Personalized oral anticoagulant treatment

dosing algorithms, drug interactions, and economic aspects

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Geïndividualiseerde orale antistollingsbehandeling

Doseringsalgoritmes, geneesmiddelinteracties en economische aspecten

(met een samenvatting in het Nederlands)

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CHAPTER 1

General Introduction

Introduction

Thrombosis is the common pathology underlying ischemic heart disease, ischemic stroke, and venous thromboembolism (VTE) [1]. Oral anticoagulation therapy with vitamin K antagonists (VKAs) has been used to prevent and treat thromboembolic disease for over seven decades. The first available VKA was dicumarol, established by Karl Paul Link in 1940 [2], but since its market introduction in the 1950s warfarin has become the most commonly used oral anticoagulant worldwide [3]. To date, warfarin remains the first choice among the VKAs for thromboembolic disorders in most countries, especially in the USA and Canada, whereas acenocoumarol and phenprocoumon are mainly used in European countries [4]. During the first decade of the 21st century, non-vitamin K oral anticoagulants, the so-called NOACs or novel oral anticoagulants have been launched [5]. These agents, also known as direct oral anticoagulants (DOACs), include dabigatran etexilate, which is a direct thrombin inhibitor [6] and rivaroxaban, apixaban, and edoxaban, which are direct factor Xa inhibitors [7].

Oral anticoagulants: clinical applications, pharmacology and mechanism of action

VKAs are indicated for the prevention and treatment of venous thromboembolism mainly in relation to non-valvular atrial fibrillation (NVAF), deep venous thrombosis (DVT), pulmonary embolism (PE), prosthetic heart valves and after a myocardial infarction [8]. DOACs have been approved for the prevention of venous thromboembolism after orthopedic surgery, of stroke and systemic embolism in adult patients with NVAF and for the treatment and prevention of DVT and PE [9-11].

The mechanisms of action of indirect (VKAs) and direct oral anticoagulants (DOACs) are presented in Figure 1 [3]. VKAs exert their anticoagulant effect by inhibiting the cyclic interconversion of vitamin K and its 2,3 epoxides (vitamin K epoxide), thereby modulating the γ-carboxylation of glutamate residues on the N-terminal regions of vitamin K-dependent proteins [12-16]. As vitamin K serves as a co-factor in the activation of coagulation factors II, VII, IX, and X, the inhibition of its recycling results in a strong anticoagulation activity [15]. On the other hand,

vitamin K also serves as a co-factor for the anticoagulant proteins C, S and Z [18], which also affects the regulation of the procoagulant-anticoagulant system. As it takes time for decarboxylated coagulation factors to appear in plasma it takes some hours for VKAs to have their anticoagulant effect.

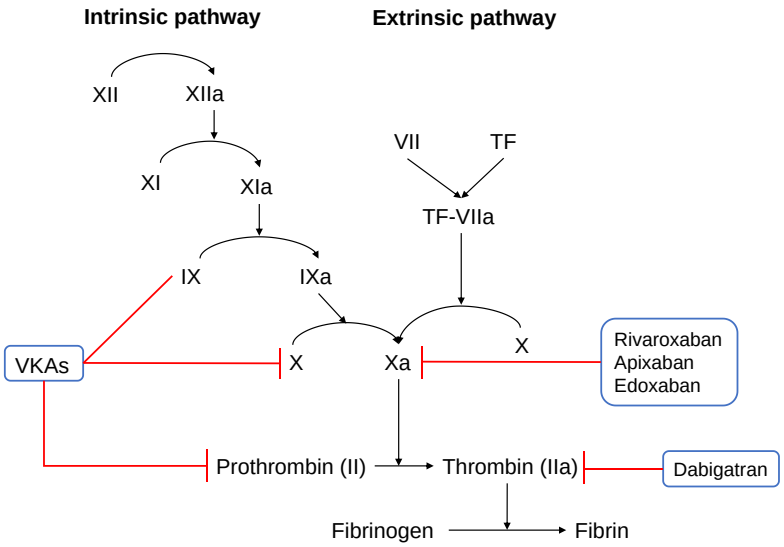

Figure 1 Mechanism of anticoagulants effect of VKAs and (DOACs), adapted from Mekaj *et al* [9].

Unlike VKAs, DOACs produce their anticoagulant effect by inhibiting one specific coagulation factor, either thrombin or active factor Xa and therefore have a direct anticoagulant effect. Dabigatran etexilate is a low molecular weight non-active pro-drug administered orally that is converted into dabigatran, a potent competitive and reversible direct thrombin inhibitor [19]. By inhibiting thrombin, dabigatran prevents a cascade of events: conversion of fibrinogen into fibrin, positive feedback amplification of coagulation activation, cross-linking of fibrin monomers, platelet activation and inhibition of fibrinolysis [19]. Other DOACs currently on the market are all factor Xa antagonists, being rivaroxaban, apixaban, and edoxaban. These

agents are reversible direct Xa antagonists. They exert their anticoagulant activity by the direct inhibition of factor Xa, which is formed by both the intrinsic and extrinsic coagulation pathways [20]. Activated factor Xa links the intrinsic and extrinsic coagulation pathways and acts as a rate-limiting step in thrombin formation. The prevention of the conversion of thrombin from prothrombin is needed to prevent the conversion of fibrinogen to fibrin [21]. Therefore, the inhibition of factor Xa activation produces a direct effect on the coagulation cascade.

Although VKAs have an acceptable benefit-risk ratio [22, 23], their use in daily practice is hampered in a few ways. They have a narrow therapeutic index necessitating frequent monitoring of their anticoagulant effect. Furthermore, there are numerous pharmacokinetic and pharmacodynamic interactions influencing the thromboembolic and bleeding risks of coumarins [23-25]. Finally, several patient characteristics such as sex, age, weight, height, genotype, disease conditions like heart failure, and dietary intake of vitamin K influence the benefit-risk ratio of these drugs [276-31]. *CYP2C9* and *VKORC1* polymorphisms have a strong impact on the dosage of VKAs that an individual patient needs for optimal anticoagulation [32, 33]. Several dosing algorithms have been developed that use information of *VKORC1* and *CYP2C9* genotypes, as well as patient characteristics such as age, gender, height and weight [33, 34] to predict the optimal initial dose and maintenance dose for VKAs.

DOACs have the advantage that they do not need monitoring of their anticoagulant effects. They can be prescribed in a standard dose, although in case of renal failure, older age and drug interactions lowering of the dose might be necessary [7]. Comparable to VKAs there are relevant genes by which pharmacokinetic interactions with other drugs can occur, especially CYP3A4 and P-glycoprotein [36, 37].

Unresolved relevant research questions

At the start of the thesis project there were several relevant research questions in relation to the use of the VKAs and DOACs in daily practice.

First, there was unclarity whether in developed dosing algorithms for VKAs genotype information has an added value compared to clinical characteristics alone.

The effects of pharmacogenetic dosing algorithms were evaluated by several large randomized clinical trials, in which dosing algorithms with genotype and clinical characteristics were compared with algorithms that only included clinical characteristics [35, 36] or a dosing adjustment strategy as normally used in daily clinical practice [37]. Examples of these trials were the European Pharmacogenetics of Anticoagulants Therapy (EU-PACT) trials which were randomized, multi-center, controlled trials conducted to assess the effects of genotype guided dosing for warfarin [37], acenocoumarol and phenprocoumon [35]. In the warfarin trial of EU-PACT an algorithm with genetic and clinical information was compared with standard care (standard dose without the use of an algorithm). It appeared that the time in the therapeutic range (TTR) of the INR was improved by the algorithm compared to standard care. In the acenocoumarol / phenprocoumon part an algorithm with genetic and clinical information was compared with an algorithm with only clinical information. For the primary outcome (TTR of the INR during the first 3 months) there were no statistically significant differences between the two dosing strategies. So, it appears that the genotype information has no added value when added to clinical information for acenocoumarol and phenprocoumon. But the question remained whether the clinical dosing algorithm without genotype would perform better than standard care (without algorithm) for these two VKAs.

Second, up to now it has not been evaluated whether age modifies the predicted value of developed dosing algorithms of VKAs. Age is associated with changes of the pharmacokinetics and pharmacodynamics of drugs; thus, it is relevant to know whether dosing algorithms can be improved by taking into account a possible age modifying effect.

Third, also for the DOACs pharmacokinetic and pharmacodynamic interactions have been determined. For some drugs these interactions have been formally studied for other drugs these interactions were not studied but anticipated because of inhibiting or inducing effects of drugs on CYP3A4 and P-glycoprotein. It is largely unknown what the bleeding risks are when a DOAC is combined with a drug known or anticipated to interact with DOACs. Furthermore, it is unknown whether in daily practice dose adjustments of DOACs are performed when these drugs are

combined with potentially interacting drugs. Such knowledge is relevant to learn whether the safe use of DOACs can be improved.

Fourth, DOACs are new and expensive and cheaper generics are not yet available. The benefit risks of DOACs appear to be better than those of the VKAs. The question arises what the costs are of DOACs compared to the costs of VKA treatment when optimally dosed (with a dosing algorithm) and how differences in costs relate to the improved benefit-risk. It is therefore important to study the cost-effectiveness of DOACs versus algorithm dosed VKAs.

Objectives and outline of this thesis

The objectives of this thesis are to:

1. further explore the relevance of dosing algorithms for VKAs compared to standard dosing and how these algorithms perform in different age groups.

2. evaluate the influence of drug interactions on the safety of DOACs in daily clinical practice and whether health care practitioners take into account drug interactions when deciding on the prescribed dose of DOACs.

3. study the cost-effectiveness of a variety of clinical and genotype-guided dosing algorithms for VKAs versus DOACs.

In **Chapter 2**, a study is presented in which we compared the anticoagulant effect of dosing algorithms for acenocoumarol and phenprocoumon including clinical patient characteristics with standard care in the Netherlands. **Chapter 3** describes a study in which we compared the effect of genotype-guided dosing of acenocoumarol or phenprocoumon in younger and older patients. **Chapter 4** of this thesis focuses on the influence of drug interactions on the safety of DOACs. We conducted a case control study in the UK Clinical Practice Research Datalink to investigate the association between concurrent use of potential pharmacokinetic or pharmacodynamic interacting drugs and major bleeding among DOAC users. In **Chapter 5**, we describe the frequency of adjustments of DOAC treatment including dose adjustment, discontinuation of use, and switching to a VKA when interacting drugs were concomitantly used. In **Chapter 6**, we assessed the cost-effectiveness of DOACs versus different clinical and genotype-guided dosing algorithms for

acenocoumarol and phenprocoumon as well as standard care in Dutch patients with atrial fibrillation. Finally, in **Chapter 7**, we summarize our main findings and discuss them in a broader perspective, discuss strengths and limitations and present recommendations for daily practice and further research.

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CHAPTER 2

Comparison of dosing algorithms for acenocoumarol and phenprocoumon using clinical factors with the standard care in the Netherlands

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Abstract:

Background: It has not been investigated how much the use of clinical factors in a dosing algorithm improves the percentage of time in therapeutic range (TTR). The present study aimed to compare the effect of dosing algorithms for acenocoumarol and phenprocoumon including clinical patient characteristics with standard care in the Netherlands.

Setting: The pre-EU-PACT study, an observational study in the Netherlands, was used to obtain standard care INR data. INR data from the Dutch patients in the EU-PACT trial (comparing the use of a clinical algorithm with and without genetic information) was used for the clinical dosing algorithm.

Methods: For both acenocoumarol and phenprocoumon, the percentage of time in, below and above therapeutic International Normalized Ratio (INR) range during 12 weeks after treatment initiation were assessed in both studies.

Results: During the weeks 2-12, the clinical dosing algorithm of acenocoumarol (80 patients) led to a higher TTR (74.3% versus 68.0% in range 2.0-3.5, 95% Confidence interval [CI] difference: 0.5% to 11.8%), and a reduced percentage of time below INR 2 and above INR 3.5, compared with standard care (272 patients). For phenprocoumon, compared with standard care (484 patients), 80 patients treated by the dosing algorithm did not obtain a significantly higher TTR in range 2.0-3.5 or a lower percentage of time above 3.5, however, they spent more time with INR below 2.

Conclusions: The use of a clinical dosing algorithm for acenocoumarol seemed to improve the quality of anticoagulation therapy during the first 2-12 weeks after treatment start. For phenprocoumon, there was no statistically significant difference in anticoagulation control.

Introduction

Vitamin K antagonists (VKA) or coumarin derivatives, such as warfarin, acenocoumarol and phenprocoumon are effective for the treatment and prevention of thromboembolic disease [1]. However, their use is challenging due to the narrow therapeutic window and high inter- and intra-individual variability in dose response. Therefore, the international normalized ratio (INR), a measurement of anticoagulation activity is regularly measured, and used to guide dosing of these drugs. To improve the management of oral anticoagulant treatment, several computerized algorithms have been developed to assist physicians with their dosing decisions and using these has been shown to be superior to traditional dosing [2-4]. In the Netherlands, the use of similar computerized algorithms (e.g. TRODIS, TDAS) are considered standard care in the anticoagulation clinics [5, 6].

The dosage of coumarin anticoagulant agents needed by an individual patient is influenced by several factors, including age [7], sex, height, weight [8], concurrent drug therapy [9], vitamin K intake [10], and genetic factors [11, 12]. In recent years, more emphasis has been put on establishing dosing algorithms that include these factors to achieve the optimal individual dosing strategy for coumarins. Several clinical trials were conducted to evaluate the effectiveness of these algorithms. The Clarification of Optimal Anticoagulation through Genetics (COAG) trial and the European Pharmacogenetics of Anticoagulants Therapy (EU-PACT) trial were randomized, multi-center, controlled trials conducted to assess the effect of genotype guided dosing for warfarin [13], acenocoumarol and phenprocoumon [14], respectively. These two trials had similar designs which compared the dosing algorithm including genetic information with an algorithm based on clinical parameters only that did not include genetic information. The result of the acenocoumarol and phenprocoumon arm of the EU-PACT trial indicated that there was no statistically significant difference between the genotype-guided algorithm and the clinical algorithm in the primary outcome of the trial (time in the therapeutic INR range during the 12 weeks of treatment) [14]. However, during the first 4 weeks of therapy, patients in the genotyped arm spend more time in therapeutic INR range. In contrast, the COAG trial showed no differences in percentage of time in therapeutic range in the initial 4 weeks of treatment between genotype-guided and

clinically dosing algorithms. The warfarin arm of the EU-PACT trial assessed the clinical utility of genotype-guided warfarin dosing by comparing an algorithm containing clinical and genetic information with standard care (standard dose) [15]. This trial did show an effect on the primary endpoint: patients in the genotyped arm spent 7% more time in therapeutic range in the first 12 weeks of treatment [15]. However, because none of the trials included three arms (standard care, clinical algorithm with and clinical algorithm without genetic information), it remains unclear what the effect of the use of the clinical dose algorithm without genetic information is versus standard care. Previously, the IWPC consortium showed that compared with a fixed dose approach, estimates from a clinical algorithm predicted warfarin actual stable dose better [16]. It is therefore hypothesized that the use of a clinical algorithm for acenocoumarol and phenprocoumon will result in a better outcome than standard care, and this might explain the different findings of the COAG trial [13], the EU-PACT acenocoumarol/phenprocoumon arm [14] and the EU-PACT warfarin arm [15]. The best way to make this comparison would of course be in a direct clinical trial. However, because it is highly unlikely that a clinical trial will be performed on this subject the aim of the present study is to compare the effect of a dose algorithm for acenocoumarol and phenprocoumon that included only clinical variables in the EU-PACT trial with a historic control group treated according to the standard care in the Netherlands.

Methods

Study design and study population

For the present study, data of acenocoumarol and phenprocoumon patients who were treated in the Netherlands were obtained from the EU-PACT trial [14] and from the pre-EU-PACT study [17]. In brief, the EU-PACT was a multicenter, single blind, randomized, controlled trial designed to test the effectiveness of three genotype guided coumarins (acenocoumarol, phenprocoumon, and warfarin) dosing respectively. The acenocoumarol trial was conducted in the Netherlands and in Greece, and the phenprocoumon trial was conducted in the Netherlands [14]. In the EU-PACT trial, patients of 18 years or older who were diagnosed with atrial fibrillation or venous thromboembolism and who had not received either acenocoumarol or phenprocoumon therapy preciously were enrolled and randomly

assigned, in a 1:1 ratio to the use of a dosing algorithm that included both clinical information (age, sex, height, weight and amiodarone use) and genotype data for *VKORC1* and *CYP2C9* or to a dosing algorithm with only clinical information. For each group, patients received a dose according to a loading algorithm during the first 3 days and a dose-revision algorithm on days 4 or 5 determined by the clinical algorithm and first INR value. After day 5, dose was adjusted according to the INR results using local procedures. The patients were followed for 3 months with a target INR range of 2.0 to 3.0 [14]. In the present study, we only included patients dosed by the clinical algorithm and only used the data that were gathered in the Netherlands.

Data of the standard care group was from the observational pre-EU-PACT study, in which patients who were using acenocoumarol and phenprocoumon during November 2009 with a target INR in the lowest intensity category (according to Dutch guidelines INR 2.0-3.5) were included. Data was obtained from the electronic registry databases of the Anticoagulation Clinic Leiden (phenprocoumon) and the Anticoagulation Clinic Medial in Hoofddorp (acenocoumarol). These patients were treated according to standard care in the Netherlands, with the help of a computerized algorithm. In the pre-EU-PACT study, patients with an INR 1.5 or greater on the first day were excluded, because their treatment probably started earlier in a hospital or another thrombosis service, and therefore they were not incident starters with coumarin therapy. The Medical Ethics Committee of the Leiden University Medical Center approved both of the study protocols and patients provided informed consent before inclusion into the study. More detailed descriptions of the two studies can be found in earlier publications [14, 17, 18].

Outcome measure

The primary outcome of the present study was the percentage of time in the therapeutic INR range (TTR) during 12 weeks after the initiation of acenocoumarol or phenprocoumon therapy. In the EU-PACT trial, all patients were treated with a target range of 2.0 to 3.0, while in pre-EU-PACT study the target was 2.0-3.5, according to standard practice in the Netherlands. Therefore, in this study, percentage of time in 2.0-3.5 was calculated. Percentage time in target range 2.0-3.0 was calculated as a sensitivity analysis. The percentage of time below (INR<2),

in (INR 2.0-3.5) and above (INR>3.0 and INR>3.5) the therapeutic range in both groups was compared. The TTR was calculated by using linear interpolation according to Rosendaal's method [19].

Statistical Analysis

Data for patients dosed according to the clinical algorithm in the present study was collected in different anticoagulation clinics. As a sensitivity analysis we performed center specific analyses using one-way ANOVA, there were no statistically significant differences in the clinical algorithm group for all outcomes in the different clinics, therefore we pooled the data in the rest of the analyses. The mean differences of the TTR between the clinical dosing algorithm from EU-PACT and standard care in the Netherlands from pre-EU-PACT with 95% confidence intervals (Cis) were calculated and compared with an independent-samples T test. The mean differences of TTR were adjusted for possible confounders using multiple linear regression. For acenocoumarol users, the adjustments were made for *CYP2C9* and *VKORC1* genotype, age, and indication. For phenprocoumon users, the adjustments were only made for *CYP2C9* and *VKORC1* genotype, and age. Indication was not used in the phenprocoumon model because it did not change the R Square in a univariate analysis.

Chi-Square Tests were used for comparison of categorical variables. Patients included in the analyses were treated at least 4 weeks. To increase power, patients with at least 10 weeks of follow-up were included for the analyses of 12 weeks except the separate analyses for the first 4 weeks and for weeks 5 through 8 which included patients with at least 4 weeks and 8 weeks follow-up, respectively. Two sensitivity analyses for the comparison of the primary outcomes were performed. In the first analysis only patients with at least 12 weeks follow-up were included. Because of the differences in study design the amount of INR measurements differed between the clinical algorithm and the standard care groups during the first month (see in the supplement Table S1). We performed another sensitivity analysis that excluded measurements in the first week and compared the TTR in week 2-12 weeks and 2-4 weeks between the groups. The number of measurements in the first week (as defined by the protocol) was much higher in the clinical trial, and this enlarges the chance of finding values outside therapeutic range. Therefore, we

show the results of both 1-12 and 2-12 weeks. All analyses were performed with IBM SPSS Statistics version 20.0 (IBM Corp., USA).

Results

Patient cohort

In this study we used data from the clinical algorithm group of the EU-PACT trial [14] and from the pre-EU-PACT study [17]. Of the 381 acenocoumarol users enrolled in the EU-PACT trial, 82 patients in the Netherlands were enrolled in the control arm and therefore eligible for the analyses in this study. Of the 471 acenocoumarol users in the pre-EU-PACT study, 272 patients were eligible for the present study. 1 pregnant patient, 113 patients who used phenprocoumon for a period of time during the first 3 months or who had a different target INR range, and 3 patients who changed anticoagulation clinics were excluded. Of the remaining patients, 65 patients who did not have a reliable start date, 14 patients who had an INR higher than 1.5 on the first day and 3 patients who had less than 2 INR measurements during the first 4 weeks were excluded. For phenprocoumon there were 167 patients in the EU-PACT trial. After excluding 83 patients treated according to genotype-guided dosing algorithm, 1 patient who withdrew the informed consent and 1 patient treated less than 4 weeks, 82 patients were included in the clinical algorithm group. Out of the 624 phenprocoumon users from the pre-EU-PACT study, 69 were excluded because they changed anticoagulation clinics, they were treated with acenocoumarol for a period of time during the first 3 months, or they had a different target INR range. Furthermore, 32 patients without a reliable start date, 37 patients with an INR greater than 1.5 on the first day, and 2 patients treated less than 4 weeks were excluded; therefore 484 patients were eligible in the present study. Patient characteristics are shown in Table 1. The selection flowchart can be found in the supplement.

There were no major differences between the clinical algorithm group and the standard care group in sex distribution or average height and weight (Table 1). The mean age in the clinical algorithm group for both acenocoumarol (65 versus 74) and phenprocoumon (67 versus 70) users was lower than that in the standard care group as shown in Table 1.

Table 1. Characteristics of included patients.

Characteristics	Acenocoumarol			Phenprocoumon		
	Clinical algorithm group (n=82)	Standard care group (n=272)	*P*-value	Clinical algorithm group (n=82)	Standard care group (n=484)	*P*-value
Male sex, *n* (%)	45 (54.9)	153 (56.3)	0.83	46 (56.1)	275 (56.8)	0.90
Age, yr, mean±SD	65± 13	74±9	0.00	67±11	70±11	0.01
Height, cm, mean±SD	175± 11	173±11	0.07	174±10	173±9	0.34
Weight, kg, mean±SD	86± 20	81± 19	0.06	83±16	81±17	0.42
Indications			0.01			0.16
Atrial fibrillation	62 (75.6%)	233 (85.7%)		68 (82.9%)	424 (87.6%)	
Venous thromboembolism	20 (24.4%)	32 (11.8%)		14 (17.1%)	52(10.7%)	
Others	-	7 (2.6%)		-	8 (1.7%)	
CYP2C9 genotype			0.07			0.97
missing	-	10 (3.7%)		2 (2 %)	21 (4.3%)	
*1*1	50 (61%)	170 (63%)		56 (68%)	309 (64%)	
*1*2	14 (18%)	53 (20%)		14 (17%)	86 (18%)	
*1*3	11 (13%)	31 (11%)		7 (9%)	47 (10%)	
*2*2	6 (7%)	3 (1%)		2 (2%)	11 (2%)	
*2*3	1 (1%)	4 (2%)		1 (1%)	7 (1%)	
*3*3	0	1 (0)		0	3 (1%)	
HWE†, *P*-value	0.02	0.94		0.77	0.33	
VKORC1 genotype			0.15			0.61
missing	-	9 (3.3%)		2 (2.4%)	20 (4.1%)	
GG	36 (44%)	91 (34%)		33 (40%)	174 (36)	
GA	33 (40%)	138 (51%)		33 (40%)	219 (45%)	
AA	13 (16%)	34 (13%)		14 (17%)	71 (15%)	
HWE, *P*-value	0.25	0.10		0.26	0.88	

† HWE denotes Hardy –Weinberg equilibrium.

TTR for acenocoumarol users and phenprocoumon users

As shown in Table 2, among acenocoumarol users, the TTR in the clinical algorithm group was higher than the standard care group both during 12 weeks (mean difference 5.0%, 95%CI: 0.0 to 10.0) and the first 4 weeks (11.1%, 95%CI: 3.6 to 18.6). The sensitivity analyses that excluded the first week showed similar results.

In adjusted analyses, the TTR differed by 6.2% (95% CI: 0.5 to 11.8) through week 2-12 and 12.2% (95%CI: 3.3 to 21.0) through week 2-4. The TTR of the clinical algorithm group in 9-12 weeks was also higher than that in standard care group (9.1 %, 95%CI: -0.2 to 18.4). During weeks 5 to 8, the TTR of the clinical algorithm group was a little lower than the standard care group.

Table 2. Percentage of time in the therapeutic range 2.0-3.5 during 12 weeks*.

	TTR in range 2.0-3.5					
Analysis	**Clinical algorithm group**	**n**	**Standard care group**	**n**	**Unadjusted Difference (95% CI)**	**Adjusted Difference # (95% CI)**
Acenocoumarol						
Exclude the first week						
Week 2-12	74.3±20.4	80	68.0±20.6	271	6.3 (1.2 to 11.5) $	6.2 (0.5 to 11.8) $
Week 2-4	68.5±33.5	82	53.2±33.0	272	15.3 (7.1 to 23.5) $	12.2 (3.3 to 21.0) $
Week 5-8	71.3±31.3	82	72.1±29.5	272	-0.8 (-8.1 to 6.7)	0.8 (-7.3 to 9.0)
Week 9-12	80.6±26.9	80	74.3±30.4	271	6.3 (-0.7 to 13.3)	6.3 (-1.9 to 14.6)
Include the first week						
Week 1-12	71.8±19.4	80	66.8±20.1	271	5.0 (0.0 to 10.0) $	4.6 (-0.9 to 10.0)
Week 1-4	62.3±28.6	82	51.2±30.5	272	11.1 (3.6 to 18.6) $	7.6 (-0.4 to 15.5)
Phenprocoumon						
Exclude the first week						
Week 2-12	75.9±21.5	80	70.1±24.7	470	5.7 (-0.03 to 11.5)	4.5 (-1.3 to 10.3)
Week 2-4	60.9±34.4	82	61.3±34.5	484	-0.4 (-8.5 to 7.7)	-1.4 (-9.6 to 6.7)
Week 5-8	75.0±29.2	82	69.6±34.2	476	5.4 (-1.7 to 12.5)	4.3 (-3.6 to 12.2)
Week 9-12	87.6±22.5	80	77.8±31.1	470	9.8 (4.1 to 15.5) $	8.3 (1.2 to 15.5) $
Include the first week						
Week 1-12	71.3±20.4	80	68.7±23.4	470	2.8 (-2.8 to 8.1)	1.5 (-3.9 to 7.0)
Week 1-4	51.2±27.3	82	58.2±29.3	484	-7.1 (-13.9 to -0.3) $	-7.9 (-14.8 to -1.0) $

*Data were expressed as: mean±SD
#Adjusted for age, CYP2C9 and VKORC1 genotype, and (for acenocoumarol only) indication.
$*P* <0.05

For phenprocoumon users, during the 12 weeks initial treatment period, the clinical algorithm group obtained a 1.5% (95% CI: -3.9 to 7.0) improvement in the TTR compared with the standard care group (71.3% versus 68.7%). However, during

the first 4 weeks, the clinical algorithm led to a clear -7.9% difference (95% CI: -14.8 to -1.0) compared with standard care. Without including the first week, the difference was 4.5% (95% CI: -1.3 to 10.3) and -1.4 % (95% CI: -9.6 to 6.7), respectively.

A sensitivity analyses was performed for the TTR in range 2.0-3.0, which gave similar results both for acenocoumarol and phenprocoumon, data are provided in the Supplementary Table S2. The sensitivity analyses including data from patients with at least 12 weeks follow up also showed similar results (Supplement Table S3 and Table S4).

Sub-therapeutic INR values

Figure 1 shows the percentage of time with an INR < 2 in patients treated with acenocoumarol and phenprocoumon. Among acenocoumarol users in the clinical algorithm group the percentage of time with INR below 2 was less than in the standard care group during all the 12 weeks (clinical algorithm 19.5% vs. standard care 22.7%, 95%CI of the mean difference: -8.1 to 1.9) and the first 4 weeks (clinical algorithm 24.6% vs. standard care 38.9%, 95%CI of the mean difference: -21.4 to -7.1). In contrast with acenocoumarol users, the patients treated with phenprocoumon according to the clinical algorithm spent more time in INR range <2 than the standard care group, both in all the 12 weeks (clinical algorithm 19.3% versus standard care 13.1%, 95%CI of the mean difference: 1.7-10.0) and in the first 4 weeks (clinical algorithm 37.6% and standard care 22.8%; 95% CI of the mean difference: 7.8- 19.8). However, when we excluded the first week, the clinical algorithm and standard care differed only 2.9 % (95%CI: -1.5 to 7.2) in week 2 to 12, and 7.0 % (95% CI: 0.2 to 13.7) in week 2 to 4 (data are shown in the supplement Table S5 and Table S6).

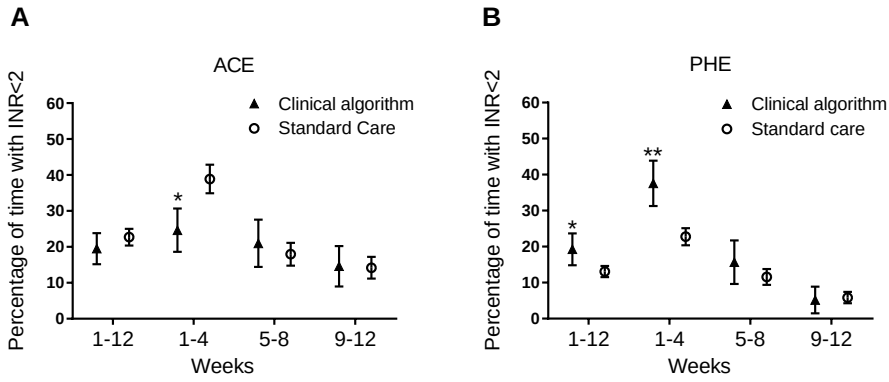

Figure 1 Percentage of time with INR below 2 in different time periods; **A**: acenocoumarol (ACE); **B**: phenprocoumon (PHE). All the data are indicated as mean±95% confidence interval (**P<0.01; *P<0.05).

Supra-therapeutic INR values

Figure 2 shows the percentage of time with INR above 3.5. During the 12 weeks treatment period, acenocoumarol patients dosed according to the clinical care algorithm spent less time in INR above 3.5 than the standard care group (Figure 2A). There is a declining trend of the percentage of time with INR above 3.5 in the clinical algorithm group, while in contrast, the time spent in INR above 3.5 increased with time in the standard care group. During the first 4 weeks the percentage of time above 3.5 in the clinical algorithm group was higher than with standard care. However, this situation was reversed in the last 4 weeks. In that period the percentage of time above 3.5 was statistically significantly lower (-6.3%, 95%CI of the mean difference: -12.0 to -0.5) in the clinical algorithm group. The sensitivity analysis that excluded the first week showed similar results and data are shown in the supplement Table S5.

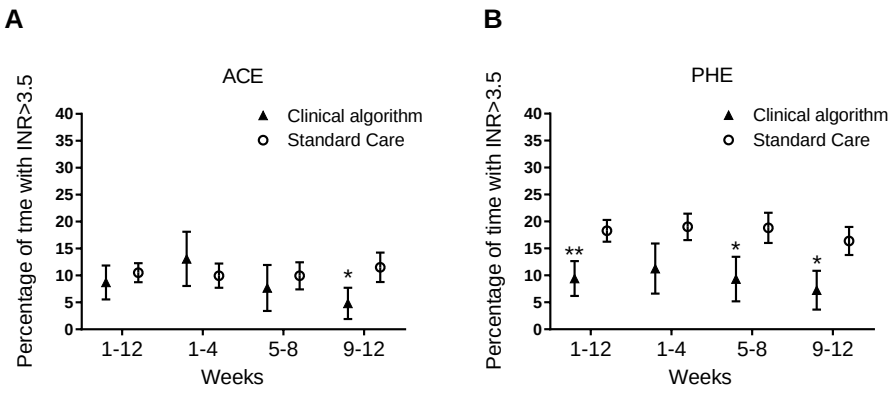

Figure 2 Percentage of time with INR above 3.5 in different time periods; **A**: acenocoumarol (ACE); **B**: phenprocoumon (PHE). All the data are indicated as mean±95% confidence interval (**P<0.01; *P<0.05).

