Loading and maintenance dose algorithms for phenprocoumon and acenocoumarol using patient characteristics and pharmacogenetic data

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Aims Polymorphisms in CYP2C9 and VKORC1 influence patients’ phenprocoumon (PHE) and acenocoumarol (ACE) dose requirements. To provide physicians with tools to estimate the patient’s individual dose, we aimed to develop algorithms for PHE and ACE.

Methods and results In two Dutch anticoagulation clinics, data on age, sex, height, weight, co-medication, coumarin derivative doses, and international normalized ratio values were obtained from 624 patients taking PHE and 471 taking ACE. Single nucleotide polymorphisms relevant to coumarin derivative dosing on the CYP2C9 and VKORC1 genes were determined. Using multiple linear regression, we developed genotype-guided and non-genotype-guided algorithms to predict the maintenance dose with patient characteristics and genetic information. In addition, loading doses were derived from the calculated maintenance doses. We performed external validation in an independent data set with 229 PHE and 168 ACE users. CYP2C9 and VKORC1 genotype, weight, height, sex, age, and amiodarone use contributed to the maintenance dose of PHE and ACE. The genotype-guided algorithms explained 55.9% (PHE) and 52.6% (ACE) of the variance of the maintenance dose, the non-genetic algorithms 17.3% (PHE) and 23.7% (ACE). Validation in an independent data set resulted in an explained variation of 59.4% (PHE) and 49.0% (ACE) for the genotype-guided algorithms and for 23.5% (PHE) and 17.8% (ACE) for the non-genotype-guided algorithms, without height and weight as parameters.

Conclusion To our knowledge, these are the first genotype-guided loading and maintenance dose algorithms for PHE and ACE using large cohorts. The utility of these algorithms will be tested in randomized controlled trials.

Keywords Dosing algorithms • Phenprocoumon • Acenocoumarol • Pharmacogenetics • CYP2C9 • VKORC1

Introduction Patients receiving coumarin therapy are at risk of thrombosis and therapy failure due to under-dosing or at risk of haemorrhage due to over-dosing,1,2 making coumarins often associated with drug-related hospitalization.3–5 This is because coumarins have a narrow therapeutic window and there is wide inter- and intra-individual variability in dose requirements.6,7 Therefore,
patients are monitored by measuring the international normalised ratio (INR). Current clinical practice is that all patients receive a standard loading dose at the start of the therapy, which is subsequently adjusted to an individual maintenance dose according to the measured INR. This leads to a mean percentage time within the target INR ranging from only 45 to 64% during the first 2 months of the anticoagulation therapy, which needs to be improved.

Developing a strategy towards more individualized dosing of coumarins has gained interest in recent years. It is known that patient characteristics such as age and body size influence the dose requirements. More recently, genetic factors, notably polymorphisms in the VKORC1 gene which expresses vitamin K epoxide reductase (the main target for coumarins) and the CYP2C9 gene which expresses cytochrome P450 2C9 (the enzyme responsible for the metabolism of coumarin), together have been shown to explain 35–50% of the inter-individual variability in dose requirements. To date, a number of studies have reported the development of pharmacogenetics-guided algorithms for warfarin. However, there are no published reports on the development and validation of algorithms in a large cohort for predicting the loading and maintenance dose of phenprocoumon (PHE) and acenocoumarol (ACE). In continental Europe, PHE and ACE are most commonly used for anticoagulant therapy; for example in 2008 in the Netherlands, >200 000 prescriptions were made for PHE and >1 million were made for ACE.

There is a definite need for the development of more refined algorithms for both these drugs if the notion of a future approach to individualized therapy is to be realized. The aim of this study was to derive algorithms to estimate the individualized loading and maintenance doses for PHE and ACE before the start of the treatment.

Methods

Study design and patients

Patients currently using either PHE or ACE were eligible to take part in the study if aged 18 years and over and with a target INR in the lowest intensity category (according to Dutch guidelines INR 2.0–3.5). Pregnant or breastfeeding women, patients who were in a nursing home, and patients participating in other clinical studies were excluded. Eligible patients who had a scheduled visit at the anticoagulation clinic from either 10 to 12 November 2009 (Anticoagulation Clinic Leiden, PHE) or from 23 to 27 November 2009 (Anticoagulation Clinic Medial, ACE) were invited to participate. We aimed to include ~1000 patients because that would increase the probability of capturing data on a reasonable number of patients (at least five patients per coumarin group) having the least frequent CYP2C9 genotypes (e.g. CYP2C9*2/*3 and *3/*3) and therefore to assure accurate dose estimates for all genotypes. The Committee Medical Ethics Leiden approved the study protocol, and patients provided informed consent before inclusion into the study.

