RESEARCH LETTER

Monitoring of anticoagulant effects of dabigatran in everyday practice: first experience in 32 Polish patients

Introduction Dabigatran etexilate, a new oral direct thrombin inhibitor, has been shown to surpass therapy with warfarin, when given for prevention of stroke and systemic embolism in patients with nonvalvular atrial fibrillation (AF).[1,2] Recently, it has been reported that higher dabigatran concentrations are associated with an elevated bleeding rate and lower concentrations, with increased thromboembolic risk.[3]

Dabigatran prolongs activated partial thromboplastin time (aPTT), thrombin time (TT), and ecarin clotting time with a slight increase in prothrombin time-international normalized ratio (PT-INR).[2,4] Routine laboratory monitoring of dabigatran using clot-based assays is currently not recommended.[4-6] However, there are situations in which measurements of the anticoagulant effect of dabigatran are desirable, for example, in patients who experienced thromboembolism or bleeding events despite regular dabigatran intake.

We investigated the variation in plasma dabigatran concentrations in AF patients with normal renal function in our outpatient clinic.

Patients and methods We studied 32 consecutive patients with permanent AF (13 women and 19 men), aged 64.7 ±9.5 years, who were treated with dabigatran for 3 months or longer and complained of no bleeding or thromboembolic events over that period of time. Patients were recruited at the Cardiology Clinic of the John Paul II Hospital, Kraków, Poland, from June to November 2013. The risk for stroke and bleeding estimated with the CHA_2DS_2VASc and HAS-BLED scores was 2.9 ±1.3 (3–6 points, n = 18) and 1.7 ±1.1 (3 points, n = 6), respectively. Twenty-eight patients (87.5%) were treated with dabigatran at a dose of 150 mg twice daily and 4 with dabigatran at a dose of 110 mg owing to high bleeding risk. The exclusion criteria were acute thromboembolism within the preceding 1 month, cancer, chronic kidney disease stage 3 or higher with an estimated glomerular filtration rate (eGFR) of less than 60 ml/min, acute infection, drugs known to interfere with dabigatran (eg, amiodarone, verapamil).

In fasting citrated plasma samples following administration of dabigatran according to an individual dosing scheme (1–24 h since the last dose), we measured aPTT, PT-INR (both Siemens, Marburg, Germany), serum creatinine with eGFR using the Chronic Kidney Disease Epidemiology Collaboration equation in a hospital laboratory. Plasma concentrations of dabigatran were determined using the Hemoclot thrombin inhibitor assay (HYPHEN BioMed, Neuville-sur-Oise, France) according to the manufacturer's instructions. Briefly, a constant amount of human thrombin was added to the diluted tested plasma mixed with a normal pooled human plasma and the measured TT was directly related to a concentration of dabigatran in the sample. In 20 nonanticoagulated healthy individuals, we determined the same parameters to obtain the reference values, ie, aPTT, 27.8 ±2.4 s (normal range, 23.1–33.8 s); PT, 11.4 ±0.5 s (10.4–13.0 s); and INR, 1.0 ±0.05 (0.85–1.15 s). All control subjects had dabigatran concentrations below 35 ng/ml (the lower limit of measuring range).

Results In AF patients with normal creatinine levels (84.4 ±16.9 µmol/l; eGFR, 76 ±13.9 ml/min), plasma dabigatran concentrations ranged from 35 to 321 ng/ml (median, 105 ng/ml; interquartile range, [IQR], 75–152 ng/ml). Patients who declared that the last dose of dabigatran was taken within 1 to 4 hours prior to blood collection (n = 23) had dabigatran concentrations from 38 to 321 ng/ml (median, 110 ng/ml; IQR, 85–177 ng/ml), while those in whom blood was drawn after 4 hours from the last dose (n = 9) had dabigatran concentrations from 35 to 140 ng/ml (median, 58 ng/ml; IQR, 50–123 ng/ml) (P = 0.036). There were 4 patients (12.5%) with dabigatran concentrations of 50 ng/ml and lower, including 3 subjects who reported 12 hours or more from the drug intake and 1 who declared