For phenprocoumon users, the percentage of time with INR above 3.5 is shown in Figure 2B. Use of the clinical algorithm led to a lower percentage of time in INR range >3.5 both in all 12 weeks (clinical algorithm 9.4% vs. standard care 18.3%, 95%CI of the mean difference: -12.3to -2.4) and in first 4 weeks (-5.9%, 95%CI of the mean difference: -11.7 to 0.0). We also calculated the percentage of time with INR above 3 and results were similar as for INR above 3.5, therefore, data are provided in the supplement Table S6.

Discussion

For the initiation of treatment with acenocoumarol or phenprocoumon the present study compared the use of a dosing algorithm that included clinical factors with standard care in the Netherlands. The clinical algorithm for both acenocoumarol and phenprocoumon led to a higher TTR during weeks 2-12 after the initiation of treatment, while only for acenocoumarol there was a significant difference.

Our data of the clinical algorithm were from the control group of the EU-PACT trial [14] which had a therapeutic INR range of 2.0-3.0, while according to clinical practice in the Netherlands, the therapeutic INR range was 2.0-3.5 for the therapy of atrial fibrillation or venous thromboembolism, therefore, we evaluated not only the TTR in both ranges but also the percentage of time below and above these ranges.

For acenocoumarol, there were no statistically significant differences between the clinical algorithm group and standard care group in the percentage of time with INR below 2 and above 3.5 during weeks 2-12. In contrast, in the clinical algorithm group, phenprocoumon users spent remarkably more time with INR below 2 but less time above 3.5 during the first 2-12 weeks. These findings indicate that using the clinical algorithm for acenocoumarol could lead to more benefit.

Because of the differences in study design the amount of INR measurements differed between the clinical algorithm and the standard care groups. According to the trial protocol all patients in the EU-PACT trial had a baseline INR measurement on the first day, with the second and the third measurement planned on day 4 and 6, respectively while in the pre-EU-PACT observational study the baseline INR measurement was not known and on average 1 INR measurement was conducted during the first 7 days. (Supplementary Table S1). Consequently, in the standard care group, the calculated TTR and the percentage of time below and above the range during the first week could not be as accurate as that in the clinical algorithm group. This might have influenced our results. We therefore performed sensitivity analyses for all the outcomes by excluding the first week.

Our study used the percentage of time in, below and above the therapeutic range which is a reflection of anticoagulation quality, to evaluate the effectiveness of the clinical dosing algorithm [20, 21]. However, earlier studies have showed that improvement in TTR led to an improvement in clinical outcomes [22, 23]. The present study suggests that a clinical dosing algorithm could improve the TTR of acenocoumarol users. However, for phenprocoumon, the clinical algorithm may not be associated with more benefit because there were no statistically significant improvements in TTR during 2-12 weeks. Furthermore, although the clinical algorithm for phenprocoumon led to remarkable less time with INR above the range, it led to more time below the range as well, which may increase the risk of thromboembolism [20, 24], especially during the initial 4 weeks of treatment.

It's interesting that we only detect a significant difference among acenocoumarol users between the clinical dosing algorithm group and the standard care. A possible explanation is that in the Netherlands, the long-acting phenprocoumon has been associated with a better quality of anticoagulation therapy than the short-acting

acenocoumarol [25, 26]. In our study, phenprocoumon users in the standard care group had a higher TTR compared with the acenocoumarol users. While in the clinical algorithm group, the TTR of phenprocoumon users was similar to the TTR of the acenocoumarol users. For the acenocoumarol users there was more to gain with the clinical algorithm. This is a plausible explanation why there was a statistically significant difference for acenocoumarol users and not for the phenprocoumon users.

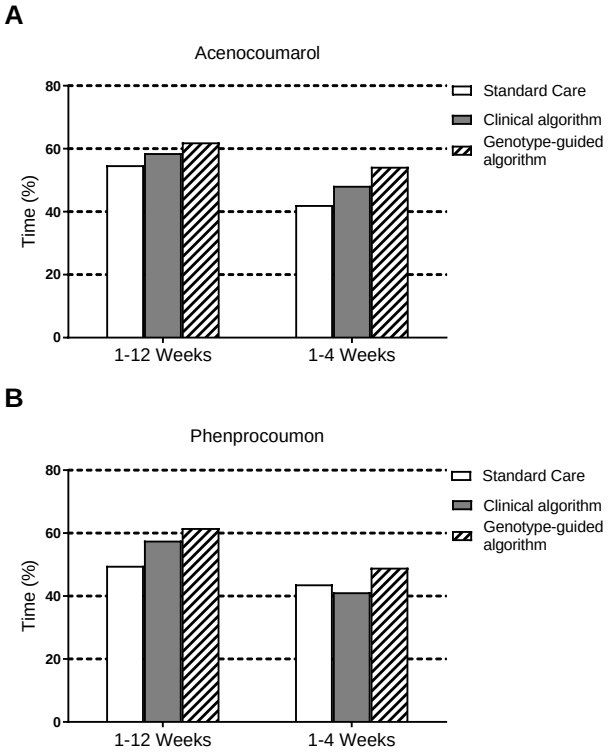

Figure 3 Percentage of time in INR range 2.0-3.0 during 12 weeks and the first 4 weeks for standard care, clinical dosing and pharmacogenetic-guided dosing.
A: acenocoumarol. **B**: phenprocoumon. Data of the genotype-guided group was from the Table 2 of the EU-PACT trial [14].

Another question to answer is whether the use of the clinical algorithm as a comparator may account for the difference in the results between the EU-PACT acenocoumarol/phenprocoumon parts and the EU-PACT warfarin part. Combining the results from the trial and this study we drew a picture that compared three

approaches for dosing acenocoumarol or phenprocoumon in the target INR range 2.0-3.0 (Figure 3). The more information is considered, the more robust the dosing algorithm will be. Data from the acenocoumarol/phenprocoumon part of the EU-PACT trial [14] indicate that during the first 12 weeks of treatment, genotype-guided dosing algorithm for acenocoumarol achieved approximately 3.4% more time in the therapeutic range (2.0-3.0) compared with the clinical algorithm, and for phenprocoumon, almost 2.5% more. However, both of the differences are not statistically significant. In our present study, this clinical dosing algorithm was compared with observational data using standard care in the Netherlands, which showed 3.5% improvement in TTR in range 2.0-3.0 for acenocoumarol and 6.5% for phenprocoumon during the first 12 weeks. Combining the genetic algorithm group of the acenocoumarol/phenprocoumon parts of the EU-PACT trial [14] and the present study, it seems that the clinical dosing algorithm led to an improvement compared with the standard care and the genetic algorithm achieved even more improvement compared with the clinical algorithm group in TTR during 12 weeks of treatment while neither of these improvement was statistically significantly different. The difference in comparator between the EU-PACT acenocoumarol /phenprocoumon arm and the warfarin arm partly account for the difference in the magnitude of the effect in both arms [15]. It is expected that compared with standard care, the use of an algorithm that includes both clinical factors and genotyping information will be the most optimal approach to predict acenocoumarol or phenprocoumon dose. However, it is unclear whether the small improvement is clinically relevant and cost-effective.

Our study has several limitations. First, the small number of patients in the clinical algorithm group caused a wide confidence interval, nevertheless we have detected a statistically significant difference. In addition, data used in the present study were derived from two studies that aimed at two different therapeutic INR ranges which will result in different way of dosing. When a higher target range is used, patient will naturally spend less time with a lower INR. Although we calculated the outcomes by using both INR ranges 2.0-3.0 and 2.0-3.5, interpretation problems remain. What's more, several variables may arise bias thus were used to correct the results. It is well known that with increasing age it is more difficult to keep the INR within

the therapeutic range [27, 28]. In our study, patients in the clinical algorithm group are on average younger than those in the standard care group, especially among acenocoumarol users. However, we do not expect that this has changed our results because we adjusted our results for age. Another variable is the genotype. However, there was no statistically significant difference in the distribution of *CYP2C9* and *VKORC1* genotypes between the clinical algorithm and the standard care groups. Therefore, the differences between groups in the present study were not caused by differences in frequencies of the *CYP2C9* and *VKORC1* genotypes but clinical factors. Finally, data of the clinical algorithm group was from a clinical trial while the standard care group was an observational study, which might have influenced our results. However, also for the observational pre-EU-PACT study an informed consent had to be signed before inclusion. Therefore, the patients in the observational study were a similar selection of the general population, and we do not expect that differences in source population will have influenced our results.

Conclusion

Using a clinical dosing algorithm for acenocoumarol resulted in more time in therapeutic range compared with standard care during the first 12 weeks of treatment in the Netherlands. For phenprocoumon effects were in the same direction, but the difference was not statistically significant. The quality of anticoagulation therapy may be improved by using a clinical dosing algorithm without knowing the genotype. Moreover, since dosing by the clinical algorithm could improve the percentage of time within the therapeutic INR range compared with the standard care, at least part of the difference between the outcome of the EU-PACT acenocoumarol/phenprocoumon arm and the EU-PACT warfarin arm can be explained by the use of the clinical dosing algorithm versus standard care.

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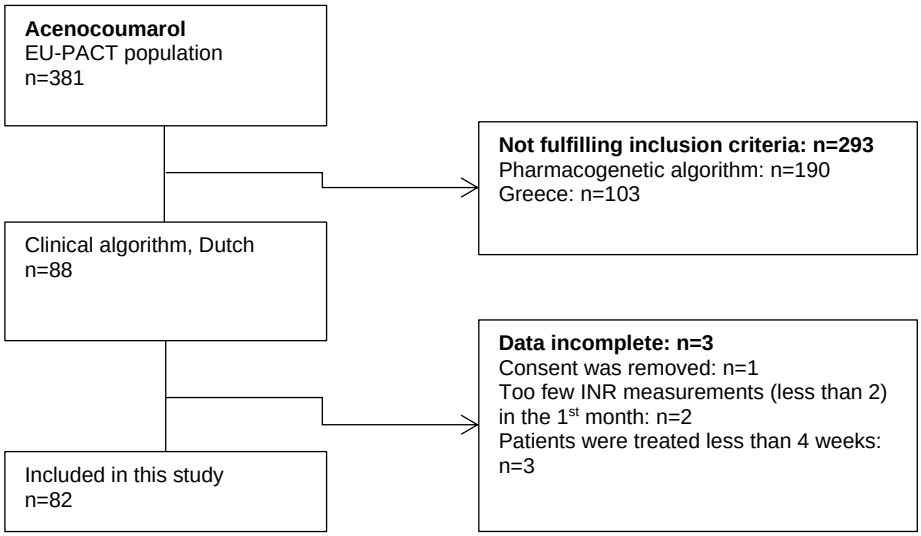
Supplement

A. Clinical algorithm group



B. Standard care group:

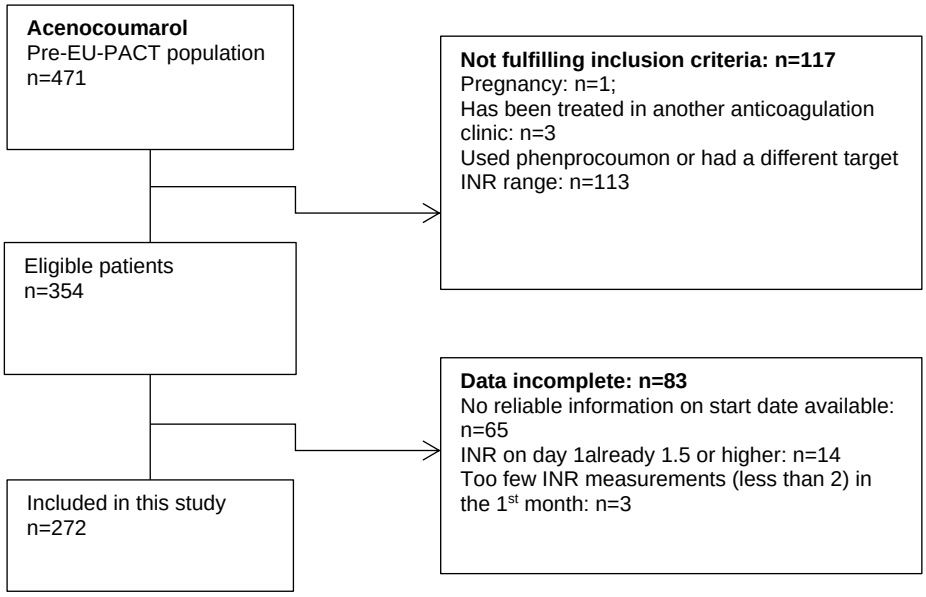
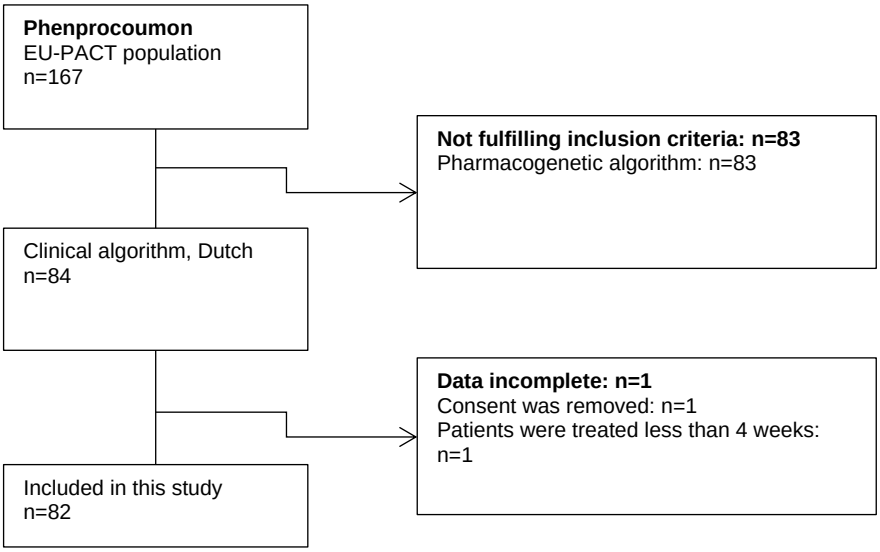


Figure S1 Patient selection process of acenocoumarol users.

A. Clinical algorithm group



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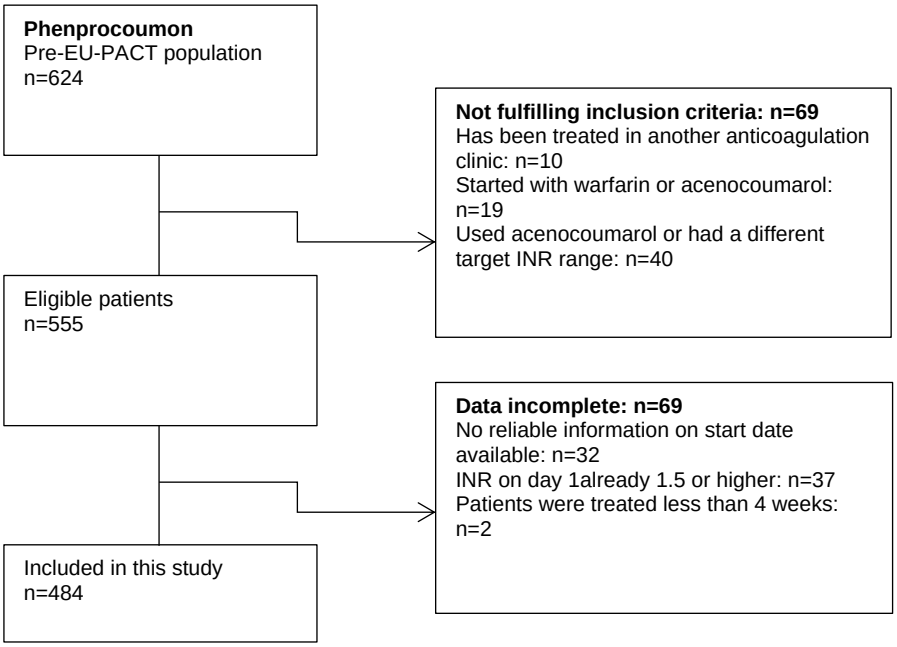

Figure S2 Patient selection process of phenprocoumon users.

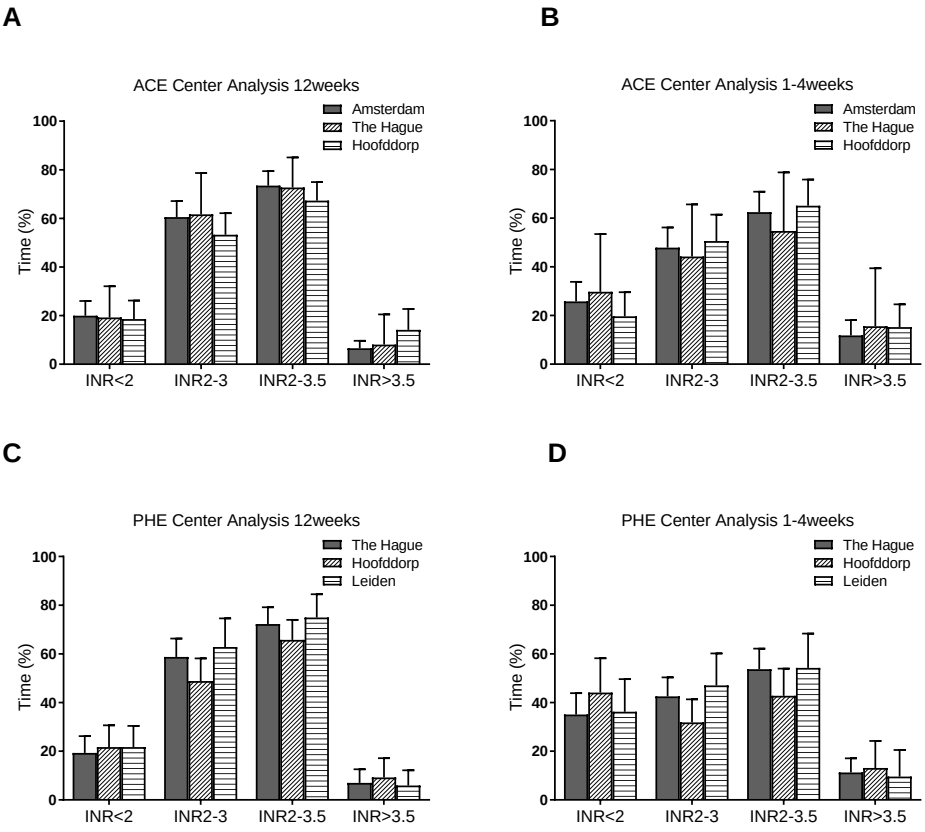

Figure S3 Center specific analysis at different centers in the clinical algorithm group. Outcomes of acenocoumarol during 12 weeks and the first 4 weeks are shown in A and B. Outcomes for phenprocoumon during 12 weeks and the first 4 weeks are shown in C and D. All the data are indicated as mean±95% confidence interval.

Table S1. Mean number of INR measurements in patients using acenocoumarol or phenprocoumon.

	Acenocoumarol			Phenprocoumon		
Number of INR measurements	**Clinical algorithm group (n=82)**	**Standard care group (n=272)**	***P*-value**	**Clinical algorithm group (n=82)**	**Standard care group (n=484)**	***P*-value**
Month1	6.2	4.6	0.00	6.4	4.3	0.00
Week1	3.1	1.2	0.00	3.1	1.4	0.00
Week2	1.3	1.3	0.38	1.3	1.1	0.00
Week3	1.0	1.1	0.17	1.1	0.9	0.00
Week4	0.0	0.0	-	0.0	0.0	-
Month2	2.5	2.7	0.07	3.0	2.7	0.00
Month3	2.2	2.1	0.44	2.4	2.0	0.00

Table S2. Mean percentage of time in the therapeutic INR range 2.0-3.0 during 12 weeks.

			TTR in range 2.0-3.0			
Analysis	**Clinical algorithm group; % mean±SD**	**n**	**Standard care group; % mean±SD**	**n**	**Unadjusted Difference (95% CI)**	**Adjusted Difference [#] (95% CI)**
Acenocoumarol						
The first week excluded						
Week 2-12	60.8±23.6	80	55.8±22.6	271	5.0 (-0.7 to 10.7)	4.8 (-1.6 to 11.1)
Week 2-4	52.1±33.6	82	43.8±32.0	272	8.3 (0.3 to 16.4) [$]	4.9 (-3.9 to 13.6)
Week 5-8	58.3±34.0	82	59.6±32.7	272	-1.3 (-9.4 to 6.9)	-0.2 (-9.2 to 8.8)
Week 9-12	68.9±32.7	80	60.6±33.8	271	8.3 (-0.1 to 16.7)	9.1 (-0.2 to 18.4)
The first week included						
Week 1-12	58.8±22.2	80	54.8±21.9	271	3.9 (-1.6 to 9.4)	3.5 (-2.6 to 9.5)
Week 1-4	48.2±27.8	82	42.1±29.3	272	6.0 (-1.1 to 13.2)	2.4 (-5.4 to 10.3)
Phenprocoumon						
The first week excluded						
Week 2-12	60.8±25.8	80	50.4±29.2	470	10.4 (4.1 to 16.7) [$]	9.1 (2.3 to 16.0) [$]
Week 2-4	48.4±33.8	82	45.0±35.2	484	3.4 (-4.9 to 11.6)	2.1 (-6.1 to 10.3)
Week 5-8	60.3±33.7	82	51.4±38.2	476	9.0 (0.8 to 17.1) [$]	8.0 (-0.8 to 16.9)
Week 9-12	69.8±35.4	80	54.1±40.0	470	15.7 (7.1 to 24.4) [$]	14.1 (4.7 to 23.6) [$]
The first week included						
Week 1-12	57.3±23.5	80	49.6±27.6	470	7.7 (1.9 to 13.5) [$]	6.5 (0.04 to 12.9) [$]
Week 1-4	41.1±25.2	82	43.7±29.4	484	-2.6 (-9.4 to 4.1)	-3.7 (-10.5 to 3.1)

CI: confidence interval; INR: International Normalized Ratio; SD: standard deviation.
[#]Adjusted for age, *CYP2C9* and *VKORC1* genotype, and (for acenocoumarol only) indications.
[$]*P* <0.05

Table S3. Sensitivity analysis for acenocoumarol of the primary outcome that is the percentage of time within the therapeutic INR range during the first 12 weeks in patients with at least 12 weeks of follow up.

	Acenocoumarol			
	Clinical algorithm group (n=68); % mean±SD	**Standard care group (n=265); % mean±SD**	**Unadjusted Difference (95% CI)**	**Adjusted Difference[#] (95% CI)**
Percentage of time within INR range 2.0-3.0				
Week 1-12	58.4±23.0	54.8±21.8	3.6 (-2.3 to 9.5)	3.4 (-3.1 to 10.0)
Percentage of time within INR range 2.0-3.5				
Week 1-12	71.4±20.2	66.9±20.1	4.5 (-0.8 to 9.9)	4.1 (-1.8 to 10.0)

CI: confidence interval; INR: International Normalized Ratio; SD: standard deviation.
[#]Adjusted for age, *CYP2C9* and *VKORC1* genotype, and indications.

Table S4. Sensitivity analysis for phenprocoumon of the primary outcome that is the percentage of time within the therapeutic INR range during the first 12 weeks in patients with at least 12 weeks of follow up.

	Phenprocoumon			
	Clinical algorithm group (n=65); % mean±SD	**Standard care group (n=468); % mean±SD**	**Unadjusted Difference (95% CI)**	**Adjusted Difference[#] (95% CI)**
Percentage of time within INR range 2.0-3.0				
Week 1-12	56.3±21.5	49.7±27.6	6.6 (-0.4 to 13.6)	5.2 (-1.8 to 12.3)
Percentage of time within INR range 2.0-3.5				
Week 1-12	70.4±19.4	68.8±23.3	1.7 (-3.5 to 6.9)	0.2 (-5.8 to 6.1)

CI: confidence interval; INR: International Normalized Ratio; SD: standard deviation.
[#]Adjusted for age, *CYP2C9* and *VKORC1* genotype

Table S5. Secondary outcomes of acenocoumarol.

Outcome	Acenocoumarol						
Time	**Clinical algorithm group; % mean±SD**	**n**	**Standard care group; % mean±SD**	**n**	**Unadjusted Difference (95% CI)**	**Adjusted Difference[#] (95% CI)**	***P*-value**
Percentage of time with INR < 2							
Week 1-12	19.5±19.5	80	22.7±19.5	271	-3.2 (-8.1 to 1.7)	-3.1 (-8.1 to 1.9)	0.23
Week 2-12	17.1±20.2	80	21.6±20.0	271	-4.4 (-9.4 to 0.6)	-4.5 (-9.8 to 0.7)	0.09
Week 2-4	17.4±30.3	82	37.1±35.6	272	-19.7 (-28.3 to -11.2) [$]	-15.8 (-24.5 to -7.2)	0.00
Week 1-4	24.6±27.3	82	38.9±33.3	272	-14.2 (-21.4 to -7.1) [$]	-10.4 (-18.2 to -2.6)	0.01
Week 5-8	21.0±30.0	82	18.0±26.7	272	3.0 (-3.8 to 9.8)	0.6 (-6.7 to 8.0)	0.87
Week 9-12	14.6±25.3	80	14.2±25.3	271	0.4 (-5.9 to 6.7)	-0.1 (-7.0 o 6.9)	0.90
Percentage of time with INR > 3							
Week 1-12	21.7±22.1	80	22.5±23.0	271	-0.7 (-6.5 to 5.0)	-0.3 (-6.2 to 5.6)	0.91
Week 2-12	22.0±22.9	80	22.6±23.7	271	-0.6 (-6.5 to 5.3)	-0.2 (-6.4 to 5.9)	0.94
Week 2-4	30.5±34.7	82	19.1±30.1	272	11.4 (3.0 to 19.8) [$]	11.0 (3.0 to 18.9)	0.01
Week 1-4	27.2±29.9	82	19.0±27.8	272	8.2 (1.2 to 15.2) [$]	8.0 (0.9 to 15.0)	0.03
Week 5-8	20.7±31.6	82	22.4±31.0	272	-1.8 (-9.5 to 6.0)	-0.5 (-8.7 to 7.8)	0.92
Week 9-12	16.5±26.2	80	25.2±32.6	271	-8.7 (-15.7 to -1.7) [$]	-9.0 (-17.6 to -0.5)	0.03
Percentage of time with INR > 3.5							
Week 1-12	8.7±14.2	80	10.5±14.9	271	-1.8 (-5.5 to 1.9)	-1.4 (-5.4 to 2.5)	0.47
Week 2-12	8.5±14.2	80	10.5±15.2	271	-1.9 (-5.7 to 1.8)	-1.7 (-5.7 to 2.4)	0.42
Week 2-4	14.1±25.4	82	9.6±20.3	272	4.5 (-1.6 to 10.6)	3.7 (-1.9 to 9.2)	0.20
Week 1-4	13.1±22.9	82	10.0±18.8	272	3.1 (-2.4 to 8.6)	2.8 (-2.3 to 7.9)	0.28
Week 5-8	7.7±19.4	82	9.9±21.0	272	-2.3 (-7.4 to 2.9)	-1.4 (-7.1 to 4.2)	0.62
Week 9-12	4.8±13.0	80	11.5±22.8	271	-6.6 (-10.7 to -2.7) [$]	-6.3 (-12.0 to -0.5)	0.03

CI: confidence interval; INR: International Normalized Ratio; SD: standard deviation.
[#]Adjusted for age, *CYP2C9* and *VKORC1* genotype, and indications.
[$]*P*<0.05

Table S6. Secondary outcomes for phenprocoumon.

Outcome	Phenprocoumon						
Time	**Clinical algorithm group; % mean±SD**	**n**	**Standard care group; % mean±SD**	**n**	**Unadjusted Difference (95% CI)**	**Adjusted Difference[#] (95% CI)**	***P*-value**
Percentage of time with INR < 2							
Week 1-12	19.3±20.0	80	13.1±17.2	470	6.2 (1.5 to 10.9) [$]	5.8 (1.7 to 10.0)	0.01
Week 2-12	14.1±20.5	80	10.9±17.8	470	3.2 (-1.7 to 8.0)	2.9 (-1.5 to 7.2)	0.19
Week 2-4	24.9±33.3	82	16.9±28.7	484	8.0 (0.3 to 15.7) [$]	7.0 (0.2 to 13.7)	0.04
Week 1-4	37.6±28.7	82	22.8±26.7	484	14.8 (8.1 to 21.6) [$]	13.8 (7.8 to 19.8)	0.00
Week 5-8	15.7±27.5	82	11.6±24.5	476	4.1 (-1.75 to 9.9)	3.7 (-2.2 to 9.6)	0.22
Week 9-12	5.2±16.7	80	5.8±17.4	470	-0.7 (-4.8 to 3.4)	-0.5 (-4.7 to 3.7)	0.83
Percentage of time with INR > 3							
Week 1-12	23.4±25.7	80	37.3±30.9	470	-13.9 (-20.2 to -7.5) [$]	-12.3 (-19.4 to -5.2)	0.00
Week 2-12	25.1±27.9	80	38.7±32.6	470	-13.6 (-20.5 to -6.8) [$]	-12.0 (-19.5 to -4.5)	0.00
Week 2-4	26.7±34.8	82	38.1±39.3	484	-11.4 (-19.8 to -3.0) [$]	-9.0 (-17.7 to -0.4)	0.04
Week 1-4	21.3±27.1	82	33.5±34.0	484	-12.2 (-18.9 to -5.5) [$]	-10.1 (-17.3 to -2.9)	0.01
Week 5-8	24.0±32.8	82	37.1±40.4	476	-13.1 (-21.1 to -5.0) [$]	-11.7 (-21.0 to -2.5)	0.01
Week 9-12	25.0±34.3	80	40.1±41.3	469	-15.8 (-23.5 to -6.6) [$]	-13.7 (-23.3 to -4.1)	0.01
Percentage of time with INR > 3.5							
Week 1-12	9.4±14.5	80	18.3±22.3	470	-8.9 (-12.7 to -5.1) [$]	-7.4 (-12.3 to -2.4)	0.00
Week 2-12	10.0±15.6	80	18.9±23.5	470	-8.9 (-13.0 to -4.9) [$]	-7.4 (-12.6 to -2.1)	0.01
Week 2-4	14.2±27.4	82	21.8±32.2	484	-7.6 (-14.2 to -1.0) [$]	-5.5 (-12.5 to 1.5)	0.13
Week 1-4	11.3±21.1	82	19.0±27.4	484	-7.7 (-13.0 to -2.5) [$]	-5.9 (-11.7 to 0.0)	0.05
Week 5-8	9.3±18.8	82	18.8±31.1	476	-9.5 (-14.5 to -4.5) [$]	-8.0 (-14.9 to -1.2)	0.02
Week 9-12	7.3±16.1	80	16.4±28.7	470	-9.1 (-13.5 to -4.7) [$]	-7.9 (-14.4 to -1.4)	0.02

CI: confidence interval; INR: International Normalized Ratio; SD: standard deviation.
[#]Adjusted for age, *CYP2C9* and *VKORC1* genotype.
[$]$P<0.05$

CHAPTER 3

Age-stratified outcome of genotype-guided dosing algorithm for acenocoumarol and phenprocoumon

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Abstract

Background: Age seemed to affect the interaction between coumarins and genotype in the acenocoumarol and phenprocoumon arm of the European pharmacogenetics of anticoagulant therapy (EU-PACT) trial.

Objectives: To investigate the effect of genotype-guided dosing stratified by age and the potential factors causing a difference.

Patients/Methods: Data from the acenocoumarol/phenprocoumon arm of the EU-PACT trial was used. The percentage of time below the therapeutic range, time above the therapeutic range, and time in the therapeutic range (TTR) during the initial 12 weeks of therapy were compared between the genotype-guided group and the control group among younger (<75 years) and older (≥75 years) patients by the use of independent *t*-tests and adjust for sex, height, weight and co-medications by the use of linear regression.

Results: Among younger phenprocoumon users, TTR during the first 12 weeks in the genotype-guided group (*n*=55) was 9.5 % (95% confidence interval (CI): 1.3 to 17.8) higher than the control group (*n*=63) with a remarkable lower percentage of time above this range (difference: -9.6%, 95%CI: -19.0 to -0.2) and similar time below this range. Older patients dosed by the genotype-guided algorithm (*n*=24) spend more time above the range (difference: 27.5%, 95%CI: 12.9 to 42.0). For acenocoumarol users, there were no significant differences between the genotype-guided and control groups for most outcomes, except for a lower percentage of time below the range among older patients.

