BMJ Open

Rationale and design of a prospective study evaluating population pharmacokinetics and pharmacodynamics of rivaroxaban in Chinese patients with non-valvular atrial fibrillation

Xiao-Qin Liu [1], Yu-Fei Zhang,[1] Hong-Yan Ding,[2] Ming-Ming Yan,[1] Ming-Kang Zhong,[1] Chun-Lai Ma[1]

¹Department of Pharmacy, Huashan Hospital Fudan University, Shanghai, China
²Department of Neurology, Huashan Hospital Fudan University, Shanghai, China

Correspondence to
Professor Chun-Lai Ma;
chunlaima@126.com

ABSTRACT

Introduction Rivaroxaban is one of the most commonly used non-vitamin K antagonists for stroke prevention in patients with non-valvular atrial fibrillation (NVAF). Different individual exposures exist for Asian and non-Asian populations, and dose selection is different for Japanese and non-Japanese subjects. Few studies have investigated the pharmacokinetics (PK) and pharmacodynamics (PD) of rivaroxaban in Chinese patients and provided a solid reference for dose selection and individualised therapy.

Methods and analysis This is a single-centre prospective study. Rivaroxaban-treated Chinese NVAF patients will be recruited according to predetermined inclusion criteria. Blood samples will be collected from both outpatients and inpatients with different sampling strategies at steady state. Rivaroxaban plasma concentration, factor Xa activity, prothrombin time and single-nucleotide polymorphisms of candidate genes will be evaluated. Follow-up will be conducted following 3 and 6 months after enrolment to collect information about the safety and efficacy outcomes. A nonlinear mixed-effects modelling strategy will be used to develop a population PK-PD model of rivaroxaban.

Ethics and dissemination The study has been approved by the Ethics Committee of Huashan Hospital, Fudan University (KY2020-016). The study findings will be submitted to peer-reviewed journals and shared with public health authorities.

Trial registration number ChiCTR2100046685.

STRENGTHS AND LIMITATIONS OF THIS STUDY

⇒ A non-linear mixed effect modelling strategy will be used to evaluate the population pharmacokinetic (PK) and pharmacodynamic (PD) characteristics of rivaroxaban in Chinese patients with non-valvular atrial fibrillation.

⇒ Rivaroxaban plasma concentration, factor Xa activity and prothrombin time will be measured.

⇒ A comprehensive evaluation of potential clinical and genetic factors affecting PK or PD of rivaroxaban will be performed.

⇒ An individualised dosing strategy of rivaroxaban for Chinese patients will be proposed based on model-informed precision dosing.

⇒ A relatively small sample size may limit the further interpretation of dose-exposure-efficacy/safety relationship.

INTRODUCTION

Atrial fibrillation (AF) is the most common arrhythmia, increasing the risk of ischaemic stroke fivefold and the risk of mortality twofold compared with patients without AF.[1 2] Non-valvular AF (NVAF), which refers to AF in the absence of a mechanical prosthetic heart valve or moderate to severe mitral stenosis, accounts for approximately 95% of all AF populations.[3] Anticoagulant treatment plays a key role in preventing stroke and embolism in patients with NVAF.[4] European guidelines have indicated a preference for non-vitamin K antagonists (NOACs) for stroke prevention in patients with NVAF.[5] Among all marketed NOACs, rivaroxaban is one of the most used NOACs.[6 7]

Rivaroxaban directly inhibits factor Xa (FXa) activity.[8] After rapid absorption, approximately two-thirds of rivaroxaban is metabolised by liver cytochrome P450 3A4/5 (CYP3A4/5), CYP2J2 and CYP-independent mechanisms. The other third is eliminated via the renal pathway as unchanged drug. P-glycoprotein and breast cancer resistance protein are involved in active renal secretion.[8] Therefore, dose reduction is necessary in patients with moderately impaired renal function.[8 9]

