CYP2C19 genotype-guided antithrombotic treatment versus conventional clopidogrel therapy in peripheral arterial disease: study design of a randomized controlled trial (GENPAD)

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Background Clopidogrel is recommended in international guidelines to prevent arterial thrombotic events in patients with peripheral arterial disease (PAD). Clopidogrel itself is inactive and metabolism is dependent on the *CYP2C19* enzyme. About 30% of Caucasian PAD patients receiving clopidogrel carry 1 or 2 *CYP2C19* loss-of-function allele(s) and do not or to a limited extent convert the prodrug into its active metabolite. As a result, platelet inhibition may be inadequate which could lead to an increased risk of adverse clinical events related to arterial thrombosis. A *CYP2C19* genotype-guided antithrombotic treatment might be beneficial for PAD patients.

Methods GENPAD is a multicenter randomized controlled trial involving 2,276 PAD patients with an indication for clopidogrel monotherapy. Patients with a separate indication for dual antiplatelet therapy or stronger antithrombotic therapy are not eligible for study participation. Patients randomized to the control group will receive clopidogrel 75 mg once daily without pharmacogenetic guidance. Patients randomized to the intervention group will be tested for carriage of *CYP2C19* *2 and *3 loss-of-function alleles, followed by a genotype-guided antithrombotic treatment with either clopidogrel 75 mg once daily for normal metabolizers, clopidogrel 150 mg once daily for intermediate metabolizers, or acetylsalicylic acid 80 mg once daily plus rivaroxaban 2.5 mg twice daily for poor metabolizers. The primary outcome is a composite of myocardial infarction, ischemic stroke, cardiovascular death, acute or chronic limb ischemia, peripheral vascular interventions, or death. The secondary outcomes are the individual elements of the primary composite outcome and clinically relevant bleeding complications.

Conclusion The aim of the GENPAD study is to evaluate the efficacy, safety, and cost-effectiveness of a genotype-guided antithrombotic treatment strategy compared to conventional clopidogrel treatment in PAD patients. (Am Heart J 2022;254:141–148.)

Background

It is estimated that globally over 200 million people are affected by peripheral arterial disease (PAD).[1] Symptoms vary and may include intermittent claudication, pain at rest, or gangrene, typically categorized according to the Rutherford classification.[2] Despite adequate secondary prevention measures, cardiovascular morbidity and mortality remain high amongst PAD patients. According to the international PAD guidelines, clopidogrel

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Submitted March 8, 2022; accepted August 4, 2022

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0002-8703

https://doi.org/10.1016/j.ahj.2022.08.001

75 mg once daily is recommended to reduce the risk of arterial thrombosis.[3,4] These guidelines do not mention the influence of *CYP2C19* genetic variants on the pharmacokinetics and the platelet inhibiting effect of clopidogrel.

Clopidogrel is a prodrug that needs to be metabolized into its active metabolite by the *CYP2C19* enzyme. About 30% of the Caucasian population carries at least one loss-of-function (LOF) allele, such as the *2 or *3 alleles. This results in a limited ability to convert the prodrug clopidogrel into its active metabolite. Studies showed that carriers of *CYP2C19* LOF alleles have lower plasma concentrations of the active metabolite of clopidogrel and increased platelet aggregation as compared to noncarriers.[5,6]

Previous studies in the fields of neurology and cardiology demonstrated the clinical relevance of *CYP2C19* LOF alleles. A meta-analyses of Pan et al regarding stroke patients indicate that carriers of *CYP2C19* LOF alleles receiving clopidogrel had a circa 2-fold increased risk of recurrent stroke as compared to noncarriers.[7] The largest body of evidence, including several meta-analyses, exists for patients with coronary artery disease (CAD). Carriers of *CYP2C19* LOF alleles who are treated with clopidogrel and undergoing percutaneous coronary intervention (PCI) have a higher risk of adverse cardiovascular events compared to noncarriers. This is more pronounced in an Asian population (RR = 1.91), where LOF-alleles are more common, compared to a Caucasian population (RR = 1.20).[8,9] Since atherosclerosis is a systemic condition, extrapolation of results from previous studies in CAD and stroke patients is possible. However, CAD patients undergoing PCI with stenting are usually treated with dual antiplatelet therapy (DAPT) instead of monotherapy clopidogrel.

