Drug-drug interactions between dicloxacillin/flucloxacillin and DOACs

Protocol

1. Introduction

Direct oral anticoagulants (DOACs) are a group of anticoagulants that include dabigatran, rivaroxaban, apixaban, and edoxaban. They are used to reduce the risk of stroke and systemic embolism (SE) in patients with non-valvular atrial fibrillation (NVAF) and as prophylaxis against deep vein thrombosis (DVT) and pulmonary embolism (PE) (1).

In vivo and *in vitro* studies have investigated two beta-lactamase-resistant penicillins, dicloxacillin and flucloxacillin, and their potential for drug-drug interactions (2–4).

Co-administration of dicloxacillin and warfarin leads to higher risk of strokes and SE (3). Whether or not a similar association between DOACs and dicloxacillin/flucloxacillin exits, has never been assessed. Administration of either flucloxacillin or dicloxacillin to patients treated with DOACs may result in lower plasma concentrations of DOACs and higher risk of strokes/SE.

2. Aim

With this cohort study we aim, to investigate if co-administration of dicloxacillin/flucloxacillin leads to increased risk of strokes or SE in patients using DOACs.

3. Methods

In a cohort of patients taking DOACs, we will investigat if short-term treatment with dicloxacillin/flucloxacillin leads to an increase in stroke or SE incidence.

3.1 Study population

Patients will enter the cohort when they receiv their first prescription for DOACs.

Patients will be censored from the cohort if they do not continuously fill a prescription for one of the DOACs, resulting in one treatment period not covering the dispensing date of the following treatment period. Only patients ≥18 years old are considered. For the main analysis we exclude patients with a diagnosis of either DVT or PE within the last five years before cohort entry and patients with a diagnose of hip- or knee arthroplasty within the last month before cohort entry.

Patients will be censored from the cohort if they have a dispensing of > 200 capsules of dicloxacillin/flucloxacillin at one occasion or receiving an osteomyelitis or endocarditis diagnosis, experiencing an outcome, death, emigration, end of study period, filled a prescription for Vitamin K antagonists (VKA) or stop filling prescriptions for DOACs (defined as one treatment period of DOACs not cover the dispensing date of the following prescription). Patients can enter the cohort if they at a later point fill a new prescription for DOACs or return to Denmark or the Netherlands after emigration.

3.2 Exposure

From the cohort of patients using DOACs, we compare the incidence of stroke and SE in patients receiving dicloxacillin/flucloxacillin to patients receiving phenoxymethylpenicillin and to patients not ingesting any antibiotics, defined as no antibiotic use 30 days before cohort entry. The group ingesting phenoxymethylpenicillin is included to control for the possible unknown effect from the infection itself. We look at patients using DOACs from 2012 to 2020.

3.3 Data sources

This study is based on secondary data collection.

We will perform the register-based study with flucloxacillin in the Netherlands and the study with dicloxacillin in Denmark. We will also analyze flucloxacillin in Denmark, however; this is only a supplementary analysis as only few patients are treated with flucloxacillin in Denmark.

We use the Danish population-based health registers to create a cohort of patients using DOACS from 2012 to 2020. Hospital-based diagnoses are acquired to elucidate the treatment indication for DOACs and are obtained from the Danish National Patient register (5).

Time for dicloxacillin/flucloxacillin prescription fill is determined using The Danish Prescription Register (6). To link data between registries we use the unique Danish Civil Registration Number (CPR) (7).

In the Netherlands we use the Dutch PHARMO network (8) to create a similar cohort of patients using DOACs in the same period. The PHARMO network contains hospital diagnostic data in ICD-10 codes and prescription claims data in ATC-codes.

3.4 Follow-up

In this study we investigated the effect on short-term dicloxacillin/flucloxacillin treatment (typically 7-10 days). The investigational period started at day 5 and ended at day 20, based on a former study

investigating the time for rifampicin to achieve induction and time after rifampicin treatment to achieve de-induction on CYP3A4 (9).

In the untreated group, the exposure period is assigned randomly to each patient in the cohort and lasts 20 days. Untreated episodes that begin within the first 20 days after dicloxacillin/flucloxacillin or phenoxymethylpenicillin start are excluded.

3.5 Outcomes

3.5.1 Primary outcome

To estimate the hazard ratios (HR) for the four DOACs with a 95% CI comparing the group treated with dicloxacillin/flucloxacillin vs. group treated with phenoxymethylpenicillin, and vs. no treatment with antibiotics.

