Archives of Medical Research 46 (2015) 265–273

Archives of Medical Research

ORIGINAL ARTICLE

Clinical Application of Genotype-guided Dosing of Warfarin in Patients with Acute Stroke

Svjetlana Šupe,[a] Zdravka Poljaković,[a] Tamara Božina,[b] Josip Ljevak,[a] Viola Macolić Šarinić,[c] and Nada Božina[d,e]

[a]*Department of Neurology, Intensive Care Unit, University Hospital Center Zagreb, Zagreb, Croatia*
[b]*Department of Medical Chemistry, Biochemistry and Clinical Chemistry, School of Medicine, University of Zagreb, Zagreb, Croatia*
[c]*Agency for Medicinal Products and Medical Devices, Zagreb, Croatia*
[d]*Department of Laboratory Diagnostics, Zagreb University Hospital Center, Zagreb, Croatia*
[e]*Department of Pharmacology, School of Medicine, University of Zagreb, Zagreb, Croatia*

Received for publication December 1, 2014; accepted May 12, 2015 (ARCMED-D-14-00706).

Background. Patients with certain types of stroke need urgent anticoagulation and it is extremely important for them to achieve fast and stable anticoagulant effect and receive individualized treatment during the initiation of warfarin therapy.

Methods. We conducted a prospective study among 210 acute stroke patients who had an indication for anticoagulation and compared the impact of *CYP2C9* and *VKORC1* genotype-guided warfarin dosing (PhG) with fixed dosing (NPhG) on anticoagulation control and clinical outcome between groups.

Results. PhG achieved target INR values earlier, i.e., on average in 4.2 (4.1–4.7, 95% CI) days compared to NPhG (5.2 days [4.7–6.4, 95% CI]) ($p = 0.0009$), spent a higher percentage of time in the therapeutic INR range (76.3% [74.7–78.5, 95% CI] vs. 67.1% [64.5–69.6, 95% CI] in NPhG), and spent less time overdosed (INR > 3.1) (PhG 0.4 [0.1–0.7, 95% CI], NPhG 1.7 [1.1–2.3, 95% CI] days; $p > 0.000$). PhG reached stable maintenance dose faster (10 [9.9–10.7, 95% CI] vs. 13.9 [13.3–14.7, 95% CI] days in controls; $p = 0.0049$) and had a better clinical outcome in relation to neurological deficit on admission as compared to NPhG.

Conclusion. We confirmed that warfarin therapy with genotype-guided dosing instead of fixed dosing reduces the time required for stabilization and improves anticoagulant control with better clinical outcome in early stages of warfarin therapy introduction among acute stroke patients, which is essential for clinical practice.

Key Words: Stroke, Warfarin, *CYP2C9*, *VKORC1*, Polymorphisms.

Introduction

Patients with cardioembolic stroke and stroke consequent to a specific condition (cerebral sinus venous thrombosis, dissection of intracranial arteries, hypercoagulable state) require urgent anticoagulation for secondary stroke prevention (1–4). This is important for prevention of arterial and venous thromboembolism, particularly of embolic stroke recurrences and to reduce a high rate of in-hospital mortality in patients with recurrent embolism (19.6%) (5,6). Despite the emergence of new anticoagulants, warfarin is a widely prescribed first-line anticoagulant for most causes of embolic stoke due to its effectiveness in the management of thromboembolism and to its low cost (7–9).

Warfarin has a narrow therapeutic index and at least 20-fold interpatient variations in dose requirements, which can lead to dose-related insufficient or excessive anticoagulation. Most dose-dependent adverse events emerge during introduction of therapy. Risk for hemorrhage or

Address reprint requests to: Nada Božina, MD, PhD, Associate Professor, Department of Laboratory Diagnostics, University Hospital Center Zagreb, Kišpatićeva 12, 10000 Zagreb, Croatia; Phone: +38512367249; FAX: +38512367395; E-mail: nbozina@kbc-zagreb.hr

0188-4409/$ - see front matter.
http://dx.doi.org/10.1016/j.arcmed.2015.05.001

thromboembolism in this initiation period is higher than during later stages (event rates range from 16–25%) (10,11). Therefore, stroke patients who require anticoagulation are also at an increased risk of warfarin-dosage side effects and hence of recurrent stroke if they are underdosed or in overdosed conditions due to warfarin-induced brain hemorrhage (12,13). Warfarin pharmacogenetics, association of *CYP2C9* and *VKORC1* gene polymorphisms and dosing algorithms that use this genetic information are well documented (14–18).There are few studies available that have investigated the effectiveness of clinical application of genotype-guided dosing, and such studies conducted among acute stroke patients are specifically scarce (19–22). For these high-risk patients, it is essential to achieve stable anticoagulant effect as early as possible in order to prevent the risk of dosage-related complications by using the initial doses based on individual genotype (17,23,24). Results of Franchini's meta-analysis that included nine trials (2812 patients) show that genotype-guided initial warfarin dosing significantly reduced the risk ratio for developing major bleeding events compared with the control group (RR = 0.47; $p = 0.04$), or reduced serious bleeding events by ~50% compared to clinically guided dosing group (25). Data obtained by meta-analysis of randomized clinical trials indicated that a genotype-guided dosing strategy was not superior to clinical dosing algorithms in terms of INR values, reduction in major bleeding or thromboembolic events (26).