Data collection and genotyping

Height, current weight (weight at the moment of inclusion), and weight at the start of the anticoagulation therapy were recorded for each participant. Data on the participants’ age, sex, history of co-medication, history of INR values, and prescribed coumarin doses were obtained from the electronic registry databases of the anticoagulation clinics. Since 1983, in the Netherlands at each visit to the anticoagulation clinic, INR measurements, prescribed doses, and co-medication are routinely collected and recorded in registry databases. Residual blood samples from INR measurements were used to genotype the patient for CYP2C9*2 (rs1799853), CYP2C9*3 (rs1057910), and VKORC1 1173C > T (rs9934438) using pre-designed Taqman assays (Applied Biosystems, Nieuwerkerk aan den IJssel, the Netherlands) and according to the manufacturers’ protocol. CYP2C9*1 and VKORC1 C genotypes were assigned if polymorphisms in the analysed corresponding Single nucleotide polymorphisms (SNPs) (CYP2C9*2, CYP2C9*3, and VKORC1 1173C > T) were lacking. Other variant alleles are rare in Caucasians. Therefore, there is a negligible risk for a misclassification of phenotypes due to other variant alleles. Genotypes were determined on LightCycler 480 (Roche Diagnostics, Almere, the Netherlands) in 384-well plates that include positive (previous established genotype) and negative controls (Tris EDTA buffer).

In addition, as quality control 10% of the samples were genotyped in duplicate.

Outcome and determinants

The mean stable coumarin maintenance dose in mg/day at the first stable period after initiation of anticoagulation therapy was used as the outcome measure. A stable period was defined as a period of at least 3 weeks with three or more consecutive INR measurements within target range with <10% change in the coumarin dose. To develop the non-genotype-guided algorithms, the a priori defined determinants were age in years, sex, amiodarone use, height in centimetres, and weight in kilograms at the start of the anticoagulation treatment (if missing, current weight was used instead). For the genotype-guided algorithm, CYP2C9 and VKORC1 genotypes were used as additional determinants.

Statistical analysis and algorithm development

Multiple linear regression was used to estimate the maintenance dose of PHE and ACE. To reduce the influence of extreme observations, the values of continuous predictive variables were truncated at approximately the 2.5th and 97.5th percentile. Either patients with missing values for at least one of the determinants or those who did not reach a stable phase within a year following the start of the therapy were excluded. The optimal transformation of the outcome (original scale, log transformation, or square root transformation) was determined by selecting the transformation with the average lowest mean squared error (which is the mean of the absolute difference between predicted and observed outcomes, the mean squared error of determination (R²) and the mean absolute error.

The algorithms were externally validated using two data sets of Schalekamp et al. These data sets contain complete data of 229 patients using PHE and 168 patients using ACE. Because weight and height were not available in the validation data sets, we validated the algorithms without height and weight. In addition, we validated the algorithms using multiple imputation methods for missing weight and height (see Supplementary material online). The R² between the algorithm predictions and the observed outcomes, the mean squared error and the mean absolute error were calculated.
The loading dosages were derived from the estimated maintenance dose. In general, only drugs with a long elimination half-life are candidates for loading dose administration at the start of the therapy for rapid achievement of the steady-state plasma drug concentration and therapeutic effect.\textsuperscript{23} ACE has a relatively short half-life of 8–14 h and therefore loading doses are not necessary. Therefore, our recommended ACE loading doses are rounded values of the calculated individual maintenance doses. PHE on the other hand has an average half-life of 160 h and therefore with loading doses a faster therapeutic response is attained.\textsuperscript{24,25}

In general, the loading dose can be calculated from the maintenance dose by using a first-order kinetics equation\textsuperscript{23}. The current standard clinical practice for PHE is to divide the loading dose over the first 3 days of therapy. The loading dose is obtained from the calculated stable maintenance dose using the equation (1)

$$MD = \frac{D_1 \cdot e^{-2k} + D_2 \cdot e^{-k} + D_3}{1 - e^{-k}} \quad (1)$$

with MD defined as the maintenance dose, $D_1$ the dose received on Day 1, $D_2$ the dose received on Day 2, and $D_3$ the dose received on Day 3, $k = \ln(2)/t_{1/2}$, where $k$ is the elimination rate constant and $t_{1/2}$ is the drug half-life in days.

