



PHARMACY CORNER

E. P. C. M. S.

Warfarin and Pharmacogenetic Testing (CYP2C9 and VKORC1)

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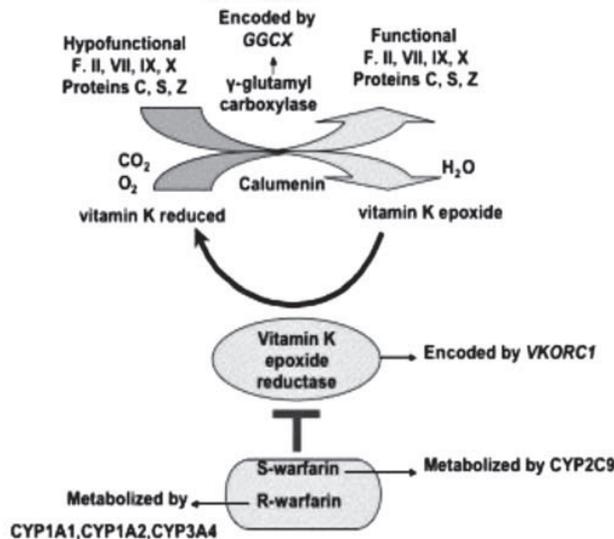
BACKGROUND

The anticoagulant effects of warfarin (3- \acute{a} -acetylnbenzyl-4-hydroxycoumarin; Coumadin[®]) have been used for more than 60 years to effectively reduce coagulopathies. The FDA has recently noted the significance of testing for genetic variations in cytochrome P450 2C9 (CYP2C9) and vitamin K epoxide reductase complex 1 (VKORC1) genes. Cytochrome P450 enzymes are responsible for drug metabolism and VKORC1 is involved in the reduction of vitamin K in the coagulation cascade. Several retrospective and prospective studies have evaluated the impact of genetic determinants in the dosing of warfarin. Warfarin is an oral vitamin K antagonist approved for prevention and treatment of venous thromboembolism (VTE) and stroke prevention in atrial fibrillation. The use of warfarin therapy is a challenge to manage due to its unpredictable dosing among individuals (up to 20-fold).¹ The international normalized ratio (INR) is a monitoring parameter used to assess adequate anticoagulation. According to the U.S. Department of Health and Human Services, about 58% of patients taking warfarin will maintain a therapeutic INR. In most patients, the therapeutic range is maintained between 2.0 and 3.0 to prevent bleeding and reduce coagulation risks. The pharmacogenetic variations and risk factors for bleeding (INR >4, \geq 65 years, history of gastrointestinal bleeding, trauma, renal insufficiency, etc.) have been considered for initiation and maintenance therapy in warfarin.^{1,2}

Warfarin inhibits the activation of clotting factors II (thrombin), VII, IX, and X, as well as protein C and S, which require vitamin K to assist in the clotting cascade (See Figure 1). The “inactive” (oxidized) form of vitamin K is reduced to its “active” form by VKORC1, which biologically activates clotting factors through γ -glutamyl carboxylation. Warfarin causes a depletion of vitamin K by competitively inhibiting the subunit 1 in the VKOR complex (See Figure 1). In warfarin metabolism, the S-enantiomer of warfarin undergoes hepatic metabolism via CYP2C9 to the 7-hydroxylated form of warfarin, and minor pathways including CYP2C8, 2C18, 2C19, and 3A4, metabolize R-warfarin. Patients may experience a risk of bleeding with heterozygous variants for CYP2C9 (*1/*2 or 1/*3) due to a 37% reduction in the clearance of S-warfarin. Whereas patients homozygous for CYP2C9 (*2/*2, *2/*3, *3/*3) experience a 70% decrease in drug elimination, resulting in a much higher plasma concentrations and increased risk of bleeding.²

According to National Surveillance for Emergency Department Visits, warfarin is considered one of the top ten medications seen in hospital admissions due to adverse events.^{4,5} Gong et al. conducted a prospective cohort study evaluating a pharmacogenetic algorithm for loading and maintenance dosing in patients with VTE and atrial fibrillation for a safer and rapid anticoagulation response. The primary outcome was to determine time to first therapeutic INR and time to first episode of potential bleeding (INR \geq 4). The sample size (N= 150) was divided into 3 groups: CYP2C9 wild-type (*1/*1), CYP2C9 that included 1 or 2 of the carriers (*1/*3, *2/*2, *3/*3, or *2/*3), and VKORC1 wild-type. The time to first stable anticoagulation was statistically significant between the VKORC1 group (P < 0.05), but no differences were found in the CYP2C9 groups (P = 0.37). VKORC1 showed more importance in determining early response to warfarin therapy compared to CYP2C9 wild-types. The time to first therapeutic INR (2.0 to 3.0) and over-anticoagulation (INR \geq 4) for the VKORC1 genotype and CYP2C9 wild-types demonstrated no statistical differences. In contrast, Voora et al. and several other studies have shown an increased risk of over-anticoagulation in patients with variant CYP2C9 genotypes.⁵

