DR MICHAEL NAGLER (Orcid ID : 0000-0003-4319-2367)

Article type : Original Article - Clinical Haemostasis and Thrombosis

Long-term outcomes of elderly patients with CYP2C9 and VKORC1 variants treated with vitamin K antagonists

M. Nagler*,†; A. Angelillo-Scherrer*,†; M. Méan‡,§§§§; A. Limacher§; C. Abbal¶; M. Righini**; J. H. Beer††; J. Osterwalder‡‡; B. Frauchiger§§; M. Aschwanden¶¶; C. M. Matter***; N. Kucher†††; J. Cornuz‡‡‡; M. Banyai§§§; M. Husmann¶¶¶; D. Staub¶¶; L. Mazzolai****, O. Hugli††††, N. Rodondi‡,‡‡‡‡; and D. Aujesky‡

Affiliations:

* Department of Haematology and Central Haematology Laboratory, Inselspital, Bern University Hospital, University of Bern, Bern, Switzerland

† Department of Clinical Research, University of Bern, Bern, Switzerland

‡ Department of General Internal Medicine, Bern University Hospital, University of Bern, Bern, Switzerland

§ CTU Bern, and Institute of Social and Preventive Medicine (ISPM), University of Bern, Bern, Switzerland

¶ Division of Hematology, Lausanne University Hospital, Lausanne, Switzerland

** Division of Angiology and Hemostasis, Geneva University Hospital, Geneva, Switzerland

†† Cantonal Hospital of Baden, Baden, Switzerland

‡‡ Emergency Department, Cantonal Hospital of St. Gallen, St. Gallen, Switzerland

§§ Department of Internal Medicine, Cantonal Hospital of Frauenfeld, Frauenfeld, Switzerland

¶¶ Division of Angiology, Basel University Hospital, Basel, Switzerland

*** Center for Molecular Cardiology, University of Zurich, and University Heart Center, University Hospital Zurich, Switzerland

††† Division of Angiology, Bern University Hospital, University of Bern, Bern, Switzerland

‡‡‡ Department of Ambulatory Care and Community Medicine, University of Lausanne, Lausanne, Switzerland

§§§ Division of Angiology, Cantonal Hospital of Lucerne, Lucerne, Switzerland

¶¶¶ Division of Angiology, Zurich University Hospital and University of Zurich, Zurich, Switzerland

**** Service of Angiology, Lausanne University Hospital, Lausanne, Switzerland

†††† Emergency Department, Lausanne University Hospital, Lausanne, Switzerland

‡‡‡‡ Institute of Primary Health Care, University of Bern, Bern, Switzerland

§§§§ Division of General Internal Medicine, Lausanne University Hospital, Lausanne, Switzerland

Correspondence to: michael.nagler@insel.ch; Department of Haematology and Central Haematology Laboratory, Inselspital University Hospital, Freiburgerstrasse, 3010 Berne, Switzerland; phone +41 31 632 2405; fax +41 31 632 5193

Running head: Clinical outcomes in CYP2C9 and VKORC1 variants

This article has been accepted for publication and undergone full peer review but has not been through the copyediting, typesetting, pagination and proofreading process, which may lead to differences between this version and the Version of Record. Please cite this article as doi: 10.1111/jth.13810

Essentials

- The long-term effects of *VKORC1* and *CYP2C9* variants on clinical outcomes remains unclear.
- We followed 774 patients ≥65 years with venous thromboembolism for a median duration of 30 months.
- Patients with CYP2C9 variants are at increased risk of death and non-major bleeding.
- Patients with genetic variants have a slightly lower anticoagulation quality only.

Abstract

Background

The long-term effect of polymorphisms of the vitamin K-epoxide reductase *(VKORC1)* and the cytochrome P450 enzyme gene *(CYP2C9)* on clinical outcomes remains unclear.

Objectives

We examined the association between *CYP2C9/VKORC1* variants and long-term clinical outcomes in a prospective cohort study of elderly patients treated with vitamin K antagonists for venous thromboembolism (VTE).

Methods

We followed 774 consecutive patients aged ≥65 years with acute VTE from nine Swiss hospitals for a median duration of 30 months. The median duration of initial anticoagulant treatment was 9.4 months. The primary outcome was the time to any

clinical event, i.e. the composite endpoint of overall mortality, major- and non-major bleeding, and recurrent VTE.

Results

Overall, 604 (78%) patients had a *CYP2C9* or *VKORC1* variant. Three hundred thirty-four patients (43.2%) had any clinical event, 119 (15.4%) died, 100 (12.9%) had major and 167 (21.6%) non-major bleeding, and 100 (12.9%) recurrent VTE. After adjustment, *CYP2C9* (but not *VKORC1)* variants were associated with any clinical event (hazard ratio [HR] 1.34; 95% confidence interval [CI] 1.08-1.66), death (HR 1.74; 95% CI 1.19-2.52), and clinically relevant non-major bleeding (sub-hazard ratio [SHR] 1.39; 95% CI 1.02-1.89), but not with major bleeding (SHR 1.03; 95% CI: 0.69-1.55) or recurrent VTE (SHR 0.95; 95% CI 0.62-1.44). Patients with genetic variants had a slightly lower anticoagulation quality.

Conclusions

CYP2C9 was associated with long-term overall mortality and non-major bleeding. While genetic variants were associated with a slightly lower anticoagulation quality, there was no relationship between genetic variants and major bleeding or VTE recurrence.

Keywords

Venous Thromboembolism; anticoagulants; phenprocoumon; cytochrome P-450 CYP2C9; mortality

Introduction

Personalized medicine based on genetic factors is seen as a promising strategy to improve patient care and save healthcare costs, in particular with regard to anticoagulation treatment.[1-7] Despite the increasing use of direct oral

anticoagulants, vitamin K antagonists (VKAs) will continue to play a major role and improving the safety of anticoagulant treatment with VKAs remains an important goal.[8, 9] Main safety issues related to VKAs are bleeding as well as thromboembolic events, which both depend on the quality of anticoagulation.[10, 11] Achieving a stable anticoagulation control is difficult because of a large variability of VKA response across individuals.[12] A major part of this variability results from polymorphisms of the vitamin K-epoxide reductase (*VKORC1*) and the cytochrome P450 enzyme gene (*CYP2C9*), which both influence VKA metabolism.[13-22] Patients with a *CYP2C9* genetic variant metabolize VKA to a lesser degree, resulting in higher levels of VKA metabolites and in decreased VKA dose requirements [6, 23]. VKORC1 encodes for Vitamin K-epoxide reductase subunit 1, which converts vitamin K into its active form [6]. Patients with the *VKORC1* G1639A variant are more sensitive to VKA and dose requirements are lower as well [6].

To date, the clinical impact of measuring genetic polymorphisms in patients receiving VKAs still remains unclear.[4, 9, 24-27] Genetics-based dosing algorithms focusing on the initiation of anticoagulation did not demonstrate a clear benefit in terms of anticoagulation quality, as shown in several recent randomized-controlled trials.[24-27] In addition, a recent systematic review did not detect relevant differences in bleeding events between patients with and without *VKORC1* variants, and only a slightly higher bleeding risk in patients with *CYP2C9*3* variants.[22] However, these studies were limited by relatively small sample sizes and/or short follow-up periods, and methodologically more rigorous studies were requested.[22, 27] Moreover, previous studies focused on the initiation of anticoagulation only and the long-term effects of genetic polymorphisms are unknown. To fill this gap of knowledge, we examined the association between *CYP2C9* and *VKORC1* variants

and long-term clinical outcomes in a prospective multicenter cohort study of elderly patients treated with VKA for venous thromboembolism (VTE).[28, 29]

Methods

Cohort sample

The study was conducted between September 2009 and December 2013 as part of the SWIss venous Thromboembolism COhort (SWITCO65+), a prospective multicenter cohort study that assessed long-term medical outcomes and quality of life in elderly patients with acute VTE from all five university hospitals and four high-volume non-university hospitals in Switzerland.[28, 29] Consecutive patients aged 65 years or older with an acute, objectively confirmed VTE were identified in the inpatient and outpatient services of all participating study sites. Patients were excluded in case of inability to provide consent (e.g., severe dementia), thrombosis at different site than DVT or PE, impossible follow-up (e.g. terminal disease), or insufficient German or French language skills. For the sake of this analysis, we considered patients treated with VKAs only. A detailed description of the study methods was published elsewhere.[28, 29] The study was approved by the ethics committees at each participating site.