3.5.2 Secondary outcomes

1. Number needed to treat for one additional patient to be harmed (NNTH)

2. Subgroup analysis (age, sex, or intake of dicloxacillin or flucloxacillin)

3. Extend the follow-up period from 5-20 days to 5-30 days

4. Subgroup analysis excluding patients with a history of diabetes, prior use of dicloxacillin/flucloxacillin, use of other antibiotics (ATC: J01) within 30 days prior to index date, hospitalization within 10 days prior to the index date and concomitant use of any of the drugs mentioned in the propensity score matching (section 3.7.1).

5. Analyze if indication for DOAC treatment has any influence

6. Performing a self-controlled case-crossover study, where patients experiencing an outcome worked as their own control and contributed with data for both the exposed and unexposed follow-up time.

3.6 Statistical analysis

We will describe the cohort using descriptive analysis.

3.6.1 Main analysis

We will calculate the 20-day risks of stroke or SE with a 95% CI for the four DOAC drugs in each exposure group. We will also calculate the hazard ratios (HR) for the four DOACs with a 95% CI comparing the group treated with dicloxacillin/flucloxacillin vs. group treated with phenoxymethylpenicillin, and vs. no treatment with antibiotics. This is done using Cox regression. We will also calculate the number needed to treat for one additional patient to be harmed (NNTH) to evaluate the risk of using dicloxacillin/flucloxacillin when patients are in concomitant DOAC treatment.

A P-value ≤ 0.05 is considered as statistically significant.

3.6.2 Sensitivity analysis

We use stratified analysis to control for confounding by creating subgroups in the sensitivity analysis. Sensitivity analyses are made to elaborate whether the rate of outcome is different in subgroups divided by age, sex, or intake of dicloxacillin or flucloxacillin. We also perform a sensitivity analysis extending the follow-up period from 5-20 days to 5-30 days to elaborate if delayed incidences occur after dicloxacillin/flucloxacillin treatment.

Furthermore, we perform subgroup analyzes excluding patients with a history of diabetes, prior use of dicloxacillin/flucloxacillin, use of other antibiotics (ATC: J01) within 30 days prior to index date, hospitalization within 10 days prior to the index date and concomitant use of any of the drugs mentioned in the propensity score matching, section 3.7.1. Furthermore, we investigate the risk of bleeding after dicloxacillin/flucloxacillin intake compared to intake of phenoxymethylpenicillin and no antibiotic intake.

We will also analyze if indication for DOAC treatment has any influence. We perform a subgroup of patients with DVT and PE. The outcome for this group is set as a new venous thromboembolism (VTE).

3.6.3 Case-cross-over study

We will perform a self-controlled case-cross-over study, where patients experiencing an outcome work as their own control and contribute with data for both the exposed and unexposed follow-up time. By using this design, we manage to control for confounders that are stable over time.

All patients experiencing either stroke or SE are included in the cohort. The time for experiencing an outcome is evaluated and defined as day 0. Day -5 to day -20 are defined as the focal window which determine the reason for the outcome. Five days before, the outcome is disregarded. We apply a wash-

out window of 15 days after the focal window. Likewise, we apply 4 reference windows after the wash-out window, all having a length of 15 days to optimize the statistical accuracy.
From data, ORs with 95% CIs are calculated when comparing the risk of outcome in the treatment period (dicloxacillin/flucloxacillin) with the non-treatment period, using conditional logistic regression. To conclude if the infection contributes to risk of experiencing an outcome, we perform the same analysis with phenoxymethylpenicillin as the exposed follow-up time.

