



EDUCATIONAL OBJECTIVE: Readers will recognize and appropriately manage warfarin resistance in patients who need higher-than-expected doses of this drug

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An algorithm for managing warfarin resistance

ABSTRACT

Some patients need higher-than-expected doses of warfarin (Coumadin) to get their international normalized ratio (INR) into the therapeutic range. The cause of warfarin resistance can be either acquired (eg, poor compliance, drug interactions, dietary interactions) or hereditary, but the genetic mechanisms of warfarin resistance are not well understood. This review offers an algorithm for the evaluation of patients with suspected warfarin resistance.

KEY POINTS

The most common cause of warfarin resistance is non-compliance. Others include poor absorption, high vitamin K intake, hypersensitivity to vitamin K, and rapid drug deactivation.

Patient education is necessary to improve compliance and to mitigate adverse effects of warfarin therapy, regardless of the dose.

In time, it may be possible to individualize anticoagulant dosing on the basis of genetic testing for patients with warfarin resistance, although currently such tests are not routinely advocated and are usually done only in specialized laboratories.

In true hereditary warfarin resistance, there are two approaches to treatment: increase the warfarin dosage (perhaps to as high as 100 mg/day or more), or switch to another anticoagulant.

WARFARIN (COUMADIN) differs from most other drugs in that the dosage required to achieve a desired therapeutic effect varies greatly among individuals. This variability can lead to therapeutic failure, potentially resulting in new thrombosis, or, at the other extreme, to life-threatening bleeding.

Further, there is no reliable means to identify patients who require unusually high doses of warfarin, although genetic testing may become available in the future.

See related patient information at
http://my.clevelandclinic.org/drugs/Coumadin/hic_Understanding_Coumadin.aspx

Warfarin, a coumarin derivative first synthesized in 1948, is still the only oral anticoagulant available for long-term use in the United States. Indications for its use include the treatment and, to a lesser extent, the prevention of arterial and venous thromboembolism. It is also used for long-term anticoagulation in patients with atrial arrhythmias (atrial fibrillation and atrial flutter) and mechanical heart valves.

In the paragraphs that follow, we review the causes of warfarin resistance and how to recognize and manage it.

WHAT IS WARFARIN RESISTANCE?

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.¹

However, a higher warfarin requirement

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Warfarin is metabolized by P450 enzymes

Warfarin is a racemic mixture of R- and S-enantiomers (mirror-image isomers), which differ in their potency and metabolism.^{5,6} The left-handed S-enantiomer is three to five times as potent as the right-handed R-enantiomer.

However, warfarin is hepatically metabolized by the cytochrome P450 complex, and although the S-isomer is more potent, the R-isomer has a longer half-life. This is because S-warfarin is metabolized faster (via 7-hydroxylation by CYP2C9) than R-warfarin (which is metabolized via 10-hydroxylation by CYP1A1, CYP1A2, and CYP3A4).⁷ Effectively, S-warfarin accounts for 60% to 70% of the overall anticoagulation response, while the R-enantiomer is responsible for approximately 30% to 40%.⁸

The steady-state concentration of warfarin

is affected by the dose, by CYP2C9-mediated metabolism of the S-enantiomer, by elimination of hydroxyl metabolites, by gastrointestinal absorption (diminished by diarrhea or vomiting), by non-CYP2C9 metabolism, by the patient's nutritional state and diet, and by drug interactions.⁹

Warfarin is rapidly absorbed from the gastrointestinal tract after oral administration, with a bioavailability of 100%,^{10,11} and its peak absorption is usually seen within 60 to 90 minutes. It can also be given intravenously and sublingually.

Warfarin is highly (97%–99%) bound to plasma proteins, primarily to albumin, with a volume of distribution of 0.12 to 0.13 L/kg.¹⁰ Its mean half-life is 44 hours (range 20–60).

does not itself establish the diagnosis of warfarin resistance. The prevalence of warfarin resistance varies by patient population and is difficult to determine. The difficulty lies largely in accounting for dietary factors and in defining normal metabolic variations among individuals.