Data from previous clinical trials and real-world studies have been used to develop a population pharmacokinetic (PK) or pharmacokinetic-pharmacodynamic (PK-PD) model of rivaroxaban to quantify the variation in PK and/or PD in NVAF patients.[10–13]
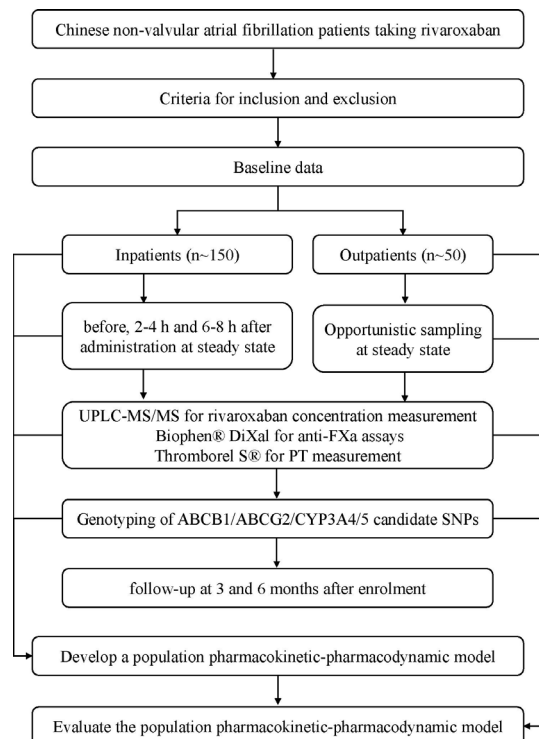
Figure 1 Study flow chart. SNPs, single-nucleotide polymorphism; UPLC-MS/MS, ultra-performance liquid chromatography with tandem mass spectrometry.

The standard rivaroxaban dose for Caucasian NVAF patients was 20 mg, whereas 15 mg was selected for Japanese patients on account of the model-based PK studies, which showed an equivalent exposure for Japanese patients taking 15 mg and Caucasian patients taking 20 mg.[12] The efficacy and safety of the two dose strategies were confirmed in previous large phase 3 trials, namely ROCKET AF and J-ROCKET AF.[14 15] Studies conducted in healthy Chinese volunteers demonstrated comparable PK and PD to healthy Caucasian volunteers. It was believed that a fixed rivaroxaban dose could be administered to all patients regardless of ethnicity.[16 17] Therefore, the standard rivaroxaban dose in China is 20 mg, according to the National Medical Products Administration and guideline of stroke prevention in Chinese patients with AF.[18]

Asian clinicians tended to prescribe a lower dose of rivaroxaban for NVAF patients than the label stated.[19–22] This may be partially attributed to the lower weight, higher prevalence of prior stroke and higher bleeding risk in NOAC-treated Asian patients than in non-Asians.[23 24] However, it remains controversial whether other Asian groups should choose a lower rivaroxaban dose. To date, few studies have explored the PK and PD characteristics of Chinese NVAF patients treated with rivaroxaban.

Here, we aim to conduct a prospective study to evaluate the population PK-PD of rivaroxaban in Chinese NVAF patients, to explore the potential variables affecting PK and PD, and to provide a reference for individualised treatment with rivaroxaban.

METHODS
Study design
This study is a single-centre prospective study to evaluate the population PK and PD profile of rivaroxaban in Chinese patients with NVAF. Patients meeting the inclusion criteria are enrolled from Huashan Hospital, Fudan University from June 2021 to June 2023. This study will not interfere with patients' medication therapy. Rivaroxaban dose selection will be performed by the clinicians. Blood samples will be taken for measurement of rivaroxaban plasma concentration, FXa activity and prothrombin time (PT). Face-to-face interviews will be conducted for each patient at 3 and 6 months after enrolment. A flow chart of this study is shown in figure 1.

All participants will provide a written informed consent. The study will comply with the principles of the Declaration of Helsinki and has been approved by the Ethics Committee of Huashan Hospital, Fudan University.

Patients and recruitment strategies
Outpatients or inpatients aged ≥18 years will be recruited if they have been diagnosed with NVAF documented based on electrocardiography and prescribed rivaroxaban. Patients will be excluded from the study if they (1) have NVAF combined with valvular heart disease; (2) are pregnant; (3) have a history of malignant tumours; (4) have severe hepatic and renal insufficiency (Child-Pugh grade B or C; creatinine clearance (CrCl) <15 mL/min); (5) suffer from abnormal coagulation function or disease predisposing to bleeding, such as thrombocytopaenia and other causes; (6) are hospitalised with an acute illness, including acute gastrointestinal haemorrhage, active endocarditis, acute clinical hepatitis, acute heart failure, acute renal failure and other diseases that researchers considered to affect PK or PD of rivaroxaban possibly and (7) are patients participating in clinical trials of other drugs at the same time.