Baseline data collection

Trained study nurses prospectively collected baseline demographics (age, sex), VTE-related information (localization of the index VTE [PE, proximal/distal DVT], type of VTE [provoked, unprovoked, cancer-related], history of prior VTE, and family history of VTE), vital signs (heart rate, systolic blood pressure), body mass index, co-morbid conditions (recent immobilization, history of major bleeding, acute

rheumatic disease, inflammatory bowel disease, cerebrovascular disease, chronic renal and pulmonary disease), laboratory findings (hemoglobin, creatinine), concomitant antiplatelet and non-steroidal anti-inflammatory therapy, polypharmacy, and VTE-related treatments (type and duration of initial parenteral anticoagulation and duration of VKA therapy) from all enrolled patients using standardized data collection forms.

Genetic analyses

A specifically designed haemostasis biobank was implemented before inclusion of patients.[29] At the day of enrollment, one EDTA-whole blood sample (2.7ml, Sarstedt, Nümbrecht, Germany) was collected. After plasma removal by centrifugation, blood cells were stored at -80°C until DNA extraction (Qiacube automated extraction plus Qiagen DNA blood mini kit, Qiagen, Hombrechtikon, Switzerland). Allelic identifications of *VKORC1* and *CYP2C9* were made by melting curve analysis of fluorescent Q-PCR amplification products. “LightMix for the detection of human *VKORC1* G1639A” and “LightMix for the detection of human *CYP2C9*2* and *CYP2C9*3*” kits (TIBMOLBIOL, Berlin, Germany), containing primers and probes, were used. PCR reactions were performed as advised by the manufacturer on a LightCycler 1.8 (Roche Diagnostics, Rotkreuz, Switzerland). Patients and managing physicians were blinded to the results of these analyses.

Study outcomes

The primary outcome was the time to any clinical event, i.e. a composite outcome of death, major bleeding, clinically relevant non-major bleeding, or recurrent VTE. Major bleeding was defined as fatal bleeding, symptomatic bleeding in a critical

organ (intracranial, intraspinal, intraocular, retroperitoneal, intraarticular, pericardial, or intramuscular with compartment syndrome), bleeding with a reduction in hemoglobin ≥20 g/l, or bleeding leading to a transfusion ≥2 units of packed red blood cells. [30] Clinically relevant non-major bleeding was defined as bleeding that did not meet the definition of major bleeding but required medical attention (i.e., a physician contact). Recurrent VTE was defined as objectively confirmed, new or recurrent proximal or distal DVT or PE, as previously described.[28, 29]

The secondary outcomes were the individual outcomes of overall mortality, major bleeding, clinically relevant non-major bleeding, and recurrent VTE. Additional outcomes were the percentage of time within the therapeutic INR range (TTR; 2.0-3.0),[31] the percentage of time above the therapeutic INR range (>3.0), the time to a therapeutic INR (defined as three consecutive INR measurements in the range between 2.0 and 3.0), and the time to first INR above 4.5.

During follow-up, patients were contacted semi-annually, alternating between face-to-face evaluations and telephone calls. Detailed information regarding the date, type and circumstance of outcomes were collected from the patient, and/or the patient's family members, primary care physician and medical chart. We also recorded all INR values measured by the family physician during the follow-up period.

All outcomes were reviewed and adjudicated by an independent committee of three blinded clinical experts. Based on the full consensus of this committee, deaths were classified as definitely due to PE (e.g., confirmed by autopsy or following severe PE), possibly due to PE (e.g., sudden death without obvious cause), due to major bleeding and due to other causes. Bleeding-related death was defined as

death following intracranial hemorrhage or hemodynamic deterioration due to major bleeding.[32]

Statistical analyses

We present Kaplan-Meier curves for any clinical event and mortality, as well as cumulative incidence curves adjusted for the competing risk of death for major bleeding, clinically relevant non-major bleeding, and recurrent VTE [33]. We examined the association between *CYP2C9* and *VKORC1* variants and any clinical outcome event and death using Cox regression using robust standard errors, adjusting for previously published predictors of the respective outcome as well as periods of anticoagulation as a time-varying covariate.[34-36]

We examined associations between *CYP2C9* and *VKORC1* variants and the time to a first VTE recurrence, major bleeding, and clinically relevant non-major bleeding using competing risk regression according to Fine and Gray,[37] accounting for non-PE- or non-bleeding-related death as a competing event. We adjusted the models for previously published predictors of bleeding and VTE recurrence.[34, 38-54] As we aimed to predict overall adverse events, we considered the full observation period from diagnosis until the end of follow-up, adjusting for periods of anticoagulation as a time-varying covariate.

We calculated the percentage of time spent within one of three specified INR ranges (<2.0, 2.0-3.0, >3.0) using the Rosendaal method [31] and compared groups using linear regression. We examined the association between *CYP2C9* and *VKORC1* variants and the time to the first therapeutic INR and the time to the first INR >4.5 using Cox-regression with robust standard errors. Only the initial VKA treatment was considered. Time was censored at stop of initial treatment, death, or

withdrawal/loss of follow-up. Analyses were adjusted for known influencing factors of anticoagulation quality, including age, sex, provoked VTE, active cancer, diabetes, heart failure, chronic pulmonary disease, chronic liver disease, renal failure (GFR <30 ml/min.), body mass index, hypertension, and smoking status.[55-57] All analyses were done using Stata 13 (Stata Corporation, College Station, Texas).

Results

Patient characteristics

Of 1863 patients screened, 1003 were included in the study cohort and 744 were analyzed for the purpose of this study (Figure 1). Analyzed patients did not differ from non-analyzed patients in terms of age (median age 75 vs. 74 years) and sex (women 46% vs. 50%).

Overall, 78% of patients (n=604) had a *CYP2C9* or *VKORC1* variant. *CYP2C9*2* polymorphisms were found in 26% of patients (n=200), *CYP2C9*3* in 12% (n=95), *VKORC1* in 66% (n=510), and both *CYP29C* and *VKORC1* variants in 24% (n=187). Patient baseline characteristics did not differ in patients with and without a genetic variant (Table 1). Although patients with a genetic variant had a shorter initial parenteral anticoagulation duration than patients without variant (9 vs. 12 days), the duration of the initial anticoagulation with vitamin K antagonists was comparable (9.7 vs. 8.4 months) (Table 1).

Clinical outcomes

The median observation time was 30 months (interquartile range 24 to 41 months). The primary outcome, the composite endpoint of death, major or non-major clinically relevant bleeding, or VTE recurrence, occurred in 43% of patients (n=334).

Overall, 15% of patients died (n=119), 13% (n=100) had major bleeding, 22% (n=167) clinically relevant non-major bleeding, and 13% (n=100) recurrent VTE.

While patients with a *CYP2C9* variant had a higher cumulative 3-year incidence of any clinical outcome event than those without these variants (52.3% vs. 41.1%; Figure 2), the cumulative incidence did not differ much in patients with or without *VKORC1* variants (47.1% vs. 41.8%). Patients with a *CYP2C9* variant had a higher cumulative 3-year incidence of death and clinically relevant non-major bleeding but not major bleeding or recurrent VTE (Figure 3).

After adjustment, *CYP2C9* variants were associated with any clinical outcome event (hazard ratio [HR] 1.34; 95% confidence interval [CI] 1.08, 1.66), death (HR 1.74; 95% CI 1.19, 2.52), and clinically relevant non-major bleeding (sub-hazard ratio [SHR] 1.39; 95% CI 1.02, 1.89), but not with major bleeding (SHR 1.03; 95% CI 0.69, 1.55) or recurrent VTE (SHR 0.95; 95% CI 0.62, 1.44) (Table 2). Patients with both *CYP2C9* and *VKORC1* variants had a higher risk of any clinical event (HR 1.40; 95% CI 1.11, 1.77) and clinically relevant non-major bleeding (SHR 1.78; 95% CI 1.30, 2.44).

Quality of anticoagulation

Associations between *CYP2C9* and *VKORC1* variants and measures of anticoagulation quality are shown in Table 3. Compared to patients without *CYP2C9* variants, patients with a *CYP2C9* variant spent less time in the therapeutic INR range (adjusted difference -3.7%; 95% CI -7.0, -0.4). Similarly, patients with *CYP2C9* variants spent more time with an INR above the therapeutic range (adjusted difference 3.3; 95% CI 0.8, 5.7), were less likely to achieve a therapeutic INR (adjusted HR 0.81; 95% CI 0.68, 0.96), and more likely to have an INR >4.5

(adjusted HR 1.42; 95% CI 1.11, 1.81). Patients with a *VKORC1* variant were more likely to have an INR >4.5 (adjusted HR 1.43; 95% CI 1.10, 1.85).

