with whites (42%) and blacks (9%).

Available pharmacogenomic tests
Rapid Genotyping Assay (ParagonD, Morrisville, NC) detects CYP2C9 and VKORC1 polymorphisms and is FDA approved. Other tests have been approved by FDA, including the Nanosphere Verigene Warfarin Metabolism Nucleic Acid Test and the Warfarin Dose/Adviser Genetic Test (Kimball Genetics, Denver, CO). The choice of laboratory and test may depend on geographic distribution and turnaround time for patients and health providers.

In a recent systematic review, the analytic validity of CYP2C9 and VKORC1 genetic testing was evaluated. More data are available for the analytic validity of CYP2C9 than of VKORC1. The sensitivity and specificity are estimated to be 98% or higher for CYP2C9 genotyping, but the sensitivity and specificity rates could not be estimated for VKORC1 genotyping due to lack of data. The authors reported that although the actual genotyping can be completed within a day, the lag time could result from slow transporting of samples or specific laboratory’s schedule for running samples.

Summary of clinically relevant literature
Presence of one or two copies of the CYP2C9*2 or CYP2C9*3 allele may require 17% to 75% lower warfarin doses. The presence of at least one variant allele was associated with an increased risk of above-average INRs (hazard ratio [HR] 1.40 [95% CI 1.03–1.90]) and increased time to achieving stable dosing (median difference 95 days [HR 0.65 [0.45–0.94]]). A two- to threefold increased risk of bleeding was observed during the initiation phase of therapy but not during long-term therapy in those with at least one variant allele of CYP2C9. Evidence is conflicting regarding the risk of bleeding events in patients during long-term therapy based on CYP2C9 genotypes.

Patients who had the AA genotype for VKORC1 SNP were more likely to have a 32% lower weekly warfarin dose compared with those with the BB genotype, who required an increased warfarin dose by 35%. The VKORC1 -1639 G>A SNP, along with CYP2C9 alleles, CYP2C9 inhibitors, and increasing age, were associated with a lower warfarin dose requirement. Recently, a multicenter trial of 4,043 patients requiring target INRs between 2 and 3 were dosed using three different algorithms: pharmacogenetic algorithm, clinical algorithm, and fixed-dose approach (35 mg/week). The pharmacogenetic algorithm was based on clinical and genetic variables that could contribute to differences in warfarin dosing such as demographics, indication for warfarin, concomitant medications, and presence of CYP2C9 and VKORC1 polymorphisms. The clinical algorithm was based on all of the same factors but did not include the genetic information. The pharmacogenetic algorithm provided more accurate dose estimates (i.e., closer to the actual doses required) than clinical or fixed-dose algorithms (Figure 3) and predicted better weekly dosing, requiring less than 21 mg/week (P < 0.001) and greater than 49 mg/week (P < 0.001).

Pharmacogenomic testing for CYP2C9 and VKORC1 polymorphisms may improve prediction of warfarin target maintenance doses. Testing may decrease time to stabilization of warfarin dose, risk of above-average INRs, and risk of serious bleeding events. Whether testing will affect long-term safety is uncertain. Furthermore, less evidence supports the association between CYP2C9 genotype and stable INR during the induction phase, when the risk of serious adverse events is the highest, and the evidence for the clinical use of VKORC1 genotyping to predict severe bleeding events is limited. A recent pharmacoeconomic study suggested that pharmacogenomic testing may be cost effective in patients who are initiating warfarin therapy and who are at high risk for bleeding events.