The association between *CYP2C19* LOF alleles and increased risk of adverse cardiovascular events in patients using clopidogrel suggests that a *CYP2C19* genotype-guided strategy could lead to improved clinical outcomes in patients with clinical manifestations of atherosclerosis. In *CYP2C19* LOF-allele carriers with cerebrovascular disease, the use of ticagrelor has proven to be superior to clopidogrel in terms of reducing risk of recurrent stroke after 90 days.[10] In CAD patients, multiple prospective trials have been performed investigating the use of a *CYP2C19* genotype-guided antiplatelet therapy.[11-14] Several large meta-analyses showed that *CYP2C19* genotype-guided strategies could reduce ischemic events in CAD patients, especially in patients undergoing PCI, compared to conventional therapy.[15-18] Pereira et al. showed that the superiority of the more potent P2Y12 inhibitors, ticagrelor, and prasugrel, in CAD patients was based primarily on *CYP2C19* LOF-alleles since these treatment strategies significantly reduced ischemic events in *CYP2C19* LOF-allele carriers, but not in non-carriers.[17]

So far, no large-scale prospective research has been performed to investigate the clinical relevance of *CYP2C19* LOF-alleles in PAD patients treated with clopidogrel, nor is there any trial evidence regarding the added value of *CYP2C19* genotype-guided treatment. We hypothesize that a *CYP2C19* genotype-guided antithrombotic treatment strategy is superior in terms of reducing the rate of adverse clinical events related to arterial thrombosis in PAD patients, and that this approach is cost-effective compared to standard clopidogrel treatment.

Methods

Objectives

The primary aim of the GENPAD study is to determine whether a *CYP2C19* genotype-guided antithrombotic treatment is superior in reducing adverse clinical events related to arterial thrombosis in comparison to standard clopidogrel treatment in PAD patients. The secondary objective is to evaluate the efficacy of *CYP2C19* genotype-guided antithrombotic treatment in reducing the separate elements of the primary composite outcome end point and to compare the occurrence of clinically relevant bleeding complications of both treatment strategies. Furthermore, we will evaluate the cost-effectiveness of a *CYP2C19* genotype-guided approach.

Design

The GENPAD study is a randomized, open label, multicenter, clinical trial. Patients will be randomized to either a *CYP2C19* genotype-guided antithrombotic treatment strategy or standard treatment with clopidogrel. Patients and health care providers are not blinded for treatment allocation.

The study is initiated by the Radboud university medical center and conducted at the Radboud university medical center, university medical center Groningen, Maastricht university medical center, Amsterdam university medical centers, Canisius Wilhelmina Ziekenhuis Nijmegen, Rijnstate Arnhem, Bernhoven Uden, Gelderse Vallei Ede, Gelre Ziekenhuizen, Máxima Medisch Centrum Veldhoven, Medisch Spectrum Twente, Ommelander Ziekenhuis Groningen, and Groene Hart Ziekenhuis Gouda. The recruitment schedule is designed to have a mean follow-up of 2 years for the entire study population. The follow-up time will range from 6 months to 3 years. The study has been approved by the regional medical ethics committee Oost-Nederland (reference number: 2020-7057), and local approval has been obtained for each participating site. This study is conducted in accordance with the latest revision of the Declaration of Helsinki and Good Clinical Practice regulations and is registered at ClinicalTrials.gov on November 6 2020 as NCT04619927 (https://www.clinicaltrials.gov/ct2/show/NCT04619927) and at the Dutch Trial Regis-

Table I. Study schedule.