The range of normally recommended daily or weekly warfarin doses to maintain a therapeutic prothrombin time or INR depends on the study population. Nevertheless, patients who need more than 105 mg per week (15 mg/day) should be considered warfarin-resistant. These patients are likely to be in the top 5% for warfarin doses within an anticoagulated cohort.

Warfarin resistance is different than warfarin failure, which is defined as a new thrombotic event despite a therapeutic prothrombin time and INR. This situation is commonly seen in patients with malignant diseases.

An important characteristic of warfarin resistance is that patients need much smaller doses of vitamin K to reverse the effect of warfarin.² Thijssen³ showed that, in warfarin-resistant rats, warfarin did not irreversibly inhibit vitamin K₁ 2,3-epoxide reductase (VKORC1) activity. This is consistent with the vitamin K hypersensitivity observed in warfarin-resistant people.^{2,3}

WHAT CAUSES WARFARIN RESISTANCE?

Warfarin resistance can be classified in practical terms as acquired vs hereditary, or in mechanistic terms as pharmacokinetic vs pharmacodynamic.

Acquired vs hereditary resistance

Hulse⁴ categorizes warfarin resistance as either acquired or hereditary.

Acquired resistance to warfarin may result from:

- Poor patient compliance (the most common cause)
- High consumption of vitamin K
- Decreased absorption of warfarin
- Increased clearance (see **WARFARIN IS METABOLIZED BY P450 ENZYMES** on this page^{5–11})
- Drug interactions (**TABLE 1**).^{12,13}

Hereditary resistance has been postulated to be caused by genetic factors that result either in faster metabolism of the drug (a form of pharmacokinetic resistance) or in lower activity of the drug (pharmacodynamic resistance). Polymorphisms may play a role, as some VKORC1 and CYP2C9 variant alleles are known to be associated with increased sensitivity to warfarin.¹⁴

However, the genetic mechanisms of warfarin resistance are not clearly understood,

**Patients
needing
> 15 mg/day
should be
considered
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resistant**

despite several case reports of hereditary resistance confirmed by similar patterns of resistance in immediate family members.¹⁵⁻¹⁹ More than one mechanism is likely. There is ample room for further insight into genetic polymorphisms underlying hereditary warfarin resistance. More on this topic is included in the sections below.

Pharmacokinetic resistance

Pharmacokinetic resistance can result from diminished absorption or increased elimination of the drug. Causes of diminished absorption include emesis, diarrhea, and malabsorption syndrome.

The mechanism of increased warfarin clearance has not been delineated, although the following have been implicated.

Genetic factors. Duplication or multiplication of cytochrome P450 enzyme genes has been described as contributing to a phenotype of ultrarapid metabolism. Some people may carry multiple copies of the CYP2C9 gene, as has already been reported for cytochrome P450 CYP2D6 and CYP2A6.^{7,8} It is also plausible that rare allelic variants of CYP2C9 exist that are associated with higher-than-normal activity, given that there are alleles known to predispose to warfarin sensitivity.

Hypoalbuminemia may increase the free fraction of warfarin, leading to enhanced rates of clearance and a shorter plasma half-life.¹⁵

Hyperalbuminemia may paradoxically also contribute to warfarin resistance via drug binding.