Testing recommendations
In August 2007, FDA updated the label for Coumadin to include information on pharmacogenomic testing and to encourage use of this information for individualized dosing. Dosing nomograms based on genotype are suggested by the manufacturer. Currently, testing for CYP2C9 and VKORC1 is not required. Health providers should not delay initiation of warfarin therapy for genetic testing. Genetic testing is not appropriate for patients already on warfarin and does not replace INR monitoring. Inform patients that not all patients with one or more polymorphisms of CYP2C9 or VKORC1 will have a serious bleeding event and that those without any polymorphisms may still be susceptible to a bleeding event. Recently, the Centers for Medicare & Medicaid Services (CMS) proposed not paying for warfarin genetic tests because insufficient evidence supports using the tests to improve patients’ health. Soon afterwards, CMS revised its decision and decided to cover pharmacogenomic testing to Medicare beneficiaries who (1) have not been previously tested for CYP2C9 or VKORC1, (2) have received fewer than 5 days of warfarin therapy, and (3) are enrolled in a clinical study that meets high standards of scientific integrity and relevance to the Medicare population.
Pharmacogenomics in clinical practice

Pharmacists must consider multiple aspects when implementing pharmacogenomic evidence into practice. These aspects include but are not limited to access, feasibility, lack of sufficient evidence, and ethical, social, legal, and economic issues.

Access, feasibility, lack of sufficient evidence

Not all practice settings have access to testing kits and laboratories for analyzing pharmacogenomics tests. Additionally, not all clinical settings have access to evidence-based practice recommendations and trained health professionals with the knowledge and skills to order and interpret pharmacogenomic test results. Furthermore, the information related to insurance coverage of pharmacogenomic tests for patients is often difficult to obtain. Feasibility information such as the turnaround time for available pharmacogenomic test results or information about the sensitivity and specificity of these tests are not readily available. For example, approximately 5 days are needed for HLA-B*5701 test results to be available (G.M. Kuo, oral communications with Labcorp, January 15 to February 9, 2009). Up to 10 days are needed to determine CYP2D6 and CYP2C9 test results from the AmpliChip CYP450 test (G.M. Kuo, oral communications with Labcorp, January 15 to February 9, 2009). Whole-blood specimen is recommended by most laboratories, although some accept buccal specimen (G.M. Kuo, oral communications with Labcorp, January 15 to February 9, 2009). The lack of quality and number of long-term studies, small sample sizes of the clinical trial, and scant information about the predictive values of laboratory tests add to the challenges faced by health professionals in keeping up with practice information from the emerging pharmacogenomic evidence at this time.

Ethical issues

Implementing pharmacogenomics into practice involves patients and health professionals overcoming ethical barriers such as concerns for loss of privacy. Selecting patients for pharmacogenomic tests must be evidence based to avoid genetic profiling, discrimination, stigmatization, or distributive injustice. Pharmacogenomic testing yields information that could help health professionals and patients make informed decisions about treatment options. Clinicians must not unduly influence patient decisions based on their own opinions; rather, it is imperative that health professionals provide patients with currently available evidence-based information to allow shared and informed decision making. The lack of information about the validity, reliability, sensitivity, and specificity makes it even more difficult for the informed consent process. Interpretation of test results may often be viewed as positive, negative, or inconclusive. How clinicians and patients assess risks in both the presence and absence of conclusive test results poses challenges in clinical practice and may produce unwanted psychological effects, especially if no alternative treatments are available. Furthermore, how and when the test results should be shared with other at-risk relatives may be difficult to assess.

Social and legal issues

Social issues derived from pharmacogenomic testing may lead to unequal treatment or undesirable outcomes such as health disparities, especially if one group of patients receives coverage for pharmacogenomic testing and others do not. Legal issues also need to be considered because they affect how pharmacogenomics is being translated into practice. A provision of the Genetic Information Nondiscrimination Act protects Americans against discrimination based on genetic information related to health insurance coverage and employment decisions. Another aspect of legal protection for clinician liability amidst many uncertainties in practice needs to be addressed as the field moves forward into practice.

Economic issues

Many unknown issues exist related to the economics of pharmacogenomic testing when translated into practice. For example, whether the patient or the insurance company should pay for the pharmacogenomic test or which tests should be covered is uncertain. Whether the insurance premium will increase as a result of increasing coverage for pharmacogenomic testing and whether test results will lead to unfair risk assessment for coverage are unclear because recommendations for pharmacogenomic testing are still evolving. Conflicting evidence to support
cost effectiveness of pharmacogenomic tests exists.\(^35\)–\(^37\) Willingness to pay from the payers’ perspective is varied, particularly because the evidence for cost effectiveness of pharmacogenomic testing is not definitive. In the case of abacavir, for which pharmacogenomic testing for HLA B\(^*\)5701 is effective in preventing HSR, the use of the pharmacogenomic test appears to be cost effective.\(^36\)–\(^39\) However, pharmacogenomic testing for CYP2C9 and VKORC1 does not appear to be cost effective for all patients taking warfarin. Continued research to assess the value of pharmacogenomics is ongoing.\(^100\)