Conclusions: The genotype-guided algorithm for phenprocoumon in the EU-PACT trial benefitted younger patients more, but for older patients the algorithm needs to be revised and tested in further research.

Introduction

Aging is one of the common causes of interindividual variation in the stable dose of coumarin derivatives [1, 2]. With increasing age, the pharmacokinetics and pharmacodynamics of coumarins change [3]. This results in the fact that elderly patients, on average, require a lower dose than younger patients to maintain the same anticoagulation effect [4]. In addition, elderly patients are more likely to have comorbidities, and they therefore receive a higher number of co-medications [5]. Both comorbidities [6] and co-medications [7] can influence the anticoagulation effect of coumarins, owing to the drug-disease interactions or drug-drug interactions. Furthermore, elderly patients usually have a high risk of bleeding even without taking coumarins [8]. Therefore, it's important to take into account the patient's age when assessing the effect of coumarin therapy.

Previously, three dosing algorithms to optimize coumarin dosing including genetic and clinical factors were investigated in the European Pharmacogenetics of Anticoagulant Therapy (EU-PACT) trial and the Clarification of Optimal Anticoagulation through Genetics (COAG) trials [9-11]. Although these dosing algorithms include age as a parameter, none of them stratified patients by age in the primary outcomes report. The mean age in the EU-PACT trial was ~ 68 years both in the acenocoumarol/phenprocoumon arm [9] and the warfarin arm [10]. In a reply to a commentary, it was shown that, among patients aged < 75 years, the group that used the genotype-guided algorithm obtained a higher percentage of time in the therapeutic INR range compared with the patients in the group dosed according to the non-genotype guided algorithm during the 12 weeks after the initiation of therapy. In contrast, patients who were aged 75 years or older did not spend more time in range in the genotyped arm [12]. Therefore, age seemed to affect the interaction between coumarins and genotype.

After this intriguing finding, we wanted to present here the further analyses of the acenocoumarol/phenprocoumon arm of the EU-PACT trial to assess the effect of genotype-guided dosing stratified by age. We also assessed the influence of potential factors such as comorbidities and concurrent drug use that may cause the differences in different age categories.

Methods

Patient selection and study design

Data from patients with at least 10 weeks of follow up in the acenocoumarol/phenprocoumon arm of the EU-PACT trial [9] were used for the present study. In brief, the EU-PACT trial was a single-blind, randomized trial comparing a genotype-guided dosing algorithm [13] that included clinical variables and genotyping for *CYP2C9* and *VKORC1* with a dosing algorithm that included only clinical variables, for the initiation of acenocoumarol or phenprocoumon treatment in patients with atrial fibrillation or venous thromboembolism [9]. Details of the study design, outcome definitions, patients and data collection, and main results of this trial are described elsewhere [9, 13, 14].

Outcome measures

The primary outcome in the present study was the percentage of time in the therapeutic INR range 2.0-3.0 (TTR) during the first 12 weeks of acenocoumarol or phenprocoumon treatment in different age groups. Rosendaal's method was used to calculate the TTR [15]. Other outcomes that we assessed were the percentage of time above and below the INR range, and the maintenance dose per day in the first stable period after initiation of anticoagulation therapy as defined in the EU-PACT trial [9].

Definition of patients group

To determine the impact of age on the primary outcome of genotype-guided dosing, the interaction between age and treatment was examined beforehand (supplement Figure S1 and Figure S2). There was a trend towards an age interaction for phenprocoumon. Patients were then categorized into two age groups: younger (<75 years) and older ((≥75 years). In each age group, the outcomes were compared between the genotype-guided group and the control group.

We also determined the outcome in three genotype strata (no variant, onw variant in either *CYP2C9* or *VKORC1* genes, and more than one variant). To evaluate the impact of the first maintenance dose, the differences of the maintenance dose

calculated with the genotype-guided algorithm and the clinical algorithm were compared.

Potential confounding factors

The baseline patient characteristics sex, height, weight, CYP2C9 genotype, VKORC1 genotype, comorbidity and concomitant medication were compared between younger and older patients. The comorbidities that we tested were hypertension, heart failure, myocardial infarction, hyperlipidemia, and diabetes mellitus, which were most common and may have an impact on the anticoagulation effect [7, 16]. The suspected concomitant drugs were defined as coumarin potentiating drugs, including statins, proton-pump inhibitors, antidepressants [17-20], antibiotics [21] non-steroidal anti-inflammatory drugs, lactulose [22], aspirin, and enzyme inducers which can reduce the effect of anticoagulation. The detailed information of the concomitant drugs used is shown in the Table S1 of supplement.

Statistical Analysis

Only patients with at least 10 weeks of follow-up were included in the analyses; however, per-protocol analyses were also performed. The independent *t*-test and Pearson's chi square test were used to compare the baseline characteristics. The primary and secondary outcomes were compared by calculating mean differences with 95% confidence intervals (95% CI), using independent-samples *t*-tests and adjusted in a linear regression model for height, weight, sex, and the concomitant used drugs (only enzyme inhibitors or inducers). The interaction of age and treatment was assessed by using ANCOVA. Genotype proportions were tested for deviations from Hardy-Weinberg equilibrium with a chi-square test. For all calculations a *P*-value of < 0.05 was considered statistically significant. All the analyses were performed with IBM SPSS STATISTICS version 20.0 (IBM Corp., Armonk, NY, USA).

Results

Patients

The present study included a total of 484 patients (325 patients treated with acenocoumarol and 159 patients treated with phenprocoumon) from the

acenocoumarol/phenprocoumon arm of the EU-PACT trail [9] which excluded 64 patients who did not have at least 10 weeks of treatment. Of these, 160 acenocoumarol users were assigned to the genotype-guided group and 165 to the control group; 79 phenprocoumon users were included in the genotype-guided group and 80 in the control group. An additional number of patients were excluded for the per-protocol analysis for the reasons outlined in the supplement Table S2. Of these, 111 acenocoumarol users were assigned to the genotype-guided group and 126 to the control group; 49 phenprocoumon users were included in the genotype-guided group and 58 in the control group.

Most of the baseline characteristics of patients were similar between the genotype-guided group and the control group in both age groups and for both acenocoumarol users and phenprocoumon users (Table 1). Only among younger phenprocoumon treated patients, a statistically significant difference was shown for weight, which was 92kg in the genotype-guided group and 85kg in the control group. The characteristics of acenocoumarol treated patients stratified by country of residence (the Netherlands and Greece) was shown in supplement Table S3 and it was similar between the genotype-guided group and the control group in both age groups.

Comorbidities and concomitant medication

There were no statistically significant differences for common comorbidities between the genotype-guided group and the control group per age group in phenprocoumon treated patients (Table 2).

The concomitant medication suspected to interact with acenocoumarol or phenprocoumon was summarized in Table 3. No statistically significant difference was shown in the distribution of concomitant drug use between the genotype-guided group and the control group in young or old age categories of acenocoumarol and phenprocoumon. During the initial therapy of phenprocoumon, for the younger age group, 33 patients (60.0%) in the genotype-guided group and 31 patients (49.2%) in the control group were taking at least one potentiating drug during the anticoagulant treatment, and for older age, it was 10 patients (41.7%) and 10 patients (58.8%), respectively in the two groups. Among younger patients treated with acenocoumarol, there were 70 patients (61.9%) concurrently using

potentiating drugs in the genotype-guided group compared with 64 (62.1%) in the control group. Only one patient used enzyme inducers, which might decrease the INR during the therapy of acenocoumarol.

We also compared the comorbidities and concomitant drug use between the young and older patients (shown in the supplement Table S4 and Table S5). As to phenprocoumon users, no statistically significant difference was shown. For acenocoumarol users, the is no statistically significant difference for most of the comorbidities except hypertension and heart failure with which the elderly group concurrent more compared with younger group. The concomitant use of potentiating drugs enzyme inhibitors or aspirin with acenocoumarol in elderly patients was also more than that in younger.

TTR during the initial 12 weeks

In all phenprocoumon treated patients, the difference in TTR between the genotype-guided and the control group was 2.5% [9]. However, the effect of genotype-guided dosing for patients under 75 years and for patients 75 years or older was different as reported in Table 4. Among patients younger than 75 years TTR during the first 12 weeks was 64.1% in the genotype-guided group, and 55.7% in the control group with an adjusted difference of 9.5 % (95% CI: 1.3 to 17.8). Younger patients treated with phenprocoumon also spent 9.6% less time (17.6% vs. 27.1%, 95% CI: -19.0 to -0.2) with INR above 3. There was no difference in the percentage of time with INR below 2. In contrast, among patients 75 years or older, genotype-guided dosing resulted in a lower TTR (the adjusted difference was -17.9%, 95%CI -31.8 to -3.9) and a high percentage of time above 3 (adjusted difference: 27.5%, 95%CI: 12.9 to 42.0) compared with the control group. The older patients with genotype-guided dosing also spent 9.7% less time with INR below 2 than that in the control group however, this was not statistically significant. A per-protocol analysis yielded similar results (shown in the supplement Table S6) though the difference of TTR between the genotype-guided group and the control was not statistically significant.

Table 1. Characteristics of patients with dosing by the genotype-guided algorithm and the control stratified by age.

Characteristics	Acenocoumarol						Phenprocoumon					
	<75 years			≥75 years			<75 years			≥75 years		
	Genotype-guided group	Control	*P*-value	Genotype-guided group	Control	*P*-value	Genotype-guided group	Control	*P*-value	Genotype-guided group	Control	*P*-value
Patient number	113	103	-	47	62	-	55	63	-	24	17	-
Age, mean±SD	62±12	62±10	1	81±4	80±4	0.42	62±12	63±9	0.76	79±3	81±4	0.24
Male sex, n (%)	82 (73)	62 (60)	0.05	23 (49)	32 (52)	0.78	36 (66)	39 (62)	0.69	13 (54)	11 (52)	0.23
Height (cm), mean±SD	175±10	174±10	0.85	166±10	165±10	0.51	175±9	176±9.5	0.81	171±9	165±9	0.06
Weight (kg), mean±SD	86±16	86±20	0.89	80±11	76±14	0.12	92±17	85±15	0.03	76±13	73±15	0.52
Race (white), n (%)	108 (95.6)	103 (100)		47 (100)	62 (100)		51 (92.7)	61 (96.8)		24 (100)	16 (94.1)	
CYP2C9 genotype, n (%)			0.62		1 missing	0.28		1 missing	0.86			0.50
*1/*1	68 (60.2)	55 (53.4)		26 (55.3)	38 (61.3)		37 (67.3)	41 (65.1)		15 (62.5)	14 (82.4)	
*1/*2	20 (17.7)	21 (20.4)		14 (29.8)	9 (14.8)		9 (16.4)	12 (6)		4 (16.7)	2 (11.8)	
*1/*3	19 (16.8)	17 (16.5)		6 (12.8)	11 (17.7)		7 (12.)	6 (9.5)		4 (16.7)	1 (5.9)	
*2/*2	3 (2.7)	7 (6.8)		0	2 (3.2)		2 (3.6)	2 (3.2)		0	0	
*2/*3	3 (2.7)	3 (2.9)		1 (2.1)	1 (1.6)		0	1 (1.6)		1 (4.2)	0	
*3/*3	0 (0)	0		0	0		0	0 (0)		0	0	
HWE for CYP2C9 genotype, *P*-value	0.52	0.09		0.54	0.38			0.52		0.54	0.38	
VKORC1 genotype, n (%)			0.58		1 missing	0.74		1 missing	0.54			0.46
GG	45 (39.8)	34 (33.0)		18 (38.3)	19 (30.6)		16 (29.1)	24 (38.1)		7 (29.2)	8 (47.1)	
GA	45 (39.8)	45 (43.7)		22 (46.8)	32 (51.6)		25 (45.5)	25 (39.7)		14 (58.3)	8 (47.1)	
AA	23 (20.4)	24 (23.3)		7 (14.9)	10 (16.1)		14 (25.5)	13 (20.6)		3 (12.5)	1 (5.9)	
HWE for VKORC1 genotype, P-value	0.07	0.23		0.95	0.57		0.07	0.23		0.95	0.57	
Atrial fibrillation, n (%)	88 (78)	82 (80)	0.76	44 (93.6)	58 (94)	0.99	45(82)	50 (79)	0.74	21(88)	16 (94.1)	0.48

HWE, Hardy–Weinberg equilibrium; SD, standard deviation.

Table 2. Common comorbidities of patients with dosing by the genotype-guided algorithm and the control group stratified by age.

	Acenocoumarol						Phenprocoumon					
	<75 years			≥75 years			<75 years			≥75 years		
	Genotype-guided group	Control group	*P*-value	Genotype-guided group	Control group	*P*-value	Genotype-guided group	Control group	*P*-value	Genotype-guided group	Control	*P*-value
Patient number	113	103		47	62		55	63		24	17	
Common comorbidities												
Hypertension, n (%)	55 (48.7)	53 (51.5)	0.68	36 (76.6)	48 (77.4)	0.92	24 (43.6)	29 (46)	0.79	13 (54.2)	12 (70.6)	0.29
Heart Failure, n (%)	14 (12.4)	9 (8.7)	0.39	9 (19.1)	18 (29.0)	0.24	2 (3.6)	0	0.13	0	0	-
Myocardial Infarction, n (%)	6 (5.3)	4 (3.9)	0.62	3 6.4)	4 (6.5)	0.99	4 (7.3)	1 (1.6)	0.13	0	1 (5.9)	0.23
Hyperlipidemia, n (%)	36 (31.9)	25 (24.3)	0.22	11 (23.4)	17 (27.4)	0.64	7 (12.7)	15(23.8)	0.12	6 (25.0)	4 (23.5)	0.91
Diabetes, n (%)	21 (18.6)	19 (18.4)	0.98	16 (34.0)	12 (19.4)	0.08	4 (7.3)	4 (6.3)	0.84	1 (4.2)	1 (5.9)	0.80

Table 3. Concomitant drug use stratified by age and coumarins for genotype-guided group and the control group separately.

	Acenocoumarol						Phenprocoumon					
	<75 years			≥75 years			<75 years			≥75 years		
	Genotype-guided group	Control group	*P*-value	Genotype-guided group	Control group	*P*-value	Genotype-guided group	Control group	*P*-value	Genotype-guided group	Control group	*P*-value
Patient number	113	103		47	62		55	63		24	17	
Comedication, n (%)	113 (100)	99 (96.1)		46 (97.9)	61 (98.4)		55 (100)	61 (96.8)		23 (95.8)	16 (94.1)	
Potentiating drugs, n (%)	70 (61.9)	64 (62.1)	0.91	38 (80.9)	47 (75.8)	0.53	33 (60.0)	31 (49.2)	0.24	10 (41.7)	10 (58.8)	0.28
Enzyme Inibitors, n (%)	50 (44.2)	43 (41.7)	0.71	28 (59.6)	35 (56.5)	0.92	27 (49.1)	22 (34.9)	0.12	9 (37.5)	8 (47.1)	0.54
Amiodarone, n (%)	7 (6.2)	11 (10.7)	0.23	8 (17.0)	7 (11.3)	0.39	0	0	-	0	0	-
PPIs, n (%)	18 (16.0)	11 (10.7)	0.26	10 (21.3)	14 (22.6)	0.87	16 (29.1)	12 (19.0)	0.20	5 (20.8)	5 (29.4)	0.53
Statins, n (%)	34 (30.1)	29 (28.2)	0.76	13 (27.7)	16 (25.8)	0.83	10 (18.2)	14 (22.2)	0.59	5 (20.8)	4 (23.5)	0.84
Antidepressants, n (%)	4 (3.5)	2 (1.9)	0.48	0	5 (8.1)	-	5 (9.1)	2 (3.2)	0.18	0	0	-
Antibacterial drugs, n (%)	11 (9.7)	8 (7.8)	0.61	5 (10.6)	10 (16.1)	0.41	6 (10.9)	6 (9.5)	0.80	1 (4.2)	0	-
Other NSAIDs, n (%)	9 (8.0)	5 (4.8)	0.35	1 (2.1)	2 (3.2)	-	2 (3.6)	3 (4.8)	0.76	0	0	-
Lactulose, n (%)	2 (1.8)	0	-	1 (2.1)	1 (1.6)	-	0	0	-	0	0	-
Aspirin, n (%)	19 (16.8)	19 (18.4)	0.75	10 (21.3)	21 (33.9)	0.15	1 (1.8)	2 (3.2)	0.64	0	2 (11.8)	-
Digoxin, n (%)	12 (10.6)	11 (10.7)	0.34	8 (17.0)	9 (14.5)	0.72	6 (10.9)	4 (6.3)	0.38	0	3 (17.6)	-
Enzyme Inducers, n (%)	0	1	-	1	0	-	0	0	-	0	0	-

NSAIDs: non-steroidal anti-inflammatory drugs; PPI: proton-pump inhibitor.

Table 4. Percentage of time in, below and above the therapeutic INR range during 12 weeks after the initiation of treatment after stratification by age.

	<75 years					≥75 years				
	Genotype-guided group; % mean±SD	Control group; % mean±SD	Difference (95%CI)	Adjusted Difference (95%CI)	*p*-value *	Genotype-guided group % mean±SD	Control group % mean±SD	Difference (95%CI)	Adjusted Difference (95%CI)	*p*-value
Phenprocoumon										
Number	55	63				24	17			
INR 2-3 (%)	64.1±19.8	55.7±24.0	8.4 (0.3 to 16.5)	9.5 (1.3 to 17.8)	0.02	50.9±21.5	63.3±21.2	-12.4 (-26.1 to 1.3)	-17.9 (-31.8 to -3.9)	0.01
INR <2 (%)	18.3±16.0	17.2±19.3	1.1 (-5.4 to 7.6)	0.1 (-6.6 to 6.8)	0.98	17.0±18.3	26.9±20.9	-9.6 (-22.3 to 2.6)	-9.7 (-22.5 to 3.3)	0.14
INR>3 (%)	17.6±2.19	27.1±26.5	-9.5 (-18.3 to -0.6)	-9.6 (-19.0 to -0.2)	0.05	32.1±27.2	9.9±16.5	22.3 (8.4 to 36.1)	27.5 (12.9 to 42.0)	<0.01
Acenocoumarol										
Number	113	103				47	62			
INR 2-3 (%)	64.5±23.7	61.3±24.4	3.2 (-3.2 to 9.7)	3.6 (-2.9 to 10.1)	0.28	57.0±25.4	61.7±21.2	-4.7 (-13.6 to 4.2)	-4.1 (-13.2 to 5.0)	0.37
INR <2 (%)	21.0±21.8	19.9±20.3	1.1 (-4.5 to 6.8)	0.1 (-5.6 to 5.6)	0.98	32.1±27.0	22.4±19.7	9.7 (0.5 to 19.0)	9.8 (0.9 to 18.7)	0.03
INR>3 (%)	14.5±18.6	18.8±21.5	-4.3 (-9.8 to 1.1)	-3.7 (-9.2 to 1.9)	0.19	10.9±15.5	15.9±16.9	-5.1 (-11.3 to 1.2)	-5.7 (-11.9 to 0.4)	0.17
Greece										
Number	48	42				32	43			
INR 2-3 (%)	66.1±27.0	65.3±26.6	0.8 (-10.5 to 12.1)	1.5 (-9.8 to 12.9)	0.79	57.3±25.7	63.0±20.8	-5.6 (-16.4 to 5.1)	-3.8 (-15.2 to 7.6)	0.51
INR <2 (%)	25.6±25.0	20.6±21.5	5.0 (-4.8 to 14.9)	4.4 (-5.5 to 14.3)	0.38	36.4±27.2	23.3±19.5	13.1 (2.4 to 23.9)	11.5 (0.1 to 2.8)	0.05
INR>3 (%)	8.3±13.9	14.1±18.3	-5.8 (-12.7 to 1.1)	-5.9 (-12.9 to 1.0)	0.09	6.3±9.5	13.8±15.6	-7.5 (-13.7 to -1.7)	-7.7 (-14.2 to -1.3)	0.02
Netherlands										
Number	65	61				15	19			
INR 2-3 (%)	63.3±21.0	58.5±22.5	4.8 (-2.9 to 12.5)	5.5 (-2.3 to 13.2)	0.17	56.4±25.7	58.9±22.4	-2.5 (-19.3 to 14.3)	-4.5 (-23.9 to 14.8)	0.63
INR <2 (%)	17.6±18.5	19.4±19.6	-1.8 (-8.5 to 4.9)	-2.5 (-9.3 to 4.4)	0.47	23.0±25.0	20.4±20.5	2.6 (-13.3 to 18.5)	5.4 (-11.6 to 22.3)	0.53
INR>3 (%)	19.0±20.9	22.0±23.1	-3.0(-10.8 to 4.7)	-3.0 (-11.0 to 5.0)	0.46	20.6±20.9	20.8±19.0	-0.1 (-14.1 to 13.9)	-0.8 (-15.3 to 13.7)	0.91

CI: confidence interval; INR: International Normalized Ratio; SD: standard deviation.

**P*-value for the difference adjusted for height, weight, sex, enzyme inhibitors, and enzyme inducers.

Table 5. Effect of genetic variants on anticoagulation control in genotyped and control patients among younger and elderly phenprocoumon users.

	<75 years						≥75 years					
	Genotype-guided group		Control group		Percentage difference (95%CI)	*P*-value	Genotype-guided group		Control group		Percentage difference (95%CI)	*P*-value
	%, mean ± SD	n	%, mean ± SD	n			%, mean ± SD	n	%, mean ± SD	n		
Percentage of time with time in therapeutic INR range												
No variation	63.3±19.2	12	53.9±24.5	18	9.4 (-7.8 to 26.6)	0.27	58.1±9.1	5	56.0±28.5	5	2.0 (-32.8 to 36.9)	0.89
One variant	62.4±19.4	21	63.0±20.2	18	-0.6 (-13.5 to 12.3)	0.93	56.7±24.6	12	67.2±18.5	11	-10.5 (-29.5 to 8.5)	0.27
Two or more variants	66.1±21.2	22	52.1±25.9	27	14.0 (0.2 to 27.8)	0.05	35.6±14.8	7	55.6	1		-
Percentage of time with INR<2.0												
No variation	22.4±18.2	12	30.0±22.8	18	-7.5 (-23.6 to 8.6)	0.42	15.6±17.2	5	30.8±24.8	5	-15.1 (-46.2 to 15.9)	0.29
One variant	18.7±17.9	21	18.3±16.3	18	0.4 (-10.8 to 11.6)	0.94	16.4±20.9	12	27.3±19.8	11	-10.8 (-28.5 to 6.9)	0.42
Two or more variants	15.6±12.6	22	8.0 ±13.3	27	7.7 (0.2 to 15.2)	0.05	19.0±16.8	7	3.5	1		-
Percentage of time with INR>3.0												
No variation	14.2±19.5	12	16.1±21.6	18	-1.9 (-17.8 to 14.0)	0.81	26.3±19.3	5	13.2±21.4	5	13.1 (-16.7 to 42.9)	0.34
One variant	18.9±23.7	21	18.8±21.8	18	0.2 (-14.7 to 15.0)	0.98	26.8±30.7	12	5.5±11.3	11	21.3 (0.9 to 41.7)	0.04
Two or more variants	18.3±22.2	22	40.0±27.6	27	-21.7 (-36.0 to -7.4)	0.00	45.3±23.7	7	40.8	1		-

CI: confidence interval; INR: International Normalized Ratio; SD: standard deviation.

For acenocoumarol, among younger patients, the genotype-guided group got a TTR of 64.5%, which was a little more time (adjusted difference: 3.6%, 95% CI: -2.9 to 10.1) than that in the control group (61.4%) while among older patients, an opposite

result (adjusted difference: -4.0%, 95% CI: -13.2 to 5.0) was shown in Table 4. However, none of these differences were statistically significant. The older patients in the genotype-guided algorithm group had a higher percentage of time with INR below 2 compared with the control group (adjusted difference: 9.9%, 95%CI: 0.9 to 18.7). There was no statistically significant difference in the percentage of time below 2 among younger patients treated with acenocoumarol. As to the percentage of time above 3, the genotype-guided group and the control group did not differ significantly both for the younger and the older patients. A per-protocol analysis shows similar results (see in the supplement Table S6).

Effect of algorithms stratified by genotype variants

As shown in Table 5, among younger patients treated with phenprocoumon, the effect of the genotype-guided dosing compared with the control group was the most remarkable (14.0% difference in TTR, P=0.04) if there were two or more variants in *CPY2C9* or *VKORC1*. There were no statistically significant differences between the two groups in patients with only 1 variant in either *CPY2C9* or *VKORC1*. Patients without variation in both *CPY2C9* and *VKORC1*, genotype-guided dosing achieved 9.4% improvement in TTR compared with the control group, however the difference was not statistically significant. Older patients without variation in *CPY2C9* and *VKORC1* dosed by genotype-guided algorithm achieved a similar TTR as the patients dosed by the clinical algorithm. However, the percentage of time above the range in the genotype-guided algorithm group was higher than in the clinical algorithm group. None of the differences were statistically significant. For the older patients with only 1 variant in either *CYP2C9* or *VKORC1*, the genotype-guide dosing led to a lower TTR and less time below an INR of 2, while 21.3% spend more time above the therapeutic range (*P*=0.04).

For acenocoumarol, both in younger and older patients without variation or with one variant of either *CPY2C9* or *VKORC1*, genotype-guided dosing led to a higher TTR;

however, the difference was not statistically significant. In contrast, among patients with two or more variants in either *CPY2C9* or *VKORC*1, the genotype-guided dosing resulted in a higher TTR in younger patients but a lower TTR in older patients, also without a statistically significant difference. These data are shown in Table S7 in the Supplement.

The initial predicted maintenance dose stratified by genotype variants was shown in Table 6. The dose calculated according to the genotype-guided algorithm was compared with the dose that was calculated according to the clinical algorithm. Generally, with the use of genotype-guided algorithm, both younger and older patients would be prescribed a higher dose if they had no variants, a similar dose if they had one variant allele, and a lower dose if they had more than one variant alleles, as compared with the dose calculated according to the clinical algorithm either for phenprocoumon users or for acenocoumarol users.

Discussion

The present study shows that there is an interaction between age and genotype-guided dosing for phenprocoumon during the initial period of use. An age cut-off point of 75 years was chosen to stratify patients in a younger and older age group. For younger patients, genotype-guided dosing increased the TTR by 9.3% and reduced the time above the therapeutic range by 9.5%. However, for patients who were aged ≥75 years, genotype-guided dosing did not show improvement as compared with patients that were treated according to a clinical algorithm (including the same factors as the genetic algorithm except for the genetic variants). For acenocoumarol users, the point estimates of the effect were in the same direction. However, there were no statistically significant differences between the age groups.

Table 6. The mean difference between the calculated dose for phenprocoumon users using the genotype guided algorithm and using the clinical algorithm*.

	Genotype-guided group					Control group				
Stratified by genotype	**n**	**Dose calculated with the genotype-guided algorithm, mean ± SD**	**Dose calculated with the clinical algorithm, mean ± SD**	**Difference**	***P*-value**	**n**	**Dose calculated with the clinical algorithm, mean ± SD**	**Dose calculated with the genotype-guided algorithm, mean ± SD**	**Difference**	***P*-value**
Age <75 years										
Pooled	55	2.2±0.6	2.3±0.3	-0.20	0.05	62	2.2±0.3	2.2±0.6	0.0	0.79
No variation	12	3.0±0.3	2.4±0.4	0.60	0.00	18	2.2±0.2	2.9±0.3	-0.7	0.00
One variant	21	2.2±0.2	2.2±0.2	0.00	0.40	18	2.2±0.3	2.3±0.2	-0.1	0.11
Two or more variants	22	1.6±0.4	2.3±0.3	-0.70	0.00	26	2.2±0.4	1.6±0.4	0.6	0.00
Age ≥75 years										
Pooled	24	1.8±0.5	1.8±0.2	0.00	0.96	17	1.7±0.2	1.9±0.4	-0.2	0.10
No variation	5	2.4±0.2	1.8±0.2	0.60	0.00	5	1.7±0.3	2.2±0.3	-0.6	0.00
One variant	12	1.9±0.3	1.9±0.3	0.00	0.65	11	1.8±0.2	1.8±0.2	0	0.35
Two or more variants	7	1.3±0.4	1.7±0.1	-0.40	0.02	0	-	-	-	-

SD: standard deviation.

Previously, the EU-PACT-trial [9] reported that genotype-guided dosing of acenocoumarol or phenprocoumon did not statistically significantly improve the TTR during the 12 weeks after the initiation of therapy. However, this outcome was the mean value based on the subjects of all ages. When patients were stratified by age groups, in the younger age group genotype-guided dosing result in a higher TTR (difference: 5.1%, *P*=0.05) than dosing according to the clinical algorithm [12]. However, that was a combined result for acenocoumarol and phenprocoumon. In the present study, by stratifying the patients by age, we provided evidence that among patients aged <75 years, genotype-guided dosing for phenprocoumon could lead to a statistically significant improvement of in the TTR. However, patients aged ≥75 years did not benefit from the current pharmacogenetic algorithm for phenprocoumon [23]. The pharmacogenetic algorithm may be considered to perform worse than the clinical algorithm in this age group, because the percentage of time spend above the target range was higher than in the clinical algorithm group,

which may cause harm to these patients. However, we should also consider the limitation of the dosing algorithm. Age might not be correctly captured. Previously, it has been shown that the clinical algorithm for phenprocoumon has a tendency to result in under-dosing relative to the genotype-guided algorithm [13]. Our present study also shows that in patients without variant alleles of *CYP2C9* and *VKORC1*, the dose predicted by the clinical algorithm would be significantly lower than that predicted by the genotype-guided algorithm in both the younger and the older group. Elderly patients in the ≥75 years group are likely to require an age-related lower dose of anticoagulant, so the lower percentage of time above the TTR in the control group of phenprocoumon users might be partly explained by the lower dose predictions of the clinical algorithm than of the genotype-guided algorithm. Therefore, the increased time above the TTR might not represent an interaction with genotype but an insufficient age-related dose correction in the genotype-guided algorithm.

Our data was a randomized controlled trial, so the baseline characteristics were similar between the genotype algorithm guided and the clinical algorithm guided groups. After stratification by age, most of the baseline characteristics of the trial population were still balanced between the genotype-guided group and the control group except the mean weight among younger phenprocoumon treated patients. However, it's unlikely that the younger patients dosed by the genotype-guided algorithm got a higher TTR was because of their higher mean weight.

We tested whether there was a difference in existing comorbidities that might influence the dose response of coumarins [7]. However, they were equally distributed between the genotype-guided group and the control group. Furthermore, our outcomes were adjusted for the co-medications, however, it did not differ between age groups, therefore, also could not explain our findings.

One suggested explanation for our findings is the different physical conditions and drug metabolism between young and old populations [3]. Although coumarins dosages were inversely related to age [1, 2], the decline rate of dose requirement was not necessarily similar between the young and old groups. For instance, among younger patients, the dose requirement decreased strongly with age increase, whereas, among elderly patients, the decrease in dose requirement with

age was less pronounced [1]. Previously, another study reported a pharmacogenetic-based dosing algorithm that failed to identify older patients who needed a lower daily dose (with two variants of *VKORC1*) of warfarin [23]. In the present study, we compared the first prescribed maintenance dose and the TTR in three genetically defined strata. The outcome in patients aged <75 years was in accordance with the predicted first prescribed dose in patients without, with one and with two or more *CYP2C9* and *VKORC1* variants. However, the dose response was not necessarily as expected among patients aged ≥75 years. For instance, dose predicted by the genotype-guided algorithm and the clinical dosing algorithm were similar for the older patients with only one variant of either *CYP2C9* or *VKORC1*, however, patients in the genotype-guided group had a higher percentage of time above the TTR. Without stratification by age, the genotype-guided algorithm was not able to accurately predict dosage for either younger or older patients.

Another possible limitation of the dosing algorithm could be we only included *CYP2C9* and *VKORC1* genotypes that are common in Caucasian populations. Besides the genetic variants and clinical factors used in the present algorithm, there might be some undetected variants that accounted for the differences in effect of pharmacogenetic dosing in older patients. For instance, if patients have variants other than *CYP2C9*2* and *CYP2C9*3* that can reduce enzyme activity, they will be misclassified as *CYP2C9 *1/*1* genotype. For those patients the dose predicted by the algorithm will be inaccurate and higher than the actual required dose. This could partly explain why, in the present study, the dosages in the patients with no variants are not accurately predicted by the genotype-guided algorithm. However, it is important to note that these variants are only expected in a small percentage of the patients. It would be important to consider the inclusion of rare genetic variants as well as making a better age adjustment for older patients when applying the dosing algorithm.