Anticoagulation response to coumarins is the result of a complex interplay between several variables, including vitamin K availability.

**Figure 1** Flowcharts of patients included in the phenprocoumon cohort (A) and the acenocoumarol cohort (B).
and the presence of functional vitamin K-dependent clotting factors. However, inhibition of the synthesis of functional vitamin K-dependent clotting factors II, VII, IX, and X is dependent on the plasma coumarin concentration, which in turn is related to the CYP2C9 enzyme activity. Therefore, on this basis equation (1) above was used for estimating PHE loading dose which to some extent reflects inter-patient CYP2C9 variability.

A number of restrictions were applied for the PHE loading dose estimation to minimize the risk of over- or under-dosing, especially for the non-genotype-guided algorithm (see Supplementary material online). We used the statistical software SPSS (PASW Statistics) version 18 for the analysis.

Results

Patient cohort

In total, 624 patients using PHE and 471 patients using ACE were included in the study. For the non-genotype-guided algorithm, complete data were available for 587 patients using PHE and 400 patients using ACE. For the genotype-guided algorithm, data on 559 PHE and 375 ACE patients were available; see flowcharts Figure 1. The median maintenance dose for PHE was 2.12 mg/day and for ACE was 2.34 mg/day. Patient characteristics are presented in Table 1. Height, weight, age, and CYP2C9 and VKORC1 genotype were not significantly different among the group with and group without missing values, except for VKORC1 genotype distribution in the ACE patients ($P = 0.037$).

Genotyping

No inconsistencies were observed for the quality controls. Allele frequencies for CYP2C9 were 0.82 for the wild-type allele, 0.12 for CYP2C9*2, and 0.07 for CYP2C9*3. Allele frequencies for VKORC1 1173C > T were 0.61 for C and 0.39 for T. All three genotype distributions followed Hardy–Weinberg equilibrium.

Phenprocoumon and acenocoumarol maintenance dose algorithm

The square root of the maintenance dose in mg/day during the first stable INR monitoring period was on average the best outcome transformation for the four algorithms and was therefore chosen as the outcome measure. Differences between this and other transformations were very small (see Supplementary material online).
The intercept and coefficients of all four algorithms as well as the univariate $R^2$ for the parameters included are presented in Table 2. There were no interactions between the determinants that improved the algorithms. The explained variance was 55.9% (PHE) and 52.6% (ACE) for the genotype-guided algorithms and 17.3% (PHE) and 23.7% (ACE) for the non-genotype-guided algorithms. The mean absolute error for the genotype-guided algorithms was 0.45 mg/day (PHE) and 0.52 mg/day (ACE) and for the non-genotype-guided algorithms 0.63 mg/day (PHE) and 0.70 mg/day (ACE).