Data collection
Data will be collected from each patient after obtaining their informed consent. Demographic information will include age, sex, body weight, height and lifestyle (smoking and drinking status). Clinical information will include comorbidities (hypertension, diabetes, hyperlipidaemia, chronic kidney disease (CKD), congestive heart failure (CHF) and vascular disease), previous history of stroke and bleeding. Laboratory information will include routine blood examination (white cell, red cell, haemoglobin, haematocrit and blood platelet count), liver function indices (alanine aminotransferase, aspartate aminotransferase, total bilirubin, direct bilirubin, total bile acid, alkaline phosphatase, gamma-glutamyl transpeptidase, total protein, albumin, globulin and prealbumin), renal function indices (blood urea nitrogen, uric acid, serum creatinine and serum cystatin C), blood glucose (fasting blood glucose, postprandial blood sugar and glycosylated haemoglobin), blood fat (cholesterol, triglycerides and low-density lipoprotein), coagulation

function (activated partial thromboplastin time, PT, thrombin time, international normalised ratio (INR) and D-dimer). Prescription information will include combined pharmacotherapy that may interact with rivaroxaban, such as antifungal agents and antiplatelet agents. Rivaroxaban dose, dosing time and coadministration with food will also be recorded for each participant.

Creatinine clearance will be estimated using the Cockcroft-Gault formula.[25] The estimated glomerular filtration rate will be estimated using a modified diet in renal disease equation[26] and CKD epidemiology collaboration equation.[27]

Patient and public involvement

Patients and/or the public were not involved in the design, or conduct, or reporting, or dissemination plans of this research.

Blood sampling and storage

Blood samples will be taken for measurement of rivaroxaban PK and PD, including rivaroxaban plasma concentration, FXa activity and PT. Different sampling strategies will be applied for inpatients and outpatients. For inpatients, three blood samples will be taken at steady state before rivaroxaban administration (defined as trough samples) and 2–4 hours (defined as peak samples) and 6–8 hours after administration. For outpatients at steady state, blood samples will be collected at each follow-up using an opportunistic sampling strategy. The sampling times will be documented exactly.

Samples (2.7 mL) will be collected into 0.109 mol/L trisodium citrate ($Na_3C_6H_5O_7$) tubes for coagulation tests (FXa activity and PT) and 2 mL samples will be collected into ethylenediaminetetraacetic acid (EDTA)-containing tubes for rivaroxaban plasma concentration measurements and DNA extraction. Blood samples will be processed within 4 hours after collection. Platelet-poor plasma and haemocytes will be prepared by centrifugation at 2500×g for 15 min at 20°C, and then frozen immediately at −80°C.

Assessment of stroke and bleeding risks

CHA2DS2-VASc (CHF, hypertension, age ≥75 years, diabetes mellitus, prior stroke or transient ischaemic attack, vascular disease, age 65–74 years, female) will be used to assess the risk of stroke. HAS-BLED (hypertension, renal/liver disease, stroke history, bleeding history or predisposition to bleeding, labile INR, age >65 years, medication usage predisposing to bleeding, and alcohol use) will be used to assess the risk of bleeding. Both CHA2DS2-VASc and HAS-BLED scores will be calculated when participants are enrolled as baseline.

Safety and efficacy

All participants will be followed in Huashan hospital at 3 and 6 months after enrolment, and safety and efficacy outcomes will be collected through face-to-face interviews. If patients could not complete the face-to-face interviews on time, the relevant data will be collected by telephone. At each visit, safety and efficacy events during the period from last visit till now will be evaluated by the investigators. Bleeding is classified based on the International Society of Thrombosis and Haemostasis criteria.[28 29] Major bleeding will be defined as fatal bleeding, symptomatic bleeding in a critical area, bleeding causing a fall in haemoglobin level of ≥20 g/L, or leading to transfusion of ≥2 U of whole cell or red cell count. Clinically relevant non-major (CRNM) bleeding refers to any sign or symptom of haemorrhage, which meet at least one of the following criteria: (1) requiring medical intervention by a healthcare professional; (2) leading to hospitalisation or increased level of care and (3) prompting a face-to-face (not just a telephone or electronic communication) evaluation. The primary safety outcomes are proportion of major or CRNM bleeding complications. Secondary outcomes are (1) the proportion of major complications and (2) the proportion of CRNM bleeding complications. Efficacy events, which defined as thromboembolic events, include ischaemic stroke, transient ischaemic attack and peripheral arterial thromboembolism. The primary efficacy outcomes are the proportion of thromboembolic events.