Study procedure	T0	T1	T2	T3	T4
Informed consent	X				
Randomization	X				
Patient characteristics	X				
Vascular state	X				
Medical history	X				
Medication use	X				
Blood sample withdrawal	X				
CYP2C19 genotyping (intervention group)	X				
Questionnaires *	X	X	X	X	X
Review of medical record	X	X	X	X	X

T0 = study start, T1 = 6 months, T2 = 12 months, T3 = 24 months, T4 = 36 months

* Questionnaires include the EQ-5D-5L, the WHOQol-Bref, an adapted combined version of the IMTA medical cost questionnaire (iMCQ) and the IMTA productivity cost questionnaire (iPCQ), and the GENPAD specific questionnaire.

ter on November 2 as NL9027 (https://www.trialregister.nl/trial/9027). The GENPAD trial is supported by a grant from ZonMw, a Dutch organization funded by the government promoting health care research, and implementation of study results in daily practice. The authors are solely responsible for the design and conduct of this study, all study analyses, the drafting and editing of the paper and its final contents.

Participants and recruitment

All patients with lower extremity PAD visiting the outpatient clinic or vascular laboratory are screened. Patients already on clopidogrel monotherapy as well as newly diagnosed patients with an indication for clopidogrel monotherapy are considered eligible. Patients with an indication for dual antiplatelet therapy or stronger anticoagulant therapy (including NOACs, vitamin K antagonists or coumarins) are not eligible for participation. Inclusion criteria for this study are:[1] previous or current ankle-brachial index < 0.9 and/or toe brachial index < 0.5,[2] previous or current symptoms due to insufficient vascularization of 1 or 2 lower extremities including intermittent claudication, pain at rest, and/or gangrene (Rutherford classification 1-6),[3] consultation of the vascular surgery department for diagnosis, treatment and/or follow-up of PAD,[4] an indication for monotherapy clopidogrel 75 mg once daily, and[5] age 16 years or older. Detailed exclusion criteria are found in appendix A. Key exclusion criteria include:[1] known *CYP2C19* genotype or metabolizer state,[2] (concomitant) treatment with other anticoagulants,[3] contraindications for clopidogrel, ASA, and/or rivaroxaban,[4] pregnant or breastfeeding women, and[5] patients who are unable to provide written informed consent.

Local computer programs select eligible patients by a pseudo-anonymous screening of the outpatient clinic and vascular laboratory of the vascular surgery departments. In case a participating hospital cannot facilitate computer-controlled screening, a physician involved in the treatment of patients with PAD will manually perform screening. Eligible patients are informed about the study and written informed consent is obtained before proceeding with any of the trial procedures. Where possible, research procedures at baseline will be combined with the participants' planned visit to the vascular laboratory or the outpatient clinic.

Study procedures

Castor EDC's (Castor Electronic Data Capture, Amsterdam, the Netherlands) validated variable block randomization model randomly assigns participants in a one-to-one ratio to either *CYP2C19* genotype-guided antithrombotic treatment or standard clopidogrel treatment, stratified by participating center. After randomization, data on baseline patient characteristics, vascular state, medical history, and medication use is collected. Height, weight, hip/waist circumference, and blood pressure is measured and blood samples are collected to perform *CYP2C19* genotyping. Patient questionnaires are obtained at baseline and at 6, 12, 24, and 36 months, respectively. These questionnaires include the EQ-5D-5L, the WHOQol-Bref, and an adapted combined version of the Institute for Medical Technology Assessment (iMTA) medical cost questionnaire (iMCQ) and the IMTA productivity cost questionnaires (iPCQ) in Dutch language versions. Additionally, a GENPAD specific questionnaire is used during follow-up, including questions about medication adherence and occurrence of adverse events. An overview of the study procedures is shown in Table I.