3.7 Confounding

3.7.1 Propensity score matching

We will make a 1:2 propensity score matching on patients receiving dicloxacillin/flucloxacillin to patients receiving phenoxymethylpenicillin and no antibiotic treatment, respectively, this to control for confounding. We will evaluate the success of propensity score matching by using standardized mean differences in patient characteristics. To estimate the propensity score, we use logistic regression. We will apply a window of 180 days when looking back at prescription data and use all available data for diagnoses. Propensity scores were calculated using logistic regression including age, sex, calendar year, season, time since first cohort entry (years), CHAD2DS2-VASc-score (congestive heart failure, hypertension, age ≥ 75 years, diabetes, prior stroke or transient ischemic attack, vascular disease, age 65-74 years, sex class (10)), HAS-BLED-score (hypertension, abnormal renal/liver function, stroke, bleeding, elderly, antiplatelets, non-steroidal anti-inflammatory drugs (NSAIDs) or alcohol use (11)); no information on INR was available), other comorbidities (thyroid disease and cancer), Charlson Comorbidity Index Score (based on comorbidities) (12), concomitant use of drugs potentially interaction with DOACs: **antiarrhythmic drugs** (amiodarone, diltiazem, dronedarone, propafenone, propranolol, quinidine, telmisartan, verapamil), **statin** (atorvastatin, lovastatin, rosuvastatin, simvastatin), **antibiotics** (clarithromycin, erythromycin, isoniazid, metronidazole, quinolones, rifampicin, trimethoprim/sulfametaoxasole), **HIV protease inhibitors, fungostatics** (fluconazole, itraconazole, ketoconazole, posaconazole, voriconazole), **immunosuppressants** (cyclosporin, tacrolimus), **NSAIDs**, and **antacids** (cimetidine, proton-pump inhibitors) (13), and drugs acting as markers of cardiovascular disease: **beta-blocker, RAAS-inhibitors, loop-diuretics, thiazides, Ca-channel blockers.**

3.7.2 Bias

In our study we exclude patients with a diagnose of DVT and PE in the main analysis. We, therefore, obtain a cohort of patients diagnosed with AF and patients where no indication of diagnose is available, named "non-indication" patients, all patients are ingesting DOACs. Based on a former study, we will

perform the main analysis including AF patients and "non-indication" patients and create a subgroup only consisting of AF patients (14). Creating two groups will minimize the risk of selection bias because "non-indication" patients might differ in terms of risk and characteristics compared to the AF-diagnosed patients.

Unmeasured confounding could occur due to confounding by indication. The infection could result in stroke and SE; however, we use an active comparator, phenoxymethylpenicillin, to compensate for this. We, therefore, do not think the infection will confound our result significantly.

By performing a self-controlled case-crossover study we managed to control for confounders that are stable over time.

3.8 Effect modifiers

To account for effect modifiers, we perform subgroup analysis dividing patients according to sex, age, history of diabetes, and indication for dicloxacillin/flucloxacillin.

3.9 Ethical aspects and data protection

In Denmark and the Netherlands register-based studies do not need approval from an ethical committee (16).
All data will be published anonymously.

4. Timeline

4.1 Start and end day of data collection

The data will be collected at 1/1 2021 in Denmark and end at the same day. Start day for data collection in the Netherlands have not yet been set.

4.2 Start of data analysis

Data analysis will begin after the end of data collection.

5. Data management

Data management and statistical programming will be performed in the Danish Health Data Agency's protected computing environment.

Data management will be performed by data managers within the Pharmacoepidemiologic Research Group at the University of Southern Denmark.
Data form the Dutch PHARMO database will stay locally at the protected environment of PHARMO.
We will publish the source codes for analysis on https://gitlab.sdu.dk/pharmacoepi/doac_diclox.

6. Dissemination

The study protocol will be registered in EU-PAS. The results obtained from the study will be published in a peer-reviewed journal regardless of the results. The results will be made available before peer-review on a preprint server, e.g. https://www.medrxiv.org.

7. Amendments and deviation

If any future amendments or deviation occur, they will be recorded in the EU-PAS.