### Table 2  Algorithms for phenprocoumon and acenocoumarol

<table>
<thead>
<tr>
<th></th>
<th>Phenprocoumon</th>
<th></th>
<th></th>
<th>Acenocoumarol</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Genotype-guided</td>
<td>Non-genotype-guided</td>
<td>Univariate $R^2$</td>
<td>Genotype-guided</td>
<td>Non-genotype-guided</td>
<td>Univariate $R^2$</td>
</tr>
<tr>
<td>Intercept</td>
<td>2.874</td>
<td>1.652</td>
<td>4.117</td>
<td>2.635</td>
<td>2.363</td>
<td>2.635</td>
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<tr>
<td>CYP2C9 genotype</td>
<td></td>
<td></td>
<td>4.6</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>*1/*1</td>
<td>0</td>
<td></td>
<td>0</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>*1/*2</td>
<td>−0.259</td>
<td></td>
<td>−0.093</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>*1/*3</td>
<td>−0.342</td>
<td></td>
<td>−0.519</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>*2/*2</td>
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<td></td>
<td>−0.435</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>*2/*3</td>
<td>−0.684</td>
<td></td>
<td>−0.466</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>*3/*3</td>
<td>−0.681</td>
<td></td>
<td>−1.375</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>VKORC1 genotype</td>
<td>34.1</td>
<td></td>
<td>27.2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CC</td>
<td>0</td>
<td></td>
<td>0</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CT</td>
<td>−0.601</td>
<td></td>
<td>−0.572</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TT</td>
<td>−1.394</td>
<td></td>
<td>−1.267</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age, in years</td>
<td>−0.015</td>
<td>−0.011</td>
<td>8.1</td>
<td>−0.027</td>
<td>−0.027</td>
<td>14.1</td>
</tr>
<tr>
<td>Sex, if female</td>
<td>0.026</td>
<td>0.105</td>
<td>2.1</td>
<td>0.271</td>
<td>0.386</td>
<td>0.2</td>
</tr>
<tr>
<td>Height, in cm</td>
<td>0.011</td>
<td>0.011</td>
<td>7.3</td>
<td>0.009</td>
<td>0.013</td>
<td>6.3</td>
</tr>
<tr>
<td>Weight, in kg</td>
<td>0.008</td>
<td>0.013</td>
<td>12.8</td>
<td>0.010</td>
<td>0.013</td>
<td>11.8</td>
</tr>
<tr>
<td>Amiodarone use, if yes</td>
<td>−0.345</td>
<td>−0.343</td>
<td>0.5</td>
<td>−0.377</td>
<td>−0.167</td>
<td>0.2</td>
</tr>
<tr>
<td>Unadjusted $R^2$</td>
<td>55.9%</td>
<td>17.3%</td>
<td>52.6%</td>
<td>23.7%</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

The formula for, for example, the genotype-guided algorithm of phenprocoumon should be read as: square root mean maintenance dose (mg/week) = 2.874 − 0 (if CYP2C9*1/*1) − 0.259 (if CYP2C9*1/*2) − 0.342 (if CYP2C9*1/*3) − 0.447 (if CYP2C9*2/*2) − 0.684 (if CYP2C9*2/*3) − 0.681 (if CYP2C9*3/*3) − 0 (if VKORC1 CC) − 34.1 (if VKORC1 CT) − 1.394 (if VKORC1 TT) − 0.015 * age (years) + 0.026 (if female) + 0.011 * height (cm) + 0.008 * weight (kg) − 0.345 (if amiodarone is used).

The outcome is the square root of the mean first stable maintenance dose in mg/week for the INR target range 2.0–3.5. If the target range 2.0–3.0 is used, all coefficients need to be divided by sqrt(1.07).

The value of this parameter is zero because it is the reference group.

### Phenprocoumon and acenocoumarol loading dose strategies

Table 3 shows the loading dose corresponding to a given maintenance dose for PHE. A dosing regimen of 9 mg on Day 1, 6 mg on Day 2, and 6 mg on Day 3 as a loading schedule (or alternatively 12-6-3 mg for the first 3 days) is currently the maximum standard loading dose for

### Table 3  Loading doses for phenprocoumon as derived from the individual maintenance dose

<table>
<thead>
<tr>
<th>Dose Day 1 (mg)</th>
<th>Dose Day 2 (mg)</th>
<th>Dose Day 3 (mg)</th>
<th>Maintenance Dose Range (mg per day)</th>
</tr>
</thead>
<tbody>
<tr>
<td>3</td>
<td>3</td>
<td>3</td>
<td>&lt;1.04</td>
</tr>
<tr>
<td>6</td>
<td>3</td>
<td>3</td>
<td>1.04–1.31</td>
</tr>
<tr>
<td>6</td>
<td>6</td>
<td>3</td>
<td>1.31–1.61</td>
</tr>
<tr>
<td>6</td>
<td>6</td>
<td>6</td>
<td>1.61–1.85</td>
</tr>
<tr>
<td>9</td>
<td>6</td>
<td>6</td>
<td>1.85–2.92</td>
</tr>
<tr>
<td>9</td>
<td>9</td>
<td>6</td>
<td>&gt;2.92b</td>
</tr>
</tbody>
</table>

The lower limit of the maintenance dose range corresponds with the given loading dose, e.g. a loading regimen of 6-3-3 leads to a monitoring dose of 1.04 mg/day.