CYP2C19 genotyping and genotype-guided prescription

For *CYP2C19* genotyping, a 6 mL blood sample is collected in an EDTA tube of each participant at baseline. Samples of participants in the intervention group are directly analyzed for the presence of *CYP2C19* LOF alleles. *CYP2C19* genotyping will take place at the de-

partment of Human Genetics of the Radboud university medical center using Taqman assays (*CYP2C19*2*: C__25986767_70, *CYP2C19*3*: C__27861809_10) according to the protocols of the manufacturer (ThermoFisher Scientific, Bleiswijk, The Netherlands).

After *CYP2C19* genotyping, participants in the intervention group will receive genotype-guided antithrombotic treatment. Patients without a *2 or *3 *CYP2C19* LOF allele are considered extensive or normal metabolizers and will be treated with clopidogrel 75 mg once daily. Patients with 1 LOF allele, either a *2 or *3 *CYP2C19* allele, are classified as intermediate metabolizers and will be treated with double-dose clopidogrel (150 mg) once daily. Finally, patients with 2 of these LOF alleles are classified as poor metabolizers and will be treated with ASA 80 mg once daily plus rivaroxaban 2.5mg twice daily. Blood samples of participants in the control group are stored at -80°C and will be analyzed for *CYP2C19* LOF alleles at the end of the study. Participants in the control group are treated with clopidogrel 75 mg once daily.

Delivery and prescription of medication

GENPAD is a pragmatic trial thus all medication will be provided via usual care and expenses are covered by the national health insurance. All eligible patients have been diagnosed with PAD and are therefore already using clopidogrel or acetylsalicylic acid. New patients are prescribed clopidogrel standard treatment upon diagnosis after which they are considered for study participation. Patients in the control group using acetylsalicylic acid are switched to clopidogrel directly after inclusion. Patients in the intervention group continue their current platelet aggregation inhibitor, clopidogrel or acetylsalicylic acid, until genotyping results are available. Results of the *CYP2C19* genotyping are available a maximum of 3 weeks after study inclusion. Requested medication modifications are directly communicated to the health care provider and monitored by the study team. Pharmacy delivery details will be checked regularly to ensure participants are treated according to protocol.

Outcomes

The primary composite outcome is the occurrence of myocardial infarction, ischemic stroke, cardiovascular death, acute or chronic limb ischemia, peripheral vascular interventions, or death. The composite of myocardial infarction, ischemic stroke or cardiovascular death is referred to as major adverse cardiovascular events (MACE). The composite of acute limb ischemia, chronic limb ischemia, and peripheral vascular interventions is referred to as major adverse limb events (MALE). Acute limb ischemia is defined as limb-threatening ischemia that is confirmed by using limb hemodynamic parameters or imaging and leading to an acute vascular intervention within thirty days of onset of symptoms. Chronic limb threatening ischemia is defined as (1) continuing ischemic limb, foot, or digit pain leading to hospitalization and intervention and not meeting the definition of acute limb ischemia, or (2) participants with Rutherford classification 4 to 6 at baseline who had a peripheral vascular intervention over the course of the trial. Peripheral vascular interventions include pharmacological interventions (eg, thrombolysis), plain balloon angioplasty with or without stenting, peripheral artery surgery/reconstruction, and major or minor lower limb amputations. Major amputation is defined as an amputation above the forefoot due to a vascular event while minor amputations are defined as more distal amputations.