Hyperlipidemia. Several observers have found that lowering serum lipids, primarily triglycerides, increases the sensitivity to warfarin irrespective of the means used to achieve this decrease.²⁰ This most likely results in a decreased pool of vitamin K, some of which is bound to triglycerides.²¹ Conversely, patients receiving intravenous lipids with total parenteral nutrition have also been diagnosed clinically with warfarin resistance,²² and rat models have shown an association between a lipid-rich diet and increased vitamin K-dependent factor activity.²³

Diuretics may decrease the response to warfarin by reducing the plasma volume, with a subsequent increase in clotting factor activity.²⁴

TABLE 1

Drugs and supplements that potentiate or inhibit warfarin

Potentiate warfarin

Acetaminophen (Tylenol)
Alcohol
Allopurinol (Zyloprim)
Amiodarone (Cordarone)
Amoxicillin-clavulanate (Augmentin)
Aspirin
Celecoxib (Celebrex)
Ciprofloxacin (Cipro)
Erythromycin
Fenofibrate (Tricor)
Fluconazole (Diflucan)
Fluvastatin (Lescol)
Garlic
Ginkgo
Levofloxacin (Levaquin)
Levothyroxine (Synthroid)
Nonsteroidal anti-inflammatory drugs
Omeprazole (Prilosec)
Paclitaxel (Taxol)
Propafenone (Rythmol)
Ritonavir (Norvir)
Tramadol (Ultram)
Trimethoprim-sulfamethoxazole (Bactrim)
Vitamin E

Inhibit warfarin

Azathioprine (Imuran)
Barbiturates
Bosentan (Tracleer)
Carbamazepine (Tegretol)
Cholestyramine
Cortisone
Dicloxacillin
Etodolac (Lodine)
Ginseng
Haloperidol (Haldol)
Mercaptopurine (Purinethol)
Multivitamins
Nafcillin (Unipen)
Oral contraceptives
Parenteral and enteral nutritional supplements
Ribavirin (Rebetol)
Rifampin (Rifadin)
Ritonavir (Norvir)
Spironolactone (Aldactone)
Trazodone (Desyrel)
Vitamin C
Vitamin K

Warfarin-resistant patients need much smaller doses of vitamin K to reverse the effect of warfarin

How warfarin works

Warfarin exerts its anticoagulant and antithrombotic activity by inhibiting the vitamin K-dependent carboxylation of clotting factors II, VII, IX, and X to a greater extent than the vitamin K-dependent natural anticoagulants, proteins C and S.^{10,27} Specifically, it inhibits an enzyme called the vitamin K₁ 2,3-epoxide reductase complex, subunit 1 (VKORC1).²⁸

In an oxidation-reduction cycle (known as the vitamin K cycle, **FIGURE 1**), vitamin K is converted to its active quinone form by two enzymes, VKORC1 (which is blocked by warfarin) and DT-diaphorase (which is not affected by warfarin). The active form is a required cofactor (along with gamma-glutamyl carboxy-

lase) in the posttranslational carboxylation of factors II, VII, IX, and X and proteins C and S.² In the process, a vitamin K epoxide forms and needs to be reduced back to vitamin K by VKORC1, completing the cycle.^{29,30}

When oral warfarin therapy is started, it begins to inhibit the synthesis of clotting factors that have a short half-life (ie, factor VII and protein C) within 12 to 24 hours. However, warfarin does not achieve its full antithrombotic effect until 5 to 7 days after initiation.^{10,27} This is the time it needs to reach a steady-state concentration and to suppress the clotting factors with the longest half-lives, namely, factor X, which has a half-life of 30 to 40 hours, and factor II, with a half-life of 60 to 70 hours.^{11,27}