8. Validity of outcome

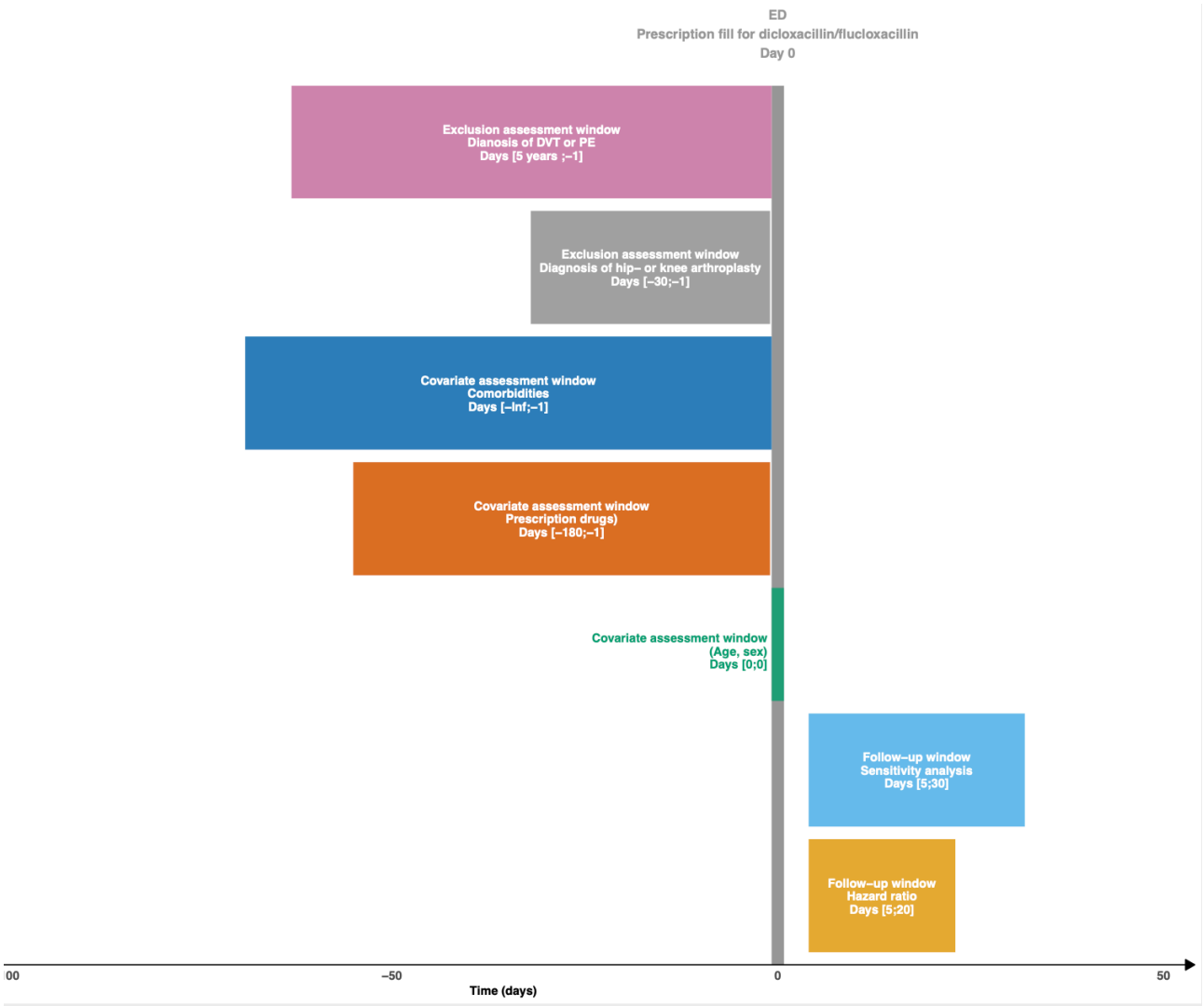
We use The Danish National Patient Registry to establish hospital-based diagnoses. Validity of stroke and SE recorded in The Danish National Patient Registry has been validated previously (17,18). The PHARMO Database Network has been validated regarding linkage against name and address information (8).