Recently, several randomized controlled trials have been published with different conclusions on the clinical utility of genotype-guided dosing of coumarin anticoagulants. A study conducted by Pirmohamed et al. (EU-PACT trial) found that pharmacogenetic-based dosing was associated with a higher percentage of time in the therapeutic INR range than standard dosing during the initiation of warfarin therapy (27), whereas Kimmel et al. (COAG trial) did not confirm these findings for the anticoagulation period of the first 4 weeks of warfarin therapy (28). Also, Verhoef et al. did not confirm the findings for acenocoumarol or phenprocoumon during 12 weeks after initiation of therapy in patients with atrial fibrillation or venous thromboembolism (29).

In this study we compared the impact of *CYP2C9* and *VKORC1* genotype-guided dosing of warfarin with a standardized, fixed dosing regarding anticoagulation control and clinical outcome in acute stroke patients during the first 3 weeks of therapy.

Patients and Methods

During 6 months, 587 Croatian Caucasian patients (EUCs) with acute ischemic stroke were hospitalized at the Department of Neurology, UHC Zagreb. We conducted a prospective trial among 210 (36% of the total number) patients with an indication for urgent anticoagulation whose initial brain CT scan was without signs of hemorrhage. Detailed inclusion and exclusion criteria are listed in Table 1.

Table 1. Inclusion and exclusion criteria for the study

Inclusion criteria
1. Previously taking warfarin due to atrial fibrillation, mechanical heart valves, deep vein thrombosis, pulmonary embolism
2. Newly discovered atrial fibrillation confirmed by HOLTER-ECG
3. Acute dissection of intracranial arteries
4. Patent foramen ovale with septal aneurysm
5. Cerebral venous sinus thrombosis
Exclusion criteria
1. Age <18 years
2. Hemorrhage in the brain detected by CT scan, except in patients with cerebral venous thrombosis
3. Malignancy, pregnancy
4. Hepatic/renal insufficiency

Power analysis was done before the study. If the statistical significance level is set to $p < 0.05$ and power to 0.95, two-tailed Fisher exact test would need the sample size of $n = 96$ in each group to detect the difference of at least 0.25 in proportion of patients with INR within the therapeutic range (2,3) at fifth day after the introduction of warfarin. Expected proportions were obtained in the pilot study done on the sample of $n = 20$ patients. The proportion of patients with INR within the therapeutic range (2,3) was 0.9 in PhG and 0.4 in NPhG group. For more conservative calculation we set the expected proportions to 0.8 in PhG and 0.55 in NPhG group. Expecting ~5–10% of missing data the initial sample size was decided to be $n = 105$ in each group.

Eligible patients ($n = 210$) were centrally registered and stratified according to gender and age and then assigned to the intervention group (*CYP2C9**2,*3 and *VKORC11173*-C>T genotype-guided dosing group, PhG [$n = 106$]) or to the control/fixed dosing group (NPhG, $n = 104$) (Figure 1).

There were no differences between groups according to gender ($p = 0.559$), age ($p = 0.669$) or indications for anticoagulation (inclusion criteria; $p = 0.591$). Among the total number of 210 patients, 32% were previously anticoagulated by using warfarin (34.9% [$n = 36$] PhG and 28.8% [$n = 30$] NPhG) and INR on admission was in all of them below the therapeutic range (INR < 1.6). Atrial fibrillation was the most common indication for anticoagulation in both groups (56% of the total number; 53 [50.5%] in PhG, 65 [62.5%] in NPhG) (Table 2).

Because the number of eligible patients, i.e., those with appropriate diagnosis for our study ($n = 210$) was relatively low and inclusion/exclusion criteria practically did not have any impact on patient selection as each patient required equal treatment and was consequently to be allocated in one of two study arms, we used a simple way of grouping (distribution) patients by alternative allocation of every second patient into one of the two study arms. Moreover, having in mind that we could not influence patient's occurrence and patient's disease status in any manner, we considered that any possible bias was absolutely negligible by using this method of allocation.