How warfarin blocks the vitamin K cycle

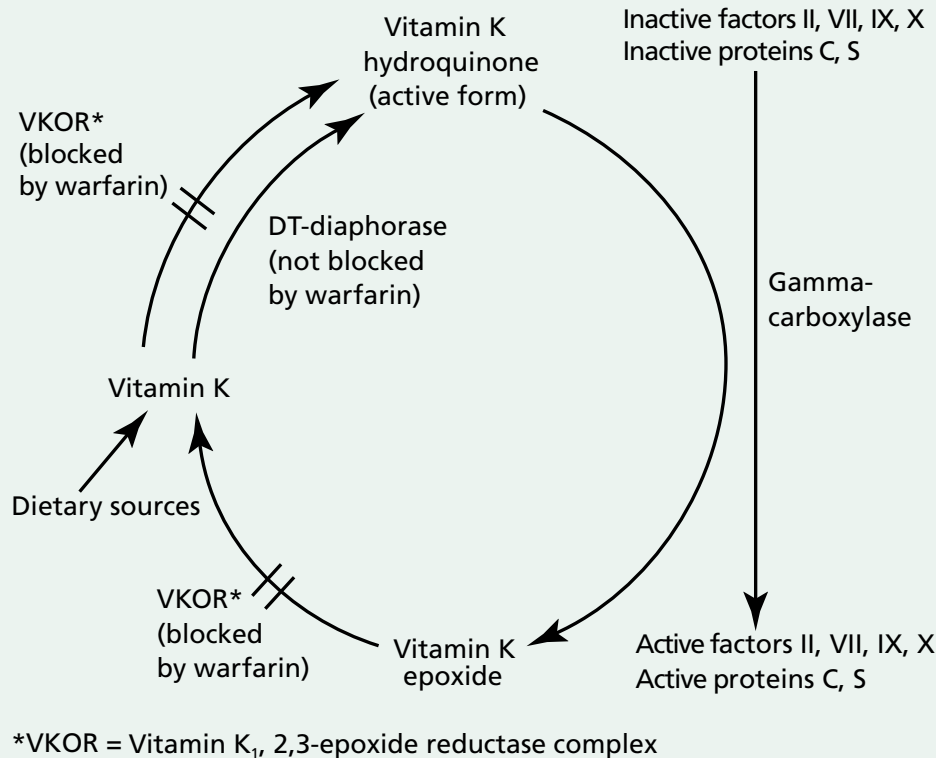


FIGURE 1

Pharmacodynamic resistance

Potential mechanisms of pharmacodynamic warfarin resistance described in rats and in people include:

- Increased affinity of vitamin K₁ 2,3-epoxide reductase complex (VKOR) for vitamin K^{25,26} (see **HOW WARFARIN WORKS** on this page^{2,10,11,27-30})

- Prolongation of normal clotting factor activity¹⁶
- Production of clotting factors that is not dependent on vitamin K¹⁶
- Decreased VKOR sensitivity to warfarin.²⁶

In rats, these mechanisms are manifested by relatively high doses of warfarin being required to achieve poisoning. In humans, they result in high doses being needed to achieve a therapeutic effect in the setting of normal warfarin pharmacokinetics, normal warfarin concentration, and normal half-lives of blood clotting proteins.

Genetics of pharmacodynamic resistance.

Pharmacodynamic warfarin resistance has also been described with inheritance of a monogenetic dominant trait. An early study by O'Reilly²⁴ traced anticoagulation resistance to a genetically linked abnormality of interaction between warfarin and a putative vitamin K receptor.

In one patient with hereditary resistance and high warfarin requirements, a heterozygous point mutation in the VKORC1 gene was identified.³¹ This results in a substitution that lies in a conserved (normally constant or unchanging DNA sequence in a genome) region of VKORC1 that contains three of four previously identified amino acid substitutions associated with warfarin resistance (Val29Leu, Val45Ala, and Arg58Gly). Further investigation is required to fully characterize the structure-function relationship for VKORC1 and to determine the relationship between the VKORC1 genotype and other pharmacogenetic determinants of warfarin dose-response.

Separately, Loebstein et al³² reported a new mutation, Asp36Tyr, which was common in Jewish ethnic groups of Ethiopian descent (in whom the prevalence is 5%) and Ashkenazi descent (prevalence 4%). In that study, Asp36Tyr carriers needed doses of more than 70 mg per week, placing them towards the high end of the usual warfarin dosing range.