Conflict of interest

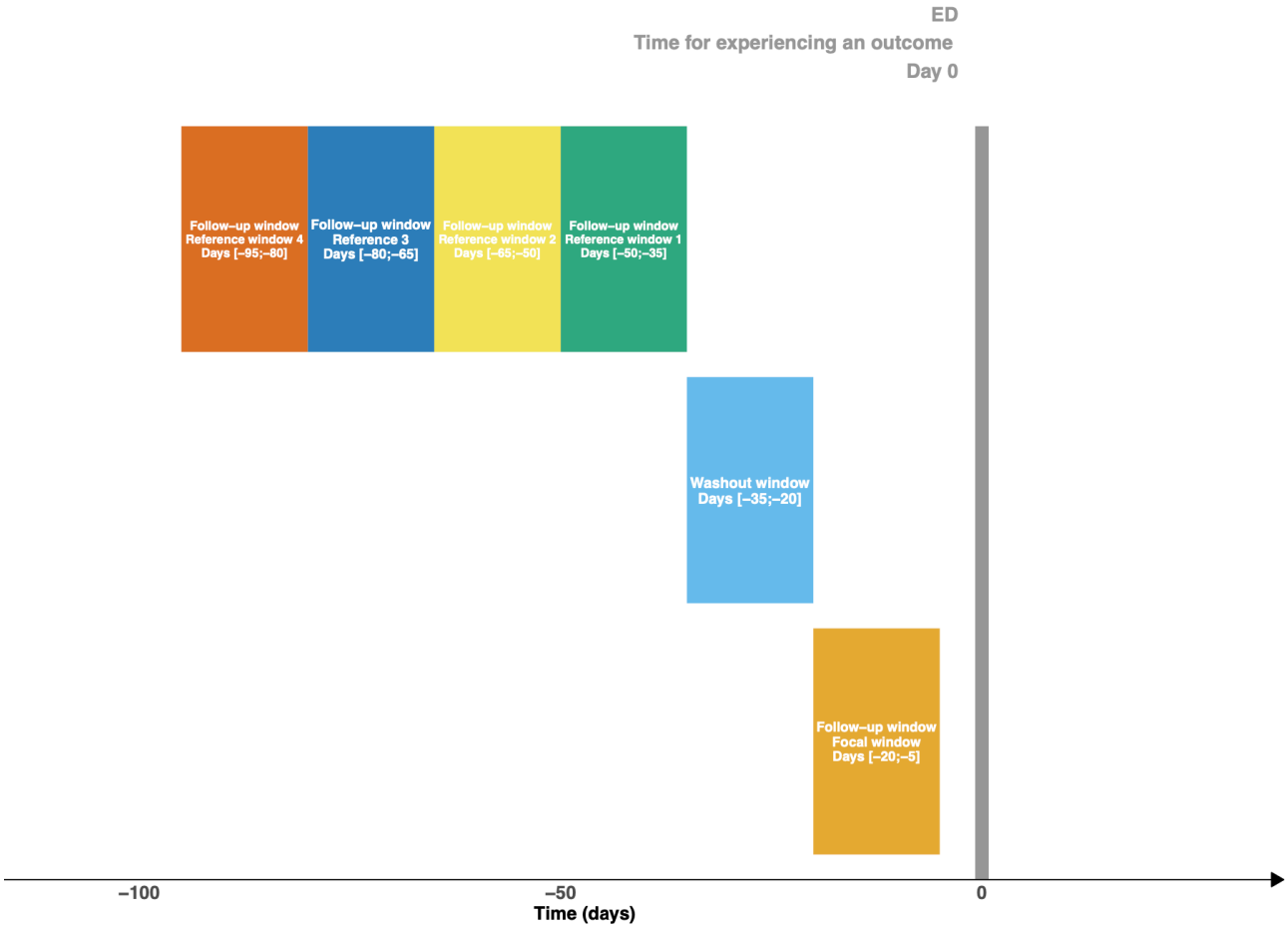
There is no conflict of interest regarding the study.

Figures

Study design diagram for the main analysis



Study design diagram for the self-controlled case-cross-over study



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Appendix

Exposure	Code	
Dicloxacillin	ATC	J01CF01
Flucloxacillin	ATC	J01CF05
Dabigatran	ATC	B01AE07
Rivaroxaban	ATC	B01AF01
Edoxaban	ATC	B01AF03
Apixaban	ATC	B01AF02
Phenoxymethylpenicillin	ATC	J01CE02
Prescription drug		
Amiodarone	ATC	C01BD01
Dilitiazem	ATC	C08DB01
Dronedarone	ATC	C01BD07
Propafenone	ATC	C01BC03
Propranolol	ATC	C07AA05
Quinidine	ATC	P01BC01
Telmisartan	ATC	C09DA07
Verapamil	ATC	C08DA01
Atorvastatin	ATC	C10AA05
Lovastatin	ATC	C10AA02
Rosuvastatin	ATC	C10AA07
Simvastatin	ATC	C10AA01
Clarithromycin	ATC	J01FA09
Erythromycin	ATC	J01FA01
Isoniazid	ATC	J04AC01
Metronidazole	ATC	D06BX01
Quinolones	ATC	J01M
Rifampicin	ATC	J04AB02
Trimethoprim/sulfametaoxasole	ATC	J01EE01