Flowers and Veenstra\(^101\) postulated that a cost-effective pharmacogenomic strategy may occur for drugs that have (1) narrow therapeutic index or high degrees of variability in interindividual response, (2) limitations in current methods for monitoring treatment responses and adverse effects, and (3) few alternative treatment options. Concern exists that the emerging genomic tools for both screening and treatment uses, when applied widely, may disrupt the current public health system. However, Garrison\(^102\) postulated that the U.S. health care system should have the flexibility to adapt with gradual and incremental progression in such a way that it will maintain its infrastructure while the quality of patient care improves with these scientific advances.

**PharmGenEd program**

Despite current limitations and uncertainties in which the standard of care will evolve, the field of pharmacogenomics is developing rapidly. At this beginning stage of implementing pharmacogenomic evidence into practice, pharmacists play a vital role in training others. Pharmacists need to obtain up-to-date pharmacogenomic information in order to interpret the literature and test results, make clinical recommendations and decisions, and provide counseling for patients. PharmGenEd is a 3-year program (2008–2011) designed to provide pharmacists, physicians, students, and other health professionals access to evidence-based pharmacogenomic information, increase their knowledge of pharmacogenomic testing for clinical application, influence their attitude to overcome barriers to discussing this topic with their patients, and improve their skills in using practice tools.

The objective of PharmGenEd is to increase awareness of current knowledge about the validity and use of pharmacogenomic tests and their potential clinical implications. The program aims to reach health professionals through collaboration with health care organizations, schools/colleges, pharmacogenomic experts, and clinicians willing to be trained and to train others. Continuing education (CE) lectures are offered through professional organizations at national/regional/local meetings and via a website. The online CE materials are available with free access to health professionals via a one-time registration (http://pharmacogenomics.ucsd.edu). Additional contents in a shared curriculum format will be available in the public domain after speakers have completed registration and training. All educational materials are designed to be relevant for clinical practice and will include case examples and vignettes for interactive learning. Resources for pharmacogenomics information are listed and updated on the website (Table 3).

The PharmGenEd program is currently seeking (1) speakers to help disseminate the CE materials, (2) reviewers to provide comments for additional modules that are being developed, and (3) trainers to teach health professionals and students. If you are interested in becoming a speaker, reviewer, or trainer, please contact the PharmGenEd office at pharmacogenomics@ucsd.edu or call 858-822-7754.

### Table 3. Informational resources on pharmacogenomics

<table>
<thead>
<tr>
<th>Topic area</th>
<th>Internet source</th>
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<td>Court M, A pharmacogenomics primer, <a href="http://jcp.sagepub.com/cgi/reprint/47/9/1087.pdf">http://jcp.sagepub.com/cgi/reprint/47/9/1087.pdf</a></td>
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<td>Pharmacogenomics Knowledge Base (PharmGKB), <a href="http://www.pharmgkb.org">www.pharmgkb.org</a></td>
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<td>FDA: valid genomic biomarkers, <a href="http://www.fda.gov/Drugs/ScienceResearch/ResearchAreas/Pharmacogenetics/ucm083378.htm">www.fda.gov/Drugs/ScienceResearch/ResearchAreas/Pharmacogenetics/ucm083378.htm</a></td>
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<td>University of Michigan Center for Public Health and Community Genomics, <a href="http://www.sph.umich.edu/genomics">www.sph.umich.edu/genomics</a></td>
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<tr>
<td></td>
<td>National Human Genome Research Institute, <a href="http://www.genome.gov/policyethics">www.genome.gov/policyethics</a></td>
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Abbreviations used: CDC, Centers for Disease Control and Prevention; SNP, single nucleotide polymorphism.