Daly and Aithal⁷ discovered that warfarin-resistant rats overexpressed a protein known as calumenin. This protein is situated in the endoplasmic reticulum and appears to interact with VKOR, decreasing the binding of warfarin. In mice, the calumenin gene is located on chromosome 7, where the gene for VKORC1 is also located.

DIAGNOSIS BY HISTORY AND LABORATORY STUDIES

A full drug and diet history is invaluable in diagnosing potential causes of warfarin resistance (TABLE 1).

Plasma warfarin levels that are subtherapeutic should raise suspicion of intestinal malabsorption or poor compliance. Poor compliance might be more appropriately seen as a mimic of warfarin resistance. Studies in humans suggest that a therapeutic total plasma warfarin level lies between 0.5 µg/mL and 3.0 µg/mL,¹⁰ though the range may vary among laboratories and patient populations.

Warfarin absorption and clearance can be evaluated by analyzing plasma levels at specific intervals after administration, eg, every 60 to 180 minutes. The drug's half-life can be determined on the basis of its concentrations in different time samples. Normally, the S-enantiomer of warfarin is cleared at twice the rate of the R-enantiomer (5.2 vs 2.5 mL/min/70 kg).⁸ A normal clearance rate confirms that resistance to warfarin is not due to enhanced elimination.

Clotting assays of factors II, VII, IX, and X may be a more precise way to assess the pharmacodynamics of warfarin,¹⁰ although there is no strong evidence to support routine use of such assays. Some studies suggest targeting factor II and factor X activity levels of 10% to 30% of normal biologic activity for a therapeutic warfarin effect in patients with an unreliable or prolonged baseline prothrombin time and INR, such as those with lupus anticoagulant.

An algorithm. Bentley et al³³ suggest using the plasma warfarin level in an algorithm to determine the type of resistance pattern. Plasma warfarin levels are typically measured by regional specialized reference laboratories with a turnaround time of 2 to 7 days, as opposed to 24 hours for factor II and X activity. Our suggested algorithm for evaluation of suspected warfarin resistance is shown in FIGURE 2.

TREAT THE CAUSE

Once the type of warfarin resistance has been determined, treatment should be oriented toward the cause.