Fluconazole	ATC	J02AC01
Itraconazole	ATC	J02AC02
Ketoconazole	ATC	J02AB02
Posaconazole	ATC	J02AC04
Voriconazole	ATC	J02AC03
Cyclosporin	ATC	L04AD01
Tacrolimus	ATC	D11AH01, L04AD02
Cimetidine	ATC	A02BA01
Non-steroidal inflammatory drug	ATC	N02BE
HIV-protease inhibitor	ATC	J05AE
Proton-pump inhibitors	ATC	A02BC01-08, A02BC53,54
Vitamin K-antagonist	ATC	B01AA03, B01AA04
Beta-blocker	ATC	C07
RAAS-inhibitors	ATC	C09
Loop-diuretics	ATC	C03C
Thiazides	ATC	C03AB, C03AA
Ca-channel blockers	ATC	C08
Antibiotics	ATC	J01
Comorbidities		
Thyroid disease	ICD-10	E00-E07
Cancer	ICD-10	C00-C97, excluding C44
Deep Vein thrombosis	ICD-10	I824
Pulmonary embolism	ICD-10	I26
Osteomyelitis	ICD-10	M86
Endocarditis	ICD-10	I33, I38, I39
Outcome		
Stroke	ICD-10	I63, I64, I693
Systemic embolism	ICD-10	I74

Bleeding	ICD-10	D62, G951A, H113, H356, H431, I60, I61, I62, I864A, I690, I691, J942, K228F, K250, K252, K254, K256, K260, K262, K264, K266, K270, K272, K274, K276, K280, K282, K284 K286 K290 K298A K625 K638B K638C K661 K868G K920-2 N02 R04 R31 R58 S063C S064 S065 S066
Charlson Comorbidity index		
Acute myocardial infarct	ICD-10	I21, I22, I252
Congestive heart failure	ICD-10	I099, I50, I110, I130, I132, I255, I420, I425, I426, I427, I428, I429, I43, P290
Peripheral vascular disease	ICD-10	440, 441, 442, 443, 444, 445, I70, I71, I731, I738, I739, I771, I790, I792, K551, K558, K559, Z958, Z959
Cerebrovascular disease	ICD-10	G45, G46, H340, I60, I61, I62, I63, I64, I65, I66, I67, I68, I69
Dementia	ICD-10	F00, F01, F02, F03, F051, G30, G311
Chronic pulmonary disease	ICD-10	I278, I279, J40, J41, J42, J43, J44, J45, J46, J47, J60, J61, J62, J63, J64, J65, J66, J67, J684, J701, J703
Rheumatologic disease	ICD-10	M05, M06, M315, M32, M33, M34, M351, M353, M360
Ulcer	ICD-10	K25 K26 K27 K28
Liver disease mild	ICD-10	B18, K700, K701, K702, K703, K709, K713, K714, K715, K717, K73, K74, K760, K762, K763, K764, K768, K769, Z944
Diabetes without chronic complication	ICD-10	E100, E101, E106, E108, E109, E110, E111, E116, E118,

		E119, E120, E121, E126, E128, E129, E130, E131, E136, E138, E139, E140, E141, E146, E148, E149
Diabetes with chronic complication	ICD-10	E102, E103, E104, E105, E107, E112, E113, E114, E115, E117, E122, E123, E124, E125, E127, E132, E133, E134, E135, E137, E142, E143, E144, E145, E147
Hemiplegia	ICD-10	G041, G114, G801, G802, G81, G82, G830, G831, G832, G833, G834, G839
Moderate to severe renal disease	ICD-10	I120, I131, N032, N033, N034, N035, N036, N037, N052, N053, N054, N055, N056, N057, N18, N19, N250, Z490, Z491, Z492, Z940, Z992
Any malignancy, incl leukaemia and lymphoma	ICD-10	C (excluding C77, C78, C79, C80)
Liver disease moderate to severe	ICD-10	I850, I859, I864, I982, K704, K711, K721, K729, K765, K766, K767
Metastatic solid tumor	ICD-10	C77, C78, C79, C80
HIV or AIDS	ICD-10	B20, B21, B22, B24
CHA_2DS_2VASc		
Congestive heart failure	ICD-10	I110, I42, I50, J819
Hypertension	ATC	C03A, C08CA, C08DB01, C09A-D
Diabetes	ICD-10	E10-14, G590, G632, H280, H360, N083, O240, O241, O242, O243
	ATC	A10
Stroke/TIA/arterial embolism	ICD-10	G458, G459, I63, I64, I693, I74
Vascular disease (Ischemic heart disease, peripheral arterial disease)	ICD-10	I20, I21, I23, I241, I249, I25, Z951, I700, I702, I708, I709
HAS-BLED score		