In the present study, unlike patients treated with phenprocoumon, the TTRs of the patients treated with the genotype-guided dose for acenocoumarol were not statistically significant different from the patients who received the clinical algorithm dose, among either the younger patients or the older patients. A possible explanation for this finding is that the half-life of acenocoumarol [24] is considerably

shorter compared with the half-life of phenprocoumon [25]. Therefore, we used a different dose adjustment strategy after the loading period in the EU-PACT trial [9]. This might account for the different stratified outcomes between acenocoumarol and phenprocoumon.

Several limitations of our study should be considered. First, this study is a subgroup analysis of a prospective randomized trial, leading to small sample sizes in different strata, especially in the older subset of the phenprocoumon patients. This reduced the power and caused a large confidence interval. Second, the stratified analysis was not part of the original study design of the EU-PACT trial and thus this *post hoc* analysis with multiple testing might cause chance findings. It is important to conduct further studies to test a separate dose algorithm for older patients. Third, our study used the TTR and the percentage of time spent below and above TTR as outcomes, which are surrogate outcomes for evaluating the quality of anticoagulation, whereas clinical events are more important in clinical practice.

In conclusion, we found, in the EU-PACT trial, that *VKORC1* and *CYP2C9* genotypes together with clinical factors could improve the accuracy in predicting the initial dose of phenprocoumon in patients aged <75 years during the initial 12 weeks of treatment. For patients aged ≥75 years, the algorithm should be revised and tested in further research.

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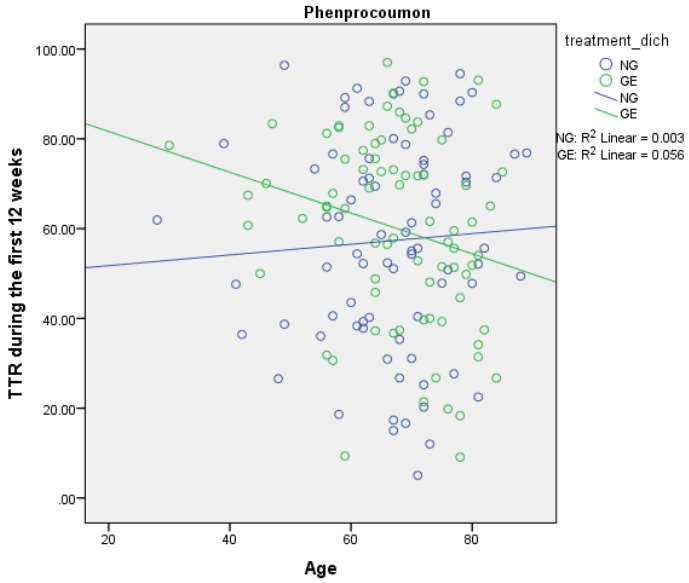
Supplement

A



B

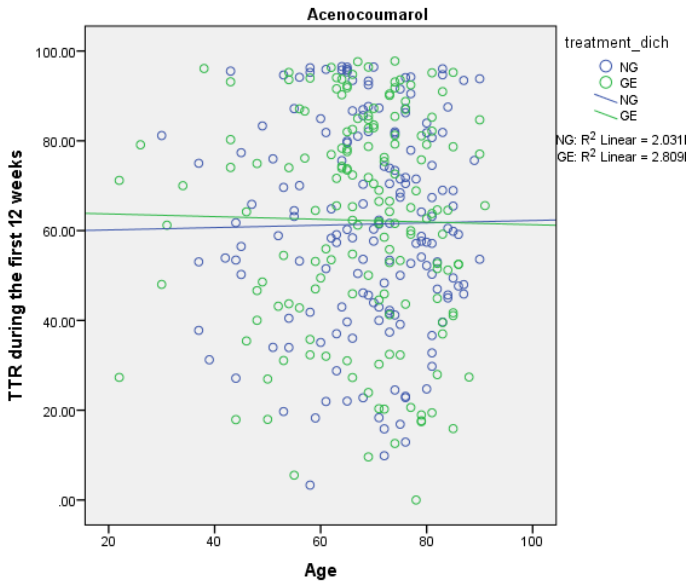


Figure S1 There is a trend towards an age interaction on the primary outcome of genotype guided dosing for phenprocoumon users (A) while not for acenocoumarol users (B). The p-value of the interaction between age and treatment was 0.08 for phenprocoumon, while 0.81 for acenocoumarol. Although not statistically significant, we considered that there is an age interaction. (GE denotes genotype-guided group; NG denotes the control group)

Per-protocol analysis

A

B

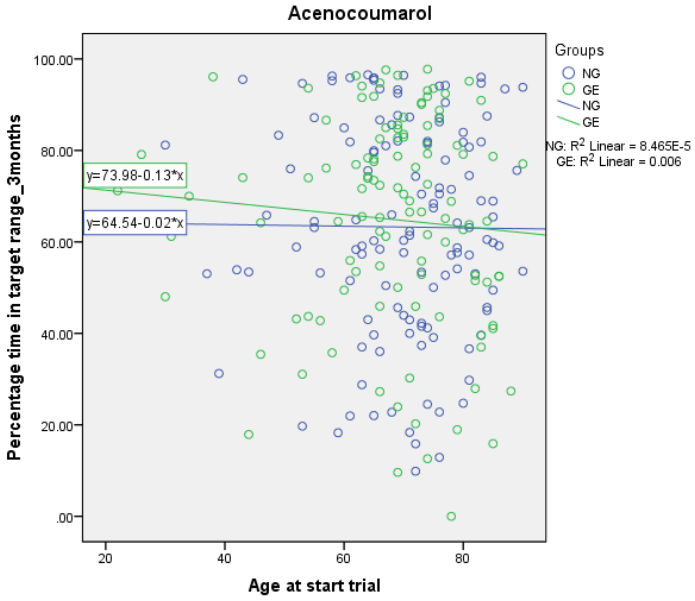


Figure S2 There is a trend towards an age interaction on the primary outcome of genotype guided dosing for phenprocoumon users (A) while not for acenocoumarol users (B). The p-value of the interaction between age and treatment was 0.095 for phenprocoumon, while 0.64 for acenocoumarol. Although not statistically significant, we considered that there is an age interaction. (GE denotes genotype-guided group; NG denotes the control group)

Table S1. Concomitant drug use stratified by age and coumarins for the genotype-guided group and the control group separately.

	Acenocoumarol						Phenprocoumon					
	<75 years			≥75 years			<75 years			≥75 years		
	Genotype-guided group (n=113)	Control (n=103)	P-value	Genotype-guided group (n=47)	Control (n=62)	P-value	Genotype-guided group (n=55)	Control (n=63)	P-value	Genotype-guided group (n=24)	Control (n=17)	P-value
Co-medications, n (%)	113 (100)	99 (96.1)		46 (97.9)	61 (98.4)		55	61 (96.8)		23 (95.8)	16 (94.1)	
Drugs with potentiating effect, n (%)	76 (67.3)	65 (63.1)	0.52	41 (87.2)	49 (79.0)	0.26	33 (60.0)	31 (49.2)		12 (50.0)	10 (58.8)	0.58
Enzyme Inhibitors, n (%)	60 (53.1)	48 (46.6)	0.34	36 (76.6)	41 (66.1)	0.24	27 (49.1)	22 (34.9)		11 (45.8)	8 (47.1)	0.94
amiodarone	7 (6.2)	13 (12.6)	0.10	8 (17.0)	7 (11.3)	0.39	0	2 (3.2)		0	0	-
PPIs, n (%)	18 (16.0)	11 (10.7)	0.51	10 (21.3)	14 (22.6)	0.87	16 (29.1)	12 (19.0)		5 (20.8)	5 (29.4)	0.53
omeprazole	9 (8.0)	5 (4.9)		6 (12.8)	7 (11.3)		11 (20.0)	7 (11.1)		5 (20.8)	3 (17.6)	
esomeprazole	9 (8.0)	6 (5.8)		4 (8.5)	7 (11.3)		5 (9.1)	5 (7.9)		0	2 (11.8)	
Statins, n (%)	34 (30.1)	29 (28.2)	0.76	13 (27.7)	16 (25.8)	0.83	10 (18.2)	14 (22.2)	0.59	5 (20.8)	4 (23.5)	0.84
simvastatin	23 (20.4)	20 (19.4)		10 (21.3)	13 (21.0)		8 (14.5)	13 (20.6)		5 (20.8)	4 (23.5)	
rosuvastatin	11 (9.7)	9 (8.7)		1 (2.1)	2 (3.2)		2 (3.6)	1 (1.6)		0	0	
Antidepressants, n (%)	4 (3.5)	2 (1.9)	0.48	0	5 (8.1)	-	5 (9.1)	2 (3.2)	0.18	0	0	-
fluoxetine	0	0		0	2 (3.2)		1 (1.8)	1 (1.6)		0	0	
venlafaxine	3 (2.7)	2 (1.9)		0	0		1 (1.8)	0		0	0	
escitalopram	1(0.9)	0		0	3 (4.8)		3 (5.5)	1 (1.6)		0	0	
Antibacterial drugs, n (%)	11 (9.7)	8 (7.8)	0.61	5 (10.6)	10 (16.1)	0.41	6 (10.9)	6 (9.5)	0.80	1 (4.2)	0	-
doxicycline	2 (1.8)	0		0	0		1 (1.8)	1 (1.6)		0	0	
ampicillin	4 (3.5)	2 (1.9)		1 (2.1)	2 (3.2)		4 (7.3)	3 (4.8)		0	0	
amoxicillin	1 (0.9)	0		0	0		0	0		0	0	
flucloxacillin	0	1 (1.0)		0	0		0	0		0	0	
piperacillin	0	0		3 (6.4)	0		0	0		0	0	
cephalosporin	3 (2.9)	2 (1.8)		1 (2.1)	4 (6.5)		0	0		0	0	
ciprofloxacine	1 (1.0)	1 (0.9)		0	4 (6.5)		0	1 (1.6)		0	0	

norfloxacin	0	0		0	0		0	1 (1.6)		0	0	
clarithromycin	0	0		0	0		1 (1.8)	0		0	0	
azithromycin	1 (0.9)	1 (1.0)		0	0		0	0		0	0	
sulfamethoxzole+trimethoprim	0	0		0	0		0	0		1 (4.2)	0	
N**SAIDs, n (%)**	9 (8.0)	5 (4.8)	0.35	1 (2.1)	2 (3.2)	1.00	2 (3.6)	3 (4.8)	0.76	0	0	-
dicolfenac	9	3 (2.9)		1 (2.1)	1 (1.6)		2 (3.6)	3 (4.8)		0	0	
naproxen	0	2 (1.9)		0	0		0	0		0	0	
meloxicam	0	0		0	1 (1.6)		0	0		0	0	
lactulose, n (%)	2 (1.8)	0	-	1 (2.1)	1 (1.6)	1.00	0	0		0	0	-
aspirin	19 (16.8)	19 (18.4)	0.75	10 (21.3)	21 (33.9)	0.15	1 (1.8)	2 (3.2)	0.64	0	2 (11.8)	0.09
digoxin	1 (0.9)	0	0.34	0	0	-	6 (10.9)	4 (6.3)	0.38	0	3 (17.6)	-
Enzyme Inducers, n (%)	0	1	-	1		-	0	0	-	0	0	-
carbamazepine	0	1	-	1		-	0	0	-	0	0	-

Table S2. Protocol violations and numbers of patients excluded from the per-protocol analyses.

	Acenocoumarol				Phenprocoumon			
	Age <75 years		Age ≥75 years		Age <75 years		Age≥75 years	
	Genotype-guided group	Control group	Genotype-guided group	Control group	Genotype-guided group	Control group	Genotype-guided group	Control group
Protocol violation	33 (29.2)	26 (25.2)	16 (34)	13 (21)	22 (40)	16 (25.4)	8 (33.3)	6 (35.3)
Error made with algorithm or genotype not available, n (%)	9 (8)	0	5 (10.6)	1 (1.6)	5 (9.1)	2 (3.2)	2 (8.3)	0
Physician changed dose or patient did not take drug as prescribed after first 2 visits, n (%)	15 (13.3)	16 (15.5)	9 (19.1)	8 (12.9)	5 (9.1)	4 (6.3)	0	0
No INR available on days 3 to 5 to calculate a revised dose, n (%)	13 (11.5)	8 (7.8)	3 (6.4)	4 (6.5)	15 (27.3)	10 (15.9)	6 (25.0)	6 (35.3)
Target INR range changed during the study, n (%)	1 (0.9)	3 (2.9)	0	0	0	1 (1.6)	0	0
Patient took more than one dose before starting on loading dose according to the algorithm, n (%)	0	0	1 (2.1)	0	0	0	0	0

Some patients had 2 or more violations.

*This result was also reported in the Supplement of our previous publication (Talitha *et al* [9]).

Table S3. Percentage of time in, below and above the therapeutic range during 12 weeks after the initiation of treatment after stratification by age* (Per protocol analysis).

	Age <75 years					Age ≥75 years				
	Genotype-guided group % mean±SD	Control group % mean±SD	Difference (95%CI)	Adjusted Difference (95%CI)	*P*-value *	Genotype-guided group % mean±SD	Control group % mean±SD	Difference (95%CI)	Adjusted Difference (95%CI)	*P*-value
Phenprocoumon										
Number	33	47				16	11			
INR 2-3 (%)	63.3±18.1	56.0±23.9	7.3 (-2.5 to 17.2)	9.1 (-1.0 to 19.1)	0.08	50.7±20.3	68.2±16.9	-17.5 (-32.9 to -2.2)	-16.7 (-33.5 to 0.1)	0.05
INR <2 (%)	17.9±16.3	19.4±20.5	-1.5 (-10.1 to 7.0)	-1.7 (-10.0 to 6.7)	0.69	20.1±19.6	25.4±18.8	-5.3 (-20.8 to 10.2)	-11.6 (-28.1 to 4.8)	0.16
INR>3 (%)	18.8±19.6	24.6±25.8	-5.8 (-16.4 to 4.8)	-7.4 (-18.5 to 3.7)	0.19	29.2±26.9	6.4±11.4	22.8 (5.0 to 40.6)	28.4 (8.9 to 47.8)	0.01
INR>4 (%)	1.9±4.2	3.2±7.0	-1.4 (-4.1 to 1.3)	-1.4 (-4.3 to 1.4)	0.31	1.4 ±2.4	1.0±3.5	0.4 (-1.9 to 2.7)	0.5 (-2.2 to 3.2)	0.71
Acenocoumarol										
Number	80	77				31	49			
INR 2-3 (%)	67.6±22.0	62.3±24.8	5.3 (-2.1to 12.7)	6.0 (-1.5 to 13.5)	0.12	58.1±25.1	64.8±20.7	-6.6 (-16.9 to 3.7)	-5.4 (-15.9 to 5.2)	0.31
INR <2 (%)	22.3±21.2	19.4±19.5	2.8 (-3.6 to 9.3)	1.2 (-5.2 to 7.6)	0.71	32.9±26.8	22.5±19.8	10.4 (-0.0 to 20.8)	9.4 (-1.2 to 20.0)	0.08
INR>3 (%)	10.1±13.9	18.3±21.3	-8.1 (-13.8 to -2.5)	-7.2 (-13.0 to -1.4)	0.02	9.0±12.8	12.7±13.7	-3.7 (-9.8 to 2.4)	-4.0 (-10.2 to 2.1)	0.19
INR>4 (%)	2.0±6.5	2.3±5.6	-0.4 (-2.2 to 1.8)	-0.2 (-2.2 to 1.8)	0.83	2.1±6.2	1.8±5.3	0.3 (-2.3 to 2.9)	0.4 (-2.2 to 3.0)	0.75

CI: confidence interval; INR: International Normalized Ratio; SD: standard deviation.

**P*-value for the difference adjusted for height, weight, sex, enzyme inhibitors, and enzyme inducers.

Table S4. Effect of genetic variants on anticoagulation control in genotyped and control patients among younger and elderly acenocoumarol users*.

	Age <75 years						Age ≥75 years					
	Genotype-guided group		Control group		Percentage difference (95%CI)	*P*-value	Genotype-guided group		Control group		Percentage difference (95%CI)	*P*-value
	%, mean ± SD	n	%, mean ± SD	n			%, mean ± SD	n	%, mean ± SD	n		
Percentage of time with time in therapeutic INR range												
No variant	65.2±22.9	33	58.9±23.8	20	6.3 (-6.9 to 19.5)	0.34	56.2±24.8	8	53.4±21.9	14	2.8 (-18.4 to 24.0)	0.79
One variant	62.4±24.6	31	65.2±21.2	33	-2.8 (-14.2 to 8.7)	0.63	56.0±26.8	21	60.9±22.5	21	-4.8 (-20.3 to 10.6)	0.53
Two or more variants	65.3±24.1	49	59.6±26.6	50	5.7 (-4.4 to 15.8)	0.27	58.5±25.5	18	66.7±19.0	27	-8.1 (-21.5 to 5.3)	0.23
Percentage of time with INR<2.0												
No variation	23.9±23.9	33	30.4±24.3	20	-6.5 (-20.4 to 7.3)	0.34	38.2±30.2	8	35.1±19.6	14	-1.0 (-24.5 to 22.6)	0.93
One variant	23.4±23.0	31	18.6±17.0	33	4.8 (-5.3 to 14.8)	0.35	31.3±25.4	21	18.0 ±18.6	21	13.3 (-6.9 to 27.2)	0.06
Two or more variants	17.0±19.3	49	16.6±19.5	50	1.0 (-6.7 to 8.8)	0.13	30.4±28.5	18	17.1±14.0	27	13.3 (-1.7 to 28.3)	0.08
Percentage of time with INR>3.0												
No variation	10.9±15.2	33	10.7±16.5	20	0.3 (-8.7 to 9.2)	0.95	5.6±8.0	8	7.4±9.3	14	-1.8 (-10.0 to 6.3)	0.65
One variant	14.2±19.3	31	16.2±19.0	33	-2.0 (-11.5 to 7.6)	0.68	12.7±19.2	21	21.2±18.5	21	-8.5 (-20.2 to 3.3)	0.15
Two or more variants	17.0±20.8	49	23.8±23.7	50	-6.8 (-15.7 to 2.1)	0.13	11.1±13.1	18	16.2±17.4	27	-5.2 (-14.9 to 4.5)	0.29

CI, confidence interval; INR, International Normalized Ratio; SD, standard deviation.
**P*-value for the difference adjusted for height, weight, sex, enzyme inhibitors, and enzyme inducers.

CHAPTER 4

Risk of major bleeding among users of direct oral anticoagulants combined with interacting drugs: a population based nested case-control study

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Abstract

Objectives: To assess the association between concurrent use of potential pharmacokinetic or pharmacodynamic interacting drugs and major bleeding among DOAC users.

Design: Nested case-control study.

Setting: UK Clinical Practice Research Datalink linked to Hospital Episode Statistics (2008-2015).

Participants: New users of DOACs (dabigatran, apixaban, and rivaroxaban) aged 18 years or older who were hospitalized having a primary diagnosis of major bleeding, matched with up to 4 controls by age, sex, index date (date of bleeding in the cases), and region.

Main outcome measure: Odds ratios (ORs) for the risk of major bleeding were assessed by conditional logistic regression analysis and adjusted for well-known covariates for the risk of bleeding.

Results: We identified 393 patients with a major bleeding from a total of 23492 new users of DOACs and matched them to 1494 controls. Most subjects were users of rivaroxaban (58.8%) on the index date. The concurrent use of pharmacodynamic interacting drugs was associated with an increased risk of major bleeding (21.6% vs. 13.5%, adjusted OR (aOR) 1.92; 95% CI, 1.40-2.66). For the antiplatelet drugs the aOR was 2.01; 95% CI, 1.29-3.11) and for the selective serotonin reuptake inhibitors (SSRIs) the aOR was 1.68; 95% CI, 1.10-2.59). We found no increased risk of major bleeding for concurrent use of pharmacokinetic interacting drugs vs. DOACs alone (45.0% vs. 51.2%; adjusted OR (aOR): 0.77; 95% CI: 0.53-1.10).

Conclusion: Among patients taking DOACs the concurrent use of antiplatelet drugs or SSRIs was associated with increased risk of major bleeding, while pharmacokinetic interacting drugs did not increase this risk.

Introduction

Oral anticoagulants are recommended for the prevention and/or treatment of thromboembolic disorders including atrial fibrillation (AF), deep venous thrombosis (DVT) and pulmonary embolism (PE), orthopedic surgery and acute myocardial infraction (MI). Vitamin-K antagonists (VKAs) were the only available oral anticoagulants in the past decades. In recent years, direct oral anticoagulants (DOACs) such as dabigatran, apixaban, rivaroxaban and edoxaban have been introduced. Compared with vitamin K antagonists, these drugs have a more predictable anticoagulant effect, without the need for routine monitoring [1]. Despite the advantages of DOACs over VKAs several uncertainties about their benefit-risk profile remain [2]. Several drugs could influence the safe use of DOACs via pharmacokinetic or pharmacodynamic interactions when used at the same time as DOACs [3-5].

The absorption of DOACs is dependent on the P-glycoprotein (P-gp) system. P-gp inhibitors, for instance, verapamil, amiodarone and quinidine, etc. can increase plasma concentrations of DOACs [6, 7], thereby enhancing the anticoagulant effect. CYP3A4-type cytochrome P450-dependent elimination is another factor involved in the metabolism of DOACs. Therefore, CYP3A4 inhibitors (e.g. verapamil) or inducers (e.g. rifampicin, carbamazepine) can influence plasma concentrations of DOACs and thereby increase bleeding risk or reduce effectiveness (less antithrombotic effect), respectively.

In addition to the pharmacokinetic interactions, there are other drugs that may increase bleeding risk via pharmacodynamic interactions such as antiplatelet drugs, non-steroidal anti-inflammatory drugs (NSAIDs) and selective serotonin re-uptake inhibitors (SSRIs) [8-10]. To date, it is not known what the clinical relevance of these drug-drug interactions is since only sporadic case reports and laboratory data of manufactures are available [6, 11]. One recent conducted study from Taiwan reported that concurrent use of drugs with pharmacokinetic interactions in DOAC users with nonvalvular atrial fibrillation was associated with an increased risk of bleeding [8]. However, it is unknown to what extent these findings can be replicated.

Therefore, the aim of our study was to evaluate the combined use of DOACs with potentially pharmacokinetic and pharmacodynamic interacting drugs on bleeding risk.

Methods

Study design and data source

We performed a case-control study nested in a cohort of new users of DOACs (dabigatran etexilate, apixaban, or rivaroxaban). Data were obtained from the Clinical Practice Research Datalink (CPRD) and linked to secondary care data from the Hospital Episode Statistics (HES) [12, 13]. CPRD is a longitudinal research database which includes more than 14 million patient records provided by general practitioners (GPs) throughout the United Kingdom. Data recorded in CPRD includes demographics, symptoms and diagnoses, prescriptions, results of diagnostic investigations, referrals to specialists and secondary care settings, feedback from other care settings and lifestyle, such as body mass index (BMI) and smoking status. Use of the CPRD as a reliable data source has been well validated [12, 14].

HES data include primary and contributory causes of patient admission to NHS hospitals in England [13]. The data includes patient demographics, and clinical and administrative details. Data are coded using the International Classification of Diseases (ICD)-10 classification. Approval of the study protocol was granted by the Independent Scientific Advisory Committee of the Medicines and Healthcare Products Regulatory Agency (protocol 17_257R).

Definition of study cohort

We identified all patients with a first prescription for a DOAC between January 1, 2008 and December 31, 2015, who were 18 years or older, had at least one year of history in CPRD prior to the date of this first prescription and were eligible for data linkage with HES. The duration of individual DOAC prescriptions was assessed by using information on the prescribed number of tablets and the dosage instruction. When such information was incomplete for the prescription, the median time between prescriptions was used. If only 1-3 prescriptions were available for an individual patient, the duration was based on the most frequently occurring

estimated prescription duration for the drug in the study population. Subsequently, periods of current use were determined for each patient by constructing treatment episodes. A treatment episode was defined as a series of subsequent prescriptions for DOAC, independent of dose changes and constructed according to the method used in our previous study [15]. We allowed for a 30-days permissible gap between the theoretical end date of one DOAC prescription and the subsequent prescription. In case a subsequent prescription for the same drug was collected before the theoretical end date of a previous prescription, the number of overlapping days was added to the theoretical end date of the subsequent prescription. The number of overlapping days was maximized at 90 days. If a subsequent prescription within the same treatment episode was for another type of DOAC, the patient was considered to have switched therapy and the remaining tablet days from the prior prescription were disregarded.

Definition of cases and controls

Cases were defined as current users of DOACs with a first hospital admission related to major bleeding after DOAC start as identified by ICD-10 codes (see Table 1 in appendix). Major bleeding events were a composite of gastrointestinal, intracranial, and other symptomatic bleeding in a critical area or organ defined use an adapted version of the definition of International Society on Thrombosis and Haemostasis [16]. The date of the major bleeding event was defined as the index date. For each case, up to 4 controls were matched from the study cohort by means of incidence density sampling on sex, age (+/-1 year), region and index date. Only controls that were using DOACs on the index date were eligible for inclusion.

Exposure to drugs with potential interaction with DOACs

For both cases and controls, therapy records for potentially interacting drugs (Table 1) were identified. These interacting drugs were obtained from the Summary of Product Characteristics (SmPCs) [8-10] and the European Heart Rhythm Association Practical Guide [17]. In the latest guide [18], many anticancer drugs were listed as potential interacting drugs, but as these are prescribed in-hospital this information is not captured in CPRD and hence were not considered in this study. We classified interacting drugs into potentially pharmacokinetic (PK)

interacting drugs (inhibitors of P-gp and/or CYP3A4) and pharmacodynamic (PD) interacting drugs (drugs which already enhance bleeding risk themselves). Drugs that can potentially cause both PK and PD interactions, including clopidogrel, ticragrelor, diclofenac, and naproxen, were categorized into PD interacting drugs because of their mild inhibitory effects on CYP3A4 and P-gp [8-10, 19-21]. We assumed concurrent use if a prescription was issued in a 30 days window prior to the index date (for antibiotics 14 days). A sensitivity analysis was performed using an extended time window of 60 days prior to the index date.

Table 1. List of drugs with potential interaction with DOACs as found to be co-prescribed in the CPRD database.

Drugs with pharmacokinetic interaction		**Drugs with pharmacodynamic interaction**
Strong CYP3A4 and/or P-gp inhibitors	**Moderate CYP3A4 and / or P-gp inhibitors**	
ketoconazole	amiodarone	**Antiplatelet drugs**
cyclosporine	posaconazole	ticlopidine
itraconazole	quinidine	clopidogrel*
dronedarone	verapamil	acetylsalicylic acid
tacrolimus	digoxin	ticagrelor*
	diltiazem	**NSAIDs**
	simvastatin	**SSRIs**
	atorvastatin	fluoxetine
	fluconazole	paroxetine
	clarithromycin	citalopram
	erythromycin	escitalopram
		sertraline
		nefazodone
		SNRIs
		venlafaxine
		duloxetine

ASA: acetylsalicylic acid; SSRIs: selective serotonin reuptake inhibitor; SNRIs: serotonin–norepinephrine reuptake inhibitor.
*Ticagrelor, and clopidogrel are also substrate of the P-glycoprotein transporter.

Potential confounding factors

We included the following covariates which are well known for the risk of bleeding: BMI, smoking status, hypertension, chronic kidney disease, hepatic impairment (moderate to severe), history of major bleeding, gastritis, cancer, peptic ulcer disease, and thrombocytopenia in the year before the index date. We included all components of the HAS-BLED score except INR value, for which data in CPRD are incomplete. Furthermore, other comedication, not belonging to the group of direct potential interacting drugs which include glucocorticoids, proton pump inhibitors, and enzyme inducers (carbamazepine, phenytoin, and rifampicin) were considered and were recorded in a 6-month period prior to the index date.

Statistical analyses

Descriptive statistics were used to assess characteristics of cases and controls. Means and standard deviations are shown for continuous variables and proportions for categorical variables. Student's t-test for continuous variables and chi-squared test for categorical variables were used as appropriate. We compared the proportion of patients having DOAC dose adjustments between index date and the last prescription prior to the index date by the use of chi-squared tests. The strength of the association between concurrent use of interacting drugs and risk of major bleeding was assessed using conditional logistic regression analysis for all DOACs together. For individual DOACs the matching of cases and controls was discarded and therefore unconditional logistic regression analyses were used. Additionally, the associations were analyzed for individual DOACs and when possible for different types of major bleeding. The associations were expressed as odds ratios (ORs) and 95% confidence intervals (95% CI). We adjusted for the above-mentioned potential confounders and type of DOAC. Additionally, when analyzing the association of potentially pharmacokinetic interacting drugs we also adjusted for potentially pharmacodynamic interacting drugs and vice versa. As mentioned above a sensitivity analysis was performed using an extended time window of 60 days (instead of 30 days) prior to the index date. A two-sided *P*-value of less than 0.05 was considered statistically significant. Data analyses were performed using SAS version 9.4 (SAS institute).

Results

The study cohort comprised of 23492 DOAC users aged 18 years or older initiating DOACs therapy between 2008 and 2015. Among these patients, we identified 393 cases with a first major bleeding event admission and matched them with 1494 controls.

Baseline characteristics of cases and controls are shown in Table 2. The mean age on the index date was 78.7 years (SD 10.6), about 62% were men, and most of the patients (73.5% in cases group vs. 81.2% in control group) used DOACs for the treatment of atrial fibrillation. In general, comorbidities were more prevalent among cases than among controls (Table 2). Use of co-medication without potential interactions was common among both cases and controls, with controls using some of the statins (with no *CYP3A4* and P-gp inhibition), angiotensin-converting-enzyme inhibitor and calcium channel blockers more frequently.

Primary analysis

Table 3 shows that use of pharmacokinetic interacting drugs on the index date occurred in 45.0% of the cases and 51.2% of controls, yielding a crude OR of 0.78 (95% CI: 0.62-0.98). After adjustment for confounders, no statistically significant association with bleeding risk was found: OR 0.77 (95 % CI: 0.53-1.10). The most frequently prescribed drugs with potential pharmacokinetic interactions with DOACs were simvastatin (cases vs controls: 19.3% vs 25.0%), followed by atorvastatin (cases vs controls: 15.0% vs 15.5%), and digoxin (cases vs controls: 13.7% vs 12.9%). When individual drugs were evaluated only verapamil and diltiazem reached statistically significant associations, however numbers of exposed patients were very low.

Concurrent use of drugs having pharmacodynamic interactions with DOACs (Table 3) was associated with a statistically significantly increased risk of bleeding: adjusted OR of 1.92 (95% CI: 1.40-2.66). The most frequently used drugs in this group were antiplatelet drugs (adjusted OR 2.01, 95%CI: 1.29-3.11) and SSRIs (adjusted OR, 1.68, 95%CI: 1.10-2.59). Acetylsalicylic acid was the most frequently used antiplatelet drug (cases vs controls: 8.1% vs 4.4%). The prevalences of all interacting drugs are presented in the appendix Table S2.

Table 2. Characteristics of included patients.

	Cases ***n*=393**	**Controls** ***n*=1494**	***P*-value**
Age, yrs			
Mean (SD)	78.7 (10.6)	78.7 (10.1)	0.76
<75, n (%)	123 (31.3)	455 (30.5)	
≥75, n (%)	270 (68.7)	1039 (69.5)	
Sex, male, n (%)	243 (61.8)	932 (62.4)	0.84
BMI (kg/m2), mean (SD)	27.3 (6.2)	27.5 (5.5)	0.48
BMI missing (%)	19 (4.8)	51 (3.4)	
Smoking status, n (%)			0.83
No	140 (35.6)	552 (37.0)	
Yes	32 (8.1)	127 (8.5)	
Former	221 (56.2)	813 (49.2)	
Type of DOAC			
Dabigatran	79 (20.1)	279 (18.7)	
Apixaban	53 (13.5)	366 (24.5)	
Rivaroxaban	261 (66.4)	849 (56.8)	
Indications			0.003
Atrial fibrillation	289 (73.5)	1213 (81.2)	
DVT/PE	62 (15.8)	153 (10.2)	
Other	56 (14.2)	185 (12.4)	
Comorbidities*			
Congestive heart failure	85 (21.6)	238 (15.9)	0.008
Diabetes	73 (18.6)	290 (19.4)	0.71
Hypertension	256 (65.1)	1012 (67.7)	0.33
COPD	73 (18.6)	165 (11)	<0.001
Peripheral vascular disease	27 (6.9)	81 (5.4)	0.27
Upper GI disease	35 (8.9)	103 (6.9)	0.17
Chronic kidney disease	28 (7.1)	75 (5.0)	0.10
Chronic kidney disease (missing)	14 (3.5)	46 (3.1)	-
Chronic liver disease	<5	<5	-
History of acute coronary disease	109 (27.7)	336 (22.5)	0.03
History of bleeding	234 (59.5)	610 (40.8)	<0.001
History of GI bleeding	93 (23.7)	212 (14.2)	<0.001
History of intracranial bleeding	16 (4.1)	38 (2.5)	0.11
Comedications#			
β-adrenergic receptor blockers	148 (37.7)	607 (40.6)	0.29
ACEI	139 (35.4)	617 (41.3)	0.03

Diuretics	127 (32.3)	474 (31.7)	0.82
Calcium channel blockers	55 (14.0)	323 (21.6)	0.001
Other statins†	145 (36.9)	648 (43.4)	0.02
Proton pump inhibitors	174 (44.3)	611 (40.9)	0.23

Note: Data are no (%) of patients unless stated otherwise. According to the policy of CPRD database, all the case less than 5 are shown as "<5".
*Comorbidities before the index date.
#Co-medications other than potentially interacting drugs.
†Excluding the potentially interacting drugs simvastatin and atorvastatin
NSAIDs: nonsteroidal anti-inflammatory drug; ACEI: angiotensin-converting-enzyme inhibitor; SD: standard deviation.