bOnly for genotype-guided algorithm.
PHE. On the basis of our data, we recommend a higher loading dose (9, 9, and 6 mg on Days 1, 2, and 3, respectively) compared with the current standard regimen only when it is 90% certain that the patient needs a higher loading dose than standard clinical care. This is valid for patients who according to the genotype-guided algorithm have a predicted maintenance dose of 2.92 mg/day or higher. In addition, 3, 3, and 3 mg as a loading schedule on Days 1, 2 and 3 (corresponding to a maintenance dose of <1.04 mg/day) is only been given to patients who are dosed according to the genotype-guided algorithm, since such low doses will not be calculated with the non-genotype-guided algorithm. Figure 2 shows the distribution of the loading dose regimens using the genotype-guided algorithm and the non-genotype-guided algorithm in the derivation cohort. Table 4 shows the loading dose corresponding to a given maintenance dose for ACE. Briefly, the predicted loading dose is rounded off to the highest number of tablets equivalent to the estimated dose, whereas for the subsequent days the dose is divided equally.

**External validation of phenprocoumon and acenocoumarol algorithms**

External validation of the genotype-guided algorithms without height and weight yielded an $R^2$ of 59.4% and a mean absolute

<table>
<thead>
<tr>
<th>Dose Day 1 (mg)</th>
<th>Dose Day 2 (mg)</th>
<th>Dose Day 3 (mg)</th>
<th>Maintenance dose range (mg per day)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1</td>
<td>1</td>
<td>&lt;1.00</td>
</tr>
<tr>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1.00–1.25</td>
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<tr>
<td>2</td>
<td>2</td>
<td>1</td>
<td>1.25–1.75</td>
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<td>1.75–2.00</td>
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<td>2</td>
<td>2</td>
<td>2.00–2.25</td>
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<td>3.00–3.25</td>
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<td>3.25–3.75</td>
</tr>
<tr>
<td>4</td>
<td>4</td>
<td>4</td>
<td>3.75–4.00</td>
</tr>
<tr>
<td>5</td>
<td>4</td>
<td>4</td>
<td>4.00–4.25</td>
</tr>
<tr>
<td>5</td>
<td>5</td>
<td>4</td>
<td>4.25–4.75</td>
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<tr>
<td>5</td>
<td>5</td>
<td>5</td>
<td>4.75–5.00</td>
</tr>
</tbody>
</table>

*The lower limit of the maintenance dose range corresponds with the given loading dose.*
error of 0.46 mg/day for PHE and an $R^2$ of 49.0% and a mean absolute error of 0.57 mg/day for ACE. For the non-genotype-guided algorithm, the $R^2$ was 23.5% (PHE) and 17.8% (ACE) and the mean absolute error was 0.62 mg/day (PHE) and 0.72 mg/day (ACE). Figure 3 shows plots of predicted vs. observed maintenance dose in the validation sets. For results of the validation using multiple imputation, see Supplementary material online.

**Discussion**

Several genotype-guided algorithms have previously been developed for warfarin. However, algorithms to estimate the PHE and ACE maintenance dose are only developed in small cohorts (<100 patients) and are non-validated. In this paper, we present genotype-guided and non-genotype-guided algorithms for the determination of the loading and maintenance phase of PHE and ACE treatment based on data derived from almost 1000 patients. The non-genotype-guided algorithms estimate the individual patient dose requirement for initiation of coumarin therapy based on patient characteristics of age, sex, height, weight, and amiodarone use. The genotype-guided algorithms include CYP2C9 and VKORC1 genotype additional to the variables included in the non-genotype-guided algorithms. These algorithms are thought to be an improvement compared with the current clinical situation, where each patient receives a standard loading dose which is subsequently adjusted to the individual dose with the INR. However, this hypothesis needs to be tested in a randomized controlled trial.

