For the treatment and prevention of thrombo-embolic disease, the most frequently used anticoagulant drug worldwide is warfarin, an oral coumarin derivative, with more than 30 million prescriptions written for this drug in the United States in 2004.\(^{(1)}\) The drug has a narrow therapeutic index and its metabolism varies by as much as a factor of 10 among individual patients, making warfarin therapy difficult to manage. Hemorrhagic complication rates of warfarin are estimated to be 5-7.9% for major (life threatening) hemorrhage and 14-36% for minor hemorrhage (e.g. nosebleeds, microscopic hematuria).\(^{(2)}\) This condition makes it difficult to establish the appropriate dose of warfarin.

Warfarin is administered as a racemic mixture of S- and R-enantiomers (mirror-image isomers), which differ in metabolism and potency, the S-enantiomer being the more potent form with a variable metabolism, resulting in the large variability in warfarin dose requirements. Of the overall anticoagulation response, 60-70% is due to the S-enantiomer, while the R-enantiomer is responsible for the remaining 30-40%. Metabolism of the enantiomers occurs in the liver via distinct cytochrome P450 (CYP) enzymes. In S-warfarin metabolism, CYP2C9 serves as the principal enzyme and CYP2C8, CYP2C18, CYP2C19 as minor metabolic pathways. R-warfarin is mainly metabolized by CYP1A2 and CYP3A4, with CYP1A1, CYP2C8, CYP2C18, CYP2C19, and CYP3A4/5 serving as minor pathways to inactive metabolites.\(^{(3)}\)

The pharmacological effect of warfarin is produced by the drug interfering with the synthesis of vitamin K-dependent clotting factors via inhibition of vitamin K\(_1\) 2,3-epoxide reductase complex, subunit 1 (VKORC1). This interferes with the post-translational gamma-carboxylation of glutamic acid residues on coagulation factors II, VII, IX, and X, as well as the anticoagulant proteins C and S. Depletion of reduced vitamin K leads to the production of nonfunctional coagulation factors, resulting in anticoagulation.\(^{(3)}\)

Resistance to warfarin has been described as the inability to prolong the prothrombin time or raise the international normalized ratio (INR) into the therapeutic range when the drug is given at normally prescribed doses. However, a higher warfarin requirement does not itself establish the diagnosis of warfarin resistance. The range of normally recommended daily or weekly warfarin doses to maintain a therapeutic prothrombin time or INR depends on the study population. Patients who need more than 105 mg per week (15 mg/day)\(^{(4)}\) should be considered warfarin resistant. It should be noted that another reference paper gives the lower dose of 70 mg per week.\(^{(3)}\) An important characteristic of warfarin resistance is that patients need much smaller doses of vitamin K to reverse the effect of warfarin.
Warfarin resistance can be classified in practical terms as acquired and hereditary. Acquired resistance to warfarin is most frequently the result of poor patient compliance, but may also be due to other causes, such as high consumption of vitamin K, decreased absorption or increased clearance of warfarin, and drug interactions. Hereditary resistance is presumably governed by genetic factors that result in faster metabolism (pharmacokinetic resistance) or in lower activity of the drug (pharmacodynamic resistance).

Genetic variability are important in causing warfarin resistance or warfarin sensitivity, the latter being associated with some VKORC1 and CYP2C9 variant alleles. The variant alleles CYP2C9*2 and *3 are more prevalent in European Americans than in African Americans, while in Asian populations, CYP2C9*2 has a frequency of 2-4%, and *3 alleles are not commonly found. In a meta-analysis of nine studies involving almost 3000 warfarin patients it was shown that carriers of CYP2C9*2 and *3 had lower mean daily doses and a significantly higher risk of hemorrhage. The relative bleeding risk for CYP2C9*2 and *3 was 1.91 (1.16-3.17) and 1.77 (1.07-2.91), respectively.

Inter-individual variability in warfarin dose among patients of European, Asian and African descent has been associated with the prevalence of variant VKORC1 1173 C>T and 1693 G/A. Several rare VKORC1 missense mutations have been found in patients with warfarin-resistance, requiring the higher doses of >70 mg/week. Although the frequency and clinical impact of these mutations at the population level is poorly documented, they may be clinically relevant in compliant patients requiring higher warfarin doses.

Warfarin resistance is diagnosed through patient history and laboratory studies, and the search for potential causes requires a full drug and dietary history. Subtherapeutic plasma warfarin levels should alert the pharmacologist to the possibility of intestinal malabsorption or poor compliance. The range of therapeutic total plasma warfarin levels is 0.5 – 3.0 µg/mL, depending on the laboratory and patient population.

To determine the type of warfarin resistance, drug absorption and clearance is evaluated by determination of plasma levels at specific intervals after administration, commonly every 60 to 180 minutes. The clearance rate of the S-enantiomer is normally twice that of the R-enantiomer (5.2 vs 2.5 mL/min/70 kg). A normal clearance rate precludes the possibility that the resistance is caused by enhanced drug elimination. In addition, Bentley et al. have devised an algorithm for using the plasma warfarin levels to determine the type of resistance. A clotting assay of factors II, VII, IX, and X has been used for determining warfarin dose. In patients with an unreliable or prolonged baseline prothrombin time and INR, some studies have targeted a factor II and X activity level of 10% - 30% of normal biologic activity for achieving a therapeutic warfarin effect. Plasma warfarin levels are typically measured with a turn-around time of 2 to 7 days, as opposed to 24 hours for factor II and X activity.

Treatment of warfarin-resistant cases should be based on the cause. Patient education is necessary to improve compliance and to reduce adverse effects of warfarin therapy, regardless of the dose. In true hereditary warfarin resistance, treatment may be effected by increasing warfarin dosage (possibly attaining 100 mg/day or more), or by switching to other types of anticoagulant. There are several alternative anticoagulants, such as subcutaneous heparins (unfractionated and low-molecular-weight heparins), fondaparinux (Arixtra, a subcutaneous factor Xa inhibitor), Dabigatran (an oral direct thrombin inhibitor, currently undergoing phase 3 studies for use in long-term anticoagulation), Rivaroxaban (a direct factor Xa inhibitor), and vitamin K antagonists (bishydroxycoumarin, phenprocoumon, acenocoumarol, phenindione).
In 2007, the Food and Drug Administration (FDA) added pharmacogenetic information to the warfarin package insert, presumably in recognition of the fact that genetic variations in the CYP2C9 and VKORC1 genes contribute significantly to the variability in dose requirements for warfarin. However, the FDA did not propose a specific method for using genetic information to predict the dose required in individual patients.\textsuperscript{(9)} One study on the use of a pharmacogenetic algorithm for estimating the appropriate initial dose of warfarin yielded recommendations that were significantly closer to the required stable therapeutic dose.\textsuperscript{(10)} On the other hand, the review by Limdi et al.\textsuperscript{(3)} did not find supporting evidence to suggest that genotype-guided therapy will improve anticoagulant control and prevent or reduce the risk of hemorrhagic or thromboembolic complications. In any case, it would be both reasonable and prudent to use CYP2C9 and VKORC1 genotypes as part of diagnostic efforts to understand unusual responses to standard medical care.

REFERENCES