Secondary analysis

Analyses of potentially interacting drugs with individual DOACs showed that the results (point estimates) were not materially different from the analyses of the DOACs as a group, although some of the associations were no longer statistically significant (see Table S4 for pharmacodynamics drugs, and Table 3 in the appendix for pharmacokinetic drugs). After stratification for different types of major bleeding only for gastrointestinal bleeding the numbers were high enough to evaluate the risks of major bleeding for DOAC users combined with interacting drugs. It appeared that major bleeding risks were similar as found for all major bleedings together, however, the associations were no longer statistically significant (Table S4 in the appendix).

Table 3. Major bleeding risk among patients taking DOACs with the concomitant use of potentially interacting drugs.

Concurrent use of*	Cases (n=393), n (%)	Controls (n=1494), n (%)	Crude OR (95% CI)	Adjusted OR# (95% CI)
Drugs with PK interaction, n (%)	177 (45.0)	765 (51.2)	0.69 (0.62-0.98)	0.69 (0.53-0.90)
amiodarone	7 (1.8)	40 (2.7)	0.66 (0.29-1.48)	0.67 (0.28-1.59)
simvastatin	76 (19.3)	374 (25.0)	0.72 (0.54-0.95)	0.69 (0.42-1.13)
atorvastatin	59 (15.0)	232 (15.5)	0.96 (0.71-1.31)	1.25 (0.83-1.88)
verapamil	5 (1.3)	12 (0.8)	1.67 (0.59-4.73)	1.76 (0.58-5.35)
digoxin	54 (13.7)	192 (12.9)	1.08 (0.78-1.50)	1.08 (0.75-1.55)
diltiazem	7 (1.8)	69 (4.6)	0.37 (0.17-0.81)	0.26 (0.11-0.61)
Drugs with PD interaction, n (%)	85 (21.6)	202 (13.5)	1.79 (1.34-2.40)	1.88 (1.36-2.61)
SSRIs	41 (10.4)	95 (6.4)	1.71 (1.14-2.54)	1.68 (1.10-2.59)
Antiplatelet drugs	41 (10.4)	90 (6.0)	1.79 (1.21-2.64)	2.01 (1.29-3.11)
ASA	28 (7.1)	62 (4.1)	1.76 (1.10-2.82)	1.94 (1.16-3.26)
CLOP	9 (2.3)	23 (1.5)	1.54 (0.70-3.41)	1.68 (0.71-3.97)
ASA+CLOP	<5	<5	-	-
NSAIDs	7 (1.8)	19 (1.3)	1.45 (0.60-3.54)	1.30 (0.50-3.41)

*All the concurrent used drugs with DOACs were compared to use DOACs but without use these drugs.

#Adjusted for smoking, history of major bleeding, history of stroke or transient ischemic attack before the bleeding event, diabetes, hypertension, myocardial infarction, congestive heart failure, chronic renal disease, hepatic impairment, peripheral vascular disease, chronic pulmonary disease, peptic ulcer disease, cancer, co-medications before the index date medications (β-adrenergic receptor blockers, ACEIs, non-P-gp inhibitor statins, proton pump inhibitors, and cytochrome P450 enzyme inducers). For analyzing the association of potentially pharmacokinetic interacting drugs we also adjusted for potentially pharmacodynamic interacting drugs and vice versa. ASA: acetylsalicylic acid; CLOP: clopidogrel; PK: P-gP inhibitors or CYP3A4 inhibitors; PD: pharmacodynamic.

According to the policy of CPRD database, all the case less than 5 are shown as "<5".

Table 4. Major bleeding risk among patients using apixaban, dabigatran, or rivaroxaban with the concomitant use of drugs with pharmacodynamics interaction.

	Apixaban				Dabigatran				Rivaroxaban			
Concurrent use of	**Cases (n=53), n (%)**	**Controls (n=279), n (%)**	**Crude OR (95% CI)**	**Adjusted OR* (95% CI)**	**Cases (n=79), n (%)**	**Controls (n=366), n (%)**	**Crude OR (95% CI)**	**Adjusted OR* (95% CI)**	**Cases (n=261), n (%)**	**Controls (n=849), n (%)**	**Crude OR (95% CI)**	**Adjusted OR* (95% CI)**
PD Interacting drugs	14 (26.4)	34 (12.2)	2.59 (1.27-5.25)	3.32 (1.43-7.71)	18 (22.8)	47 (12.8)	2.00 (1.09-3.68)	3.00 (1.48-6.1)	53 (20.3)	121 (14.3)	1.53 (1.07-2.19)	1.39 (0.93-2.06)
SSRIs	6 (11.3)	13 (4.7)	2.22 (0.60-8.21)	2.78 (0.78-9.91)	10 (12.7)	20 (5.5)	2.51 (1.12-5.59)	2.70 (1.09-6.70)	25 (9.6)	62 (7.3)	1.35 (0.83-2.19)	1.23 (0.73-2.07)
Antiplatelet dugs	6 (11.3)	19 (6.8)	2.30 (0.60-8.86)	2.07 (0.66-6.49)	9 (11.4)	24 (6.6)	1.83 (0.82-4.11)	2.81 (1.05-7.51)	26 (10.0)	47 (5.5)	1.89 (1.14-3.12)	1.69 (0.97-2.94)
ASA	<5	12 (4.3)	-	-	5 (6.3)	17 (4.6)	1.44 (0.51-4.02)	1.90 (0.58-6.25)	18 (6.9)	33 (3.9)	1.86 (1.03-3.37)	1.71 (0.89-3.29)
CLOP	<5	6 (2.2)	-	-	<5	6 (1.6)	1.63 (0.32-8.24)	3.05 (0.44-21.34)	6 (2.3)	11 (1.3)	1.86 (0.68-5.09)	1.66 (0.56-4.89)
ASA+ CLOP	0	1 (0.4)	-	-	<5	<5	-	-	<5	<5	-	-
NSAIDs	<5	<5	-	-	<5	5 (1.4)	-	-	<5	10 (1.2)	1.31 (0.41-4.20)	1.24 (0.33-4.73)

*All the concurrent used drugs with DOACs were compared with DOAC use without these drugs.

#Adjusted for age, gender, BMI, smoking, history of major bleeding, history of stroke or transient ischemic attack before the bleeding event, diabetes, hypertension, myocardial infarction, congestive heart failure, chronic renal disease, hepatic impairment, peripheral vascular disease, chronic pulmonary disease, peptic ulcer disease, cancer, co-medications before the index date medications (β-adrenergic receptor blockers, ACEIs, non-P-gp inhibitor statins, proton pump inhibitors, and cytochrome P450 enzyme inducers). For analyzing the association of potentially pharmacokinetic interacting drugs we also adjusted for potentially pharmacodynamic interacting drugs and vice versa. ASA: acetylsalicylic acid; CLOP: clopidogrel; PD: pharmacodynamic.

According to the policy of CPRD database, all the case less than 5 are shown as “<5”.

Sensitivity analysis

Sensitivity analyses were conducted with an extended time window (prescription of potential interacting drugs within 60 days prior to the index date). Although more prescriptions for most of the interacting drugs were found (Table S5 in appendix), results did not materially change (Table S6 in appendix).

Discussion

In this nested case-control analysis using real-world primary care data, we found the risk of major bleeding leading to hospitalization increased by approximately 100% and 70% when antiplatelet drugs or SSRIs, respectively were combined with DOACs. For the drugs that inhibit CYP3A4 and/or P-gp no increased bleeding risk was found.

Nearly half (45%) of the patients with a major bleeding admission in our study was using a drug which can potentially cause a pharmacokinetic interaction with DOACs on the index date. Simvastatin, atorvastatin, and digoxin were the most commonly co-administered interacting drugs. It is reassuring that prescriptions of strong CYP3A4 and/or P-gp inhibitors like antifungal azoles and cyclosporine, which are advised to be avoided in DOAC users [22] were not found in this study.

Comparison with other studies

We found a statistically significant, nearly two-fold increased risk of major bleeding for drugs that pharmacodynamically interact with DOACs. Both antiplatelet drugs and SSRIs inhibit platelet aggregation, and thus primary haemostasis while the DOACs inhibit fibrin formation and thus secondary haemostasis. Similar increased risks in randomized controlled trials and observational studies were observed when antiplatelet drugs were combined with DOACs [9, 23, 24]. The increased bleeding risk when combining SSRIs with DOACs has not been published before although there are warnings in the SmPCs of dabigatran and rivaroxaban [9, 10]. In line with our findings, for the combination of coumarins and SSRIs an increased bleeding risk has been reported before [25, 26].

An important finding in our study is that despite a substantial combined use of moderate CYP3A4 and/or P-gp inhibitors and DOACs and the well-known effect of

these inhibitors to increase DOAC plasma levels, no increased major bleeding risk was observed. Also in post-hoc analyses of the combined use of amiodarone (moderate CYP3A4 inhibitor) with apixaban or rivaroxaban in the randomized trials ARISTOTLE [27] and ROCKET AF [28] respectively, no significant interactions on bleeding risk were found. This discrepancy between confirmed pharmacokinetic interactions and clinically relevant major bleeding may indicate the limitations of pharmacokinetic data in predicting clinical outcomes. Obviously, we cannot say anything about minor bleeding risks. In contrast to the probably limited impact of moderate CYP3A4 and/or P-gp inhibitors in bleeding risk as described above, other observational studies did report an increased risk for major bleedings when amiodarone, simvastatin or lovastatin were combined with DOACs [3, 29]. An explanation for these contrasting findings might be different behavior of prescribers to adjust (lower) the dose of DOACs when a pharmacokinetic interacting drug is co-prescribed as advised in the SmPCs. In a study presented in Chapter 5 we found that when a DOAC was combined with a potential interacting drug (CYP3A4 and/or P-gp inhibitor) there were not more dose adjustments of the DOACs than when DOACs were used without interacting drugs. However, the proportion of patients that received a lower DOAC dose was low. Thus, the fact that we did not find an increased major bleeding risk when DOACs were combined with pharmacokinetic interacting drugs cannot be explained by lowering of DOAC dosages. The unexpected decreased risk on major bleeding as found in our study when diltiazem was combined with DOACs is probably a chance finding due to a low number of patients exposed to diltiazem.

The rate of major bleeding observed in this study is lower than that reported in other study [30-32]. However, the rates are not comparable because we only included patients with first hospital admission related to major bleeding.

Implications for clinicians and policymakers

Although the combination of a platelet inhibitor and a DOAC increases the risk for major bleeding compared to DOACs alone or platelet inhibitors alone [23, 24] this increased risk is acceptable when a patient with atrial fibrillation develops an acute coronary syndrome [18] and can be considered in a patient with a recent acute coronary syndrome [23, 24]. Such combination of a platelet inhibitor and a DOAC

should be carefully considered depending on the bleeding risk of a patient (for instance by using the HAS-BLED score). Also, the guidelines should be strictly followed when the platelet inhibitor or DOAC needs to be discontinued (e.g. discontinuation of the platelet inhibitor after twelve months after an acute coronary syndrome with stent placement). Based on our findings of the increased bleeding risks when a DOAC is combined with a SSRI, prescribers should try to prevent such a combination in patients with a high bleeding risk. For instance, by considering a tricyclic antidepressant in the case of depression. Although we did not find an increased risk of major bleeding when drugs that moderately inhibit CYP3A4 and/or P-gp are combined with a DOAC, we advise to strictly follow the dose recommendations in the SmPCs of the specific DOACs.

Strength and Limitations

The strength of this study is that we used population-based data a from primary care setting, thereby reflecting the risk of major bleeding of the combined use of potentially interacting drugs with DOACs in daily practice. CPRD is a well-known research database of which the medical information entered is monitored for validity and completeness. However, some limitations need to be addressed. Firstly, our study might not include all concurrent exposure to interacting drugs at the index date, as we defined concurrent use based on a prescription in a 30-day time window prior to the index date. However, in sensitivity analyses where we expanded this period to 60 days provided similar results and therefore information bias caused by misclassification of the exposure is expected to be low. NSAIDs are available over the counter and therefore we expect misclassification to be present and therefore biased effect estimates in our study. Further research is necessary to evaluate the bleeding risk of the combined use of NSAIDs and DOACs. As we used the CPRD we might miss patients in the database that use a DOAC prescribed by hospital specialists. These patients probably have more complex diseases and treatments than patients prescribed DOACs by primary care physicians and therefore may have other bleeding risks when DOACs are combined with drugs known to interact with them. Furthermore, we did not have information on patient adherence and the identification of an adjustment of drug treatment can only be seen at the time a next prescription is issued. Due to the limited sample size it was only partly possible to

evaluate subgroups stratified by type of bleeding (e.g. intracranial, gastrointestinal bleeding), type of DOAC (dabigatran, rivaroxaban, apixaban) and individual interacting drugs.

Conclusion

Our study showed that drugs with pharmacodynamic interactions, mainly SSRIs and antiplatelet drugs, were used frequently in patients using DOACs and were associated with an increased risk of major bleeding. Although inhibitors of CYP3A4 and/or P-gp influence the pharmacokinetics of DOACs we did not find that these drugs increased the risk of major bleeding.

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Supplement

Table S1. International Classification of Diseases 10th revision (ICD-10) Codes used to identify major bleeding cases.

Condition	ICD-10 code
Major bleeding	
Haemorrhagic stroke/ intracranial	I60 I61 I62
Bleeding	
Extracranial or unclassified major bleeding	D62, J942, H113, H313, H356, H431 N02
	N95 R04 R31 R58
Gastrointestinal bleeding	K25.0, K25.2,
	K25.4, K25.6, K26.0, K26.2, K26.4, K26.6, K27.0, K27.2, K27.4, K27.6,
	K28.0, K28.2, K28.4, K28.6, K29.0,
	K62.5
	K92.0, K92.1, K92.2
Traumatic intercranial bleeding	S063C S064 S065 S066

Table S2. Interacting drugs prescribed to cases and controls on DOAC therapy in a 30-day time window prior to the index date.

	Cases n=393, n (%)	Controls n=1494, n (%)
Concomitant use of at least 1 drug with PK interaction	177 (45.0)	765 (51.2)
Strong CYP3A4 and/or P-gp inhibitor co-medication		
ketoconazole,	0	0
cyclosporine	0	0
itraconazole	0	1
dronedarone	<5	<5
tacrolimus	0	0
Moderate CYP3A4 and/or P-gp inhibitors		
amiodarone	7 (1.8)	40 (2.7)
posaconazole	0	0
quinidine	0	0
verapamil	5 (1.3)	12 (0.8)
digoxin	54 (13.7)	192 (12.9)
diltiazem	7 (1.8)	69 (4.6)
simvastatin	76 (19.3)	374 (25.0)
atorvastatin	59 (15.0)	232 (15.5)
ticagrelor	<5	0
fluconazole	<5	<5
clarithromycin	<5	7 (0.5)
erythromycin	<5	<5
Concomitant use at least 1 drug with PD interaction	85 (21.6)	202 (13.5)
Antiplatelets	41 (10.4)	90 (6.0)
ticlopidine	<5	0
clopidogrel	13 (3.3)	27 (1.8)
Low dose Acetylsalicylic acid (ASA)	32 (8.1)	66 (4.4)
NSAIDs	7 (1.8)	19 (1.3)
diclofenac	0	0
naproxen	<5	5 (0.3)
SSRIs	41 (10.4)	95 (6.4)
SNRI	6 (1.5)	16 (1.1)
Enzyme inducers		
rifampicin	0	0
carbamazepine	<5	<5
phenytoin	<5	<5
other inducers	0	0

Data are no (%) of patients unless stated otherwise; PK: P-gP inhibitors or CYP3A4 inhibitors; PD: pharmacodynamic, ACE: angiotensin-converting-enzyme; NSAIDs: nonsteroidal anti-inflammatory drug; SSRIs: Selective serotonin reuptake inhibitor; SNRIs: Serotonin–norepinephrine reuptake inhibitor. The SSRI we assessed were fluoxetine, paroxetine, sertraline, citalopram, escitalopram, sertraline, and nefazodone.

According to the policy of CPRD database, all the case less than 5 are shown as "<5".

Table S3. Association between the concurrent use of potentially interacting drugs and DOACs and gastrointestinal bleeding.

Concurrent use of	Cases (n=157), n (%)	Controls (n=594), n (%)	Crude OR (95% CI)	Adjusted OR* (95% CI)	*P*-value
Drugs with PK interaction,	68 (43.3)	314 (52.9)	0.66 (0.45-0.95)	0.53 (0.26-1.06)	0.07
amiodarone	<5	19 (3.2)	-	-	-
simvastatin	33 (21.0)	153 (25.8)	0.75 (0.49-1.16)	0.90 (0.47-1.72)	0.75
atorvastatin	22 (14.0)	87 (14.6)	0.95 (0.57-1.59)	1.11 (0.85-2.13)	0.75
verapamil	<5	<5	-	-	-
digoxin	19 (12.1)	91 (15.3)	0.76 (0.45-1.30)	0.62 (0.34-1.13)	0.12
diltiazem	<5	26 (4.4)	-	-	-
Drugs with PD interaction	28 (17.8)	84 (14.1)	1.28 (0.80-2.07)	1.27 (0.75-2.12)	0.37
SSRIs	14 (8.9)	41 (6.9)	1.29 (0.67-2.48)	1.25 (0.62-2.53)	0.50
Antiplatelet	14 (8.9)	38 (6.4)	1.40 (0.74-2.66)	1.39 (0.38-2.83)	0.36
ASA	10 (6.4)	28 (4.7)	1.37 (0.65-2.88)	1.29 (0.57-2.92)	0.55
CLOP	<5	8 (1.3)	1.93 (0.55-6.70)	2.48 (0.65-9.45)	0.19
ASA+ CLOP	0	<5	-	-	-
NSAIDs	0	6 (1.0)	-	-	-

Data are no (%) of patients unless stated otherwise; PK: pharmacokinetic, PD: pharmacodynamic, ACE: angiotensin-converting-enzyme; NSAIDs: nonsteroidal anti-inflammatory drug; SSRIs: Selective serotonin reuptake inhibitor; SNRIs: Serotonin–norepinephrine reuptake inhibitor. The SSRIs we assessed were fluoxetine, paroxetine, sertraline, citalopram, escitalopram, sertraline, and nefazodone.
According to the policy of CPRD database, all the case less than 5 are shown as "<5".

Table S4. Interacting drugs prescribed to cases and controls on DOAC therapy in a 60-day time window prior to the index date.

	Cases n=393, n (%)	Controls n=1494, n (%)
Concomitant use of at least 1 drug with PK interaction	224 (57.0)	936 (62.7)
Strong CYP3A4 and/or P-gp inhibitor co-medication		
ketoconazole,	0	0
cyclosporine	0	0
itraconazole	0	<5
dronedarone	<5	<5
tacrolimus	0	0
Moderate CYP3A4 and/or P-gp inhibitors		
amiodarone	10 (2.5)	51 (3.4)
posaconazole	0	0
quinidine	0	0
verapamil	5 (1.3)	18 (1.2)
digoxin	65 (16.5)	228 (15.3)
diltiazem	11 (2.8)	82 (5.5)
simvastatin	95 (24.2)	467 (31.3)
atorvastatin	76 (19.3)	294 (19.7)
ticagrelor	<5	<5
fluconazole	0	0
clarithromycin	0	0
erythromycin	<5	9 (0.6)
Concomitant use at least 1 drug with PD interaction	123 (31.3)	278 (18.6)
Antiplatelets	70 (17.8)	146 (9.8)
ticlopidine		
clopidogrel	23 (5.9)	40 (2.7)
acetylsalicylic acid (ASA)	55 (14.0)	110 (7.4)
NSAIDs	9 (2.3)	29 (1.9)
diclofenac	0	0
naproxen	5 (1.3)	11 (0.7)
SSRIs	53 (13.5)	113 (7.6)
SNRI	8 (2.0)	17 (1.1)
Enzyme inducers		
rifampicin	0	0
carbamazepine	<5	<5
phenytoin	<5	<5
other inducers	0	0

Data are no (%) of patients unless stated otherwise; PK: P-gP inhibitors or CYP3A4 inhibitors; PD: pharmacodynamic; ACE: angiotensin-converting-enzyme; NSAIDs: nonsteroidal anti-inflammatory drug; SSRIs: Selective serotonin reuptake inhibitor; SNRIs: Serotonin–norepinephrine reuptake inhibitor. The SSRIs we assessed were fluoxetine, paroxetine, sertraline, citalopram, escitalopram, sertraline, and nefazodone.
According to the policy of CPRD database, all the case less than 5 are shown as "<5".

Table S5. Association between use of concomitant drugs in current users of DOACs and risk of major bleeding (Sensitivity 60days).

Concurrent use of*	Cases (n=393), n (%)	Controls (n=1494), n (%)	Crude OR (95% CI)	Adjusted OR# (95% CI)
Drugs with PK interaction, n (%)	224 (57.0)	936 (62.7)	0.80 (0.635-1.00)	0.94 (0.64-1.39)
amiodarone	10 (2.5)	51 (3.4)	0.74 (0.38-1.48)	0.71 (0.34-1.48)
simvastatin	95 (24.2)	467 (31.3)	0.71 (0.55-0.92)	0.84 (0.58-1.20)
atorvastatin	76 (19.3)	294 (19.7)	1.00 (0.75-1.33)	1.18 (0.82-1.70)
verapamil	5 (1.3)	18 (1.2)	1.11 (0.41-3.04)	1.94 (0.64-5.83)
digoxin	65 (16.5)	228 (15.3)	1.09 (0.80-1.48)	1.06 (0.76-1.48)
diltiazem	11 (2.8)	82 (5.5)	0.49 (0.26-0.94)	0.37 (0.18-0.74)
Drugs with PD interaction, n (%)	123 (31.3)	278 (18.6)	2.02 (1.56-2.61)	2.10 (1.58-2.78)
SSRIs	53 (13.5)	113 (7.6)	1.94 (1.35-2.79)	1.91 (1.29-2.83)
Antiplatelet drugs	70 (17.8)	146 (9.8)	1.97 (1.45-2.69)	2.00 (1.36-2.95)
ASA only	46 (11.7)	103 (6.9)	1.82 (1.25-2.64)	1.88 (1.25-2.84)
CLOP only	14 (3.6)	33 (2.2)	1.74 (0.92-3.30)	1.59 (0.78-3.21)
> 1 drug	9 (2.3)	7 (0.5)	5.26 (1.95-14.15)	6.19 (2.12-18.03)
NSAIDs	9 (2.3)	29 (1.9)	1.16 (0.54-2.48)	1.16 (0.51-2.64)

*All the concurrent used drugs with DOACs were compared to use DOACs but without use these drugs.

#Adjusted for smoking, alcohol abuse, history of stroke or transient ischemic attack before the bleeding event, diabetes, hypertension, myocardial infarction, congestive heart failure, chronic renal disease, hepatic impairment, peripheral vascular disease, chronic pulmonary disease, peptic ulcer disease, cancer, comedications before the index date medications (β-adrenergic receptor blockers, ACEIs, non-PgP inhibitor statins, proton pump inhibitors, and cytochrome P450 enzyme inducers). For evaluating the association between pharmacodynamic interacting drugs and the major bleeding, co-medications with potential pharmacokinetic interactions were adjusted for. For evaluating the association between the combination use of pharmacokinetic interactions and DOAC and the major bleeding, drugs with potential pharmacodynamic interactions were adjusted for. ASA: Acetylsalicylic acid; CLOP: clopidogrel. PK: P-gP inhibitors or CYP3A4 inhibitors, PD: pharmacodynamic.
According to the policy of CPRD database, all the case less than 5 are shown as "<5".

Table S6. Major bleeding risk among patients taking apixaban, dabigatran, or rivaroxaban with the concomitant use of drugs with pharmacokinetic interactions.

	Apixaban				Dabigatran				Rivaroxaban			
Concurrent use of	**Cases (n=79), n (%)**	**Controls (n=366), n (%)**	**Crude OR (95% CI)**	**Adjusted OR* (95% CI)**	**Cases (n=79), n (%)**	**Controls (n=366), n (%)**	**Crude OR (95% CI)**	**Adjusted OR* (95% CI)**	**Cases (n=79), n (%)**	**Controls (n=366), n (%)**	**Crude OR (95% CI)**	**Adjusted OR* (95% CI)**
Drugs with PK interaction	24 (45.3)	137 (49.1)	0.86 (0.48-1.55)	0.80 (0.37-1.73)	38(48.1)	201 (54.9)	0.76 (0.47-1.24)	0.61 (0.33-1.71)	115 (44.1)	427 (50.3)	0.78 (0.60-1.03)	0.73 (0.52-1.02)
amiodarone	<5	10 (3.6)	-	-	<5	9 (2.5)	-	-	5 (1.9)	21 (2.5)	0.77 (0.29-2.06)	0.77 (0.27-2.19)
simvastatin	8 (15.1)	58 (20.8)	0.68 (0.30-1.52)	0.77 (0.31-1.92)	13 (16.5)	95 (26.0)	0.56 (0.30-1.07)	0.42 (0.21-0.87)	55 (21.1)	221 (26.0)	1.03 (0.69-1.54)	0.74 (0.51-1.07)
atorvastatin	9 (17.0)	52 (18.6)	0.89 (0.41-1.94)	0.61 (0.23-1.65)	14 (17.7)	66 (18.0)	0.98 (0.52-1.85)	1.05 (0.51-2.14)	225 (86.2)	735 (86.6)	1.10 (0.69-1.74)	1.10 (0.69-1.74)
verapamil	<5	<5	-	-	0	<5	-	-	<5	6 (0.7)	-	-
digoxin	9 (17.0)	34 (12.2)	1.47 (0.66-3.29)	1.72 (0.66-4.49)	11 (13.9)	56 (15.3)	0.90 (0.45-1.80)	0.88 (0.41-1.93)	34 (13.0)	102 (12.0)	1.10 (0.72-1.66)	1.09 (0.68-1.73)
diltiazem	<5	<5	-	-	<5	24 (6.6)	-	-	6 (2.3)	33 (3.9)	0.58 (0.24-1.40)	0.68 (0.25-1.84)

*All the concurrent used drugs with DOACs were compared to use DOACs but without use these drugs.

[#]Adjusted for smoking, alcohol abuse, history of stroke or transient ischemic attack before the bleeding event, diabetes, hypertension, myocardial infarction, congestive heart failure, chronic renal disease, hepatic impairment, peripheral vascular disease, chronic pulmonary disease, peptic ulcer disease, cancer, comedications before the index date medications (β-adrenergic receptor blockers, ACEIs, non-PgP inhibitor statins, proton pump inhibitors, and cytochrome P450 enzyme inducers). For evaluating the association between pharmacodynamic interacting drugs and the major bleeding, co-medications with potential pharmacokinetic interactions were adjusted for. For evaluating the association between the combination use of pharmacokinetic interactions and DOAC and the major bleeding, drugs with potential pharmacodynamic interactions were adjusted for. ASA: Acetylsalicylic acid; CLOP: clopidogrel. PK: P-gP inhibitors or CYP3A4 inhibitors .

According to the policy of CPRD database, all the case less than 5 are shown as “<5”.

CHAPTER 5

Dose adjustments or discontinuation of direct oral anticoagulants when combined with potential interacting drugs

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Manuscript ready for submission.

Abstract

Background: Direct oral anticoagulants (DOACs) have fixed dosing regimens, but dose adjustments may be needed when combined with potentially interacting drugs.

Aim: To evaluate whether needed and/or recommended dose adjustments or discontinuation of DOACs related to the combined use with interacting drugs are being followed in daily practice.

Methods: We performed a descriptive study using data from the UK Clinical Practice Research Datalink. We identified patients with a first prescription of a DOAC (apixaban, dabigatran, and rivaroxaban) between 2008 and 2015. The starters of DOACs were stratified for yes or no already using a pharmacokinetic and/or pharmacodynamic interacting drug (PKID and/or PDID). Furthermore, we studied starters of PKID and/or PDID during DOAC treatment. Dose adjustments of DOACs, discontinuation of DOACs and a switch to vitamin K antagonists (VKAs) were evaluated.

Results: A total of 23492 patients starting with a DOAC was identified. After excluding patients in which the prescribed daily dose (PDD) of the DOAC and/or creatinine clearance was not available 1400 patients starting with apixaban, 911 with apixaban and 2889 with rivaroxaban were available for further analyses. Between 38% to 63% of the patients were already using an PKID and/or PDID when DOAC treatment was started. It appeared that the DOAC starting dose was not reduced in these patients more often than in patients without interacting drugs at the start of DOAC treatment. When an interacting drug was started during DOAC treatment in only a small percentage of patients (<11%) the dose of DOACs was reduced, the DOAC discontinued or switched to a VKA.

Conclusion: Concurrent use of interacting drugs is high among DOACs users. It appears that the recommended dose adjustments of DOACs when combined with an interacting drug are largely ignored.

Introduction

Direct oral anticoagulants (DOACs), such as dabigatran, apixaban, rivaroxaban and edoxaban have recently entered the market and are rapidly becoming the most commonly prescribed oral anticoagulants for prevention or treatment of thromboembolic disorders including atrial fibrillation, acute myocardial infarction, deep venous thrombosis, pulmonary embolism, and other thrombotic diseases [1]. Compared with vitamin K antagonists, these drugs have a more predictable anticoagulant effect, and therefore are not subject to routine monitoring [2].

Despite the advantages of DOACs over VKAs, some prescribers may be unaware of potential drug-drug interactions and necessary dose adjustments based on renal function [3], because these agents are relatively new. Recent literature evaluating prescribing patterns of dabigatran, apixaban, and rivaroxaban in the US suggests that inappropriate prescribing of DOACs occurs frequently. The most common errors are under-dosing or over-dosing and incorrect administration (e.g. rivaroxaban ingested without meal) and are associated with increased risk of thromboembolic or bleeding events [3]. It has not been studied yet whether the recommended dose adjustments of DOACs when combined with an interacting drug are being followed.

Therefore, the present study was conducted to evaluate whether needed and/or recommended dose adjustments or discontinuation of DOACs related to the combined use with interacting drugs are being followed in daily practice.

Methods

Setting

Data for this study were obtained from the Clinical Practice Research Datalink (CPRD). CPRD is a longitudinal research database which includes more than 14 million patient records provided by general practitioners (GPs) throughout the United Kingdom [4]. Data recorded in CPRD include demographics, symptoms and diagnoses, prescriptions, results of diagnostic investigations, referrals to specialists and secondary care settings, feedback from other care settings and lifestyle, such as body mass index (BMI) and smoking status. Use of the CPRD as a reliable data

source has been well validated [5, 6]. The protocol of this study was reviewed and approved by the independent scientific advisory committee (ISAC) of CPRD (protocol number: 18_258R).

Study design and population

A descriptive cohort study was employed that included all patients with a first prescription for a DOAC between January 1, 2008 and December 31, 2015, who were 18 years or older, and had at least one year of history in CPRD prior to the date of this first prescription. We excluded those patients with missing information on renal function, prescribed daily dose and/or using drugs inducing CYP3A4 and/or P-glycoprotein (P-gp) inhibitors (this last group was excluded because of very low numbers).