Therapeutic warfarin levels may be 0.5–3.0 µg/mL

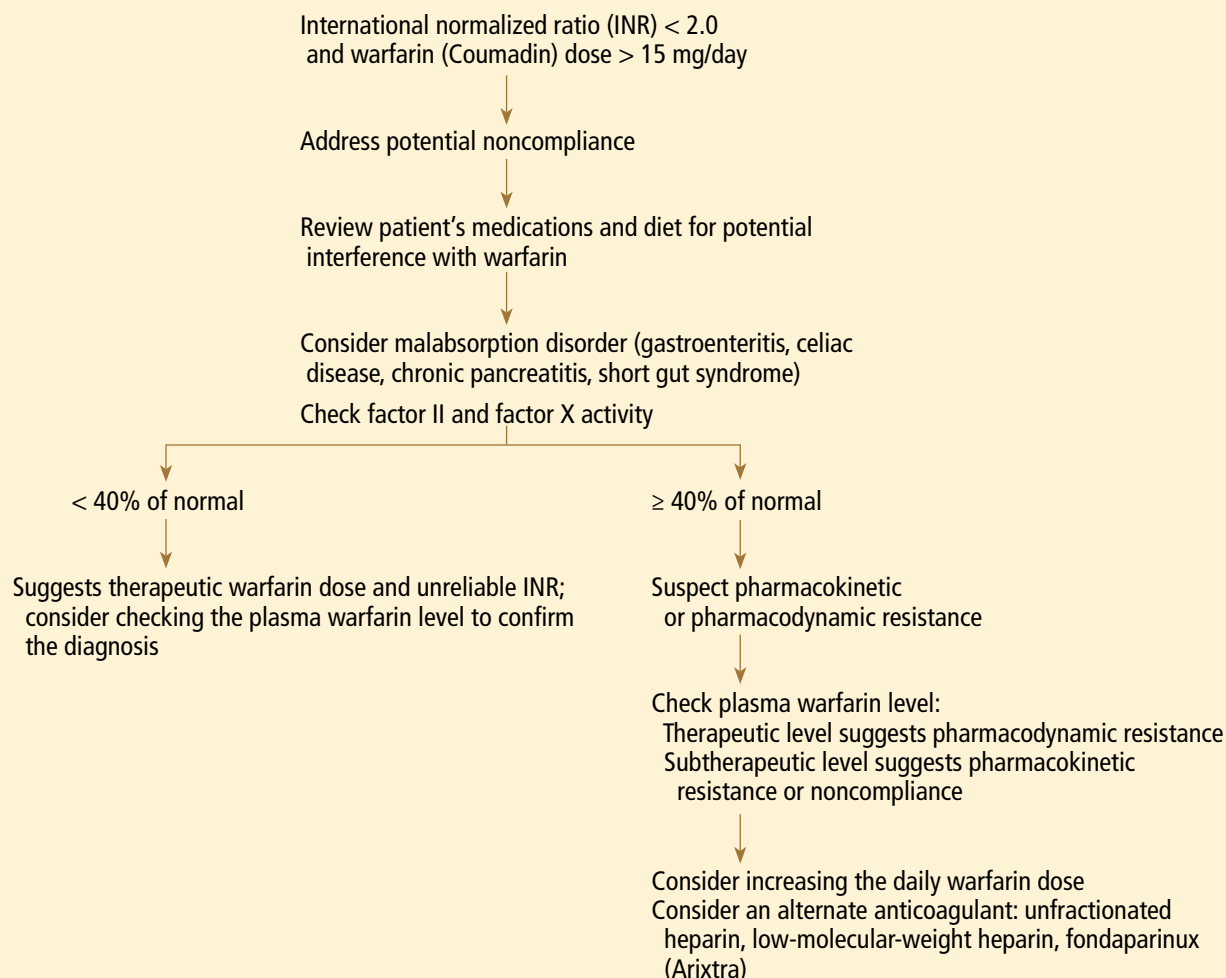


FIGURE 2. Algorithm for evaluating suspected warfarin resistance

Educate the patient

The importance of compliance should be reinforced. Educating the patient about diet and other medications that may interact with warfarin is also important. (See an example of patient education material at http://my.clevelandclinic.org/drugs/Coumadin/hic_Understanding_Coumadin.aspx.)

Increase the warfarin dose

If the patient truly has hereditary resistance, there are two approaches to treatment.

The first is to increase the warfarin dose until the prothrombin time and INR are in the therapeutic ranges. When indicated, the warfarin dose can be safely titrated upward to more than 100 mg per day in patients

who are monitored regularly—as all patients on chronic warfarin therapy should be—and whose other medications are otherwise stable. One such example is reported in a warfarin-resistant patient who needed 145 mg/day to maintain a therapeutic prothrombin time.²²

Try other anticoagulants?

The second approach is to change to another type of anticoagulant. However, there is no strong evidence in favor of this approach over prescribing larger dosages of warfarin.

Other anticoagulant drugs currently available in the United States include subcutaneous heparins (unfractionated and low-molecular-weight heparins) and the subcutaneous factor Xa inhibitor fondaparinux (Arixtra).

Agents not available in the United States include the following.

Dabigatran, an oral direct thrombin inhibitor, is undergoing phase 3 studies of its use for long-term anticoagulation.

Rivaroxaban (a direct factor Xa inhibitor) and dabigatran have been approved in Canada and the European Union to prevent venous thromboembolism after knee and hip arthroplasty, based on prospective comparisons with enoxaparin (Lovenox).³⁴⁻³⁷

Vitamin K antagonists other than warfarin that are not available in the United States include bishydroxycoumarin (which has limitations including slow absorption and high frequency of gastrointestinal side effects), phenprocoumon, and acenocoumarol. Another is phenindione, which has been associated with serious hypersensitivity reactions, some of which proved fatal and occurred within a few weeks of initiating therapy.