Figure 1. Mechanism of action and metabolism of warfarin^{2,3}



Pharmacogenetic Variations among Ethnic Populations

Relating to the El Paso and surrounding communities, pharmacogenetic variations in VKORC1 and CYP2C9 alleles have been studied in Hispanics. Since warfarin is the most commonly prescribed anticoagulant, Palacio et al. were interested in the impact of VKORC1 and CYP2C9 variants influencing warfarin dose requirements in the Hispanic population. A total of 191 patients were recruited and three groups were identified based on the 25th and 75th percentiles

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Table 1. Studies related to Pharmacogenomics Testing in Warfarin^{5,6,10}

Author/Year	Gong et al. 2011	Palacio et al. 2010	Patrick et al. 2009
Study Design	Prospective cohort study	Retrospective study	Cohort Study
Objective	To refine the loading and maintenance dose algorithm after monitoring an initial cohort of patients for application in a final cohort	To provide a pharmacogenomics model outlining the response to warfarin therapy in Hispanic initiation could be cost-effective	To evaluate under what circumstances CYP2C9 and VKORC1 genotyping before warfarin
Outcomes	Primary: Time to first therapeutic INR, time to first overanticoagulation (INR \geq 4); Secondary: Time to first stable anticoagulation, time spent in therapeutic range, and time spent above therapeutic range during the first 30 days and after 30 days	Primary: Mean stable daily dose, mean daily dose based on age and weight, mean stable daily dose in sensitive, intermediate, and resistant groups, warfarin dose requirements in VKORC1 and CYP2C9 genotypes Secondary: Influence of concurrent medications on warfarin	Primary: Threshold analysis to assess the test characteristics under which genetically-guided dosing would be cost-effective
Number of Subjects	150	191	-
Duration	60 days	6 months	-
Results	<ul style="list-style-type: none"> No significance to time required to reach the first therapeutic INR or time required to obtain an INR \geq 4 in VKORC1 (P=0.52; P=0.64) or CYP2C9 wild-types (P=0.28; P=0.96) Time to stable anticoagulation was significantly different between VKORC1 (P<0.05) and no differences between CYP2C9 groups (P=0.37) 	<ul style="list-style-type: none"> Mean stable daily warfarin dose was 4.47 ± 2.09 mg with a mean INR of 2.58 ± 0.29 (range 1.93-3.26) Mean daily dose for sensitive, intermediate, and resistant groups (2.28 ± 0.49, 4.2 ± 0.76, and 7.4 ± 1.54 mg/day) and average age were statistically significant (63.16 ± 16.13, 52.88 ± 16, and 47.10 ± 17.07 years; P < 0.001) Warfarin dose was significantly and inversely correlated with age ($r=-0.45$); P<0.001) BSA and BMI were not related to daily dose of warfarin dose requirement Atrial fibrillation carried the lowest dosing requirement among diagnoses (P=0.02) VKORC1 heterozygous had a significant effect on daily warfarin requirement (P<0.001) 	<ul style="list-style-type: none"> If genotyping increases time spent in range by <5 percentage points, incremental cost-effective ratio (ICER) would be <\$100,000 per QALY and <\$50,000 per QALY if increased to time to range by 9 percentage points. 2.8% of patient-time shifted from high INRs: 2.3% from the INR 3 to 4 range and 0.5% from the INR >4 range. When the amount of patient-time shifted from the high INR range was increased to 3.4% (2.4% from the INR 3 to 4 range and 1% from the INR >4 range), the ICER of genotyping fell to \$31,000/QALY. When the patient-time shifted from the high INR range was reduced to 2.1% (1.9% from the 3 to 4 range and 0.2% from the >4 range) the ICER increased to \$82,000/QALY
Conclusion	First prospective study to demonstrate the utility of a genotype-guided warfarin initiation algorithm and can be used for outpatient management in patients with genomic variants requiring warfarin	Consider pharmacogenomics testing in Hispanics upon the initiation of warfarin. Mean warfarin daily dose was significantly lower in patients with the wild-type allele and VKORC1 G/A variant had a significant effect on daily warfarin dose requirements. Homozygous (GG) and heterozygous (GA) patients required higher doses than patients with AA genotype	The investigators found that the test would be cost-effective if it showed a 10% improvement in time to therapeutic range. Based on the results, if approximately 300,000 patients were started on warfarin, pharmacogenetic testing would prevent 300 major bleeding events with a cost of more than \$113 million per year
Limitations	Study not powered to detect secondary outcomes and lack of control group	Inclusion of patients with a stable INR; study not powered to detect significant differences in BMI, BSA, or gender	Specific patient population, based bleeding data from a previous study in patients starting warfarin up to one year, the results only pertain to the model-based analysis