Dose adjustments of DOACs at start of treatment

At the start of DOAC treatment patients were categorized into patients using a pharmacokinetic interacting drug (PKID) or a pharmacodynamic interacting drug (PDID) or both and a control group not using an interacting drug. In these subgroups we evaluated the starting prescribed daily dose (PDD). According to the SmPC of the three DOACs the starting doses were categorized into the recommended doses (See in the Supplement Table S1) in patients not using an interacting drug or the recommended reduced doses when an interacting drug (inhibitor of P-gp, inhibitor of CYP3A4, platelet inhibitor, NSAID, SSRI, SNRI) was being used at the start of DOAC treatment (see Table 1 for the recommendations and warnings). For both situations (yes or no using an interacting drug) dose recommendations based on kidney function and age were taking into account (see Table 1). We assessed the PDD separately for patients with AF or VTE since the dose schedules recommended in the SmPCs are different during the initial period [7-9].

Table 1. Recommendations for dose adjustments and contraindications for use of DOACs according to their SmPCs.

	Apixaban	Rivaroxaban	Dabigatran
Reduce dose	age ≥ 80 years; CrCl 15-29 ml/min	CrCl 15-29 ml/min	age ≥ 75 years CrCl 30~50 ml/min Concomitant use of amiodarone, quinidine, or verapamil
Contraindications			severe renal impairment (CrCL < 30 mL/min) **Strong P-gp inhibitors:** ketoconazole, itraconazole, cyclosporine, dronedarone
Use not recommended	**CrCL<15 ml/min** **Strong CYP3A4 and P-gp inhibitors:** ketoconazole, itraconazole, voriconazole, posaconazole, ritonavir, dronedarone*		**Strong P-gp inhibitors:** ritonavir, tacrolimus
Use with cautions	**Strong inducers of both CYP3A4 and P-gp:** phenytoin, carbamazepine, rifampicin		
	Moderate CYP3A4 and P-gp inhibitors: amiodarone, quinidine, verapamil, digoxin, diltiazem, simvastatin#, atorvastatin, fluconazole, clarithromycin, erythromycin, tacrolimus		**Moderate P-gp inhibitors:** posaconazole, digoxin, diltiazem, simvastatin#, atorvastatin, fluconazole, clarithromycin, erythromycin
	Drugs with pharmacodynamic interactions: aspirin, NSAIDs, clopidogrel, ticagrelor, prasugrel, SSRIs/SNRIs#		

*Not referred in the SmPC of apixaban.
#Interaction was found in clinical study but not listed in the SmPC.
NSAIDs, nonsteroidal anti-inflammatory drug; SSRIs, selective serotonin reuptake inhibitors; SNRIs, selective noradrenaline reuptake inhibitors.

Dose adjustments or discontinuation of DOACs during treatment

During DOAC treatment we evaluated when an interacting drug was started whether the prescribed dose was adjusted, DOAC treatment was discontinued without a switch to another anticoagulant or DOAC treatment was discontinued with a switch to another anticoagulant (another DOAC or coumarin). The time span in which these changes were evaluated were from 30 days prior to the start of the interacting drug until 30 days after the start. The dose adjustments were

categorized as an increase or decrease of the PDD during the presented time span compared to the prescription prior to the dose adjusted prescription.

Treatment episodes of DOAC use

In our cohort of DOAC starters patients might stop using a DOAC without restart of the same or another anticoagulant, might discontinue with a restart of another anticoagulant and/or might be more or less nonadherent. Therefore, in order to be able to select from our cohort of DOAC starters those patients that started an interacting drug during DOAC treatment we had to construct treatment episodes of DOAC use.

Treatment episodes of DOACs were constructed allowing for a 30-days permissible gap between the theoretical end date of a prescription and the subsequent prescription. When the gap was more than 30 days, we assumed patients had discontinued their DOAC treatment. Treatment episodes were defined as a series of subsequent prescriptions for DOAC, independent of dose changes and constructed according to the method used in our previous study [10]. In case a subsequent prescription for the same drug is collected before the theoretical end date of a previous prescription, the number of overlapping days is added to the theoretical end date of the subsequent prescription. The number of overlapping days was maximized at 90 days. If a subsequent prescription within the same treatment episode was for another type of DOAC, the patient was considered to have switched therapy and the remaining tablet days from the prior prescription were disregarded. Duration of individual DOAC prescriptions were assessed by using information on the prescribed number of tablets and the dosage instruction. When such information was not available, the median time between prescriptions was used. When only 1-3 prescriptions were available for an individual patient the duration was based on the most frequently occurring estimated prescription duration for the drug in the study population.

Other reasons for dose changes of a DOAC

Factors that need dose adjustment or discontinuation of DOACs except concurrent use of potentially interacting drugs were collected as covariates: indications, renal

function, liver impairment, age and major bleeding events. Read codes or laboratory values were used to identify these covariates.

Data analysis

Patients with reduced or increased doses, discontinuations or switch to another anticoagulant within a subgroup of patients with or without an interacting drug are presented as percentages of the total group of patients within that subgroup. The Chi-square test was used to compare the percentages of treatment changes of DOACs between patients with and without the use of an interacting drug. The analyses were stratified for age (cut of 75 years) and kidney function (creatinine clearance (CrCL) <30 ml/min, 30-50 ml/min and >50 ml/min). This stratification was not done for the analyses in which interacting drugs were started during DOAC treatment. A *P*-value <0.05 was considered as statistically significant. All the data analyses were carried out using IBM SPSS Statistics, version 24 (IBM Corp., Armonk, N.Y. USA).

Results

After the selection procedure as presented in Figure 1, in total we identified 1400, 911 and 2889 new users of apixaban, dabigatran, and rivaroxaban, respectively. Baseline characteristics of the study population are summarized in Table 2. The mean age of the DOAC users was approximately 74. The distribution of the characteristics was similar in the three groups.

The proportions of patients with a reduced initial PDD of DOACs in patients with or without the use of potentially interacting drugs which started before DOAC treatment are summarized in Table 3. For atrial fibrillation only in the case of dabigatran there was a statistically significant higher reduced starting dose (expressed as PDD) of 56.5% when at the start of dabigatran patients already were using a PKID and PDID compared to 46.8% among patients without the use of interacting drugs at the start of dabigatran. Furthermore, for atrial fibrillation in the case of apixaban the PDD at the start of therapy was less reduced when patients were using a PKID compared to patients not using an interacting drug (25.5% versus 33.9%, p=0.02). For the other comparisons the percentages of reduced PDDs were comparable with or without the use of interacting drugs at the start of

DOAC treatment. For VTE only for rivaroxaban the numbers of patients were enough to evaluate the percentages of reduced PDD in patients yes or not using interacting drugs at the start of rivaroxaban. These percentages did not show significant differences.

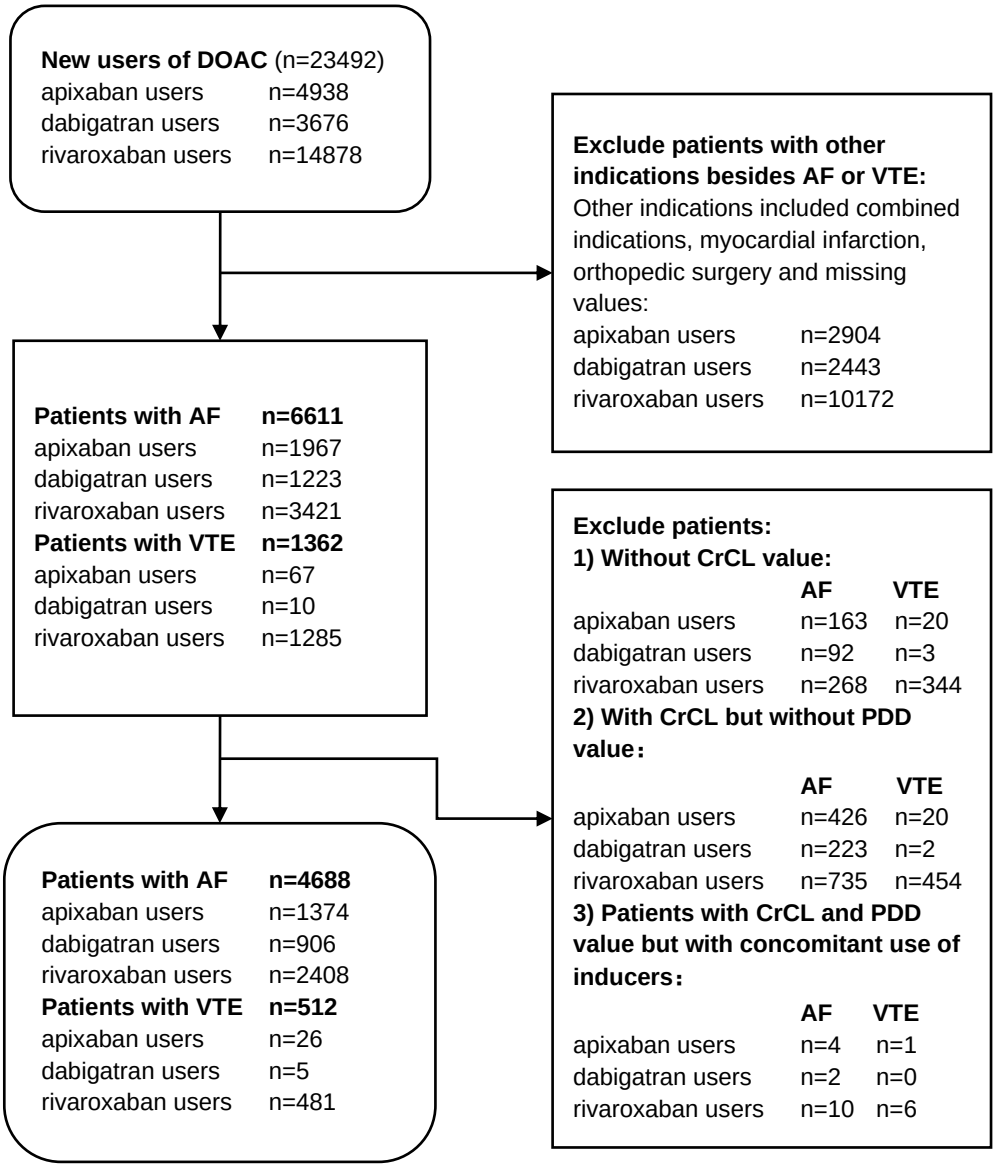

Figure 1. Flow chart of patient selection

Table 2. Baseline characteristics of starters of apixaban, dabigatran, and rivaroxaban

	Apixaban	Dabigatran	Rivaroxaban
	n=1400	n=911	n=2889
Age, yr, mean (SD)	74.9 (10.7)	73.5 (11.0)	73.72 (12.2)
<75, n (%)	619 (44.2)	461 (50.6)	1345 (46.6)
≥75, n (%)	781 (55.8)	450 (49.4)	1544 (53.4)
Sex, male (%)	765 (54.6)	571 (62.7)	1527 (52.9)
Indications, n (%)			
AF only	1374 (98.1)	906 (99.5)	2408 (83.4)
VTE only	26 (1.9)	5 (0.5)	481 (16.6)
Creatinine clearance, n (%)			
<15ml//min	<5	<5	<5
15-29 ml/min	28 (2.0)	7 (0.8)	44 (1.5)
30-49ml/min	369 (26.4)	220 (24.1)	776 (26.9)
50-79ml/min	743 (53.1)	490 (53.8)	1559 (54.0)
≥80ml/min	259 (18.5)	193 (21.2)	507 (17.5)
Comedication use at baseline, n (%)			
β-adrenergic receptor blockers	620 (44.3)	448 (49.2)	1147 (39.7)
ACEI/ARB	709 (50.6)	418 (45.9)	1344 (46.5)
Diuretics	505 (36.1)	336 (36.9)	951 (32.9)
Calcium channel blockers	441 (31.5)	262 (28.8)	933 (32.3)
Statins	62 (4.4)	32 (3.5)	101 (3.5)
Proton pump inhibitors	545 (38.9)	356 (39.2)	1188 (41.1)
Comorbidities, n (%)			
Chronic heart failure	164 (11.7)	116 (12.6)	274 (9.5)
Diabetes	291 (20.8)	178 (19.5)	547 (18.9)
Hypertension	884 (63.1)	559 (61.4)	1787 (61.9)
COPD	141 (10.1)	82 (9.0)	302 (10.5)

AF: atrial fibrillation; VTE: venous thromboembolic events; ACEI: angiotensin-converting-enzyme inhibitor; ARB: angiotensin receptor blocker.
According to the policy of CPRD database, all the case less than 5 (except 0) are shown as “<5”.

Table 3. Percentages of reduced prescribed daily dose at the start of apixaban, dabigatran and rivaroxaban treatment among patients with atrial fibrillation or venous thromboembolic events already using a potential interacting drug.

Concomitant with	Apixaban			Dabigatran			Rivaroxaban		
	n	Reduced PDD n (%)	*P*-value	n	Reduced PDD n (%)	*P*-value	n	Reduced PDD n (%)	*P*-value
AF	1374			906			2408		
Control	504	171 (33.9)		344	161 (46.8)		944	163 (17.3)	
PKID	286	73 (25.5)	0.01	200	95 (47.5)	0.88	507	96 (18.9)	0.43
PDID	187	54 (28.9)	0.21	114	57 (50.9)	0.55	283	47 (16.6)	0.80
PKID+PDID	397	135 (33.8)	0.96	248	140 (56.5)	0.02	674	132 (19.6)	0.23
VTE	26			5			481		
Control	16	5 (31.3)		<5	-		248	15 (6.0)	
PKID	<5	-	-	<5	-	-	104	10 (9.6)	0.24
PDID	<5	0	-	0	0	-	51	<5	-
PKID+PDID	<5	-	-	0	0	-	78	6 (7.7)	0.61

PKID: Pharmacokinetic interacting drugs - P-gP inhibitors or CYP3A4 inhibitors; PDID: Pharmacodynamic interacting drugs; PDD: prescribed daily dose; AF: atrial fibrillation; VTE: venous thromboembolism

According to the policy of CPRD database, all the case less than 5 (except 0) are shown as "<5".

When the percentages reduced PDDs were stratified for age and renal function the general picture was that when the patients were older and the kidney function was lower more patients had a reduced PDD at the start of DOAC treatment (Table 4). The statistically significant higher reduced PDDs were only observed in the patients above 75 years of age and CrCL lower than 30 ml/min although the numbers in this subgroup were very low.

Table 4. Percentages of reduced prescribed daily dose at the start of apixaban, dabigatran and rivaroxaban treatment among patients with atrial fibrillation already using a potential interacting drug stratified by age and CrCL.

	Apixaban			Dabigatran			Rivaroxaban		
Concomitant use	n	Reduced PDD n (%)	*P*-value	n	Reduced PDD n (%)	*P*-value	n	Reduced PDD n (%)	*P*-value
Age<75									
Normal function to mildly reduced (CrCl >50 ml/min)									
Control	198	25 (12.6)		173	38 (22.0)		368	22 (6.0)	
PKID	114	5 (4.4)	0.02	98	17 (17.3)	0.36	190	5 (2.6)	0.08
PDID	62	<5	-	47	7 (14.9)	0.29	93	<5	-
PKID +PDID	135	10 (7.4)		86	11 (12.8)	0.08	213	9 (4.2)	0.37
Moderately reduced (CrCl 30~50 ml/min)									
Control	26	<5	-	20	8 (40.0)		69	15 (21.7)	
PKID	23	<5	-	16	7 (43.8)	0.82	31	7 (22.6)	0.93
PDID	12	<5	-	8	<5	-	13	<5	-
PKID +PDID	33	<5	-	11	<5	-	48	11 (22.9)	0.88
Severely reduced and renal failure (CrCl <30 ml/min)									
Control	0	-	-	0	-	-	0	0	-
PKID	<5	<5	-	0	-	-	<5	<5	-
PDID	0	-	-	0	-	-	<5	<5	-
PKID +PDID	<5	<5	-	0	-	-	<5	<5	-
Age≥75									
Normal function to mildly reduced (CrCl >50 ml/min)									
Control	187	85 (45.5)		99	74 (74.7)	-	307	45 (14.7)	-
PKID	83	35 (42.2)	0.62	48	40 (83.3)	0.24	179	29 (16.2)	0.65
PDID	71	24 (33.8)	0.09	39	30 (76.9)	0.79	112	15 (13.4)	0.74
PKID +PDID	133	58 (43.6)	0.74	89	75 (84.3)	0.11	235	31 (13.2)	0.63
Moderately reduced (CrCl 30~50 ml/min)									
Control	85	51 (60.0)		51	41 (80.4)		186	74 (39.8)	
PKID	59	24 (40.7)	0.02	34	27 (79.4)	0.91	96	45 (46.9)	0.25
PDID	39	20 (51.3)	0.36	19	17 (89.5)	0.37	61	25 (41.0)	0.87
PKID +PDID	86	52 (60.5)	0.95	60	50 (83.3)	0.69	166	72 (43.4)	0.50
Severely reduced and renal failure (CrCl <30 ml/min)									
Control	8	6 (75.0)		<5	0	-	14	7 (50.0)	
PKID	6	6 (100.0)	0.19	<5	<5	0.03	8	8 (100.0)	0.02
PDID	<5	<5	-	<5	<5	-	<5	<5	-
PKID +PDID	9	9 (100.0)	0.11	<5	<5	0.08	11	8 (72.7)	0.25

PKID: Pharmacokinetic interacting drugs - P-gP inhibitors or CYP3A4 inhibitors; PDID: Pharmacodynamic interacting drugs; PDD: prescribed daily dose; CrCL: creatinine clearance.
According to the policy of CPRD database, all the case less than 5 (except 0) are shown as "<5".

Table 5. The number and percentages of patients* with atrial fibrillation that have dose adjustments, discontinue the use of a DOAC or switch to warfarin in relation to the start of a potential interacting drug.

	Prescribed n=1838	Adjustment n (%)	DOACs dose increase n (%)	DOACs dose decrease n (%)	Discontinuation n (%)	Switch to VKA n (%)
PKID (at least 1)	1154	122 (10.6)	19 (1.6)	18 (1.6)	83 (7.23)	33 (2.9)
PDID (at least 1)	370	31 (8.4)	<5 (1.1)	<5 (0.8)	28 (7.6)	<5
PKID+PDID	314	20 (6.4)	10 (3.2)	7 (2.2)	17 (9.0)	<5

* Presented are all patients with a start of an interacting drug during DOAC treatment. Patients that already used an interacting drug at the start of DOAC treatment and did not discontinue this drug during DOAC treatment were excluded.
PKID: Pharmacokinetic interacting drugs - P-gP inhibitors or CYP3A4 inhibitors; PDID: Pharmacodynamic interacting drugs.
According to the policy of CPRD database, all the case less than 5 (except 0) are shown as "<5".

In Table 5 it can be seen that only in a small percentage (<11%) of patients the DOAC treatment is changed when an interacting drug is started during DOAC treatment.

Discussion

The results of this population-based cohort study among new users of DOACs showed that the proportion of patients that have a dose adjustment when at the start of the DOAC treatment one or more interacting drugs were being used is low and similar to dose adjustments observed when no interacting drugs were being used at the start of DOAC treatment. Furthermore, when during DOAC treatment an interacting drug was started also in only a small percentage of patients (<11%) the dose of the DOAC was adjusted or the DOAC was discontinued. Irrespective of the combined use of interacting drugs the lowest DOAC doses were observed in patients above 75 years of age with severely reduced and renal failure (CrCL <30 ml/min).

This is the first study to systematically evaluate dose adjustments of DOACs when combined with interacting drugs that potentially increase bleeding risk. An important finding was that between 38% and 63% of patients already use a potentially interacting drug at the start of DOAC treatment. We found that there was a higher

percentage of reduced doses in patients with atrial fibrillation already using PKID and PDID at the start of dabigatran compared to no use of interacting drugs (56.5% versus 46.8%; *P*=0.02), however this might be a chance finding. After all, we performed multiple comparisons, this finding was not observed for the other DOACs and in the atrial fibrillation patients using apixaban the percentage of reduced doses was lower when PKIDs were already being used at the start of apixaban treatment compared to patients not using interacting drugs at the start of apixaban treatment (25.5% versus 33.9%; *P*=0.01). It is reassuring that in the elderly with reduced kidney function prescribers appear to be cautious to reduce the standard dose of DOACs [11].

An important question is whether the lack of reducing the dose of DOACs when combined with interacting drugs potentially increasing the bleeding risk has clinical consequences. In a previous study (submitted and presented in this thesis in Chapter 4) we showed that the risk of major bleeding is increased twice when DOACs are combined with platelet inhibitors and/or SSRIs and that there was no increased risk when DOACs were combined with drugs inhibiting CYP3A4 and/or P-gp inhibitors. Although in theory reduction of the dose of a DOAC when combined with a platelet inhibitor in situations of an acute coronary syndrome in patients with atrial fibrillation will reduce the increased major bleeding risk it should be balanced against less effectiveness for a recurrent acute coronary syndrome or the prevention of ischaemic stroke and systemic embolism [12]. Although there was no increased major bleeding risk when DOACs prescribed in standard doses were combined with PKIDs this does not mean that there is also no increased risk for minor bleeds [13-15].

The strength of our study is that it is population based and presents the prescribing behavior in daily practice in the United Kingdom. It gives an impression of the way prescribers deal with interaction problems, renal function and age. A limitation was that for many patients there was no information available on renal function and PDD of the DOACs. As the patients excluded for the latter reasons were comparable with included patients for baseline characteristics (suggesting a random selection of excluded patients) we expect that the analysis of only a part of the total population will not have a substantial influence on our findings.

In conclusion, our study demonstrated that prescribers in the United Kingdom do not more often adjust doses of or discontinue DOACs when combined with a PKID and/or PDID. More research is needed to evaluate the clinical consequences of the lack of dose adjustments.

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Supplement

Table S1. Dose recommendation of DOACs based on SmPC.

		Total daily dose		
Indications		**apixaban**	**dabigatran**	**rivaroxaban**
AF		10 mg	300 mg	20 mg
VTE	Treatment DVT or PE	20 mg for the first 7 days; followed by 10 mg	220 mg	30 mg (Day 1-21) 20 mg (Day 22 onwards)
	Prevention of recurrent DVT or PE	5 mg		10mg or 20 mg

AF: atrial fibrillation; VTE: venous thromboembolism; DVT: deep venous thrombosis; PE: pulmonary embolism.

CHAPTER 6

Cost-effectiveness of genotype-guided dosing versus alternative anticoagulation in atrial fibrillation in the Netherlands

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Abstract

Objective: To evaluate the cost-effectiveness of direct oral anticoagulants (DOACs) and a clinical and genotype-guided dosing algorithm for phenprocoumon and acenocoumarol in patients with atrial fibrillation in the Netherlands.

Methods: A decision-analytic Markov model was used to estimate the cost-effectiveness of genotype-guided dosing and clinical dosing versus the standard dosing practice, and was compared with apixaban, dabigatran, and rivaroxaban.

Results: For phenprocoumon, genotype-guided and clinical algorithm guided-dosing increased the quality-adjusted life-years (QALYs) by 0.005 and 0.003 respectively compared to the standard care in the Netherlands. The incremental cost-effectiveness ratios (ICERs) were €12777, and €10998 per QALY gained. The use of apixaban, dabigatran and rivaroxaban increased health by 0.377, 0.366, and 0.139 QALYs compared with the Dutch standard care. The ICERs were €14241, €15918, and €42140 per QALY gained. Apixaban had the highest chance (30 %) of being cost effective at a threshold of €20,000. Compared with the Dutch standard care of acenocoumarol, genotype guided dosing and the use of apixaban, dabigatran and rivaroxaban got an ICER of €8956, €14241, €15918, and €42140 per QALY gained, respectively.

Conclusions: Apixaban could be the most cost-effective alternative in the Netherlands. The pharmacogenetic dosing algorithm for phenprocoumon was not likely to be cost effective compared with the standard care in the Netherlands, at a willingness-to-pay threshold of €20,000 per QALY.

Introduction

The risk of stroke and other thromboembolic events are increased among patients with atrial fibrillation (AF). Long term anticoagulation is an effective treatment for stroke prevention in AF patients [1]. Vitamin K antagonists (VKAs) have been used for decades as oral anticoagulants for the prevention of stroke and systemic embolism in AF. Because of their narrow therapeutic window and large inter-and intra-individual variability in dose-response among users, routine monitoring is required to manage the intensity of anticoagulation, which is measured as an international normalized ratio (INR) [2].

A number of approaches have been proposed for increasing the effectiveness and safety of VKAs, including the usage of genetic information into a dosing algorithm for instance. Since polymorphisms in the *VKORC1* gene and *CYP2C9* gene together account for approximately one third of the variability in dose requirement, several dosing algorithms have been developed that include the information of these two genotypes and patient characteristics such as age, gender, height and weight [3, 4]. Three large randomized controlled trials examined the effect of using these algorithms. One of these trials included acenocoumarol and phenprocoumon users from the Netherlands [5]. In this trial, a dosing algorithm based on age, sex, height, weight and *VKORC1* and *CYP2C9* genotype was compared to an algorithm with the same patient characteristics but without including the genotype information. The genotype-guided dosing for phenprocoumon improved the time in therapeutic range (TTR) in the first 4 weeks compared with the clinical dosing algorithm [6], however, there was no significant difference between the groups after 12 weeks.

Recently, direct oral anticoagulants (DOACs) dabigatran, rivaroxaban, and apixaban have become available for the prevention of stroke and systemic embolism in patients with AF. These drugs have shown to be non-inferior or even superior to warfarin in randomized controlled trails [7-9]. The DOACs also seem to be cost effective compared to the standard care with VKAs [10, 11]. However, it is not clear whether it is still cost effective when dosing algorithms are used to improve the quality of anticoagulation control. The aim of this study is therefore to investigate the cost-effectiveness of DOACs and a variety of clinical and genotype-guided

dosing algorithms for acenocoumarol and phenprocoumon compared with standard care in Dutch patients with atrial fibrillation.

Methods

Model structure

A previously published decision-analytic Markov model (Figure 1) was adapted to evaluate the cost effectiveness of the alternative anticoagulation treatment strategies in patients with AF [12, 13]. The model was developed using Microsoft Excel 2013. For the base case analysis, patients were assumed to be a cohort of Dutch patients with AF, aged 70 years and were treated with VKAs per Dutch guideline (standard care). Using this model, we examined the cost-effectiveness of the treatment with dabigatran (150 mg twice daily), rivaroxaban (20 mg), apixaban (5 mg twice daily), with the standard care in the Netherlands. We also evaluated the cost-effectiveness of the use of a genotype-guided algorithm and the clinical algorithm dosed phenprocoumon and acenocoumarol.

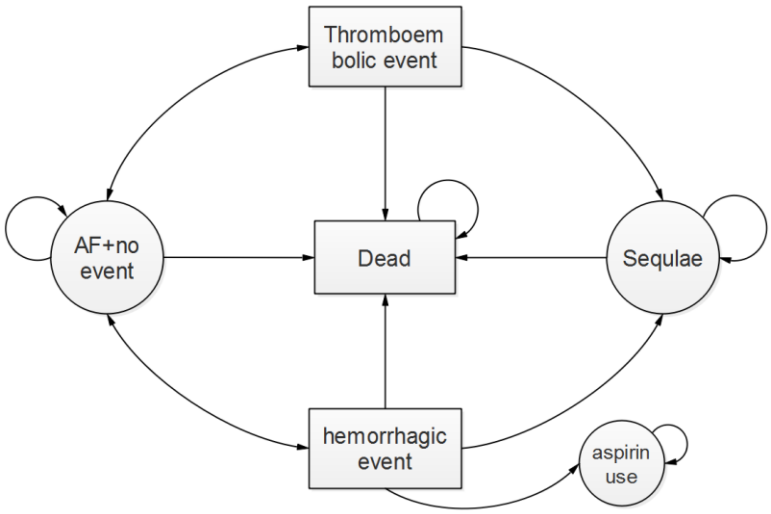

Figure 1. Markov model health states.

Patients entered the model in the “AF (atrial fibrillation) +no event” state and can move to other states at monthly intervals. The thromboembolic event includes ischaemic stroke, systemic embolism, transient ischaemic attack, myocardial infarction; the hemorrhagic thromboembolic event includes intracranial hemorrhage, extracranial hemorrhage.

In the decision-analytic Markov model, patient health states were defined as: no event, ischaemic stroke (IS), systemic embolism (SE), transient ischaemic attack (TIA), myocardial infarction (MI), intracranial hemorrhage (ICH), extracranial hemorrhage (ECH), sequelae after IS or ICH and death. All patients entered the model in the no event state and could move to one of the other states at monthly intervals. Patient would stay in a state for 1 month and then move to no event, sequelae or death after an event occurred. Patients who recovered from an event went back to the no event and could have a recurrent event. All input parameters were equal for both treatment strategies after stroke or ICH went to the sequelae state, except the time spent in INR range and the costs of genotyping.

The input parameters of the model are shown in Table 1. About 28% of the thromboembolic events were assumed to be transient ischaemic attacks (TIA) [14, 15]. Patients with ischaemic stroke had a 15% chance of dying and 40% chance of disability [8, 9]. Patients with TIA were assumed to be recovered. The probability that an ICH would result in sequelae was 50% [16]and the chance that it would be fatal was 45% [17]. Patients after an ICH were assumed to be switched to aspirin [18]. The chance that MI and ECH would be fatal is 16% and 7%, respectively [19, 20]. The chance of dying among patients with sequelae is 5.6% [21]. For all patients, the age-specific mortality rates [22] were included in the model. These rates can be found in supplement Table S1.

Model outcomes were presented in monthly cycles and included quality-adjusted life-years (QALYs) and costs in euros (€). Incremental cost-effectiveness ratios (ICER) were calculated. The effects and costs were discounted at an annual rate of 1.5% and 4% respectively as recommended in the national guidelines in the Netherlands [23].

Clinical input

The percentage of time in different INR ranges (<2.0, 2.0–3.5, 3.5–5.0 and >5.0) were used to determine the probability of an event for phenprocoumon treated patients and were shown in the supplement Table S2. During the first 3 months of the treatment according to standard practice in the Netherlands, the time spent in these ranges was calculated by using data from the pre-EU-PACT study [3, 24].

The EU-PACT trial [5] data was used to determine the percentage of time in different INR ranges in the first 3 months of treatment with phenprocoumon in patients dosed by the genotype-guided algorithm and the clinical algorithm. In the EU-PACT trial, the therapeutic INR range is 2.0-3.0 while according to standard practice in the Netherlands, the therapeutic range is 2.0-3.5. Therefore, in this study, we reanalyzed the data to get the percentage of time in four ranges (<2.0, 2.0–3.5, 3.5–5.0 and >5.0). We used data from the Dutch Federation of Thrombosis Services [25] to estimate the average percentage time spent in the four different ranges after the first 3 months and assumed that this percentage is the same in all phenprocoumon treated patients.

The specific event rates of clinical events of coumarins were calculated by multiplying the risk of an event at a specific INR range by the proportion of the event and by the percentage of time spent at the INR range. The incidence of the thromboembolism and hemorrhagic events at different levels of INR were obtained from the meta-analysis by Oake *etal* [26] and were shown in Table 1. In this study, the investigated INR range was 2.0-3.0. We assumed that the risk of these events associated with an INR in these therapeutic ranges was similar to the risk with INR range 2.0-3.5 which is used in the Netherlands.

The annualized clinical event rates for apixaban, dabigatran, and rivaroxaban were summarized in Table 2. These rates were obtained from the ARISTOTLE [8], RE-LY [7, 27], and ROCKET-AF [9] trial. An indirect comparison method was used to adjust for the differences in baseline risks between the three trials [28, 29].

For the standard care of phenprocoumon therapy, the frequency of INR measurements is assumed to be 4 in the first month based on the pre-EU-PACT study [3, 30]. For the genotype-guided dosed and clinical algorithm dosed patients, we assume the frequency of INR measurements to be 6 in the first month and 2.5 per month in the second and third months based on the EU-PACT trial [5]. In the Netherlands, the frequency of INR measurements was approximately 21 per year [17], therefore, we assume the 1.75 times INR measurements per month after the first month for the standard care group and after the first 3 months for the algorithm dosed group.

Utilities and costs

Utility values of health states (Table 2) were based on the preference-based EQ-5D index scores reported by US [31, 32] and the cost-effectiveness studies in UK [33]. A decrement in quality of life of 0.013 was used for phenprocoumon use and 0.002 for aspirin use [34].

Costs of drugs or other interventions are shown in Table 2. Monthly drug costs were derived from the Dutch healthcare insurance board [35]. The cost of a point-of -care genotyping test was estimated to be approximately €40 [25].

Sensitivity analyses

One-way sensitivity analyses were performed to measure the influence of input parameters on the economic results. The parameters were varied over 95% confidence intervals or decreased and increased by 20% if the value of the confidence interval was not available (Table 1 and Table 2). The cost of genotyping was varied by ±50%.