Bioanalysis

Rivaroxaban plasma concentration will be determined using a sensitive and validated ultraperformance liquid chromatography with tandem mass spectrometry assay. The calibration range will be from 1 ng/mL (lower limit of quantification, LLOQ) to 1000 ng/mL. The accuracies of the low, medium and high control samples will be 85.9%–105%. Intraday and interday precision will be below 5%.

Additionally, the FXa assay will also be performed using the Biophen DiXal kit (Hyphen BioMed, France). This assay is based on the inhibition of a constant and excess quantity of FXa. Residual FXa hydrolyses the FXa-specific chromogenic substrate. Paranitroaniline, then released from the substrate, can be measured by measuring absorbance at 405 nm and is inversely proportional to the concentration of rivaroxaban in the sample. Biophen DiXal calibrators and controls will be used for all samples. For samples with a quantitative measurement <80 ng/mL, repeated measurements by Biophen DiXal Low will be viewed as the final test result. PT will be tested by Thromborel S reagent kit (Siemens Healthcare Diagnostic, Erlangen, Germany). Both FXa assay and PT will be measured using a CN6000 coagulation Analyser (TOA Medical Electronics, Kobe, Japan) according to the protocols provided by the manufacturers.

Genotyping

Single-nucleotide polymorphisms (SNPs) of four genes involved in the PK process, namely ATP-binding cassette subfamily B member 1 (ABCB1), ATP-binding cassette transporter G2 (ABCG2), cytochrome P450 (CYP) 3A4/5, will be analysed. Information from the 1000 Genome Project (https://www.internationalgenome.org/) and

the dbSNP database of the National Centre for Biotechnology Information (https://www.ncbi.nlm.nih.gov/snp/) will be combined to select the candidate SNP and analysed using Haploview software (V.4.20, https://www.broadinstitute.org/haploview/haploview). The criteria will include: (1) the minor allele frequency is >20% in the HapMap database of Han Chinese in Beijing and Southern Han Chinese populations and (2) a r^2 threshold of 0.8.

Haemocytes extracted from EDTA tubes will be used for genotyping. Genomic DNA will be extracted using the TIANGEN DNA Kit (DP348, TIANGEN Biotech, Beijing, China) according to the manufacturer's protocol. Genotype detection will be analysed using the MALDI-TOF Mass Array assay.

Rivaroxaban population PK-PD evaluation

Rivaroxaban population PK and PD analyses will be performed sequentially. Rivaroxaban concentration will be modelled as PK data; FXa activity and PT will be modelled as PD data.

The PK profile of rivaroxaban will be described by an oral one-compartment model based on previous studies.[10 30] PK parameters, including the first-order absorption rate constant (k_a), apparent oral clearance (CL/F) and apparent volume of distribution (V/F) will be estimated. The previously identified variables and potential covariates, such as age, body weight, serum creatinine (SCr) and CrCl, that will influence PK parameters will be included to assess their influence. Concomitant drugs with potential influence will be tested for comedications >20% in all patients. Continuous variables will be tested via a proportional, exponential or power function, and discontinuous variables will be tested using a scale function. The selection of covariates will be in accordance with the stepwise approach and will be physiologically reasonable.

Individual PK parameters will be estimated from the final population PK model using the POSTHOC method and will be included in the sequential PD analysis. A direct-linear, near-linear or E_{max} model will be used to correlate the rivaroxaban concentration with PD markers based on their relationship. The selection of the structural PD model will be based on the Akaike information criterion, Bayesian information criterion and the precision of estimates. The selection strategies of covariates for PD markers are similar to those for the rivaroxaban PK model. The final model will be evaluated by goodness-of-fit, visual predictive check and non-parametric bootstrap.

Determination of sample size

There is no reference range for the number of patients and sample size for population PK and PK-PD studies. Referring to the sample size in previous studies,[13 31 32] we aim to enrol at least 50 outpatients and 150 inpatients in total. In view of the reality that the timing of medication administration and adherence is better controlled for inpatients, only the data from inpatients will be used to perform population PK-PD analysis. Besides, data from outpatients will be used for model evaluation.