Discussion

Our results demonstrated a association between *CYP2C9* genetic variants and the composite endpoint of death, major bleeding, clinically relevant non-major bleeding, and recurrent VTE. This result was driven by a 74% higher mortality and a 39% higher risk of clinically relevant non-major bleeding in patients with *CYP2C9* variants. While patients with genetic variants, in particular *CYP2C9,* had a slightly lower anticoagulation quality, we found no relationship between genetic variants and major bleeding or recurrent VTE.

To our knowledge, our work is the first study to demonstrate a association between *CYP2C9* variants and mortality. Interestingly, the increased mortality does not appear to be driven by a higher rate of major bleeding or recurrent VTE. Several explanations for the higher mortality rate in patients with *CYP2C9* are possible. First, genetic variants may affect mortality based on mechanisms not related to anticoagulation quality. A small observational study demonstrated a higher risk of stroke and myocardial infarction in patients with *VKORC1* variants, the underlying mechanism for the increased risk of cardiovascular complications remaining unexplained.[58] Second, although we adjusted for many co-variates, we cannot fully exclude the possibility that the higher mortality in patients with *CYP2C9* variants is caused by residual confounding.

We did not observe an increased risk of major bleeding in patients with *CYP2C9* or *VKORC1* variants. This finding is consistent with previous studies focusing on short-term clinical outcomes. In a systematic review by Jorgensen et al.,

which pooled data from several smaller-sized observational studies, no association between genetic variants and major bleeding was reported.[22] Similarly, in a sub-study of the ENGAGE AF-TIMI 48 phase III clinical trial (edoxaban for stroke prophylaxis in patients with atrial fibrillation), no association between *CYP2C9/VKORC1* variants and major bleeding was found.[59] However, as demonstrated in several smaller observational studies,[60-62] we observed a association between *CYP2C9* variants and clinically relevant non-major bleeding (adjusted SHR 1.39; 95% CI 1.02, 1.89).

We did not find an association between genetic variants and recurrent VTE. This result is consistent with the findings of a small retrospective study conducted in 120 patients with mechanical heart valves, which did not report an increased risk of VTE in patients with genetic variants.[58]

In our cohort, genetic variants, in particular *CYP2C9*, were associated with a lower anticoagulation quality, but the absolute differences in anticoagulation quality measures were small. Even though prior studies have shown a consistent association between genetic variants and VKA dose requirements,[14, 61, 63-69] the evidence with regard to anticoagulation quality is much more limited. In a systematic review, the magnitude of associations between *CYP2C9* and *VKORC1* genetic variants and different measures of anticoagulation quality was moderate at best.[22] Randomized-controlled trials comparing genetic-based dosing schedules for initiation of anticoagulation to standard care did not demonstrate a relevant benefit in terms of quality of anticoagulation or clinical outcomes.[24-27] Overall, the results from our cohort of elderly patients indicate that although patients with genetic variants appear to have a somewhat lower anticoagulation quality and a higher risk of non-major bleeding (*CYP2C9)*, but they do not have an increased risk of major bleeding. Thus,

the costly measurement of *CYP2C9* and *VKORC1* variants is unlikely to have a relevant beneficial effect on anticoagulation quality and clinical outcomes of elderly patients with VTE. Interestingly, associations between genetic variants and anticoagulation quality as well as clinical outcomes were much more prominent in case of CYP2C9. This observation suggests that fluctuations in VKA concentrations are more important than VKA sensitivity.

Our study has several strengths. In contrast to prior studies, our study had a long-term follow-up, larger sample size, and clinically relevant outcome variables. [14, 22, 24-27, 59-69] In addition, we focused on elderly patients, a population at risk for both bleeding complications and death.[70] The fact that we enrolled patients from university and non-university hospitals increases the generalizability of our findings. Because patients and managing physicians were blinded to the results of the genetic analyses, a performance bias is unlikely.

Our study has several potential limitations. First, our study included patients aged 65 years or older only. Thus, the results may not be generalizable to younger patients. Second, our study included exclusively patients with VTE and therefore, our results may not be generalizable to other populations, such as patients with atrial fibrillation. Finally, even though we adjusted our analyses for many covariates, we might have missed important predictor variables.

In conclusion, we found a association between *CYP2C9* genetic variants and long-term clinical outcomes, notably overall mortality and clinically relevant non-major bleeding. In contrast, we found no relationship between *VKORC1* variants and clinical outcomes. While genetic variants, in particular *CYP2C9*, appear to be associated with a somewhat lower anticoagulation quality, there was no relationship between genetic variants and major bleeding or recurrent VTE.

Addendum

M. Nagler, A. Limacher, A. Angelillo-Scherrer, and D. Aujesky developed the protocol and the analysis plan, conducted the analyses, and drafted the manuscript. C. Abbal conducted the genetic analysis, and intellectually reviewed the manuscript. M. Méan, A. Angelillo-Scherrer, M. Righini, B. Frauchiger, J. Osterwalder, N. Kucher, and N. Rodondi organized data collection, intellectually reviewed the manuscript, and participated in funding procedure. J. H. Beer, C. M. Matter, J. Cornuz, M. Banyai, M. Aschwanded, M. Husmann, D. Staub, L. Mazzolai, and O. Hugli organized data collection, and intellectually reviewed the manuscript. D. Aujesky was principle investigator and was responsible for planning of the study, data collection, drafting of the manuscript, and obtaining funding.

Acknowledgements

The SWITCO65+ cohort study was funded by grants of the Swiss National Science Foundation (no. 33CSCO-122659/139 470). We gratefully acknowledge all collaborators.

Disclosure of Conflict of Interests

M. Nagler has received research grants or lecture fees from Bayer, Roche diagnostics, and CSL Behring. O. Hugli participated in an advisor board for Novartis. M. Husmann received lecture or travel fees from Abbott Endovascular, Medtronic, EV3, Cordis, Sanofi Aventis, Daiichi Sankyo, Bayer, Boeringer Ingelheim, and AstraZeneca. C. M. Matter reports grants from MSD, AstraZeneca, EliLilly, Bayer, and SIrtris; and personal fees from MSD and Amgen, outside the submitted work.

References

1 Charlab R, Zhang L. Pharmacogenomics: historical perspective and current status. *Methods in molecular biology*. 2013; **1015**: 3-22. 10.1007/978-1-62703-435-7_1.

2 Hatz MH, Schremser K, Rogowski WH. Is individualized medicine more cost-effective? A systematic review. *PharmacoEconomics*. 2014; **32**: 443-55. 10.1007/s40273-014-0143-0.

3 Pacanowski MA, Leptak C, Zineh I. Next-generation medicines: past regulatory experience and considerations for the future. *Clinical pharmacology and therapeutics*. 2014; **95{Pacanowski, 2014 #4}**: 247-9. 10.1038/clpt.2013.222.

4 Zineh I, Pacanowski M, Woodcock J. Pharmacogenetics and coumarin dosing--recalibrating expectations. *The New England journal of medicine*. 2013; **369**: 2273-5. 10.1056/NEJMp1314529.

5 Waldman SA, Terzic A. Pharmacoeconomics in the era of individualized medicine. *Clinical pharmacology and therapeutics*. 2008; **84**: 179-82. 10.1038/clpt.2008.142.

6 Shaw K, Amstutz U, Kim RB, Lesko LJ, Turgeon J, Michaud V, Hwang S, Ito S, Ross C, Carleton BC, Group CCR. Clinical Practice Recommendations on Genetic Testing of CYP2C9 and VKORC1 Variants in Warfarin Therapy. *Therapeutic drug monitoring*. 2015; **37**: 428-36. 10.1097/FTD.0000000000000192.

7 Abohelaika S, Wynne H, Avery P, Kamali F. Influence of CYP2C9 polymorphism on the fall in International Normalized Ratio in patients interrupting warfarin therapy before elective surgery. *Journal of thrombosis and haemostasis : JTH*. 2015; **13**: 1436-40. 10.1111/jth.13014.

8 Kirley K, Qato DM, Kornfield R, Stafford RS, Alexander GC. National trends in oral anticoagulant use in the United States, 2007 to 2011. *Circulation Cardiovascular quality and outcomes*. 2012; **5**: 615-21. 10.1161/CIRCOUTCOMES.112.967299.

9 Furie B. Do pharmacogenetics have a role in the dosing of vitamin K antagonists? *The New England journal of medicine*. 2013; **369**: 2345-6. 10.1056/NEJMe1313682.