A probabilistic sensitivity analysis was performed through 10000 Monte-Carlo simulations to evaluate the combined impact of multiple model parameters on the estimated cost effectiveness of the anticoagulation alternatives. Dirichlet distributions were used to vary the probabilities of different outcomes of stroke and ICH (more than two possible results). Beta distributions were used for all other probabilities and QALYs, and gamma distributions for the costs. A normal distribution was used to vary the frequency of INR measurements, the age of the patients and the percentage time spent in the therapeutic INR range. Probabilities of cost-effectiveness are presented through multiple cost-effectiveness acceptability curves which were plotted to depict the incremental costs and effects of every simulation.

Table 1. Clinical input parameters used in the model.

Input parameter	Base case	Range	Distribution	Reference
Risk of Ischemic stroke, yearly, %				
INR<2	2.68	1.36-5.28	Beta	[26]
INR within range	0.57	0.26-1.19	Beta	[26]
INR 3.0-5.0	0.88	0.26-2.77	Beta	[26]
INR>5	1.98	1.14-3.48	Beta	[26]
Aspirin	2.16	1.44-3.17	Beta	[18]
Apixaban	0.70 [a]	0.56-0.85	Beta	[8]
Rivaroxaban	0.71 [a,b]	0.52-0.96	Beta	[8, 9]
Dabigatran	0.57 [a,b]	0.41-0.79	Beta	[7, 8]
Risk of TIA, yearly, %				
INR<2	1.04	0.53-2.04	Beta	[26]
INR within range	0.22	0.10-0.46	Beta	[26]
INR 3.0-5.0	0.34	0.10-1.07	Beta	[26]
INR>5	0.77	0.44-1.34	Beta	[26]
Aspirin	0.84	0.56-1.23	Beta	[18]
Apixaban	0.27 [a]	0.22-0.33	Beta	[8]
Rivaroxaban	0.27 [a,b]	0.20-0.37	Beta	[8, 9]
Dabigatran	0.22 [a,b]	0.16-0.31	Beta	[7, 8]
Risk of Systemic embolism, yearly, %				
INR<2	0.37	0.19-0.72	Beta	[26]
INR within range	0.08	0.04-0.16	Beta	[26]
INR 3.0-5.0	0.12	0.04-0.38	Beta	[26]
INR>5	0.27	0.16-0.47	Beta	[26]
Aspirin	0.40	0.15-3.33	Beta	[18]
Apixaban	0.09	0.04-0.18	Beta	[8]
Rivaroxaban	0.03 [b]	0.01-0.09	Beta	[8, 9]
Dabigatran	0.08 [b]	0.03-0.22	Beta	[7, 8]
Risk of Myocardial infraction, yearly, %				
INR<2	2.01	1.02-3.96	Beta	[26]
INR within range	0.43	0.20-0.89	Beta	[26]
INR 3.0-5.0	0.66	0.20-2.08	Beta	[26]
INR>5	1.49	0.86-2.61	Beta	[26]
Aspirin	0.90	0.54-1.60 [37]	Beta	[18]
Apixaban	0.53	0.40-0.71	Beta	[8]
Rivaroxaban	0.49 [b]	0.33-0.72	Beta	[8, 9]
Dabigatran	0.76 [b]	0.51-1.16	Beta	[7, 8]
Risk of ICH, yearly, %				
INR<2	0.09	0.07-0.12	Beta	[26]
INR within range	0.30	0.12-0.78	Beta	[26]
INR 3.0-5.0	1.40	0.39-5.18	Beta	[26]
INR>5	5.91	2.23-15.62	Beta	[26]

Input parameter	Base case	Range	Distribution	Reference
Risk of ICH, yearly, %				
Aspirin	0.40	0.21-1.05	Beta	[18]
Apixaban	0.33	0.24-0.46	Beta	[8]
Rivaroxaban	0.53 [b]	0.33-0.85	Beta	[8, 9]
Dabigatran	0.32 [b]	0.19-0.53	Beta	[7, 8]
Risk of ECH, yearly, %				
INR<2	0.31	0.23-0.39	Beta	[26]
INR within range	1.00	0.39-2.62	Beta	[26]
INR 3.0-5.0	4.70	1.31-17.33	Beta	[26]
INR>5	19.79	7.47-52.28	Beta	[26]
Aspirin	0.90	0.54-1.49	Beta	[18]
Apixaban	1.79	1.54-2.11	Beta	[8]
Rivaroxaban	2.60[b]	2.12-3.19	Beta	[8, 9]
Dabigatran	2.42[b]	1.95-3.01	Beta	[7, 8]
Outcomes of events (if occurs), %				
Fatal stroke	15	11.2-18.9	Dirichlet	[8, 9]
Disabling stroke	40	36.3-43.7	Dirichlet	[8, 9]
Fatal transient ischemic attack	0	-	-	-
Fatal systemic embolism	7	5.6-8.4	Beta	assumption
Fatal myocardial infraction	16	13-19	Beta	[19, 20]
Fatal extracranial hemorrhage	7	5.6-8.4	Beta	assumption
Fatal intracranial hemorrhage	45	36.0-48.5	Dirichlet	[17]
Disabling intracranial hemorrhage	50	46.4-53.6	Dirichlet	[16]
Transient ischemic attack	28	25-31	Beta	[14, 15]
Death in case of sequelae, monthy	5.6	4.5-6.7	Beta	[21]
INR measureements				
First month (algorithm)	6	4.27-7.73	Normal	[5]
First month (standard dosed)	4	2-6	Normal	[30]
Months 2 and 3, per month	2.5	1.15-4.85	Normal	[5]
Consecutive months, per month	1.75	1.31-2.12	Normal	[17]
Age				
Age at start of treatment, years	70	50-90	Normal	[3, 5]

[a]28% of ischaemic strokes were assumed to be TIA

[b]Adjusted event rates for rivaroxaban and dabigatran were calculated by multiplying the hazard ratios, by the event rates of apixaban in the ARISTOTLE trial

Table 2. Utilities and costs.

Parameter	Base case	Range	Distribution	Reference
Utilities				
Atrial fibrillation	0.81	0.7784 to 0.8430	Beta	[32]
VKA use	-0.013	-0.002 to -0.0033	Beta	[21, 34]
Use of DOAC	-0.006	-0.004 to -0.007	Beta	[15]
Aspirin use (after ICH)	-0.002	0.000 to -0.006	Beta	[15, 34]
Myocardial infraction	-0.1247	-0.1065 to -0.1436	Beta	[32]
Systemic embolism	-0.1199	-0.1022 to -0.1388	Beta	[32]
Extracranial hemorrhage	-0.06	-0.02 to -0.1	Beta	[21]
Intracranial hemorrhage	-0.1814	-0.1550 to -0.2089	Beta	[32]
Transient ischemic attack	-0.1032	-0.0991 to -0.1189	Beta	[32]
Stroke	-0.1385	-0.1184 to -0.1560	Beta	[32]
Sequelae	-0.374	-0.160 to -0.588	Beta	[32]
Costs (€)				
Phenprocoumon per month	2.17	1.74 to 2.60	Gamma	[35]
Apixaban, 5mg bid., per month	73.14	58.51 to 87.77	Gamma	[35]
Dabigatran, 150mg bid., per month	73.14	58.51 to 87.77	Gamma	[35]
Rivaroxaban, 20mg daily, per month	68.69	54.95 to 82.43	Gamma	[35]
Aspirin tablets per month	3.12	2.5 to 3.74	Gamma	[35]
Genotyping	40	20 to 60	Gamma	[35]
INR measurement and visit to anticoagulant clinic	12.07	9.66 to 14.48	Gamma	[13, 40]
ECH	13690	10952 to 16428	Gamma	[40]
ICH	25047	20037 to 30057	Gamma	[40]
TIA	987	790 to 1184	Gamma	[41]
Stroke	18075	14460 to 21690	Gamma	[42, 43]
Sequelae, first month	9254	7403 to 11105	Gamma	[6, 13]
Sequelae, subsequent months	480	384 to 576	Gamma	[13, 30]
Discount rate (yearly, %)				
Costs	4	0-8	-	[23]
Effects	1.5	0-3	-	[23]

Results

Base case

The results of the cost-effectiveness analyses for patients treated with phenprocoumon and DOACs were reported in Table 3. Compared with the standard care, using genotype-guided dosing or clinical algorithm-dosed phenprocoumon increased the QALYs by 0.005 and 0.003 respectively. The ICER of genotype-guided versus the standard treatment with phenprocoumon was € 12777 per QALY gained.

Apixaban, dabigatran, and rivaroxaban could extend the QALYs by 0.361, 0.350, and 0.140, respectively, as compared with the standard dosed phenprocoumon. Treatment with apixaban, dabigatran got an ICER of € 14241 and € 15918 per QALY gained respectively. Treatment with rivaroxaban resulted in lower increments of QALY compared with apixaban and dabigatran, while the costs were higher, thus resulted in an ICER of €42140 per QALY gained.

Sensitivity analyses

The results of probabilistic sensitivity analysis were shown in Figure 2. DOACs and genotype-guided dosing were more effective than the standard care of phenprocoumon however cost more. The incremental costs per QALY gained were below €20,000 in 63% of the simulations for genotype-guided dosing and in 55% in the simulations for clinical algorithm group. The incremental costs per QALY gained were below €20,000 in 54%, 50%, and 28% respectively for apixaban, dabigatran, and rivaroxaban.

The probability of these anticoagulation alternatives would be the most cost-effective in the Netherlands over a range of likely willingness-to-pay thresholds as depicted in Figure 3. Apixaban had the highest probability (30%) of being cost effective at a willingness-to-pay threshold of €20,000 per QALY gained or higher. The probability that dabigatran and rivaroxaban were cost effective options was 24%, and 2%, respectively. Genotype-guided doing for phenprocoumon was cost effective with a probability of 13% which is higher than the clinical algorithm dosed (9%), however, lower than the standard care (21%).

Table 3. Quality-adjusted life-years, total costs, and incremental cost-effectiveness ratios for patients with atrial fibrillation.

Treatment	Total costs (€)	Total QALYs	Δ Costs (€)	Δ QALYs	ICER (€/QALY gained)
Acenocoumarol					
Standard care	8742	9.832			
Clinical algorithm	8740	9.835	-2*	0.003	
Genotype guided algorithm	8780	9.836	38*/	0.004	8956
Rivaroxaban	15032	9.953	6290*/	0.121*	51933*/ dominated
Apixaban	14157	10.172	5415*/	0.341*/0.336	15895*/15982#
Dabigatran	14897	10.182	6155*/	0.351*/0.01	17545*/72750$
Phenprocoumon					
Standard care	8372	9.823			
Clinical algorithm	8405	9.825	3*	0.003	10998*
Genotype guided algorithm	8376	9.828	33*/19#	0.005*/0.002#	12777*/8295#
Rivaroxaban	15016	9.921	6644*/	0.140*	42140*/dominated
Apixaban	14114	10.141	5741*/5066#	0.251*/0.347#	14241*/13968#
Dabigatran	14859	10.152	6487*/769$	0.262/0.011$	15918*/70921$

QALY denote quality-adjusted life year

ICER incremental cost-effectiveness ratio

All the values were discounted.

*Compared with the standard care of acenocoumarol or phenprocoumon

#Compared with the genotype-guided algorithm dosed acenocoumarol or phenprocoumon

$Compared to apixaban.

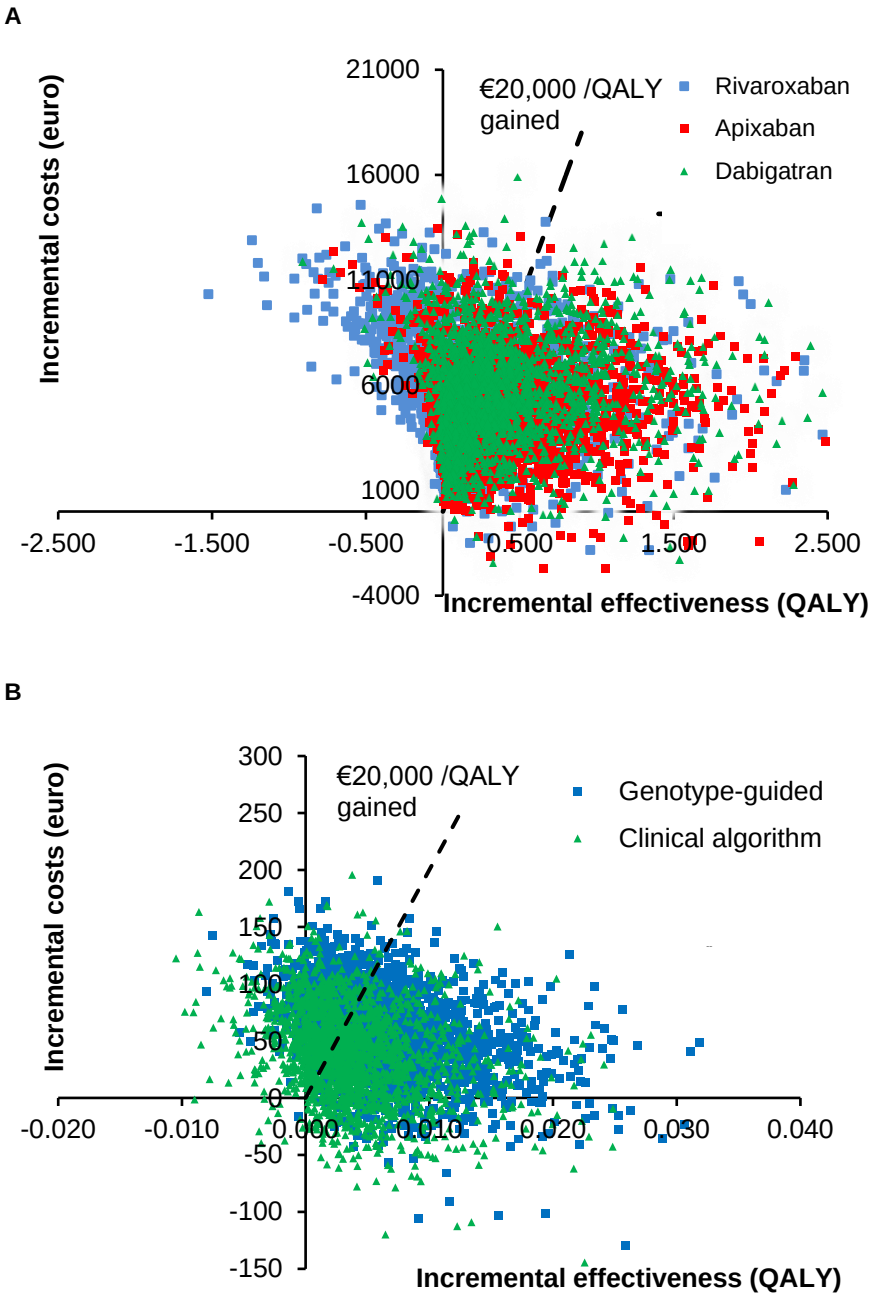

Figure 2 Scatter plot reflecting the uncertainty in the differences in costs and effectiveness. **A**. Difference between apixaban, dabigatran, and rivaroxaban. **B**. Difference between pharmacogenetic dosing and clinical dosing for phenprocoumon.

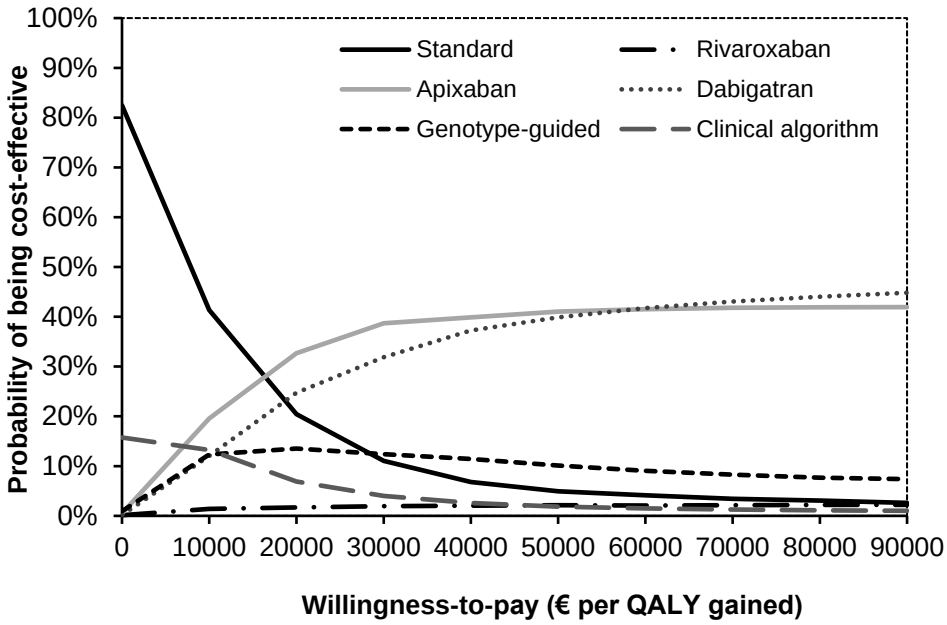

Figure 3. Cost-effectiveness acceptability curves for the base case analysis.

The results of the univariable sensitivity analysis were summarized in the Supplement Figure S1. For genotype-guided dosing and clinical algorithm dosed phenprocoumon, age at the start of treatment has the largest influence on the ICER. The cost-effectiveness would be more favorable for younger patients than for older patients in the cases of genotype guided phenprocoumon therapy, and apixaban or dabigatran treatment. For the apixaban and dabigatran, TTR after the initial period of phenprocoumon therapy (dosed according the usual care in the Netherlands) had the greatest impact on the cost-effective results. This factor also had an important impact on the cost-effectiveness of genotype-guided dosing and clinical algorithm dosed phenprocoumon.

Discussion

The present study shows that apixaban is the most cost-effective alternative among the DOACs compared with phenprocoumon in the Netherlands. Although the genotype-guided dosing algorithm of phenprocoumon also appears to be cost effective as compared with the clinical algorithm dosed phenprocoumon and the

standard care of phenprocoumon, the probability of being cost effectiveness is smaller than the standard care at cost-effectiveness at a threshold of €20,000 per QALY gained. The increase in health is very small, while costs are relatively high. The health gained with each dosing method was only 0.002 QALY.

It is confirmed that the probability of DOACs being cost-effectiveness is depending on the quality of anticoagulation control of VKAs in numerous studies [12, 36]. Our previous study also showed that apixaban and dabigatran could be cost effective alternatives compared with VKAs in the Netherlands while are largely dependent on the setting and quality of local anticoagulant facilities [12]. Another economic evaluation in the Netherlands [37] demonstrated the apixaban is likely to be a cost-effective alternative to VKAs however the impact of different coagulation monitoring levels was small. Even though the mean TTR in the Netherlands is higher (approximately 79%), the present study shows that apixaban has a higher probability of cost-effectiveness at a threshold of €20,000 per QALY gained compared with phenprocoumon treatment regardless of how this is dosed. However, it should be noted that the percentage of TTR has the highest influence on the cost-effectiveness of the DOACs in our study.

A number of studies have assessed the cost-effectiveness of genotype-guided dosing [38] compared with usual care or using clinical dosing for warfarin. A recent study using the EU-PACT trial data suggest that pharmacogenetic-guided dosing is cost-effective strategy in atrial fibrillation patients treated with warfarin in UK and Sweden. However, another study investigated the cost-effectiveness of a pharmacogenetic-guided algorithm for coumarin anticoagulants in the Netherlands shows the pharmacogenetic dosing slightly increase health but is unlikely to be cost effective compared to the clinical algorithm [6]. In our study, the genotype-guided dosing for phenprocoumon was not likely to be cost-effective in a model with DOACs. However, if we only evaluated genotype-guided dosing and the standard care in the Netherlands, the genotype-guided dosing will be more cost-effective (Figure 2) at a threshold of € 20000/QALYs gained.

Findings of our study are similar to another analysis in the UK setting, in which Pink *et al* [39] estimated the cost-effectiveness of clinical and pharmacogenetic dosing algorithms for warfarin and compared with DOACs in one model. In that study,

apixaban is estimated as the most likely option to be cost effective at a threshold of £ 20000/QALYs gained by the National Institute for Health and Care Excellence in UK, while genotype-guided warfarin has a higher probability of being cost-effectiveness than clinical algorithm-dosed warfarin above threshold of £6700 /QALYs gained.

In the Netherlands, both acenocoumarol and phenprocoumon are used in daily practice, while in our study, only phenprocoumon data were used in the model. However, we do not expect too much difference in our results if we use acenocoumarol treatment as the base case since the effect of genotype-guided dosing for acenocoumarol is similar to phenprocoumon and the cost.

The strength of our study is that we evaluated the cost-effectiveness of all the potential anticoagulant alternatives in one model specifically for the Netherlands. Another strength is that we used several country-specific parameters, such as cost, mortality and events rates from the Netherlands. The estimates of genotyping in the Netherlands were more reliable than the previous studies, because our parameters are estimated based on the EU-PACT trial [5], which is the only larger clinical trial on genotyping for phenprocoumon and acenocoumarol.

A major limitation of the present study is the difference of therapeutic INR range used in the trial data and the standard care in the Netherlands. The EU-PACT trial used 2.0-3.0 as the therapeutic range, which is relatively narrow. We reanalyzed the data to get the adverse events by using therapeutic range 2.0-3.5 per Dutch guideline. This might relatively cause an overestimation on the effect of the two dosing methods. Another limitation is the use of INR as a surrogate parameter. The association between INR and the risk of adverse events is an important uncertainty in our study. We have evaluated the influence of this uncertainty on the risk of bleeding and thromboembolic events.

A potential limitation of our study is that the data for the alternative strategies are all from the clinical trials that were used to model the incidence of events, which may cause an overestimation for cost-effectiveness in this study. However, the pharmacogenetic dosing has not been implemented thus no real-world data were available for this option.

Conclusion

Our study suggests apixaban to be the most cost-effective alternative as compared with the standard treatment with phenprocoumon., dabigatran, and rivaroxaban. The genotype-guided dosing algorithm was not likely to be cost effective compared with the standard care in the Netherlands, at a willingness-to-pay threshold of €20,000 per QALY.

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Supplement

Table S1. Age specific mortality rates in The Netherlands, excluding cerebrovascular deaths [1]. Linear interpolation was used for the missing age groups.

Age	Mortality rate
47	0.0014
52	0.0025
57	0.0043
62	0.0069
67	0.0106
72	0.0174
77	0.0285
82	0.0521
87	0.0966
92	0.1780
97	0.2908
100	0.3282

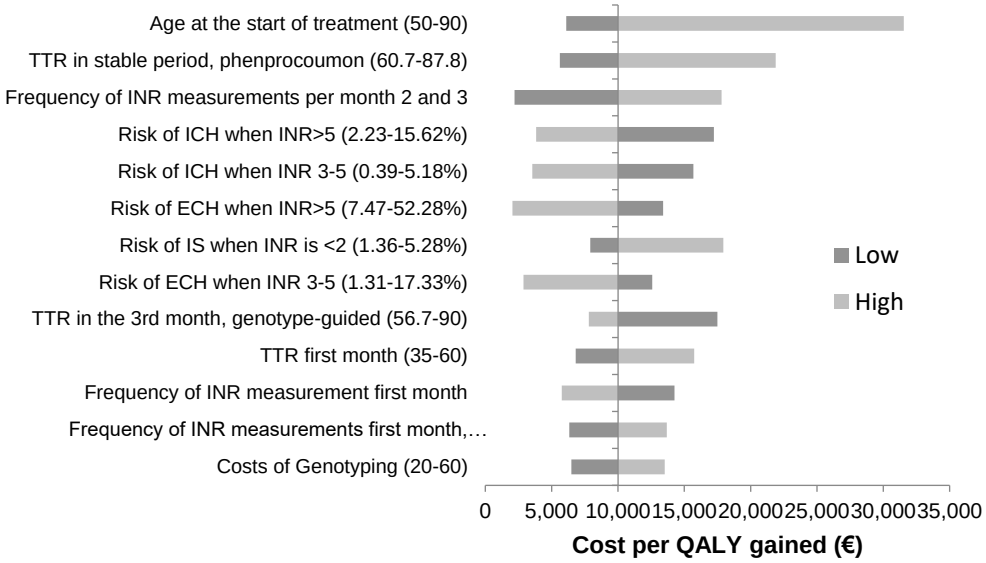
Table S2. Proportion of time spent in different INR ranges during the first 3 months of treatment.

	Standard Care Base case (range*)	Clinical algorithm Base case (range*)	Genotype guided Base case (range*)
Month1			
<2	24.0 (21.4-26.6)	38.8 (31.2-45.6)	36.9 (30.3-43.5)
2-3.5	57.3 (54.5-60.1)	50.0 (43.5-56.5)	58.7 (52.3-65.1)
3.5-5	15.1 (13.0-17.2)	10.2 (5.4-15.0)	4.4 (1.7-7.1)
>5	3.6 (2.5-4.7)	1.0 (0.0-2.27) #	0.0 (0.0-0.0)
Month2			
<2	12.1 (9.7-14.4)	15.3 (8.9-21.7)	12.8 (7.2-18.5)
2-3.5	69.1 (65.8-72.4)	75.5 (68.6-82.4)	79.6 (73.6-85.6)
3.5-5	17.2 (14.4-19.9)	8.7 (4.5-13.0)	7.4 (3.4-11.4)
>5	1.7 (0.9-2.5)	0.4 (0.0-1.0) #	0.2 (0.0-0.5)
Month3			
<2	12.8 (2.8-22.8)	6.5 (2.0-11.0)	6.6 (3.2-10.0)
2-3.5	79 (69.3-88.8)	85.9 (80.0-91.8)	81.4 (75.3-87.5)
3.5-5	7.4 (6.4-8.4)	6.4 (2.8-9.9)	11.8 (6.1-17.5)
>5	0.2 (0.1-0.3)	1.2 (0.0-2.5) #	0.2 (0.0-0.5)

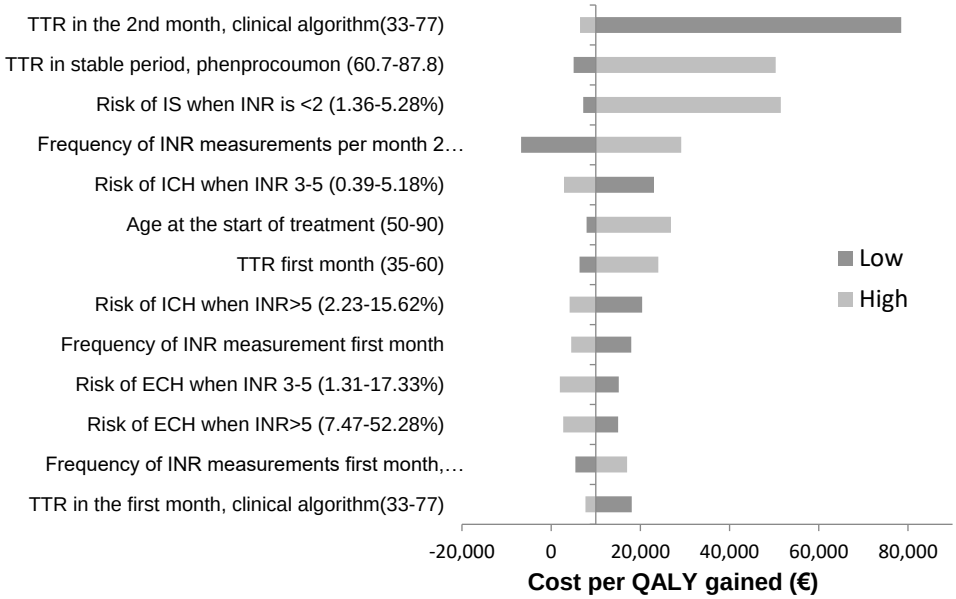
*Range was defined as 95% confidence intervals.
#The lower bound was assumed to be zero.

Figure S1. Tornado diagrams of the incremental cost–effectiveness ratios of pharmacogenetic dosing versus clinical dosing (excluding parameters regarding the effect of genotyping).

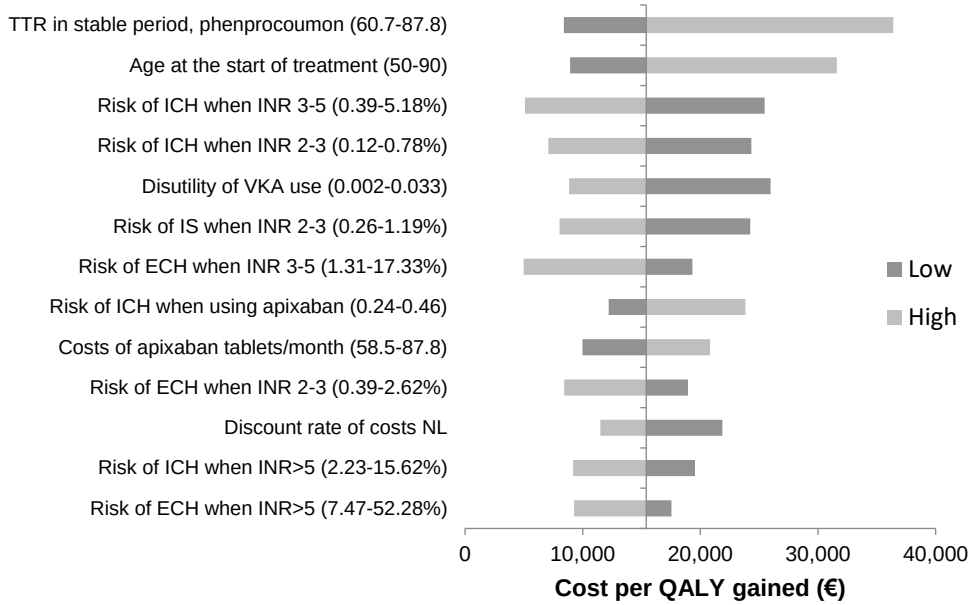
A. Genotype-guided dosing vs. standard care



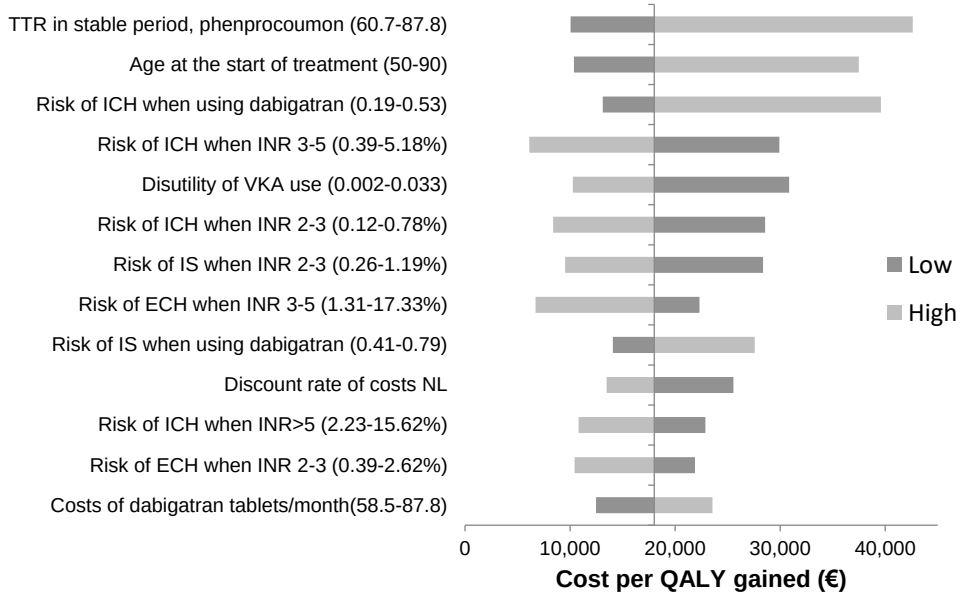
B. Clinical dosing vs. phenprocoumon (standard care)



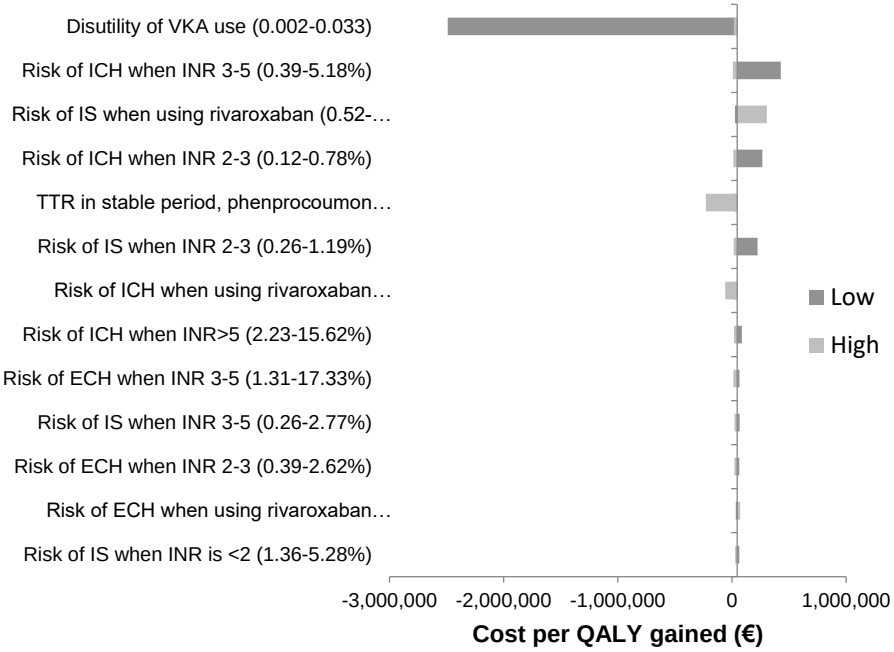
C. Apixaban vs. phenprocoumon (standard care)



D. Dabigatran vs. phenprocoumon (standard care)



E. Rivaroxaban vs. phenprocoumon (standard care)



CHAPTER 7

General discussion

Introduction

For the past 70 years Vitamin K antagonists (VKAs) have been widely used to treat patients at risk of thromboembolic disorders. Dosing of VKAs is difficult because many variables can impact the degree of anticoagulation achieved by individual patients and the therapeutic window of these drugs is narrow. Factors contributing to the variability in anticoagulation effect are drug-drug interactions, drug-food interactions, comorbidities, age, body weight, treatment adherence, and genetic variation. Genetic variants in the vitamin K oxidoreductase complex 1 (*VKORC1*) and in cytochrome P450 (*CYP2C9*) are the ones most commonly associated with pharmacogenetic interactions with VKAs [1]. Several dosing algorithms including genetic and non-genetic information (clinical factors) were developed to calculate the right dose for individual patients to improve anticoagulation control and decrease the risk of thromboembolic or bleeding events [2, 3].