REFERENCES

1. Lefrere JJ, Horellou MH, Conard J, Samama M. Proposed classification of resistance to oral anticoagulant therapy. *J Clin Pathol* 1987; 40:242.
2. Linder MW. Genetic mechanisms for hypersensitivity and resistance to the anticoagulant warfarin. *Clin Chim Acta* 2001; 308:9-15.
3. Thijssen HH. Warfarin resistance. Vitamin K epoxide reductase of Scottish resistance gene is not irreversibly blocked by warfarin. *Biochem Pharmacol* 1987; 36:2753-2757.
4. Hulse ML. Warfarin resistance: diagnosis and therapeutic alternative. *Pharmacotherapy* 1996; 16:1009-1017.
5. Hirsh J, Dalen JE, Deykin D, Poller L, Bussey H. Oral anticoagulants. Mechanism of action, clinical effectiveness, and optimal therapeutic range. *Chest* 1995; 108(suppl 4):2315-2345.
6. Daly AK, King BP. Pharmacogenetics of oral anticoagulants. *Pharmacogenetics* 2003; 13:247-252.
7. Daly AK, Aithal GP. Genetic regulation of warfarin metabolism and response. *Semin Vasc Med* 2003; 3:231-238.
8. Takahashi H, Echizen H. Pharmacogenetics of warfarin elimination and its clinical implications. *Clin Pharmacokinet* 2001; 40:587-603.
9. Retti AE, Wienkers LC, Gonzalez FJ, Trager WF, Koreczekwa KR. Impaired (S)-warfarin metabolism catalysed by the R144C allele variant of CYP2C9. *Pharmacogenetics* 1994; 4:39-42.
10. Porter RS, Sawyer WR. Warfarin. In: Evans WE, Shentag JJ, Jusko WJ, editors. *Applied Pharmacokinetics. Principles of Therapeutics Drug Monitoring*, 3rd ed. Washington, DC: Applied Therapeutics, 1992:31.1-31.46.
11. Warrell DA, Cox TM, Firth JD. *Oxford Textbook of Medicine*, 4th ed. Oxford University Press, 2003:734.
12. Holbrook AM, Pereira JA, Labiris R, et al. Systematic overview of warfarin and its drug and food interactions. *Arch Intern Med* 2005; 165:1095-1106.
13. Medical Economics Staff. *Physicians' Desk Reference*, 55th Ed. Medical Economics, 2001:1139-1140.
14. Schwarz UI, Ritchie MD, Bradford Y, et al. Genetic determinants of response to warfarin during initial anticoagulation. *N Engl J Med* 2008; 358:999-1008.
15. Diab F, Feffer S. Hereditary warfarin resistance. *South Med J* 1994; 87:407-409.
16. O'Reilly RA. The second reported kindred with hereditary resistance to oral anticoagulant drugs. *N Engl J Med* 1970; 282:1448-1451.
17. O'Reilly RA, Aggeler PM, Hoag MS, Leong LS, Kropatkin ML. Hereditary transmission of exceptional resistance to coumarin anticoagulant drugs. The first reported kindred. *N Engl J Med* 1964; 271:809-815.
18. Alving BM, Strickler MP, Knight RD, Barr CF, Berenberg JL, Peek CC. Hereditary warfarin resistance. Investigation of rare phenomenon. *Arch Intern Med* 1985; 145:499-501.
19. Warrior L, Brennan CA, Lusher JM. Familial warfarin resistance in a black child. *Am J Pediatr Hematol Oncol* 1986; 8:346-347.
20. Nikkila EA, Pelkonen R. Serum lipid-reducing agents and anticoagulant requirement. *Lancet* 1963; 1:332.
21. Robinson A, Liao FO, Routledge PA, Backhouse G, Spragg BP, Bentley DP. Lipids and warfarin requirements. *Thromb Haemost* 1990; 63:148-149.