10 Connolly SJ, Pogue J, Eikelboom J, Flaker G, Commerford P, Franzosi MG, Healey JS, Yusuf S, Investigators AW. Benefit of oral anticoagulant over antiplatelet therapy in atrial fibrillation depends on the quality of international normalized ratio control achieved by centers and countries as measured by time in therapeutic range. *Circulation*. 2008; **118**: 2029-37. 10.1161/CIRCULATIONAHA.107.750000.

11 Gallagher AM, de Vries F, Plumb JM, Hass B, Clemens A, van Staa TP. Quality of INR control and outcomes following venous thromboembolism. *Clin Appl Thromb Hemost*. 2012; **18**: 370-8. 10.1177/1076029611426139.

12 Voora D, McLeod HL, Eby C, Gage BF. The pharmacogenetics of coumarin therapy. *Pharmacogenomics*. 2005; **6**: 503-13. 10.2217/14622416.6.5.503.

13 Aithal GP, Day CP, Kesteven PJ, Daly AK. Association of polymorphisms in the cytochrome P450 CYP2C9 with warfarin dose requirement and risk of bleeding complications. *Lancet*. 1999; **353**: 717-9. 10.1016/S0140-6736(98)04474-2.

14 Sconce EA, Khan TI, Wynne HA, Avery P, Monkhouse L, King BP, Wood P, Kesteven P, Daly AK, Kamali F. The impact of CYP2C9 and VKORC1 genetic polymorphism and patient characteristics upon warfarin dose requirements: proposal for a new dosing regimen. *Blood*. 2005; **106**: 2329-33. 10.1182/blood-2005-03-1108.

15 Aquilante CL, Langaee TY, Lopez LM, Yarandi HN, Tromberg JS, Mohuczy D, Gaston KL, Waddell CD, Chirico MJ, Johnson JA. Influence of coagulation factor,

vitamin K epoxide reductase complex subunit 1, and cytochrome P450 2C9 gene polymorphisms on warfarin dose requirements. *Clinical pharmacology and therapeutics*. 2006; **79**: 291-302. 10.1016/j.clpt.2005.11.011.
16 Higashi MK, Veenstra DL, Kondo LM, Wittkowsky AK, Srinouanprachanh SL, Farin FM, Rettie AE. Association between CYP2C9 genetic variants and anticoagulation-related outcomes during warfarin therapy. *Jama*. 2002; **287**: 1690-8.
17 D'Andrea G, D'Ambrosio RL, Di Perna P, Chetta M, Santacroce R, Brancaccio V, Grandone E, Margaglione M. A polymorphism in the VKORC1 gene is associated with an interindividual variability in the dose-anticoagulant effect of warfarin. *Blood*. 2005; **105**: 645-9. 10.1182/blood-2004-06-2111.
18 Hillman MA, Wilke RA, Caldwell MD, Berg RL, Glurich I, Burmester JK. Relative impact of covariates in prescribing warfarin according to CYP2C9 genotype. *Pharmacogenetics*. 2004; **14**: 539-47.
19 Carlquist JF, Horne BD, Muhlestein JB, Lappe DL, Whiting BM, Kolek MJ, Clarke JL, James BC, Anderson JL. Genotypes of the cytochrome p450 isoform, CYP2C9, and the vitamin K epoxide reductase complex subunit 1 conjointly determine stable warfarin dose: a prospective study. *Journal of thrombosis and thrombolysis*. 2006; **22**: 191-7. 10.1007/s11239-006-9030-7.
20 Rieder MJ, Reiner AP, Gage BF, Nickerson DA, Eby CS, McLeod HL, Blough DK, Thummel KE, Veenstra DL, Rettie AE. Effect of VKORC1 haplotypes on transcriptional regulation and warfarin dose. *The New England journal of medicine*. 2005; **352**: 2285-93. 10.1056/NEJMoa044503.
21 Wadelius M, Chen LY, Downes K, Ghori J, Hunt S, Eriksson N, Wallerman O, Melhus H, Wadelius C, Bentley D, Deloukas P. Common VKORC1 and GGCX polymorphisms associated with warfarin dose. *The pharmacogenomics journal*. 2005; **5**: 262-70. 10.1038/sj.tpj.6500313.
22 Jorgensen AL, FitzGerald RJ, Oyee J, Pirmohamed M, Williamson PR. Influence of CYP2C9 and VKORC1 on patient response to warfarin: a systematic review and meta-analysis. *PloS one*. 2012; **7**: e44064. 10.1371/journal.pone.0044064.
23 Gong IY, Schwarz UI, Crown N, Dresser GK, Lazo-Langner A, Zou G, Roden DM, Stein CM, Rodger M, Wells PS, Kim RB, Tirona RG. Clinical and genetic determinants of warfarin pharmacokinetics and pharmacodynamics during treatment initiation. *PloS one*. 2011; **6**: e27808. 10.1371/journal.pone.0027808.
24 Kimmel SE, French B, Kasner SE, Johnson JA, Anderson JL, Gage BF, Rosenberg YD, Eby CS, Madigan RA, McBane RB, Abdel-Rahman SZ, Stevens SM, Yale S, Mohler ER, 3rd, Fang MC, Shah V, Horenstein RB, Limdi NA, Muldowney JA, 3rd, Gujral J, Delafontaine P, Desnick RJ, Ortel TL, Billett HH, Pendleton RC, Geller NL, Halperin JL, Goldhaber SZ, Caldwell MD, Califf RM, Ellenberg JH, Investigators C. A pharmacogenetic versus a clinical algorithm for warfarin dosing. *The New England journal of medicine*. 2013; **369**: 2283-93. 10.1056/NEJMoa1310669.
25 Pirmohamed M, Burnside G, Eriksson N, Jorgensen AL, Toh CH, Nicholson T, Kesteven P, Christersson C, Wahlstrom B, Stafberg C, Zhang JE, Leathart JB, Kohnke H, Maitland-van der Zee AH, Williamson PR, Daly AK, Avery P, Kamali F, Wadelius M, Group E-P. A randomized trial of genotype-guided dosing of warfarin. *The New England journal of medicine*. 2013; **369**: 2294-303. 10.1056/NEJMoa1311386.
26 Verhoef TI, Ragia G, de Boer A, Barallon R, Kolovou G, Kolovou V, Konstantinides S, Le Cessie S, Maltezos E, van der Meer FJ, Redekop WK, Remkes

M, Rosendaal FR, van Schie RM, Tavridou A, Tziakas D, Wadelius M, Manolopoulos VG, Maitland-van der Zee AH, Group E-P. A randomized trial of genotype-guided dosing of acenocoumarol and phenprocoumon. *The New England journal of medicine*. 2013; **369**: 2304-12. 10.1056/NEJMoa1311388.
27 Stergiopoulos K, Brown DL. Genotype-guided vs clinical dosing of warfarin and its analogues: meta-analysis of randomized clinical trials. *JAMA internal medicine*. 2014; **174**: 1330-8. 10.1001/jamainternmed.2014.2368.
28 Mean M, Righini M, Jaeger K, Beer HJ, Frauchiger B, Osterwalder J, Kucher N, Lammle B, Cornuz J, Angelillo-Scherrer A, Rodondi N, Limacher A, Trelle S, Matter CM, Husmann M, Banyai M, Aschwanden M, Egloff M, Mazzolai L, Hugli O, Bounameaux H, Aujesky D. The Swiss cohort of elderly patients with venous thromboembolism (SWITCO65+): rationale and methodology. *Journal of thrombosis and thrombolysis*. 2013; **36**: 475-83. 10.1007/s11239-013-0875-2.
29 Mean M, Aujesky D, Lammle B, Gerschheimer C, Trelle S, Angelillo-Scherrer A. Design and establishment of a biobank in a multicenter prospective cohort study of elderly patients with venous thromboembolism (SWITCO65+). *Journal of thrombosis and thrombolysis*. 2013; **36**: 484-91. 10.1007/s11239-013-0922-z.
30 Schulman S, Kearon C, Subcommittee on Control of Anticoagulation of the S, Standardization Committee of the International Society on T, Haemostasis. Definition of major bleeding in clinical investigations of antihemostatic medicinal products in non-surgical patients. *Journal of thrombosis and haemostasis : JTH*. 2005; **3**: 692-4. 10.1111/j.1538-7836.2005.01204.x.
31 Rosendaal FR, Cannegieter SC, van der Meer FJ, Briet E. A method to determine the optimal intensity of oral anticoagulant therapy. *Thrombosis and haemostasis*. 1993; **69**: 236-9.
32 Jakobsson C, Jimenez D, Gomez V, Zamarro C, Mean M, Aujesky D. Validation of a clinical algorithm to identify low-risk patients with pulmonary embolism. *Journal of thrombosis and haemostasis : JTH*. 2010; **8**: 1242-7. 10.1111/j.1538-7836.2010.03836.x.
33 Coviello V, Boggess M. Cumulative incidence estimation in the presence of competing risks. *The Stata Journal*. 2004; **4**: 103-12.
34 Gussoni G, Frasson S, La Regina M, Di Micco P, Monreal M, Investigators R. Three-month mortality rate and clinical predictors in patients with venous thromboembolism and cancer. Findings from the RIETE registry. *Thrombosis research*. 2013; **131**: 24-30. 10.1016/j.thromres.2012.10.007.
35 Jimenez D, Aujesky D, Moores L, Gomez V, Lobo JL, Uresandi F, Otero R, Monreal M, Muriel A, Yusen RD, Investigators R. Simplification of the pulmonary embolism severity index for prognostication in patients with acute symptomatic pulmonary embolism. *Archives of internal medicine*. 2010; **170**: 1383-9. 10.1001/archinternmed.2010.199.
36 Spirk D, Husmann M, Hayoz D, Baldi T, Frauchiger B, Engelberger R, Amann-Vesti B, Baumgartner I, Kucher N. Predictors of in-hospital mortality in elderly patients with acute venous thrombo-embolism: the SWIss Venous ThromboEmbolism Registry (SWIVTER). *European heart journal*. 2012; **33**: 921-6. 10.1093/eurheartj/ehr392.
37 Fine JP, Gray RJ. A proportional hazards model for the subdistribution of a competing risk. *J Am Stat Assoc*. 1999: 496-509.
38 Kuijer PM, Hutten BA, Prins MH, Buller HR. Prediction of the risk of bleeding during anticoagulant treatment for venous thromboembolism. *Archives of internal medicine*. 1999; **159**: 457-60.