Statistical analysis

Statistical analysis will be conducted using R software (V.3.6.0). Continuous variables will be shown as mean±SD. Groups will be compared using the t-tests. Categorical variables will be shown as numbers and percentages. Groups will be compared using Fisher's exact test or the χ^2 test. Non-linear mixed effect modelling will be performed using NONMEM software (V.7.4; ICON Development Solutions, Ellicott City, Maryland, USA) with first-order conditional estimation with interaction method. The selection of covariates will be based on the likelihood ratio test and a stepwise approach. The criterion is a drop of objective function value of at least 3.84 (p<0.05, df=1) for forward inclusion, and at least 6.63 (p<0.01, df=1) for backward elimination. Statistical analysis and diagnostic plots will be completed using R (V.4.1.1) and Xpose (V.4.3.2) software packages. All SNPs will be analysed using the χ^2 test for the assessment of Hardy-Weinberg equilibrium.

Ethics and dissemination

This study has been approved by the Ethics Committee of the Huashan Hospital, Fudan University. Informed consent will be obtained from all participants. The findings of this study will be summarised in the manuscript of structural preclinical PKPD analysis and submitted to peer-view journals. Original data will be stored at the Huashan Hospital, Fudan University.

ORCID iD
Xiao-Qin Liu http://orcid.org/0000-0002-3615-3274

REFERENCES

1 Lip GYH, Lane DA. Stroke prevention in atrial fibrillation: a systematic review. *JAMA* 2015;313:1950–62.

2 Kovacs RJ, Flaker GC, Saxonhouse SJ, *et al*. Practical management of anticoagulation in patients with atrial fibrillation. *J Am Coll Cardiol* 2015;65:1340–60.

3 Shafrin J, Bruno A, MacEwan JP, *et al*. Physician and patient preferences for nonvalvular atrial fibrillation therapies. *Value Health* 2016;19:451–9.

4 Heidbuchel H, Verhamme P, Alings M, *et al*. Updated European heart rhythm association practical guide on the use of non-vitamin-K antagonist anticoagulants in patients with non-valvular atrial fibrillation: Executive summary. *Eur Heart J* 2017;38:2137–49.

5 Steffel J, Collins R, Antz M, *et al*. 2021 European heart rhythm association practical guide on the use of non-vitamin K antagonist oral anticoagulants in patients with atrial fibrillation. *Europace* 2021;23:1612–76.

6 Zhu J, Alexander GC, Nazarian S, *et al*. Trends and variation in oral anticoagulant choice in patients with atrial fibrillation, 2010-2017. *Pharmacotherapy* 2018;38:907–20.

7 Maggioni AP, Dondi L, Andreotti F, *et al*. Four-Year trends in oral anticoagulant use and declining rates of ischemic stroke among 194,030 atrial fibrillation patients drawn from a sample of 12 million people. *Am Heart J* 2020;220:12–19.

8 Mueck W, Stampfuss J, Kubitza D, *et al*. Clinical pharmacokinetic and pharmacodynamic profile of rivaroxaban. *Clin Pharmacokinet* 2014;53:1–16.

9 Food and drug administration, updated 2021-01XARELTO® [insert package]. Available: https://www.accessdata.fda.gov/drugsatfda_docs/label/2021/215859s000lbl.pdf.

10 Girgis IG, Patel MR, Peters GR, *et al*. Population pharmacokinetics and pharmacodynamics of rivaroxaban in patients with non-valvular atrial fibrillation: results from rocket AF. *J Clin Pharmacol* 2014;54:917–27.

11 Kaneko M, Tanigawa T, Hashizume K, *et al*. Confirmation of model-based dose selection for a Japanese phase III study of rivaroxaban in non-valvular atrial fibrillation patients. *Drug Metab Pharmacokinet* 2013;28:321–31.

12 Tanigawa T, Kaneko M, Hashizume K, *et al*. Model-Based dose selection for phase III rivaroxaban study in Japanese patients with non-valvular atrial fibrillation. *Drug Metab Pharmacokinet* 2013;28:59–70.

13 Suzuki S, Yamashita T, Kasai H, *et al*. An analysis on distribution and inter-relationships of biomarkers under rivaroxaban in Japanese patients with non-valvular atrial fibrillation (CVI ARO 1). *Drug Metab Pharmacokinet* 2018;33:188–93.

14 Patel MR, Mahaffey KW, Garg J, *et al*. Rivaroxaban versus warfarin in nonvalvular atrial fibrillation. *N Engl J Med* 2011;365:883–91.