Even though novel anticoagulants have recently entered the market, PHE and ACE are anticipated to remain commonly prescribed anticoagulants because new anticoagulants are more expensive and have no antidote, and the experience is limited at the moment. To avoid increased costs, it is likely that authorities will encourage continued use of coumarin derivatives. Therefore, it is crucial to improve the safety of the treatment with coumarin derivatives. Randomized clinical trials are needed to investigate whether the safety of the anticoagulation treatment will improve by developing individualized dosing regimens as shown in this paper.
Before the anticoagulation therapy starts, the maintenance dose can be calculated by filling in the algorithm. Then the loading dose belonging to the calculated maintenance dose can be found in Table 3 (PHE) and Table 4 (ACE). For example, a 78-year-old man who starts PHE therapy, with weight of 91 kg, height 180 cm, and genotype CYP2C9*1/*2, and VKORC1 CT, who does not use amiodarone would need a maintenance dose of 1.89 mg/day. The maintenance dose was calculated as follows: square root of the mean weekly dose in mg = 2.874−0.259 (CYP2C9*1/*2) − 0.601(VKORC1 CT) − 0.015* 78 (age) + 0.011* 180 (height) + 0.008*91 (weight) = 3.64 √mg/week. The week dose is in milligram is = 3.64^2 = 13.26 mg/week, which equals 1.89 mg/day. This maintenance dose corresponds (Table 3) to a loading dose of 6 mg on Day 1, 6 mg on Day 2, and 6 mg on Day 3.

Both square root and log transformations are common for the warfarin algorithms.12–17 We used the square root transformation of the maintenance dose since this was on average the best outcome measurement (see Supplementary material online). Explained variability of our algorithms are comparable with earlier developed warfarin algorithms and to the small cohort ACE algorithm.12–16 In addition, the correlations between various prediction scores for the maintenance dose given in the literature and our two algorithms are high (see Supplementary material online), showing similarity between our algorithms and the earlier developed warfarin algorithms.

Based on theory, it could be expected that polymorphisms in the CYP2C9 gene influence the maintenance doses less for PHE than for ACE. However, it was shown in this paper that the effects of these polymorphisms are comparable for PHE and ACE; CYP2C9 explains 4-6 and 4-5% of the dose variability, respectively. It is supported by other studies that polymorphisms in the CYP2C9 gene does influence PHE doses; some studies found lower PHE doses or increased bleeding risks26–28 for patients having an SNP in the CYP2C9 gene, where that of Visser et al.29 did not.

We have considered the use of the CYP2C9 genotype to individualize the elimination half-life of coumarins to calculate the genotype-specific accumulation indexes. However, the available data of the CYP2C9 effect on the elimination half-lives are too limited30 to be used in our algorithm, and additional assumptions would have to be made. In addition, the CYP2C9 genotype is already used as a parameter to calculate the maintenance dose, which is used to derive the loading dose. Therefore, CYP2C9 genotype is indirectly used to estimate the loading dose.

Our study has some limitations. First, some bias might have been introduced. We collected data from current PHE and ACE users. This approach may introduce some selection bias because long-term users are more likely to be selected. However, the distribution of allele frequencies, amiodarone use, sex, and age is similar in cohorts of other studies.13,16,17 Furthermore, we had no data about patient compliance and non-compliance and that could be a source of bias. Nevertheless, non-compliance would reduce the R^2 of the algorithms, because it dilutes the effects. Therefore, we anticipate that the selection and compliance bias have a minor effect. Secondly, it is possible that the estimated height and weight are slightly off due to errors in these variables. Thirdly, we did not include drugs other than amiodarone use, ethnicity, smoking status, and diet as factors in the algorithm, although other drugs have been shown to affect dose requirements in some studies. Interacting drugs other than amiodarone were not significantly associated with the maintenance dose or did not increase the explained dose variation and therefore were not included in the algorithms. The main reason that ethnicity, smoking status, and diet were excluded from the algorithms is because these factors are challenging to assess accurately and objectively. Our aim was to develop a clinically applicable algorithm with the most important determinants that can be easily implemented in a routine care setting.

The strengths of this study are that the algorithms were developed using large patient populations and that the algorithms performed equally well when validated in independent prospective data sets.

The aim of this study was to develop genotype-guided and non-genotype-guided algorithms to determine the maintenance dose of PHE and ACE, and to derive the individualized loading algorithms for PHE. The question of whether the use of these algorithms will improve clinical care will be answered in the upcoming EU-PACT trial, a two-armed, single-blinded, randomized controlled trial taking place in six European countries.31 The main outcome of the trial will be whether a pharmacogenetics-guided algorithm increases the time into the therapeutic range during the first 3 months of therapy. In addition, the cost-effectiveness of pre-treatment genotyping will be assessed looking at adverse events, i.e. thrombo-embolic events and haemorrhages, and the quality-adjusted life-years.

### Supplementary material

Supplementary material is available at European Heart Journal online.

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### Conflict of interest

none declared.

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