39 Fang MC, Go AS, Chang Y, Borowsky LH, Pomernacki NK, Udaltsova N, Singer DE. A new risk scheme to predict warfarin-associated hemorrhage: The ATRIA (Anticoagulation and Risk Factors in Atrial Fibrillation) Study. *Journal of the American College of Cardiology*. 2011; **58**: 395-401. 10.1016/j.jacc.2011.03.031.
40 Pisters R, Lane DA, Nieuwlaat R, de Vos CB, Crijns HJ, Lip GY. A novel user-friendly score (HAS-BLED) to assess 1-year risk of major bleeding in patients with atrial fibrillation: the Euro Heart Survey. *Chest*. 2010; **138**: 1093-100. 10.1378/chest.10-0134.
41 Olesen JB, Lip GY, Hansen PR, Lindhardsen J, Ahlehoff O, Andersson C, Weeke P, Hansen ML, Gislason GH, Torp-Pedersen C. Bleeding risk in 'real world' patients with atrial fibrillation: comparison of two established bleeding prediction schemes in a nationwide cohort. *Journal of thrombosis and haemostasis : JTH*. 2011; **9**: 1460-7. 10.1111/j.1538-7836.2011.04378.x.
42 Prandoni P, Lensing AW, Piccioli A, Bernardi E, Simioni P, Girolami B, Marchiori A, Sabbion P, Prins MH, Noventa F, Girolami A. Recurrent venous thromboembolism and bleeding complications during anticoagulant treatment in patients with cancer and venous thrombosis. *Blood*. 2002; **100**: 3484-8. 10.1182/blood-2002-01-0108.
43 Ruiz-Gimenez N, Suarez C, Gonzalez R, Nieto JA, Todoli JA, Samperiz AL, Monreal M, Investigators R. Predictive variables for major bleeding events in patients presenting with documented acute venous thromboembolism. Findings from the RIETE Registry. *Thrombosis and haemostasis*. 2008; **100**: 26-31. 10.1160/TH08-03-0193.
44 White RH, Beyth RJ, Zhou H, Romano PS. Major bleeding after hospitalization for deep-venous thrombosis. *The American journal of medicine*. 1999; **107**: 414-24.
45 Lip GY, Frison L, Halperin JL, Lane DA. Comparative validation of a novel risk score for predicting bleeding risk in anticoagulated patients with atrial fibrillation: the HAS-BLED (Hypertension, Abnormal Renal/Liver Function, Stroke, Bleeding History or Predisposition, Labile INR, Elderly, Drugs/Alcohol Concomitantly) score. *Journal of the American College of Cardiology*. 2011; **57**: 173-80. 10.1016/j.jacc.2010.09.024.
46 Beyth RJ, Quinn LM, Landefeld CS. Prospective evaluation of an index for predicting the risk of major bleeding in outpatients treated with warfarin. *The American journal of medicine*. 1998; **105**: 91-9.
47 Shireman TI, Mahnken JD, Howard PA, Kresowik TF, Hou Q, Ellerbeck EF. Development of a contemporary bleeding risk model for elderly warfarin recipients. *Chest*. 2006; **130**: 1390-6. 10.1378/chest.130.5.1390.
48 van der Meer FJ, Rosendaal FR, Vandenbroucke JP, Briet E. Bleeding complications in oral anticoagulant therapy. An analysis of risk factors. *Archives of internal medicine*. 1993; **153**: 1557-62.
49 Torn M, Bollen WL, van der Meer FJ, van der Wall EE, Rosendaal FR. Risks of oral anticoagulant therapy with increasing age. *Archives of internal medicine*. 2005; **165**: 1527-32. 10.1001/archinte.165.13.1527.
50 Kyrle PA, Rosendaal FR, Eichinger S. Risk assessment for recurrent venous thrombosis. *Lancet*. 2010; **376**: 2032-9. 10.1016/S0140-6736(10)60962-2.
51 Cannegieter SC, van Hylckama Vlieg A. Venous thrombosis: understanding the paradoxes of recurrence. *Journal of thrombosis and haemostasis : JTH*. 2013; **11 Suppl 1**: 161-9. 10.1111/jth.12263.

52 Kearon C, Akl EA, Ornelas J, Blaivas A, Jimenez D, Bounameaux H, Huisman M, King CS, Morris TA, Sood N, Stevens SM, Vintch JR, Wells P, Woller SC, Moores L. Antithrombotic Therapy for VTE Disease: CHEST Guideline and Expert Panel Report. *Chest*. 2016; **149**: 315-52. 10.1016/j.chest.2015.11.026.
53 Lijfering WM, Rosendaal FR, Cannegieter SC. Risk factors for venous thrombosis - current understanding from an epidemiological point of view. *British journal of haematology*. 2010; **149**: 824-33. 10.1111/j.1365-2141.2010.08206.x.
54 Iorio A, Kearon C, Filippucci E, Marcucci M, Macura A, Pengo V, Siragusa S, Palareti G. Risk of recurrence after a first episode of symptomatic venous thromboembolism provoked by a transient risk factor: a systematic review. *Archives of internal medicine*. 2010; **170**: 1710-6. 10.1001/archinternmed.2010.367.
55 Schaefer C, Wuillemin WA, Kessels A, Jacobson A, Nagler M. Predictors of anticoagulation quality in 15 834 patients performing patient self-management of oral anticoagulation with vitamin K antagonists in real-life practice: a survey of the International Self-Monitoring Association of Orally Anticoagulated Patients. *British journal of haematology*. 2016. 10.1111/bjh.14273.
56 Nilsson H, Grove EL, Larsen TB, Nielsen PB, Skjoth F, Maegaard M, Christensen TD. Sex differences in treatment quality of self-managed oral anticoagulant therapy: 6,900 patient-years of follow-up. *PloS one*. 2014; **9**: e113627. 10.1371/journal.pone.0113627.
57 Apostolakis S, Sullivan RM, Olshansky B, Lip GY. Factors affecting quality of anticoagulation control among patients with atrial fibrillation on warfarin: the SAMe-TT(2)R(2) score. *Chest*. 2013; **144**: 1555-63. 10.1378/chest.13-0054.
58 Giansante C, Fiotti N, Altamura N, Pitacco P, Consoloni L, Scardi S, Mazzone C, Grassi G, Pandullo C, Di Lenarda A. Oral anticoagulation and VKORC1 polymorphism in patients with a mechanical heart prosthesis: a 6-year follow-up. *Journal of thrombosis and thrombolysis*. 2012; **34**: 506-12. 10.1007/s11239-012-0740-8.
59 Mega JL, Walker JR, Ruff CT, Vandell AG, Nordio F, Deenadayalu N, Murphy SA, Lee J, Mercuri MF, Giugliano RP, Antman EM, Braunwald E, Sabatine MS. Genetics and the clinical response to warfarin and edoxaban: findings from the randomised, double-blind ENGAGE AF-TIMI 48 trial. *Lancet*. 2015; **385**: 2280-7. 10.1016/S0140-6736(14)61994-2.
60 Lund K, Gaffney D, Spooner R, Etherington AM, Tansey P, Tait RC. Polymorphisms in VKORC1 have more impact than CYP2C9 polymorphisms on early warfarin International Normalized Ratio control and bleeding rates. *British journal of haematology*. 2012; **158**: 256-61. 10.1111/j.1365-2141.2012.09150.x.
61 Gaikwad T, Ghosh K, Kulkarni B, Kulkarni V, Ross C, Shetty S. Influence of CYP2C9 and VKORC1 gene polymorphisms on warfarin dosage, over anticoagulation and other adverse outcomes in Indian population. *European journal of pharmacology*. 2013; **710**: 80-4. 10.1016/j.ejphar.2013.04.006.
62 Tomek A, Matoska V, Kolarova T, Neumann J, Sramek M, Sarbochova I, Taborsky L, Bojar M, Goetz P, Serebruany VL. The bleeding risk during warfarin therapy is associated with the number of variant alleles of CYP2C9 and VKORC1 genes. *Cardiology*. 2013; **125**: 182-91. 10.1159/000350407.
63 Limdi NA, Brown TM, Yan Q, Thigpen JL, Shendre A, Liu N, Hill CE, Arnett DK, Beasley TM. Race influences warfarin dose changes associated with genetic factors. *Blood*. 2015; **126**: 539-45. 10.1182/blood-2015-02-627042.
64 Tatarunas V, Lesauskaite V, Veikutiene A, Grybauskas P, Jakuska P, Jankauskiene L, Bartuseviciute R, Benetis R. The effect of CYP2C9, VKORC1 and