Secondary outcomes are the occurrence of the separate elements of the primary composite outcome, and the occurrence of major and clinically relevant bleeding complications. Bleeding complications are defined according to the International Society on Thrombosis and Haemostasis criteria.[19] Major bleeding complications are defined as (1) fatal bleeding, (2) symptomatic bleeding into a critical organ, (3) bleeding causing a fall in hemoglobin level of 20 g L^{-1} (1.24 mmol L^{-1}) or more or leading to transfusion of 2 or more units of whole blood or red blood cells, or (4) surgical site bleeding requiring reoperation.[19] Clinically relevant minor bleeding complications are bleedings leading to (1) hospitalization (including presentation to an acute care facility without an overnight stay), (2) a physician-guided medical or surgical treatment for bleeding, or (3) a change in antithrombotic treatment. An independent, blinded clinical end point committee will determine and grade all adverse clinical events. Other outcomes of interest include patient-reported health-related quality of life (WHOQol-Bref), patient-reported health state (EQ-5D-5L), amount of medical costs (iMCQ), and costs due to productivity loss (iPCQ).

Data collection and management

To ensure privacy, all study participants are identified by a unique subject number used in all correspondence and in the study database. Baseline values such as gender, age, comorbidities, vascular state, and medication use are collected from the medical records and stored on the secured Castor (Castor Electronic Data Capture, Amsterdam, the Netherlands) servers. Case report forms are used to obtain relevant information about ethnicity, smoking behavior, alcohol consumption, and family history of cardiovascular disease. Current height, weight, hip/waist ratio, and blood pressure are measured at baseline. Study-related correspondence, patient records, signed informed consent forms, and source documents with an exception for the questionnaires, will be preserved at the participating site for 15 years. All questionnaires will be sent to and preserved at Radboud University medical center. Source data will be entered in the online database Castor and exported for statistical analyses afterward.

Power calculation

Based on previous literature we assume a risk ratio of 1.8 between carriers of *CYP2C19* *2 and/or *3 allele(s) and noncarriers.[8,9,18,20-26] It is known that 30% percent of the Caucasian population is carriers of 1 or 2 of these LOF alleles.[27-29] Van Mil et al showed that 32% of PAD patients consulting a vascular surgeon at Radboud University Medical Center will develop an adverse clinical event within 1 year.[21] For the power calculation we used a more conservative percentage of 25% over 2 years follow-up to avoid insufficient power due to a lack of events. The 2-year risk for noncarriers to develop an adverse clinical event is 20%. We hypothesize that the risk of an adverse clinical event related to arterial thrombosis in patients with a relevant *CYP2C19* variant that receive genotype-guided antithrombotic therapy will be reduced to the risk of patients without a genetic variant. With an alpha of 5% and a power of 80% a total of 1089 patients need to be included per group. To account for a drop-out rate of 5% we will include 2,276 patients in total.

Statistical analysis

Baseline characteristics for continuous data are described as mean and standard deviation while categorical data will be described as number and percentage. Data is analyzed according to intention-to-treat principle. Primary and secondary outcomes in the intervention and control groups are compared. Additionally, subgroup analyses are performed for intermediate and poor metabolizers.

For both the primary composite outcome as well as the secondary outcomes, Cox proportional-hazards model is used to analyze the primary and secondary time-to-event. Kaplan-Meier estimates of the cumulative proportion of patients with events are performed. Explorative subgroup analyses are performed to assess differences in efficacy between gender, age groups, and ethnic groups. The economic evaluation is embedded in the design of the study and will be undertaken as cost-effectiveness analysis (CEA) with the costs per adverse event avoided over a 2-year period. Additionally, an empirical cost-utility analysis (CUA) will be performed with the costs per quality-adjusted life-year (QALY) as outcome over a 2-year time period. A long-term scenario will be explored by decision analytical modeling according the International Society for Pharmacoeconomics and Outcomes Research and the Netherlands Health Care Institute guidelines for economic evaluations.[30] The CEA closely relates to the results concerning the primary composite outcome, the CUA is performed to enable priority setting during health care policy making across patient groups, interventions, and health care settings. Both analyses will be performed from a societal perspective (as base-case) and the time for empirical data collection is set at 24 months, after which extrapolation to long term will be applied. With the 24 months, horizon discounting of costs and effects are unnecessary. In case of confounding the net monetary benefit (NMB) approach will be applied to incorporate the confounders in the regression model with NMB as dependent variable. In case of QALY as efficiency outcome results will be displayed graphically by means of cost-effectiveness planes and acceptability curves according to the Dutch guideline for economic evaluations.[30] For all statistical analyses, *P*-values of .05 or less are considered significant.