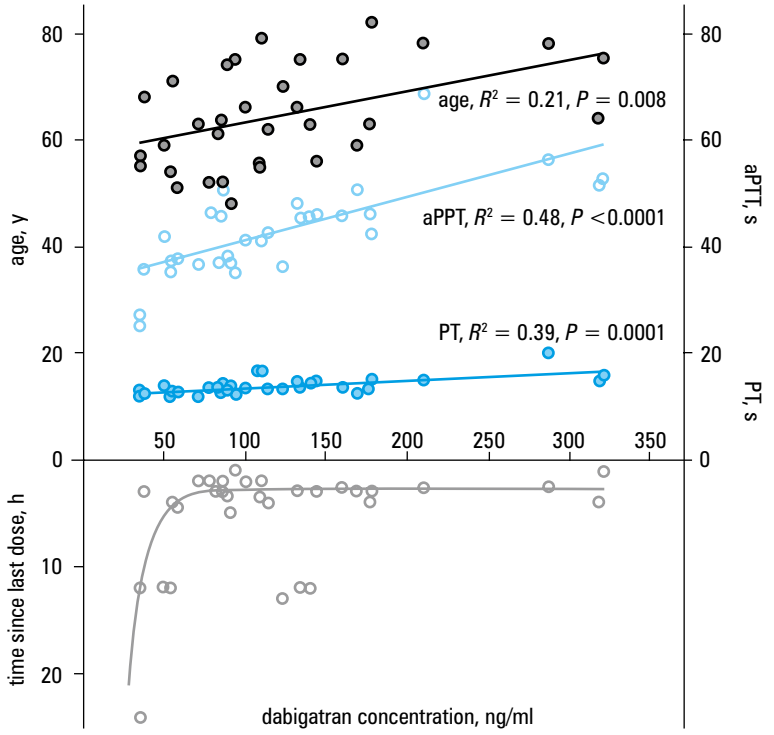

FIGURE Associations of age and time since the last dose with plasma dabigatran concentrations and their correlations with coagulation times
Abbreviations: aPTT – activated partial thromboplastin time, PT – prothrombin time

taking dabigatran 3 hours before the visit. Two patients with AF (6.3%) declared that more than 12 hours elapsed since the last dose of dabigatran despite the fact that this anticoagulant should be taken every 12 hours. Dabigatran concentrations were weakly correlated with age (R^2 = 0.21; P = 0.008) but not with eGFR, sex, and the time since the last dose (**FIGURE**). By analysis of covariance, after adjustment for sex (P = 0.4), the time of blood collection (P = 0.14), and creatinine levels (P = 0.54), age alone was an independent predictor of the dabigatran concentration (F = 5.07, P = 0.033).

The median dabigatran concentration tended to be higher in patients with a CHA_2DS_2VASc of 3 and higher as compared with those with a CHA_2DS_2VASc of 2 and lower (128 ng/ml vs. 85 ng/ml, P = 0.06, respectively). There were no differences in patients with a HAS-BLED score of 0 to 1 and ≥2.

Routine coagulation tests showed prolonged aPTT (43.1 ±8.8 s), PT (13.9 ±1.7 s), and INR (1.24 ±0.16) compared with controls (all P <0.0001). All patients on dabigatran except the 2 subjects with the drug concentration of 35 ng/ml had prolonged aPTT. Positive linear correlations between dabigatran concentrations and aPTT (R^2 = 0.48; P <0.0001) as well as PT (R^2 = 0.39, P = 0.0001) were observed (**FIGURE**).

To our knowledge, this study is the first to show the results of dabigatran monitoring using the Hemoclot assay recommended for this purpose in the European Union since 2013 in everyday practice in a Polish hospital. In typical stable outpatients with permanent AF and an eGFR exceeding 60 ml/kg/min, not requiring reduction in dabigatran dosage for this reason, the levels were similar to the expected values[5] with the peak/trough ratio of about 2:1 as in the study by Reilly et al.,[3] in which a validated high-performance liquid chromatography–tandem mass spectrometry was used to determine drug concentrations. We found a large variability in dabigatran concentrations when determined in AF patients with the highest levels 1 to 4 hours after the last dose and an overlap of the results for samples taken at the peak anticoagulant activity versus the later time. The blood sampling time is crucial for the interpretation of the test results[6] as confirmed in our study. Patients' age was identified to be the only independent predictor of dabigatran concentrations in our group. aPTT shows a linear relationship with dabigatran concentrations up to the very high levels typical of overdose, after which a flattering of the curve is observed.[4,5] Despite a significant, though moderate, correlation between aPTT and dabigatran concentrations estimated using the Hemoclot assay, aPTT does not allow quantitative assessment of dabigatran levels.[2-5] Nevertheless, prolonged aPTT indicates the presence of the drug in the blood and, given the fact that all subjects (control and tested) with dabigatran levels of 35 ng/ml and less had normal aPTT, it could be useful in patients scheduled for major surgery with high bleeding risk in whom physicians are uncertain as to whether anticoagulant effects of dabigatran subsided.[2,5] Although prolonged PT was observed within the first hours after the dabigatran intake, this parameter is much less sensitive to thrombin inhibition than aPTT.[5]

Of note, we found that about 6% of patients with AF on dabigatran reported that the intervals between the 2 consecutive doses were longer than the recommended 12 hours. Good compliance is of key importance for good and safe anticoagulation, especially in patients on new oral anticoagulants with a relatively short half-times.[2,6]

Our findings indicate that the measurement of plasma dabigatran concentrations is feasible in everyday practice and may support patient management if necessary, though the therapeutic range has not been determined yet.

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