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of mean daily doses, which ranged from ≤ 2.86 mg/d and ≥ 5.54 mg/d. The groups identified patients as sensitive (2.28 +/- 0.50 mg/d), intermediate (4.2 +/- 0.76 mg/d), and resistant (7.40 +/- 1.54 mg/d; $P < 0.001$). Higher warfarin dosing was required in patients with CYP2C9 allele *1 ($P = 0.006$) and VKORC1 genotypes ($P < 0.001$) and an increase in age demonstrated an inverse relationship ($P < 0.001$; effective dose diminished 0.56 mg/d/decade). A linear regression analysis demonstrated that 38.2% of warfarin responses was influenced by age (20.9%), VKORC1 (11.3%), and CYP2C9 *1, *2, and *3 variants (7.1%). One study found that a Caucasian population showed lower dose requirements in patients with heterozygous variations in either CYP2C9 *2 and *3 alleles. While another study found that Chinese and Malaysian patients required higher warfarin doses with a genetic variation in CYP2C9 *1/*1 genotype (93.2%) and lower dosing in CYP2C9 *1/*3 genotype (6.7%).⁶

Cost of Pharmacogenetic Testing

There is a lack of data exploring economic implications of pharmacogenetic testing, and it is a relatively new technology that is still being studied. Several private companies have emerged that offer polymerase chain reaction (PCR) tests for CYP2C9 and VKORC1 genes. These tests can cost between \$250-\$600 per patient. Also, the question remains whether health insurance providers will cover this testing. Currently, coverage varies among health plans and certain criteria must be met for a patient to be eligible. For example, one insurance provider states that "the testing method has been scientifically proven to show a relationship between a specific gene biomarker or gene mutation and a specific therapeutic drug target," in order for this to be eligible for insurance coverage.⁷ An estimated 2 million patients start warfarin therapy each year, which could translate to approximately \$1 billion in healthcare costs. However, this estimate does not take into account the cost of pharmacogenetic testing. With inconsistent evidence supporting the use of such testing, at this time it seems that the cost is not warranted.^{8,9}

The COUMAGEN trial investigated the effectiveness of comparing two pharmacogenetic algorithms and standard care for dosing of warfarin. The model used in the study to estimate cost-effectiveness found a minor improvement in Quality Adjusted Life Years (QALY) (0.003) at the cost of \$162. A prior analysis showed cost-effectiveness in elderly patients diagnosed with atrial fibrillation to reduce over-anticoagulation. Patrick et al. used a Markov model to determine if pharmacogenetic testing was indeed cost-effective. The investigators found that the test would be cost-effective if it showed a 10% improvement in time to therapeutic range. Based on the results, if approximately 300,000 patients were started on warfarin, pharmacogenetic testing would prevent 300 major bleeding events with a cost of more than \$113 million per year. The investigators concluded that the need for testing should be considered in patients with inconsistent INR levels.^{4,10}

Implications for Clinical Practice

Even though pharmacogenomic testing requires further evaluation, healthcare professionals should be well aware of the possible impact that CYP2C9 and VKORC1 variants may have on warfarin therapy. Indications for genetic testing may be for individualized dosing of warfarin, increased safety and efficacy of therapy, improvement in quality, and decreased expenses of managing adverse events. Arguments against genetic testing include availability, cost, time-consumption, and unlikelihood of life-threatening

bleeding.⁸ For further insight to pharmacogenetic testing, pharmacists can provide information about the appropriate use and availability of genetic testing. It is necessary for the provider or pharmacist to educate the patient regarding the signs and symptoms of over-coagulation and clotting risks in warfarin therapy. For the time being, continue to follow the current guidelines on initiation and maintenance dosing of warfarin. The American College of Chest Physicians has recently updated warfarin-dosing requirements in atrial fibrillation, VTE, and other indications requiring anticoagulation. Guidelines can be accessed through the following website: <http://journal.publications.chestnet.org/ss/guidelines.aspx>. Most of the studies discussed throughout the article are described in detail below (refer to Table 1).

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