CYP4F2 polymorphism and of clinical factors on warfarin dosage during initiation and long-term treatment after heart valve surgery. *Journal of thrombosis and thrombolysis*. 2014; **37**: 177-85. 10.1007/s11239-013-0940-x.
65 Miyagata Y, Nakai K, Sugiyama Y. Clinical significance of combined CYP2C9 and VKORC1 genotypes in Japanese patients requiring warfarin. *International heart journal*. 2011; **52**: 44-9.
66 Moreau C, Pautas E, Gouin-Thibault I, Golmard JL, Mahe I, Mulot C, Loriot MA, Siguret V. Predicting the warfarin maintenance dose in elderly inpatients at treatment initiation: accuracy of dosing algorithms incorporating or not VKORC1/CYP2C9 genotypes. *Journal of thrombosis and haemostasis : JTH*. 2011; **9**: 711-8. 10.1111/j.1538-7836.2011.04213.x.
67 Vear SI, Ayers GD, Van Driest SL, Sidonio RF, Stein CM, Ho RH. The impact of age and CYP2C9 and VKORC1 variants on stable warfarin dose in the paediatric population. *British journal of haematology*. 2014; **165**: 832-5. 10.1111/bjh.12817.
68 Biss TT, Avery PJ, Brandao LR, Chalmers EA, Williams MD, Grainger JD, Leathart JB, Hanley JP, Daly AK, Kamali F. VKORC1 and CYP2C9 genotype and patient characteristics explain a large proportion of the variability in warfarin dose requirement among children. *Blood*. 2012; **119**: 868-73. 10.1182/blood-2011-08-372722.
69 Nowak-Gottl U, Dietrich K, Schaffranek D, Eldin NS, Yasui Y, Geisen C, Mitchell LG. In pediatric patients, age has more impact on dosing of vitamin K antagonists than VKORC1 or CYP2C9 genotypes. *Blood*. 2010; **116**: 6101-5. 10.1182/blood-2010-05-283861.
70 Lip GY, Clementy N, Pericart L, Banerjee A, Fauchier L. Stroke and major bleeding risk in elderly patients aged >/=75 years with atrial fibrillation: the Loire Valley atrial fibrillation project. *Stroke*. 2015; **46**: 143-50. 10.1161/STROKEAHA.114.007199.

Figure Legends

Figure 1. Patient flow chart. *Multiple reasons may apply.

Figure 2. Kaplan-Meier estimates of a first clinical event (death, major or clinically relevant non-major bleeding, or VTE recurrence) according to presence of *CYP2C9* or *VKORC1* variants. The cumulative 3-year incidence of a first clinical event was 52.3% for patients with a *CYP2C9* variant vs. 41.1% for patients without variant and 47.1% for patients with a *VKORC1* variant vs. 41.8% for patients without variant.

Figure 3. Cumulative incidences of (A) death, (B) major bleeding, (C) clinically relevant non-major bleeding, and (D) VTE recurrences according to the presence of *CYP2C9* or *VKORC1* variants. Compared to patients without variant, patients with a *CYP2C9* variant had a higher cumulative 3-year incidence of death (20.4% vs. 12.4%) and non-major clinically relevant bleeding (27.5% vs. 21.3%). Graphs were adjusted for the competing risk of death if appropriate.

Table 1. Baseline characteristics by presence of any genetic variant

	All *(N=774)*	Any genetic variant* *(N=604)*	No genetic variant *(N=170)*	Missing values
	n (%) or median (IQR)			*n (%)*
Age, years	75.0 (69.0; 81.0)	75.0 (69.0; 81.0)	74.5 (69.0; 81.0)	0 (0)
Female sex	355 (46)	280 (46)	75 (44)	0 (0)
BMI, kg/m²	26.8 (24.3; 29.9)	26.8 (24.3; 29.8)	26.9 (24.3; 30.6)	4 (1)
Smoking status				2 (1)
current smoker	58 (8)	47 (8)	11 (6)	
past smoker	315 (41)	248 (41)	67 (39)	
never smoker	399 (52)	307 (51)	92 (54)	
Localization of thrombosis				0 (0)
distal DVT only	53 (7)	44 (7)	9 (5)	
proximal DVT	165 (21)	127 (21)	38 (22)	
PE	556 (72)	433 (72)	123 (72)	
Prior VTE	229 (30)	181 (30)	48 (28)	0 (0)
Family history of PE/DVT	140 (18)	112 (19)	28 (16)	7 (1)
Provoked index VTE	214 (28)	171 (28)	43 (25)	0 (0)
Active cancer†	82 (11)	60 (10)	22 (13)	0 (0)
Arterial hypertension	500 (65)	392 (65)	108 (64)	0 (0)
Diabetes mellitus	120 (16)	89 (15)	31 (18)	0 (0)
Chronic heart failure	91 (12)	72 (12)	19 (11)	0 (0)
Acute rheumatic disease during the last 3 months	25 (3)	21 (3)	4 (2)	0 (0)
Inflammatory bowel disease	27 (3)	19 (3)	8 (5)	0 (0)
History of major bleeding‡	72 (9)	52 (9)	20 (12)	1 (0)
Cerebrovascular disease (stroke, TIA)	72 (9)	59 (10)	13 (8)	0 (0)
Chronic renal disease	142 (18)	113 (19)	29 (17)	0 (0)
Chronic pulmonary disease	104 (13)	87 (14)	17 (10)	0 (0)

Chronic liver disease	11 (1)	9 (1)	2 (1)	0 (0)
Anemia[§]	267 (37)	214 (38)	53 (34)	49 (6)
Creatinine >107 µmol/L	185 (26)	146 (26)	39 (26)	63 (8)
Systolic BP <100 mm Hg	27 (4)	20 (3)	7 (4)	13 (2)
Heart rate ≥110/min.	73 (10)	214 (38)	53 (34)	15 (2)
Immobilization during the last 3 months	154 (20)	122 (20)	32 (19)	0 (0)
Antiplatelet therapy	254 (33)	189 (31)	65 (38)	0 (0)
NSAID	72 (9)	62 (10)	10 (6)	0 (0)
Polypharmacy[¶]	388 (50)	309 (51)	79 (46)	0 (0)
Duration of initial parenteral AC, days	9.0 (6.0; 16.0)	9.0 (6.0; 14.0)	12.0 (7.0; 19.0)	0 (0)
Duration of initial VKA therapy, months	9.4 (5.3; 29.1)	9.7 (5.2; 29.0)	8.4 (5.4; 29.2)	0 (0)

