Warfarin Therapy in a Patient Homozygous for the CYP2C9*3 Allele

Jacob Ablin MD[1], Shaltiel Cabili MD[1], Ayala Lagziel PhD[2] and Hava Peretz PhD[2]

[1] Department of Internal Medicine F, Tel Aviv Sourasky Medical Center and Sackler Faculty of Medicine, Tel Aviv University, Ramat Aviv, Israel
[2] Clinical Biochemistry Laboratory, Tel Aviv Sourasky Medical Center and Sackler Faculty of Medicine, Tel Aviv University, Ramat Aviv, Israel
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A component of cytochrome P450, CYP2C9, is an enzyme of major importance in the metabolism of S-warfarin. Three allelic variants of this enzyme have been identified: CYP2C9*1, CYP2C9*2 and CYP2C9*3. Alleles 2 and 3 differ from the wild-type allele 1 by amino acid substitutions Arg144Cys and Ile359Leu, respectively. Both allelic variants possess decreased enzymatic activity and hence metabolize warfarin at a slower rate. Clinically, these two mutations have been shown to cause increased susceptibility to warfarin, manifested by abnormally low dose requirements, as well as by an increased rate of hemorrhagic complications during treatment [1,2]. Surprisingly however, while individuals heterozygous for alleles 2 and 3 were over-represented among a group of warfarin-treated individuals who required exceptionally low doses for the achievement of anticoagulation, very few patients homozygous for the CYP2C9*3 have been reported. One possible explanation for this apparent paradox is that patients homozygous for the CYP2C9*3 variant allele are so sensitive to the effect of warfarin that therapy is withheld at an early stage.

We present a case in which achieving effective warfarin therapy was difficult, associated with a homozygous genotype for the CYP2C9*3 allele. The potential clinical utility of this novel genetic test is discussed.

Patient Description

An 83 year old man was hospitalized due to pulmonary congestion. He had a history of ischemic heart disease and congestive heart failure, and had undergone coronary bypass surgery 5 years prior to his hospitalization. In addition, he suffered from chronic atrial fibrillation and 2 years previously had sustained a right hemiparesis with motor aphasia. Additional medical problems included hypertension, partial gastrectomy due to leiomyoma, and colectomy due to carcinoma. The patient was receiving chronic treatment with warfarin at a weekly dose of 17.5 mg, as well as verapamil, enalapril, aspirin, furosamide and famotidine.

On physical examination fine crepitations were noted on both lung fields. There was no evidence of bleeding. Laboratory tests showed a hemoglobin concentration of 10 g/dl, and normal levels of serum electrolytes. The INR value on admission was 6.0. During hospitalization the patient was treated for congestive heart failure. Despite discontinuation of warfarin therapy, INR values continued to increase with no accompanying deterioration in liver function. There was no bleeding. The

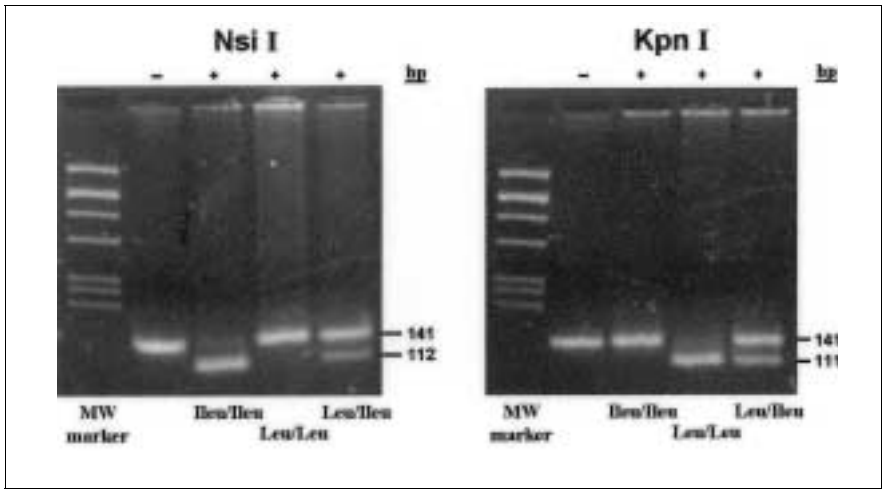

Figure 1. Polymerase chain reaction-restriction fragment length polymorphism analysis of CYP2C9 Ileu359Leu polymorphism. Ethidium bromide-stained agarose gels showing separation of Nsi I (left panel) and Kpn I (right panel) digestion products of amplified DNA. **Lane 1:** pBR322/Alu I molecular weight marker. **Lane 2:** undigested polymerase chain reaction product of 141 base pairs. **Lane 3:** control DNA sample with the wild-type genotype, Ileu/Ileu, **Lane 4:** DNA of patient. Note lack of digestion by Nsi I and full digestion by Kpn I, demonstrating homozygosity for the variant, 359Leu allele. **Lane 5:** DNA sample of a heterozygous carrier of the variant, 359Leu allele.