Present status

The first patient was enrolled on March 16, 2021. Currently, 10 participating study centers are actively enrolling patients and a total number of 749 patients have been included so far. Patient recruitment is expected to be completed in 2023.

Discussion

The GENPAD study is, to the best of our knowledge, the first randomized clinical trial to evaluate the efficacy of a *CYP2C19* genotype-guided antithrombotic treatment strategy in PAD patients.

We hypothesize that PAD patients with *CYP2C19* LOF alleles who are treated with clopidogrel are at increased risk of ischemic events. However, large-scale research on PAD patients is lacking, and the existing evidence regarding this association in PAD patients is conflicting. The EUCLID study was a large randomized trial investigating the effects of ticagrelor vs clopidogrel in 13,885 PAD patients.[31] The main conclusion of this trial was that ticagrelor did not reduce ischemic events in patients with PAD. During the EUCLID trial 6,955 patients were randomized to clopidogrel treatment, and 30% of these patients were found to have one1 *CYP2C19* LOF-allele. All poor metabolizers were excluded, and major adverse limb events were not taken into account. In a secondary subgroup analyses of the EUCLID trial, published as a research letter, carriage of one *CYP2C19* LOF allele was not associated with an increased risk of the primary efficacy end point of cardiovascular death, MI, or ischemic stroke.[32] However, the studies of Guo et al and Lee et al in PAD patients do suggest a higher risk of adverse cardiovascular and limb events in *CYP2C19* LOF-allele carriers treated with clopidogrel,[33,34] which is in line with results from previous studies in the fields of cardiology and neurology that showed an increased risk of recurrent ischemic events in carriers of *CYP2C19* LOF allele(s) with coronary or cerebral artery disease that were treated with clopidogrel.

Optimizing clopidogrel treatment is fundamental since clopidogrel is the mainly used antithrombotic drug in the treatment of symptomatic PAD patients. The CAPRIE trial showed that PAD patients receiving clopidogrel monotherapy had a lower risk of cardiovascular events than those using ASA.[35] The previously mentioned EU-

CLID trial compared the use of ticagrelor vs clopidogrel in symptomatic PAD, and ticagrelor was not shown to be superior to clopidogrel in reducing adverse cardiovascular events.[31] Both ticagrelor and prasugrel are not registered for PAD patients, so currently treatment option with an alternative P2Y12 inhibitor is lacking. More recent, the COMPASS trial, although increasing bleeding complications, showed superiority of dual pathway inhibition using low-dose rivaroxaban plus ASA compared to ASA alone in reducing adverse cardiovascular events in PAD patients.[36] Direct comparisons between clopidogrel and low-dose rivaroxaban plus ASA have not been made yet. Therefore clopidogrel remains a pillar in the treatment of symptomatic PAD patients, which is why optimizing clopidogrel treatment is of great importance.

During the GENPAD trial, alternative treatment strategies are used for intermediate and poor metabolizers in the intervention group. Double-dose clopidogrel will be prescribed for intermediate metabolizers since previous research found that clopidogrel 150 mg once daily leads to better platelet inhibition in patients with low clopidogrel responsiveness.[13,14,37-48] Double dose clopidogrel is therefore recommended by the Dutch Pharmacogenetics Working Group (DPWG) of the Dutch Royal Pharmacist Association for patients with an intermediate metabolizer phenotype after PCI or after experiencing a stroke or TIA when use of alternative treatment options are lacking.[49] However, trial evidence for the effectiveness of this approach in patients with PAD is currently lacking which is a potential limitation of our study. Another potential limitation is that an interim analysis is not included in the protocol, so if our power calculation proves inaccurate our study might be underpowered.