Abbreviations: IQR= interquartile range, BMI=body mass index, DVT= deep vein thrombosis; PE= pulmonary embolism; VTE= venous thromboembolism; TIA= transient ischemic attack; BP= blood pressure; NSAID= nonsteroidal anti-inflammatory drug; AC= anticoagulation; VKA= vitamin K antagonist

* *CYP2C9*2*, *CYP2C9*3*, or *VKORC1 G1639A* genetic variant

† Solid or hematologic cancer requiring chemotherapy, radiotherapy, surgery, and/or palliative care during the last 3 months

‡ Bleeding that led to a hospital stay or transfusions

§ Serum hemoglobin <130 g/L for men or <120 g/L for women

¶ **Pharmacotherapy with >4 different drugs**

Table 2. Associations between CYP2C9 and VKORC1 variants and clinical outcomes

Clinical outcome / genetic variant (s)	*No. of events/ patients (%)*	*No. of events/ patients (%)*	*Unadjusted HR or SHR (95%-CI)*	*Adjusted HR or SHR (95%-CI)*[§]
	With variant(s)	*Without variant(s)*		
Any clinical event (Death, VTE recurrence, major bleeding, or clinically relevant non-major bleeding)				
***CYP2C9* variant**[†]	138/ 281 (49.1)	196/ 493 (39.8)	1.29 (1.04, 1.60)	1.34 (1.08, 1.66)
***VKORC1* variant**[‡]	228/ 510 (44.7)	106/ 264 (40.2)	1.16 (0.92, 1.46)	1.12 (0.88, 1.41)
***CYP2C9* and *VKORC1* variants**	99/ 187 (52.9)	235/ 587 (40.0)	1.44 (1.14, 1.81)	1.40 (1.11, 1.77)
Death				
***CYP2C9* variant**[†]	55/ 281 (19.6)	64/ 493 (13.0)	1.55 (1.08, 2.22)	1.74 (1.19, 2.52)
***VKORC1* variant**[‡]	76/ 510 (14.9)	43/ 264 (16.3)	0.90 (0.62, 1.31)	0.82 (0.55, 1.22)
***CYP2C9* and *VKORC1* variants**	34/ 187 (18.2)	85/ 587 (14.5)	1.25 (0.84, 1.86)	1.29 (0.84, 1.96)
Major bleeding				
CYP2C9 variant[†]	37/ 281 (13.2)	63/ 493 (12.8)	1.03 (0.69, 1.54)	1.03 (0.69, 1.55)
VKORC1 variant[‡]	64/ 510 (12.5)	36/ 264 (13.6)	0.92 (0.61, 1.38)	0.93 (0.61, 1.40)
CYP2C9 and *VKORC1* variants	23/ 187 (12.3)	77/ 587 (13.1)	0.93 (0.58, 1.48)	0.91 (0.57, 1.46)
Clinically relevant non-major bleeding				
CYP2C9 variant[†]	72/ 281 (25.6)	95/ 493 (19.3)	1.38 (1.01, 1.87)	1.39 (1.02, 1.89)
VKORC1 variant[‡]	119/ 510 (23.3)	48/ 264 (18.2)	1.32 (0.95, 1.85)	1.32 (0.94, 1.85)
CYP2C9 and *VKORC1* variants	58/ 187 (31.0)	109/ 587 (18.6)	1.81 (1.32, 2.50)	1.78 (1.30, 2.44)
VTE recurrence				
CYP2C9 variant[†]	35/ 281 (12.5)	65/ 493 (13.2)	0.93 (0.62, 1.40)	0.95 (0.62, 1.44)
VKORC1 variant[‡]	66/ 510 (12.9)	34/ 264 (12.9)	1.00 (0.66, 1.51)	1.11 (0.72, 1.70)
CYP2C9 and *VKORC1* variants	25/ 187 (13.4)	75/ 587 (12.8)	1.03 (0.65, 1.62)	1.15 (0.72, 1.84)

Abbreviations: HR= hazard ratio; CI= confidence interval; SHR= sub-hazard ratio

[*] Death, VTE recurrence, major bleeding, or clinically relevant non-major bleeding

[†] *CYP2C9*2* and *CYP2C9*3* genetic variants

[‡] *VKORC1 G1639A* genetic variant

[§] Adjusted for age, sex, active cancer, provoked VTE, prior VTE, heart failure, chronic pulmonary disease, overt PE, history of major bleeding, heart rate ≥110/min, systolic BP <100 mm Hg, anemia, creatinine >107 µmol/L, antiplatelet treatment, and periods of anticoagulation as time-varying covariate

[¶] Adjusted for age, sex, active cancer, immobilization, heart failure, chronic pulmonary disease, overt PE, history of major bleeding, heart rate ≥110/min, systolic BP <100 mm Hg, anemia, creatinine >107 µmol/L, and periods of anticoagulation as a time-varying covariate

[**] Adjusted for age, sex, active cancer, history of major bleeding, overt PE, anemia, creatinine >107 µmol/L, antiplatelet treatment, and periods of anticoagulation as time-varying covariate

[††] Adjusted for age, sex, active cancer, provoked VTE, prior VTE, and periods of anticoagulation as time-varying covariate

Table 3. Associations between CYP2C9 and VKORC1 variants and quality of anticoagulation

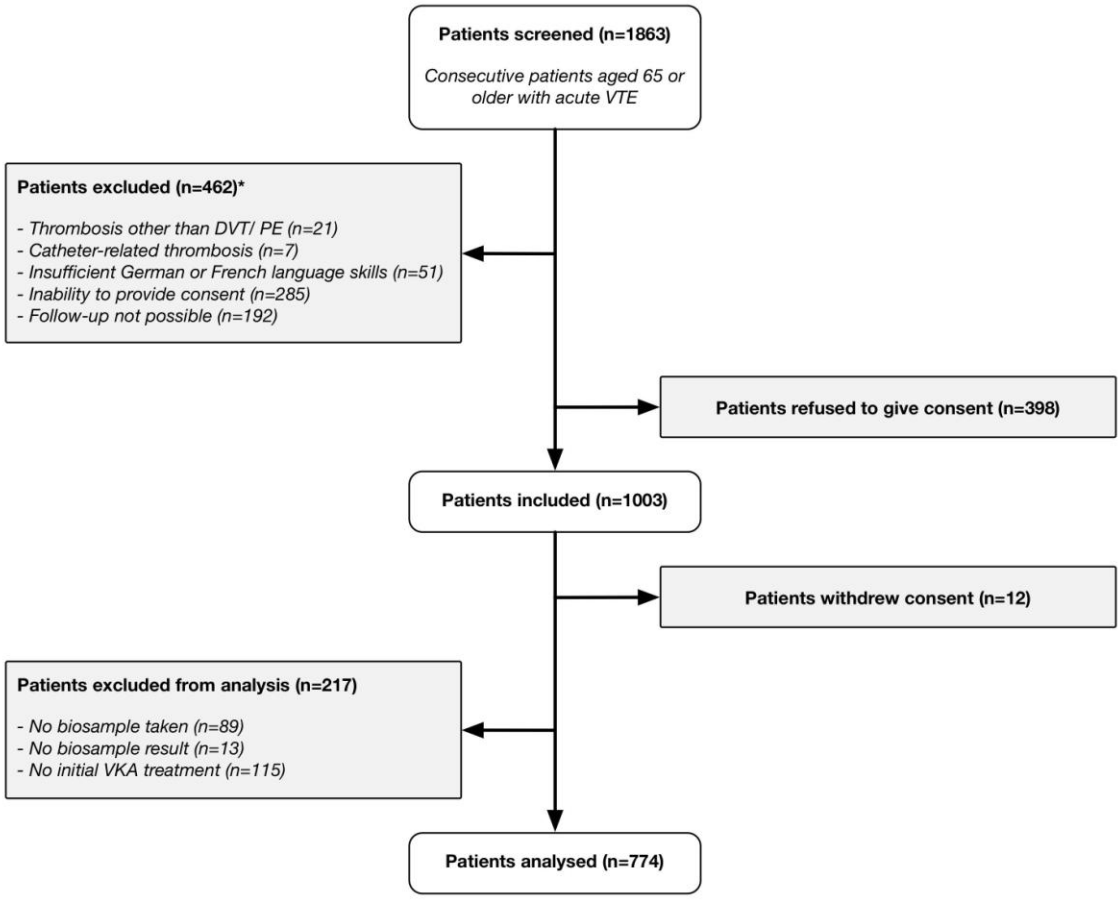
Genetic variants	Percentage of time in the therapeutic INR range (2.0-3.0)		Percentage of time above therapeutic INR range (>3.0)		Time to a first therapeutic INR (≥2.0)	Time to first INR >4.5
	Mean (SD)	*Adjusted difference (95% CI)* ‡	*Mean (SD)*	*Adjusted difference (95% CI)* ‡	*Adjusted hazard ratio (95%-CI)* ‡	*Adjusted hazard ratio (95%-CI)* ‡
CYP2C9* variant						
No (n=481)	61.1 (22.8)		13.8 (15.3)		1 (reference)	1 (reference)
Yes (n=274)	57.4 (23.7)	-3.7 (-7.0, -0.4)	17.1 (18.7)	3.3 (0.8, 5.7)	0.81 (0.68, 0.96)	1.42 (1.11, 1.81)
***VKORC1* variant**†						
No (n=257)	58.4 (23.7)		13.5 (15.6)		1 (reference)	1 (reference)
Yes (n=498)	60.4 (22.9)	2.4 (-1.0, 5.7)	15.8 (17.1)	2.3 (-0.2, 4.8)	1.01 (0.85, 1.20)	1.43 (1.10, 1.85)
***CYP2C9* and *VKORC1* variant**						
No (n=571)	60.2 (23.2)		13.9 (15.4)		1 (reference)	1 (reference)
Yes (n=184)	58.3 (23.4)	-1.5 (-5.2, 2.2)	18.4 (19.7)	4.4 (1.7, 7.2)	0.80 (0.65, 0.97)	1.47 (1.13, 1.93)