patient was discharged with careful explanations on the monitoring of warfarin therapy including testing of INR values twice a week, and with a recommended weekly dose of 14 mg. Nevertheless, several days later he was readmitted due to non-measurably high INR values. The patient was subsequently discharged without warfarin therapy. Due to the unusual difficulty observed in achieving stable anticoagulation in this patient, genetic analysis was undertaken to determine his CYP2C9 genotype as previously described [3]. As can be seen in Figure 1, the patient was found to be homozygous for the variant, 359Leu allele.

In view of this finding it was decided not to reinstate warfarin therapy. Four months later the patient was doing well, and the INR was normal (1.07).

Comment

Oral anticoagulation with warfarin is the standard treatment for diverse situations, ranging from classical indications, such as deep vein thrombosis, atrial fibrillation and prosthetic heart valves – to the recently expounded ones, such as ischemic heart disease. Unfortunately, despite growing recognition of the importance of anticoagulant therapy, treatment is withheld from many patients who need it [4]. Evidently, ongoing concern regarding the risk of hemorrhage, and the notorious variability in individual response to treatment, as well as a narrow therapeutic index, continue to hamper appropriate use of warfarin.

The genetic variability in warfarin metabolism is a recently identified factor assumed to influence the clinical goal of reaching and maintaining therapeutic INR levels [1,2]. Cytochrome P-450 CYP2C9 catalyzes the conversion of the more potent enantiomeric form, S-warfarin, to inactive metabolites. Two allelic variants, CYP2C9*2 and CYP2C9*3, reduced 12% and 5%, respectively, the *in vitro* capacity to metabolize S-warfarin compared to that of the wild-type allele. In a study by Aithal et al. [1], patients heterozygous for both CYP2C9*2 and CYP2C9*3 were more likely than controls to be among those requiring very low doses of warfarin for maintenance of anticoagulation, and were significantly more likely to suffer from complications upon initiation of therapy. Unpredictably, although 28% of a group of patients receiving very low doses of warfarin were found to be heterozygous for CYP2C9*3, no such patient was found to be homozygous for this allele. As the authors suggest, this apparent under-representation of homozygous patients may be explained by the fact that homozygous patients are so difficult to maintain on stable anticoagulation that such therapy is invariably withheld. Similarly, in a recent study testing 561 warfarin-treated patients, among whom the gene frequency of CYP2C9*3 was determined to be 0.05%, no patients homozygous for this allele were identified [2]. The authors similarly suggest that homozygosity may preclude warfarin therapy due to increased complications, although no such finding was identified among patients homozygous for the CYP2C9*2 allele or among heterozygous individuals. The fact that our patient – although treated with a low dose – was eventually withdrawn from warfarin therapy, despite an ongoing indication, tends to support the assumption that the CYP2C9*3 is rarely found among patients experiencing difficulty with warfarin therapy simply because it makes such treatment unfeasible.

The importance of acquiring the capability to analyze the CYP2C9 genotype pertains to the novel possibility of performing pharmacogenetic matching between a particular drug and the patient for whom the drug is considered. Though some patients have absolute indications for anticoagulation, in cases of less compelling necessity physicians may choose to abstain from administering warfarin to patients known to be carriers of the CYP2C9 variant alleles, as was done in the present case. On the other hand, genetic information on the CYP2C9 variants may prevent hemorrhagic complications during the initiation of therapy by dictating the use of low doses, careful adjustments, and possibly increased frequency of INR measurements. Particular attention could be given to the interaction of warfarin with other drugs known to be metabolized by the CYP2C9 system, such as losartan and celecoxib. The patient's age should also

be taken into account, since it was shown that the clearance of warfarin decreases with age and might be half or less than normal by the age of 80 years [5]. Possibly even more significantly, knowledge that a patient is a carrier of the wild-type allele may serve to reduce concern about the risk of hemorrhage and thus increase the proportion of patients receiving this important mode of therapy. A cost analysis is called for in order to determine whether the potential savings attainable through this strategy justify the widespread implementation of such genetic testing for patients who are candidates for anticoagulation.

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Correspondence: Dr. H. Peretz, Clinical Biochemistry Laboratory, Tel Aviv Sourasky Medical Center, Tel Aviv 64239, Israel.
Phone: (972-3) 697-3406
Fax: (972-3) 691-8599.
email: hperetz@tasmc.health.gov.il