22. MacLaren R, Wachsmann BA, Swift DK, Kuhl DA. Warfarin resistance associated with intravenous lipid administration: discussion of propofol and review of the literature. *Pharmacotherapy* 1997; 17:1331-1337.
23. DeCurtis A, D'Adamo MC, Amore C, et al. Experimental arterial thrombosis in genetically or diet induced hyperlipidemia in rats—role of vitamin K-dependent clotting factors and prevention by low-intensity oral anticoagulation. *Thromb Haemost* 2001; 86:1440-1448.
24. O'Reilly RA. Drug interaction involving oral anticoagulation. In: Melmon KL, editor. *Cardiovascular Drug Therapy*, Philadelphia; FA Davis, 1975:23-41.
25. O'Reilly RA, Pool JG, Aggeler PM. Hereditary resistance to coumarin anticoagulation drugs in man and rat. *Ann N Y Acad Sci* 1968; 151:913-931.
26. Cain D, Hutson SM, Wallin R. Warfarin resistance is associated with a protein component of the vitamin K 2,3-epoxide reductase enzyme complex in rat liver. *Thromb Haemost* 1998; 80:128-133.
27. Rodvold KA, Quandt CM, Friedenberg WR. Thromboembolic disorders. In: DiPiro JT, Talbert RL, editors. *Pharmacotherapy. A Pathophysiologic Approach*, 2nd ed. New York: Elsevier, 1992:312-335.
28. Park BK. Warfarin: metabolism and mode of action. *Biochem Pharmacol* 1988; 37:19-27.
29. Cain D, Hutson SM, Wallin R. Assembly of the warfarin-sensitive vitamin K 2,3-epoxide reductase enzyme complex in the endoplasmic reticulum membrane. *J Biol Chem* 1997; 272:29068-29075.
30. Gallop PM, Lian JB, Hauschka PV. Carboxylated calcium binding proteins and vitamin K. *N Engl J Med* 1980; 302:1460-1466.
31. Rost S, Fregin A, Ivaskevicius V, et al. Mutations in VKORC1 cause warfarin resistance and multiple coagulation factor deficiency type 2. *Nature* 2004; 427:537-541.
32. Loebstein R, Dovskin I, Halkin H, et al. A coding VKORC1 Asp36-Tyr polymorphism predisposes to warfarin resistance. *Blood* 2007; 109:2477-2480.
33. Bentley DP, Backhouse G, Hutchings A, Haddon RL, Spragg B, Routledge PA. Investigation of patients with abnormal response to warfarin. *Br J Clin Pharmacol* 1986; 22:37-41.
34. Eriksson BI, Borris LC, Friedman RJ, et al; RECORD1 Study Group. Rivaroxaban versus enoxaparin for thromboprophylaxis after hip arthroplasty. *N Engl J Med* 2008; 358:2765-2775.
35. Kakkar AK, Brenner B, Dahl OE, et al; RECORD2 Investigators. Extended duration rivaroxaban versus short-term enoxaparin for the prevention of venous thromboembolism after total hip arthroplasty: a double-blind, randomised controlled trial. *Lancet* 2008; 372:31-39.
36. Lassen MR, Ageno W, Borris LC, et al; RECORD3 Investigators. Rivaroxaban versus enoxaparin for thromboprophylaxis after total knee arthroplasty. *N Engl J Med* 2008; 358:2776-2786.
37. Klowacz SE, Roskell NS, Plumb JM, Caprini JA, Eriksson BI. Efficacy and safety of dabigatran etexilate for the prevention of venous thromboembolism following total hip or knee arthroplasty. A meta-analysis. *Thromb Haemost* 2009; 101:77-85.

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