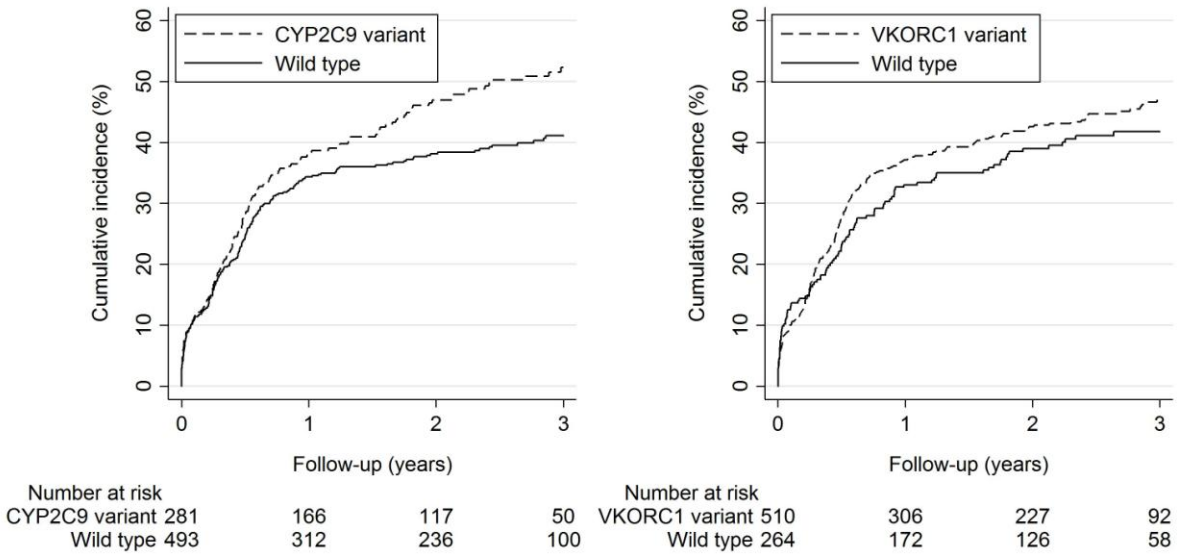
Abbreviations: INR= international normalized ratio; CI= confidence interval

* *CYP2C9*2* and *CYP2C9*3* genetic variants

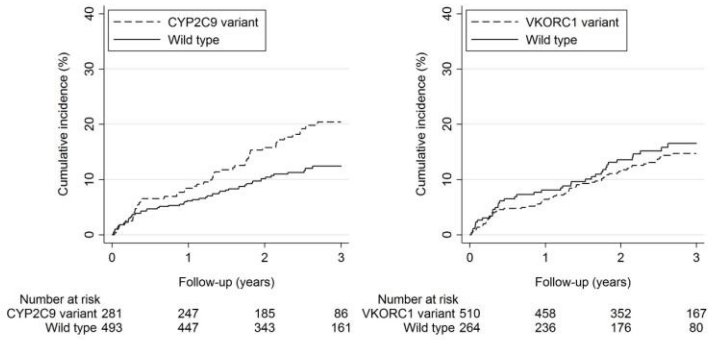
† *VKORC1 G1639A* genetic variant

‡ Adjusted for age, sex, provoked VTE, active cancer, diabetes, heart failure, chronic pulmonary disease, chronic liver disease, severe renal failure (GFR <30 ml/min.), body mass index, hypertension, and smoking status





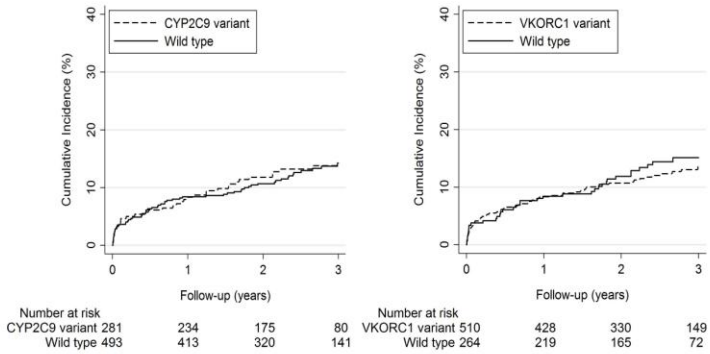
A **Death**



Number at risk				
CYP2C9 variant	281	247	185	86
Wild type	493	447	343	161

Number at risk				
VKORC1 variant	510	458	352	167
Wild type	264	236	176	80

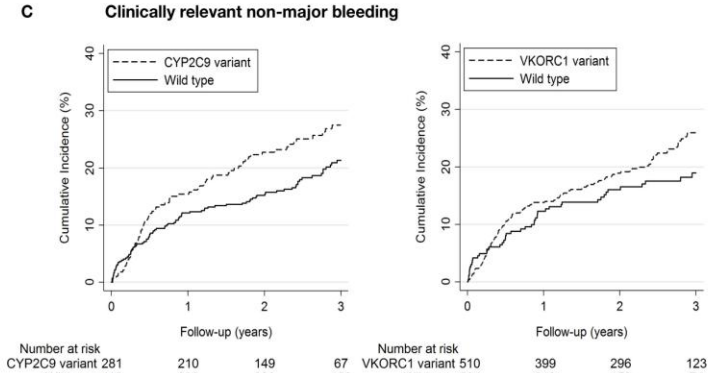
B **Major bleeding**



Number at risk				
CYP2C9 variant	281	234	175	80
Wild type	493	413	320	141

Number at risk				
VKORC1 variant	510	428	330	149
Wild type	264	219	165	72

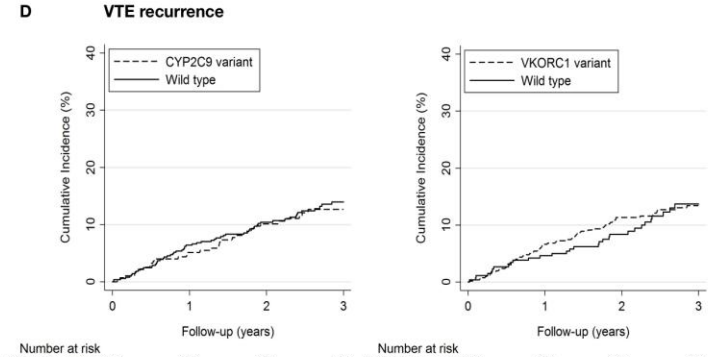
C **Clinically relevant non-major bleeding**



Number at risk				
CYP2C9 variant	281	210	149	67
Wild type	493	399	303	130

Number at risk				
VKORC1 variant	510	399	296	123
Wild type	264	210	156	74

D **VTE recurrence**



Number at risk				
CYP2C9 variant	281	238	167	75
Wild type	493	425	314	139

Number at risk				
VKORC1 variant	510	434	316	144
Wild type	264	229	165	70