The GENPAD trial aims to evaluate the efficacy, safety, and cost-effectiveness of a genotype-guided antithrombotic treatment strategy compared to conventional clopidogrel treatment in PAD patients. We expect to see a higher rate of ischemic events in patients with LOF-allele(s) who are treated with standard clopidogrel dose as compared to patients with LOF-allele(s) treated according to a *CYP2C19* genotype-guided approach. Furthermore, we expect the genotype-guided strategy to be cost-effective. If so, pharmacogenetic testing should be implemented in daily practice and treatment strategies used in the GENPAD trial may be used to optimize treatment of *CYP2C19* LOF-carriers. This knowledge is required to enhance treatment of PAD patients, possibly leading to improved outcomes and reduced cardiovascular morbidity and mortality while reducing health care costs.

Authors' contributions

MCW has contributed to the concept of the study. LHW, JK, RJV, MC, EA, RD, CJZ, VHMD, MMPJR, CK and MCW have contributed to the trial design. JK wrote the first draft of the manuscript. LHW, JK, RJV, MC, EA, RD, CJZ, VHMD, MMPJR, CK and MCW have read, revised, and approved the final manuscript.

Ethics approval and consent to participate

The protocol has been approved by the regional medical ethics committee 'METC Oost-Nederland' on February 2, 2021 (version 2, reference number 2020-7057). Written informed consent will be obtained from participants before proceeding with any of the trial procedures.

Consent for publication

Not applicable.

Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Funding

The study is supported by a grant (grant number: 10330022010007) from ZonMw, an independent Dutch organization for Health Research and Health Innovation. The funding source reviewed and approved the study protocol, but had no role in the design of the study. Neither will they have any role in collection, analysis, and interpretation of data, nor in writing or submitting the manuscript.

Declaration of competing interest

The authors declare that they have no competing interests.

Acknowledgements

Not applicable.

Appendix A: Exclusion Criteria

A potential subject who meets any of the following criteria will be excluded from participation in this study:

- Patients with a known *CYP2C19* genotype or metabolizer state
- Patients treated with coumarins, Non-vitamin K Oral Anti-Coagulants (NOACs), unfractionated heparin (UFH), low molecular weight heparins (LMWH) or double antiplatelet therapy (DAPT) with ASA and a P2Y12 inhibitor for other indications
- Patients having one of the following contraindications for clopidogrel, ASA and/or rivaroxaban
    - Hypersensitivity to clopidogrel, ASA or rivaroxaban

- History of asthma attacks, caused by salicylates or nonsteroidal anti-inflammatory drugs (NSAIDs)
- Patients at significant risk for major bleeding
  - Current gastrointestinal ulceration
  - Presence of malignant neoplasms, with the exception of non-melanoma skin cancer
  - Recent (<2 months) brain or spinal injury
  - Recent (<3 months) brain or spinal surgery
  - Recent (<3 months) intracranial, gastrointestinal or pulmonary hemorrhage
  - Presence of arteriovenous malformations,
  - Major intraspinal or intracerebral vascular abnormalities
  - Congenital or acquired bleeding disorders
  - Uncontrolled severe arterial hypertension (180 mmHg or more systolic, or 110 mmHg or more diastolic)
- Patients with severe hepatic disease: Child-Pugh classification B or C.
- Patients with severe kidney failure: Patients with an estimated glomerular filtration rate < 15 mL/min or requiring dialysis
- Patients with severe heart failure: Patients with a known ejection fraction of < 30% or New York Heart Association class III or IV symptoms
- Patients using methotrexate at a weekly dose of 15 mg or more
- Concomitant treatment with medication with a strong pharmacokinetic interaction with rivaroxaban, leading to contra-indication according to the 'Regionale richtlijn DOAC'[50]

- Patients who are pregnant or breastfeeding
- Patients who are unable to give informed consent, including not being able to understand the Dutch language

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