15 Hori M, Matsumoto M, Tanahashi N, *et al*. Rivaroxaban vs. warfarin in Japanese patients with atrial fibrillation – the J-ROCKET AF study –. *Circ J* 2012;76:2104–11.

16 Zhao X, Sun P, Zhou Y, *et al*. Safety, pharmacokinetics and pharmacodynamics of single/multiple doses of the oral, direct factor Xa inhibitor rivaroxaban in healthy Chinese subjects. *Br J Clin Pharmacol* 2009;68:77–88.

17 Jiang J, Hu Y, Zhang J, *et al*. Safety, pharmacokinetics and pharmacodynamics of single doses of rivaroxaban - an oral, direct factor Xa inhibitor - in elderly Chinese subjects. *Thromb Haemost* 2010;103:234–41.

18 Zhang S, Yang YM, Huang CX. Guideline of stroke prevention in Chinese patients with atrial fibrillation (2017). *Chin J Cardiac arrhyth* 2018;22:17–30.

19 Cheng W-H, Chao T-F, Lin Y-J, *et al*. Low-Dose rivaroxaban and risks of adverse events in patients with atrial fibrillation. *Stroke* 2019;50:2574–7.

20 Lee SR, Lee YS, Park JS, *et al*. Label adherence for non-vitamin K antagonist oral anticoagulants in a prospective cohort of Asian patients with atrial fibrillation. *Yonsei Med J* 2019;60:277–84.

21 Cho MS, Yun JE, Park JJ, *et al*. Outcomes after use of Standard- and low-dose non-vitamin K oral anticoagulants in Asian patients with atrial fibrillation. *Stroke* 2018:STROKEAHA118023093.

22 Cho MS, Yun JE, Park JJ, *et al*. Pattern and impact of off-label underdosing of non-vitamin K antagonist oral anticoagulants in patients with atrial fibrillation who are indicated for standard dosing. *Am J Cardiol* 2020;125:1332–8.

23 Wong KSL, Hu DY, Oomman A, *et al*. Rivaroxaban for stroke prevention in East Asian patients from the rocket AF trial. *Stroke* 2014;45:1739–47.

24 Chan Y-H, Lee H-F, Wang C-L, *et al*. Comparisons of rivaroxaban following different dosage criteria (rocket AF or J-ROCKET AF trials) in Asian patients with atrial fibrillation. *J Am Heart Assoc* 2019;8:e013053.

25 Cockcroft DW, Gault MH. Prediction of creatinine clearance from serum creatinine. *Nephron* 1976;16:31–41.

26 Levey AS, Coresh J, Greene T, *et al*. Using standardized serum creatinine values in the modification of diet in renal disease study equation for estimating glomerular filtration rate. *Ann Intern Med* 2006;145:247–54.

27 Levey AS, Stevens LA, Schmid CH, *et al*. A new equation to estimate glomerular filtration rate. *Ann Intern Med* 2009;150:604–12.

28 Schulman S, Kearon C, Subcommittee on Control of Anticoagulation of the Scientific and Standardization Committee of the International Society on Thrombosis and Haemostasis. Definition of major bleeding in clinical investigations of antihemostatic medicinal products in non-surgical patients. *J Thromb Haemost* 2005;3:692–4.

29 Kaatz S, Ahmad D, Spyropoulos AC, *et al*. Definition of clinically relevant non-major bleeding in studies of anticoagulants in atrial fibrillation and venous thromboembolic disease in non-surgical patients: communication from the SSC of the ISTH. *J Thromb Haemost* 2015;13:2119–26.

30 Willmann S, Zhang L, Frede M, *et al*. Integrated population pharmacokinetic analysis of rivaroxaban across multiple patient populations. *CPT Pharmacometrics Syst Pharmacol* 2018;7:309–20.

31 Zdovc J, Petre M, Pišlar M, *et al*. Downregulation of ABCB1 gene in patients with total hip or knee arthroplasty influences pharmacokinetics of rivaroxaban: a population pharmacokinetic-pharmacodynamic study. *Eur J Clin Pharmacol* 2019;75:817–24.

32 Goto E, Horinaka S, Ishimitsu T, *et al*. Factor Xa inhibitors in clinical practice: comparison of pharmacokinetic profiles. *Drug Metab Pharmacokinet* 2020;35:151–9.