Direct oral anticoagulants (DOACs), an important development in oral anticoagulation, are now on the market for 10 years. These alternative oral anticoagulant drugs do not need routine monitoring. They are at least as effective as the VKAs and appear to have a lower risk for intracranial bleeding [4]. However, their safety needs further follow up in daily clinical practice as patients with different morbidity and drug use than were evaluated in pre-registration studies will receive these new anticoagulants.

In this chapter, we will elaborate on the results of this thesis. First, the main findings will be presented, and their relevance discussed in a broader perspective. Second, we will discuss methodological aspects of this thesis and address several considerations for the implications of our findings for clinical practice and future research.

Main findings and relevance

Dosing algorithm to improve anticoagulation

The first two chapters of this thesis explored personalized VKA dosing algorithms. To achieve an optimal individual dosing strategy for VKAs, algorithms were developed in the past that made use of information on the *VKORC1* and *CYP2C9*

polymorphisms, as well as non-genetic information. However, because none of the clinical trials included three arms (standard care, clinical algorithm with and clinical algorithm without genetic information), it remained unclear what the effect of the use of the clinical dose algorithm without genetic information is versus standard care.

To further clarify this lack of knowledge a study was performed in which we compared INR data obtained during standard care (from the pre-EU-PACT study [4]) with INR data obtained from a group of patients that were dosed based on a clinical algorithm in the EU-PACT trial. This comparison was done for both acenocoumarol and phenprocoumon (**Chapter 2**). Compared to standard care, clinical dosing algorithms increased the time in therapeutic range (TTR) of the INR during a 2 to 12-weeks period after coumarin start with approximately 6% and 4% for acenocoumarol and phenprocoumon, respectively with only the difference for acenocoumarol being statistically significant. Our results suggest that the quality of anticoagulation therapy may already be improved by using a clinical dosing algorithm without knowing the genotype. The implication of these findings is that when standard care is compared with a dosing algorithm including both clinical and genetic factors as was done in part of the EU-PACT trial (warfarin arm of the trial) it is not possible to differentiate the contributions of clinical versus genetic factors to the observed improvement [5, 6]. Furthermore, it appears that a clinical algorithm without genotype information is already enough to improve dosing of coumarins compared to standard care dosing.

The study in **Chapter 3** demonstrated that in the EU-PACT trial for phenprocoumon the effect of the dosing algorithm, including clinical and genetic factors, compared to the algorithm without genetic factors was modified by age. Among younger (<75 years) phenprocoumon users, TTR during the first 12 weeks in the genotype-guided group was 9.5% (95% CI 1.3 to 17.8) higher than in the control group with a remarkably lower percentage of time above the therapeutic range (difference: -9.6%, 95% CI -19.0 to -0.2) and a similar time below this range. In the older group (≥75 years) we found that patients dosed by the genotype-guided algorithm spent more time above the therapeutic range (difference: 27.5%, 95% CI 12.9 to 42.0). For acenocoumarol users, there was no clear effect modification by age and there

were no significant differences between the genotype-guided and control groups for most outcomes, except for a lower percentage of time below the therapeutic range among older patients.

Safe use of DOACs: focus on drug interactions

We performed two studies concerning the safety of DOACs using data from the UK Clinical Practice Research Datalink (CPRD). First, we assessed the association between concurrent use of potential pharmacokinetic or pharmacodynamic interacting drugs and the risk of major bleeding events among DOAC users (**Chapter 4**). This was a case-control study nested in a cohort of new users of DOACs (dabigatran, apixaban, and rivaroxaban), who were at least 18 years old with a first hospital admission for a major bleeding. We identified 393 patients with a major bleeding from a total of 23492 new users of DOACs and matched them to 1494 controls. We found a 2-fold increased risk of major bleeding among patients taking DOACs when there was concurrent use of antiplatelet drugs, and a 1.7-fold increased risk for patients using SSRIs in combination with DOACs. Combined use of DOACs with pharmacokinetic interacting drugs (inhibitors of CYP3A4 and P-glycoprotein) was not associated with an increased risk of major bleeding. In **Chapter 5** a cohort study is presented in which we studied whether the prescribing of DOACs was adjusted when combined with drugs known to interact with DOACs. Dose adjustments of DOACs, discontinuation of DOACs, and switches to vitamin K antagonists (VKAs) were evaluated. It appeared that DOACs were often combined with potentially interacting drugs (PKIDs and/or PDIDs), between 38% and 63%. Furthermore, it appeared that the DOAC starting dose was not reduced in these patients more often than in patients without interacting drugs at the start of DOAC treatment. When an interacting drug was started during DOAC treatment in only a small percentage of patients (<11%) the dose of DOACs was reduced, the DOAC discontinued or switched to a VKA (**Chapter 5**).

Cost-effectiveness analysis

Currently, there are several treatment options when oral anticoagulation treatment is needed. The recently introduced DOACs are costly drugs but may reduce overall costs by being safer (less intracranial bleeding). To support the choices between

anticoagulant drugs it is important to study the cost-effectiveness of DOACs versus VKAs. In **Chapter 6** we evaluated the cost-effectiveness of patients using DOACs versus patients using phenprocoumon or acenocoumarol guided by a clinical and genotype-guided dosing algorithm in patients with atrial fibrillation in the Netherlands. We found that compared with the Dutch standard care of acenocoumarol, genotype guided dosing and the use of apixaban, dabigatran and rivaroxaban the ICERs were €8956, €14241, €15918, and €42140 per QALY gained, respectively. Apixaban could be the most cost-effectiveness alternative in the Netherlands.

Methodological considerations

For the studies described in **Chapter 2** and **Chapter 3** we used data from the EU-PACT trial [5, 7]. As this was a randomized controlled trial (RCT) the risk of selective information and confounding bias is lower compared with observational epidemiological studies. However, patients participating in an RCT may not be representative for patients treated in daily clinical practice. RCTs are usually powered to have sufficient patients to study the main effect (i.e. the primary outcome). Therefore, when subgroup analyses are performed often the power in these subgroups is too low. In **Chapter 3** we performed subgroup analyses with data form the EU-PACT trial. Preferably when evaluating the quality of anticoagulation, one would like to study thromboembolic, as well as bleeding events. These events were collected in this trial, but the numbers were already too low to be studied in the main analysis and thus certainly for our subgroup analyses. To overcome this problem, we decided to evaluate the TTR of the INR as a surrogate endpoint (**Chapter 2** and **Chapter 3**). INRs below the therapeutic range are associated with a higher risk for thromboembolic events and values above this range are associated with a higher risk for bleeding events [8]. INR values were not studied as a dichotomous parameter, but as a continuous parameter. The use of a continuous outcome provides more statistical power; however, the interpretation of the clinical relevance is more difficult. This is because when a clinically relevant cut-off point is chosen results are easier to interpret.

The studies in **Chapter 4** and **Chapter 5** were conducted with data from the CPRD. CPRD is a large real-life longitudinal database of UK primary care. It is an often-

used research database of which the medical information entered is monitored for validity and completeness [9]. The strength of using population-based data from primary care is that it is representative of daily practice and when a large database is being used it allows to evaluate different subgroup of patients often not participating in RCTs. However, some limitations need to be addressed. First, there are some methodological challenges related to the classification of exposure. We defined concurrent use based on a prescription in a 30 days' time window prior to the index date which may lead to misclassification of concurrent exposure. In **Chapter 4**, we conducted sensitivity analyses and found this did not impact our results. Another limitation of CPRD is that not all drugs that patients are using are available. Drug-dispensing data and information on over the counter drugs is not included in CPRD; therefore, uncertainty remains about patients filling prescriptions, and misclassification of drug exposure cannot be completely ruled out. Furthermore, in CPRD drugs prescribed by hospital specialists are not consequent recorded by general practitioners. In our studies we therefore missed patients prescribed DOACs by hospital specialists. These patients probably have more complex diseases and treatments than patients prescribed DOACs by primary care physicians and therefore may have other bleeding risks when DOACs are combined with drugs known to interact with these DOACs.

Implications

Implementation of genotype-guided dosing

Personalized dosing of VKAs by using a genotype-guided algorithm could be a good treatment option among patients for whom VKAs are preferable. Based on our studies especially in patients younger than 75 years the use of a dosing algorithm for phenprocoumon might improve the safety of phenprocoumon during the first weeks after treatment start. For acenocoumarol, such recommendations cannot be given. As warfarin is not marketed in the Netherlands the positive results of the EU-PACT warfarin arm have no implications for Dutch patients. However, the phenprocoumon/acenocoumarol arm of the trial did show a statistically significant difference with respect to time in therapeutic range in the first four weeks of treatment. Furthermore, our finding that this effect is stronger in younger patients does suggest that it is important that genotyping becomes routinely available (now

mostly available in research environments) and health insurance companies reimburse these tests.

Use of DOACs

DOACs have proven to be an effective and safe alternative to VKAs for prevention of stroke and systemic embolism in patients with AF and patients with VTE both in trials and real-life studies [4, 10-15]. However, patients usually have complex risk profiles in real-life, among them drugs that might potentially influence the safety of DOACs. In **Chapter 4** we presented that the combination of a platelet inhibitor or SSRI with a DOAC increases the risk for major bleeding compared to use of DOACs alone with approximately 100% [13, 16]. The combination of a platelet inhibitor and a DOAC should be carefully considered depending on the bleeding risk of a patient (for instance by using the HAS-BLED score). Also, the guidelines should be strictly followed when the platelet inhibitor or DOAC needs to be discontinued (e.g. discontinuation of the platelet inhibitor after twelve months after an acute coronary syndrome with stent placement). For the increased major bleeding risk when a DOAC is combined with a SSRI, prescribers should try to prevent this combination. For instance, this could be achieved by considering a tricyclic antidepressant when a patient with a depression is using a DOAC. Interestingly, our studies in this thesis did not show an increased risk of major bleeding when drugs that moderately inhibit CYP3A4 and/or P-gp are combined with a DOAC. As this finding does not exclude an increased risk for minor bleeding events it is still important to strictly follow the dose recommendations in the SmPCs of the specific DOACs.

Future perspectives

VKAs will still be on the center stage of oral anticoagulation treatment for a long time despite the fact that DOACs are becoming a main treatment option for patients that have an indication for oral anticoagulation [17-19]. Therefore, it is still worthwhile to further explore genotype-guided dosing algorithms to optimize the individualized treatment with VKAs. Currently, there are numerous algorithms, but still there is no perfect algorithm because not all factors that determine VKAs dosing have been identified [20]. There is still a 20% to 40% unexplained variability in dose requirement of VKAs. Thus, there is a room for increasing the percentage of

explained variability. Besides the factors included in the existing dosing algorithm, there is still a lack of factors representing drugs known to interact with VKAs [21] and undetected gene variations. Recent evidence indicate that the CYP4F2*3 polymorphism was consistently associated with an increase in mean coumarin dose, with a stronger effect in females, in patients taking acenocoumarol [22]. It is important when new factors predicting dose needed are being found that the clinical relevance of adjusted algorithms are first studied before implementation in routine daily clinical practice.

In the future, it may be possible to personalize the use of all the oral anticoagulants. There are already attempts to develop algorithms based on clinical features or by patterns of risk factors and comorbidities to identify patients to be treated with a particular oral anticoagulant to promote optimal clinical outcomes [23, 24]. To further improve such algorithms more research is needed on factors influencing the benefit risk of DOACs. For instance, it is important to further study the influence of mutations in CYP3A4 and P-glycoprotein on the pharmacokinetics of DOACs [25]. Also, it is relevant to evaluate the effectiveness and safety of a lower dose of DOACs when doses are adjusted to interacting drugs. The SmPCs of the DOACs advise to be cautious when DOACs are combined with interacting drugs. Further research will help to change these warnings in practical advices what to do in certain situations. Our study demonstrated that the call in the SmPC to be cautious when a DOAC is concomitantly used with an interacting drug does not lead to dose adjustments of DOACs in daily clinical practice and teaches us that further education of health care professionals/prescribers/practitioners is needed how to safely handle DOACs.

Conclusion

Our studies on VKAs and DOACs contribute to the knowledge on factors that influence the benefit risk of those oral anticoagulants and thereby will contribute to the safe use of these medicines in daily practice. Furthermore, our cost-effectiveness analysis showed that DOACs are an acceptable alternative of VKAs.

For the VKAs dosing algorithms were studied and it was shown that also dosing algorithms with clinical information but without genotype information already might

improve the time patients are within the therapeutic INR range. Furthermore, it appears that age is an important determinant for the performance of a dosing algorithm. Especially in patients younger than 75 years a dosing algorithm for phenprocoumon appears to be beneficial. The studies on DOACs showed that the combination of a DOAC and a platelet inhibitor or SSRI increases the risk on major bleeding while the combination with an inhibitor of CYP3A4 and/or P-glycoprotein does not increase this bleeding risk. Finally, our study on dose adjustments of DOACs when combined with an interacting drug taught us that further education of health care practitioners on the safe use of DOACs is needed.

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APPENDICES

APPENDIX I

SUMMARY

Summary

Vitamin K antagonists (VKAs) and direct oral anticoagulants (DOACs) are commonly used for prophylaxis and treatment of thromboembolic disorders. Dosing of VKAs is difficult because the degree of anticoagulation achieved by individual patients can be influenced by genetic variation such as the variants of VKORC1 and CYP2C9 and non-genetic factors like drug-drug and drug-food interactions, comorbidities, age, body weight and treatment adherence. The dose of VKAs is guided by measurements of blood coagulation (INR measurements). Several dosing algorithms including genetic and non-genetic information were developed to calculate the right dose for individual patients to improve anticoagulation control and decrease the risk of thromboembolic and bleeding events.

Direct oral anticoagulants (DOACs), the alternative drugs of VKAs, do not need routine monitoring of anticoagulation and are at least as effective as the VKAs and appear to have a lower risk for intracranial bleeding.

At the start of this PhD project there were several aspects of the use of VKAs and DOACs in daily practice that needed further exploration especially aspects related to individualization of anticoagulation treatment.

In **Chapter 1** we provide a general introduction on oral anticoagulants and describe the aims of this thesis. We aimed to explore the relevance of dosing algorithms for VKAs compared to standard dosing and how these algorithms perform in different age groups. We also aimed to evaluate the influence of drug interactions on the safety of DOACs in daily clinical practice and whether health care practitioners take into account drug interactions when deciding on the prescribed dose of DOACs. Finally, we aimed to assess the cost-effectiveness of a variety of clinical and genotype-guided dosing algorithms for VKAs versus DOACs.

In **Chapter 2**, we compared the anticoagulant effect of dosing algorithms for acenocoumarol and phenprocoumon including clinical patient characteristics with standard care in the Netherlands. We compared INR data obtained during standard care (from the pre-EU-PACT study) with INR data obtained from a group of patients that were dosed based on a clinical algorithm in the randomized EU-PACT trial. Compared to standard care, clinical dosing algorithms increased the time in

therapeutic range (TTR) of the INR during a 2 to 12-weeks period after coumarin start with approximately 6% and 4% for acenocoumarol and phenprocoumon, respectively, with only the difference for acenocoumarol being statistically significant. Our results suggest that the quality of anticoagulation treatment may already be improved by using a clinical dosing algorithm without knowing the genotype. In **Chapter 3** we described a sub-analysis of the EU-PACT acenocoumarol/ phenprocoumon data by evaluating the effect of dosing algorithms on anticoagulation after stratification by age. Among younger (<75 years) phenprocoumon users, the time in therapeutic INR range during the first 12 weeks in the genotype-guided group was 9.5% (95% CI 1.3 to 17.8) higher than in the control group with a remarkably lower percentage of time above the therapeutic INR range (difference: -9.6%, 95% CI -19.0 to -0.2) and a similar time below this range. In the older group (≥75 years) we found that patients dosed by the genotype-guided algorithm spent more time above the therapeutic INR range (difference: 27.5%, 95% CI 12.9 to 42.0). For acenocoumarol users, there was no clear effect modification by age and there were no significant differences between the genotype-guided and control groups for most outcomes, except for a lower percentage of time below the therapeutic INR range among older patients. We thus demonstrated that in the EU-PACT trial for phenprocoumon the effect of the dosing algorithm, including clinical and genetic factors, compared to the algorithm without genetic factors was modified by age.

We performed two studies (**Chapter 4** and **Chapter 5**) concerning the safety of DOACs based on data of the UK Clinical Practice Research Datalink (CPRD). We conducted a case-control study nested in a cohort of new users of DOACs who were at least 18 years old with a first hospital admission for a major bleeding (**Chapter 4**). We assessed the association between concurrent use of potential pharmacokinetic or pharmacodynamic interacting drugs and the risk of major bleeding events among DOAC users. We found a 2-fold increased risk of major bleeding among patients taking DOACs when there was concurrent use of antiplatelet drugs, and a 1.7-fold increased risk for patients using SSRIs in combination with DOACs. Combined use of DOACs with pharmacokinetic interacting drugs (inhibitors of CYP3A4 and P-glycoprotein) was not associated

with an increased risk of major bleeding. In **Chapter 5,** a cohort study is presented in which we studied whether the prescribing of DOACs was adjusted when combined with drugs known to interact with DOACs. Dose adjustments of DOACs, discontinuation of DOACs, and switches to vitamin K antagonists (VKAs) were evaluated. DOACs were often combined with potentially interacting drugs (pharmacokinetic and/or pharmacodynamic interacting drugs), between 38% and 63%. It appeared that the DOAC starting dose was not reduced in these patients more often than in patients without interacting drugs at the start of DOAC treatment. When an interacting drug was started during DOAC treatment in only a small percentage of patients (<11%) the dose of DOACs was reduced, the DOAC discontinued or switched to a VKA. In **Chapter 6**, we conducted cost-effectiveness analyses on DOAC users versus patients using phenprocoumon or acenocoumarol guided by a clinical and genotype-guided dosing algorithm in patients with atrial fibrillation in the Netherlands. We found that compared with the Dutch standard care of acenocoumarol, genotype guided dosing and the use of apixaban, dabigatran and rivaroxaban the ICERs were €8956, €14241, €15918, and €42140 per QALY gained, respectively. Apixaban could be the most cost-effective alternative in the Netherlands.

In **Chapter 7** we discussed the main findings described in this thesis and their relevance. Furthermore, implications, strengths and limitations of the studies, and future prospectives were discussed. Personalized dosing of VKAs by using a genotype-guided algorithm could be a good treatment option among patients for whom VKAs are preferable. DOACs are becoming more and more the main treatment option for patients that have an indication for oral anticoagulation. Our studies show that compared to VKAs, also for DOACs drug interactions can cause serious adverse drug reactions. More attention should be given to possible dose adjustments of DOACs when combined with a potential interacting drug, especially pharmacodynamic interacting drugs (platelet inhibitors and SSRIs).

APPENDIX II

SAMENVATTING

SAMENVATTING

Vitamine K-antagonisten (VKA's) en directe orale anticoagulantia (DOAC's) worden veel gebruikt voor de behandeling and profylaxe van trombo-embolische aandoeningen. Het doseren van VKA's is lastig, omdat de mate van stolling binnen patiënten beïnvloed kan worden door genetische variatie van VKORC1 en CYP2C9 polymorfismen en door niet-genetische factoren, zoals interacties met andere geneesmiddelen, voedingsstoffen, co-morbiditeiten, leeftijd, lichaamsgewicht en therapietrouw. De dosering van VKA's wordt bepaald op basis van bloedstollingsmetingen (INR-bepalingen). Verschillende doseringsalgoritmes met zowel genetische als niet-genetische informatie zijn ontwikkeld om de juiste dosering voor individuele patiënten te berekenen en zodoende de controle van de antistolling te verbeteren en het risico op trombo-embolische en bloedings gebeurtenissen te verlagen.

DOAC's, hetgeen therapeutische alternatieven zijn voor VKA's, hebben een dergelijke routinematige monitoring van de bloedstolling niet nodig, zijn minstens zo effectief als VKA's en lijken tevens een lager risico te geven op intracraniële bloedingen.

Bij de aanvang van dit promotietraject waren er diverse aspecten bij gebruik van VKA's en DOAC's in de dagelijkse praktijk die nader uitgezocht dienden te worden, in het bijzonder rondom de individualisering van de antistollingsbehandeling.

In Hoofdstuk1 hebben we een algemene inleiding over orale anticoagulantia gegeven en zijn de verschillende doelen van dit proefschrift beschreven. Een eerste doel was om de relevantie van dosisalgoritmes voor VKA's te vergelijken met een standaarddosering en te onderzoeken hoe deze algoritmes presteren in verschillende leeftijdsgroepen. Daarnaast was een doel om het effect van geneesmiddelinteracties op de veiligheid van DOACs in de dagelijkse praktijk te evalueren en te onderzoeken of zorgverleners bij het doseren van DOACs rekening houden met geneesmiddelinteracties. Als laatste was het doel om de kosteneffectiviteit van een aantal klinische en op genotype gebaseerde doseringsalgoritmes van VKA's te vergelijken met DOAC's.

In Hoofdstuk 2 hebben we het antistollingseffect van het gebruik van doseringsalgoritmes, inclusief klinische patiëntkenmerken van acenocoumarol en fenprocoumon vergeleken met het antistollingseffect na gebruik van de standaarddoseringen van deze middelen in Nederland. We vergeleken INR-data verkregen uit de standaardzorg (middels gegevens uit de pre-EU-PACT-studie) met INR-gegevens van een groep patiënten die gedoseerd werden volgens een klinisch algoritme in de gerandomiseerde EU-PACT trial. Vergeleken met de standaardbehandeling, verhoogde het gebruik van een klinisch algoritme de tijd in therapeutisch INR-bereik gedurende een 2 tot 12 weekse periode na start van coumarine therapie met ongeveer 6% en 4% voor acenocoumarol en fenprocoumon, respectievelijk, waarbij alleen het verschil voor acenocoumarol statistisch significant was. De resultaten suggereren dat de kwaliteit van de antistollingsbehandeling al verbeterd kan worden door een klinisch doseringsalgoritme te gebruiken zonder kennis te hebben van het genotype.

In Hoofdstuk 3 wordt een sub-analyse van de EU-PACT acenocoumarol/ fenprocoumon gegevens gepresenteerd door het effect van doseringsalgoritmes op antistolling te evalueren na stratificatie op leeftijd. Onder jongere (<75 jaar) fenprocoumon gebruikers was de tijd in therapeutisch INR-bereik in de eerste 12 weken in de op genotype-gebaseerde groep 9,5% (95% betrouwbaarheidsinterval [BI] 1,4 tot 17.8) hoger dan in de controle groep met een opmerkelijk lager percentage tijd boven het therapeutisch INR-bereik (verschil: -9,6%, 95%BI -19,0 tot -0,2) en een vergelijkbare tijd onder dit bereik. In de oudere leeftijdsgroep (≥75 jaar) vonden we dat patiënten die gedoseerd werden middels het genotype-gebaseerde algoritme meer tijd boven het therapeutisch INR-bereik hadden met een verschil van 27,5% (95% BI 12,9 tot 42,0). Voor acenocoumarol gebruikers was er geen duidelijke effect-modificatie door leeftijd en waren er geen significante verschillen tussen de genotype-gebaseerde en controle groepen voor de meeste uitkomsten, behalve een lager percentage tijd onder het therapeutisch INR-bereik bij oudere patiënten. Met dit onderzoek hebben we laten we zien dat in de EU-PACT trial voor fenprocoumon het effect van het doseringsalgoritme, met klinische en genetische factoren, vergeleken met het algoritme zonder genetische factoren werd veranderd door leeftijd.

Er zijn twee onderzoeken (Hoofdstuk 4 en Hoofdstuk 5) gedaan naar de veiligheid van DOAC's waarbij gebruik is gemaakt van gegevens van de Britse Clinical Practice Research Datalink (CPRD). We verrichtten een patiënt-controle onderzoek binnen een cohort van nieuwe gebruikers van DOAC's die minstens 18 jaar oud waren en een eerste opname voor een ernstige bloeding hadden (Hoofdstuk 4). We onderzochten de associatie tussen gelijktijdig gebruik van mogelijk farmacokinetisch of farmacodynamisch interacterende geneesmiddelen en het risico op ernstige bloedingen onder DOAC gebruikers. We vonden een tweemaal verhoogd risico op ernstige bloedingen bij patiënten die DOAC's gebruikten in combinatie met plaatjesaggregatieremmers en een 1,7-voudig verhoogd risico voor patiënten die DOAC's combineerden met SSRI's. Gecombineerd gebruik van DOAC's met farmacokinetisch interacterende geneesmiddelen (remmers van CYP3A4 en P-glycoproteïne) bleek niet geassocieerd met een verhoogd risico op ernstige bloedingen. In Hoofdstuk 5 is een cohort onderzoek gepresenteerd waarin werd nagegaan of het voorschrijven van DOAC's werd aangepast wanneer een combinatie optrad met geneesmiddelen waarvan bekend is dat deze een interactie hebben met DOAC's. Doseringsaanpassingen van DOAC's, het discontinueren van DOAC's en het switchen van DOAC's naar VKA's werden onderzocht. DOAC's werden vaak – tussen de 38% en 63%- in combinatie gebruikt met potentieel interacterende geneesmiddelen (farmacokinetisch en/of farmacodynamisch). Het leek er op dat de startdosis van DOAC's niet werd verminderd bij dergelijke patiënten ten opzichte van patiënten die geen interacterende geneesmiddelen gebruikten ten tijde van de start van de behandeling met DOAC's. In het geval dat een interacterend geneesmiddel werd gestart gedurende de behandeling met DOAC's werd slechts bij een klein percentage patiënten (< 11%) de DOAC-dosering verminderd, de DOAC gestopt of geswitcht naar een VKA.

In Hoofdstuk 6 werden kosteneffectiviteitsanalyses uitgevoerd bij gebruikers van DOAC's ten opzichte van gebruikers van fenprocoumon of acenocoumarol met een klinisch en op genotype gebaseerd doseringsalgoritme bij patiënten met atriumfibrilleren in Nederland. We vonden dat vergeleken met de standaardbehandeling met acenocoumarol, een op genotype gebaseerde dosering

van acenocoumarol en het gebruik van apixaban, dabigatran en rivaroxaban de ICER's respectievelijk €8.956, €14.241, €15.918 en €42.140 per QALY waren. Apixaban zou hiermee het meest kosteneffectieve alternatief in Nederland kunnen zijn.

In hoofdstuk 7 bediscussieerden we de belangrijkste bevindingen van dit proefschrift en hun relevantie. Daarnaast werden de implicaties, sterktes en beperkingen van de onderzoeken en toekomstperspectieven beschouwd. Geïndividualiseerde dosering van VKA's door middel van gebruik van op genotype gebaseerde algoritmes zou een goede behandelingsoptie kunnen zijn voor patiënten waarbij behandeling met VKA's de voorkeur heeft. DOAC's worden meer en meer de belangrijkste behandeloptie voor patiënten die een indicatie voor het gebruik van orale anticoagulantia hebben. De onderzoeken in dit proefschrift laten zien dat vergeleken met VKA's ook voor DOAC's geldt dat geneesmiddelinteracties kunnen resulteren in ernstige geneesmiddelbijwerkingen. Meer aandacht zou moeten worden gegeven aan mogelijke doseringsaanpassingen bij DOAC gebruik wanneer deze worden gecombineerd met een potentieel interacterend geneesmiddel, met name farmacodynamisch interacterende geneesmiddelen (plaatjesaggregatieremmers en SSRI's).

APPENDIX III

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APPENDIX IV

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APPENDIX V

LIST OF PUBLICATIONS

List of publications

Publications related to this thesis

Zhang Y, de Boer A, Verhoef TI, van der Meer FJ, le Cessie S, Manolopoulos VG, Maitland-van der Zee AH, group EU-PACT. Age-stratified outcome of a genotype-guided dosing algorithm for acenocoumarol and phenprocoumon. **J Thromb Haemost.** 2017; 15 (3): 454-64.

Zhang Y, de Boer A, Verhoef TI, van der Meer FJ, le Cessie S, Maitland-van der Zee AH, group EU-PACT. Comparison of dosing algorithms for acenocoumarol and phenprocoumon using clinical factors with the standard care in the Netherlands. **Thromb Res**. 2015; 136 (1): 94-100.

Publication unrelated to this thesis

Danese E, Raimondi S, Montagnana M, Tagetti A, Langaee T, Borgiani P, Ciccacci C, Carcas AJ, Borobia AM, Tong HY, Davila-Fajardo C, Rodrigues Botton M, Bourgeois S, Deloukas P, Caldwell MD, Burmester JK, Berg RL, Cavallari LH, Drozda K, Huang M, Zhao LZ, Cen HJ, Gonzalez-Conejero R, Roldan V, Nakamura Y, Mushiroda T, Gong IY, Kim RB, Hirai K, Itoh K, Isaza C, Beltran L, Jimenez-Varo E, Canadas-Garre M, Giontella A, Kringen MK, Haug KBF, Gwak HS, Lee KE, Minuz P, Lee MTM, Lubitz SA, Scott S, Mazzaccara C, Sacchetti L, Genc E, Ozer M, Pathare A, Krishnamoorthy R, Paldi A, Siguret V, Loriot MA, Kutala VK, Suarez-Kurtz G, Perini J, Denny JC, Ramirez AH, Mittal B, Rathore SS, Sagreiya H, Altman R, Shahin MHA, Khalifa SI, Limdi NA, Rivers C, Shendre A, Dillon C, Suriapranata IM, Zhou HH, Tan SL, Tatarunas V, Lesauskaite V, **Zhang Y**, Maitland-van der Zee AH, Verhoef TI, de Boer A, Taljaard M, Zambon CF, Pengo V, Zhang JE, Pirmohamed M, Johnson JA, Fava C. Effect of CYP4F2, VKORC1, and CYP2C9 in Influencing Coumarin Dose: A Single-Patient Data Meta-Analysis in More Than 15,000 Individuals. **Clin Pharmacol Ther**. 2019; 105(6):1477-91.

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Yumao Zhang was born in Anhui, China, on May 10th, 1987. In 2005 he completed his pre-university education at Anhui. In the same year, he started his study in Clinical Pharmacy at Dalian Medical University and received the medical bachelor's degree in 2010. Afterwards, he was enrolled into China Pharmaceutical University as a master student and obtained his master's degree in Clinical Pharmacy in 2013. In the same year, with the support from the China Scholar Council he started his PhD research at the Division of Pharmacoepidemiology and Clinical Pharmacology of Utrecht University, under the supervision of Prof. A. de Boer, Prof. A.H. Maitland-van der Zee and Dr. P.C. Souverein. His main research focused on the personalized use of oral anticoagulants, which include the prediction algorithms for vitamin K antagonists and drug interactions on the safety use of direct oral anticoagulants, as described in this thesis.