Hypertension	ATC	C03A C08CA C08DB01 C09A-D
Abnormal renal function	ICD-10	E102, E112, E122, E132, E142, I12 (÷I129), N01, N03, N083, N085, N118C, N14, N150, N16 (÷ N160), N18 (÷N181), N19, N26, P960, Q601, Q602, Z992
Abnormal liver function	ICD-10	D684C, I850, I859, I982B, K701, K703, K704, K720, K721, K729, K743, K744, K745, K746, K767
Stroke	ICD-10	I63, I64, I693
Bleeding	ICD-10	D62, G951A, H113, H356, H431, I60, I61, I62, I864A, I690, I691, J942, K228F, K250, K252, K254, K256, K260, K262, K264, K266, K270, K272, K274, K276, K280, K282, K284 K286 K290 K298A K625 K638B K638C K661 K868G K920-2 N02 R04 R31 R58 S063C S064 S065 S066
Drugs	ATC	B01AC06, B01AC30, B01AC04, B01AC22, B01AC24, M01A (÷M01AX05), N02BA01
Alcoholism	ICD-10	E244, E529A, F10, G312, G405B, G621, G721, I426, K292, K70, K860, O354, P043, T519, Z502, Z714, Z721
	ATC	N07BB

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European Network of Centres for Pharmacoepidemiology and Pharmacovigilance

ENCePP Checklist for Study Protocols (Revision 4)

Adopted by the ENCePP Steering Group on 15/10/2018

The European Network of Centres for Pharmacoepidemiology and Pharmacovigilance (ENCePP) welcomes innovative designs and new methods of research. This Checklist has been developed by ENCePP to stimulate consideration of important principles when designing and writing a pharmacoepidemiological or pharmacovigilance study protocol. The Checklist is intended to promote the quality of such studies, not their uniformity. The user is also referred to the ENCePP Guide on Methodological Standards in Pharmacoepidemiology, which reviews and gives direct electronic access to guidance for research in pharmacoepidemiology and pharmacovigilance.

For each question of the Checklist, the investigator should indicate whether or not it has been addressed in the study protocol. If the answer is "Yes", the section number of the protocol where this issue has been discussed should be specified. It is possible that some questions do not apply to a particular study (for example, in the case of an innovative study design). In this case, the answer 'N/A' (Not Applicable) can be checked and the "Comments" field included for each section should be used to explain why. The "Comments" field can also be used to elaborate on a "No" answer.

This Checklist should be included as an Annex by marketing authorisation holders when submitting the protocol of a non-interventional post-authorisation safety study (PASS) to a regulatory authority (see the Guidance on the format and content of the protocol of non-interventional post-authorisation safety studies). The Checklist is a supporting document and does not replace the format of the protocol for PASS presented in the Guidance and Module VIII of the Good pharmacovigilance practices (GVP).

Study title:
Drug-drug interactions between dicloxacillin/flucloxacillin and DOACs.

EU PAS Register® number:
Study reference number (if applicable):

Section 1: Milestones	**Yes**	**No**	**N/A**	**Section Number**
1.1 Does the protocol specify timelines for				
1.1.1 Start of data collection[1]	X	☐	☐	4.1
1.1.2 End of data collection[2]	X	☐	☐	4.1
1.1.3 Progress report(s)	☐	X	☐	
1.1.4 Interim report(s)	☐	X	☐	
1.1.5 Registration in the EU PAS Register®	X	☐	☐	6
1.1.6 Final report of study results.	X	☐	☐	6

[1] Date from which information on the first study is first recorded in the study dataset or, in the case of secondary use of data, the date from which data extraction starts.
[2] Date from which the analytical dataset is completely available.

Comments:

Section 2: Research question		**Yes**	**No**	**N/A**	**Section Number**
2.1	Does the formulation of the research question and objectives clearly explain:	X	☐	☐	
	2.1.1 Why the study is conducted? (e.g. to address an important public health concern, a risk identified in the risk management plan, an emerging safety issue)	X	☐	☐	1
	2.1.2 The objective(s) of the study?	X	☐	☐	2
	2.1.3 The target population? (i.e. population or subgroup to whom the study results are intended to be generalised)	X	☐	☐	3.1
	2.1.4 Which hypothesis(-es) is (are) to be tested?	☐	X	☐	
	2.1.5 If applicable, that there is no *a priori* hypothesis?	☐	☐	X	

Comments:

Section 3: Study design		**Yes**	**No**	**N/A**	**Section Number**
3.1	Is the study design described? (e.g. cohort, case-control, cross-sectional, other design)	X	☐	☐	3
3.2	Does the protocol specify whether the study is based on primary, secondary or combined data collection?	X	☐	☐	3.3
3.3	Does the protocol specify measures of occurrence? (e.g., rate, risk, prevalence)	X	☐	☐	3.6
3.4	Does the protocol specify measure(s) of association? (e.g. risk, odds ratio, excess risk, rate ratio, hazard ratio, risk/rate difference, number needed to harm (NNH))	X	☐	☐	3.6
3.5	Does the protocol describe the approach for the collection and reporting of adverse events/adverse reactions? (e.g. adverse events that will not be collected in case of primary data collection)	☐	X	☐	

Comments:

Section 4: Source and study populations		**Yes**	**No**	**N/A**	**Section Number**
4.1	Is the source population described?	X	☐	☐	3.3
4.2	Is the planned study population defined in terms of:				
	4.2.1 Study time period	X	☐	☐	3.3
	4.2.2 Age and sex	X	☐	☐	3.1
	4.2.3 Country of origin	X	☐	☐	3.3
	4.2.4 Disease/indication	X	☐	☐	3.1
	4.2.5 Duration of follow-up	X	☐	☐	3.4

Section 4: Source and study populations	**Yes**	**No**	**N/A**	**Section Number**
4.3 Does the protocol define how the study population will be sampled from the source population? (e.g. event or inclusion/exclusion criteria)	X	☐	☐	3.1

Comments:

Section 5: Exposure definition and measurement	**Yes**	**No**	**N/A**	**Section Number**
5.1 Does the protocol describe how the study exposure is defined and measured? (e.g. operational details for defining and categorising exposure, measurement of dose and duration of drug exposure)	X	☐	☐	3.2
5.2 Does the protocol address the validity of the exposure measurement? (e.g. precision, accuracy, use of validation sub-study)	X	☐	☐	3.2
5.3 Is exposure categorised according to time windows?	X	☐	☐	3.4, 3.6.2, 3.6.3
5.4 Is intensity of exposure addressed? (e.g. dose, duration)	☐	X	☐	
5.5 Is exposure categorised based on biological mechanism of action and taking into account the pharmacokinetics and pharmacodynamics of the drug?	☐	X	☐	
5.6 Is (are) (an) appropriate comparator(s) identified?	X	☐	☐	3.2

Comments:

Section 6: Outcome definition and measurement	**Yes**	**No**	**N/A**	**Section Number**
6.1 Does the protocol specify the primary and secondary (if applicable) outcome(s) to be investigated?	X	☐	☐	3.5
6.2 Does the protocol describe how the outcomes are defined and measured?	X	☐	☐	3.4
6.3 Does the protocol address the validity of outcome measurement? (e.g. precision, accuracy, sensitivity, specificity, positive predictive value, use of validation sub-study)	X	☐	☐	8
6.4 Does the protocol describe specific outcomes relevant for Health Technology Assessment? (e.g. HRQoL, QALYs, DALYS, health care services utilisation, burden of disease or treatment, compliance, disease management)	☐	X	☐	

Comments:

Section 7: Bias	**Yes**	**No**	**N/A**	**Section Number**
7.1 Does the protocol address ways to measure confounding? (e.g. confounding by indication)	X	☐	☐	3.7
7.2 Does the protocol address selection bias? (e.g. healthy user/adherer bias)	X	☐	☐	3.7.2
7.3 Does the protocol address information bias? (e.g. misclassification of exposure and outcomes, time-related bias)	☐	X	☐	

Comments:

Section 8: Effect measure modification	**Yes**	**No**	**N/A**	**Section Number**
8.1 Does the protocol address effect modifiers? (e.g. collection of data on known effect modifiers, sub-group analyses, anticipated direction of effect)	X	☐	☐	3.8

Comments:

Section 9: Data sources	**Yes**	**No**	**N/A**	**Section Number**
9.1 Does the protocol describe the data source(s) used in the study for the ascertainment of:				
9.1.1 Exposure? (e.g. pharmacy dispensing, general practice prescribing, claims data, self-report, face-to-face interview)	X	☐	☐	3.2, 3.3
9.1.2 Outcomes? (e.g. clinical records, laboratory markers or values, claims data, self-report, patient interview including scales and questionnaires, vital statistics)	X	☐	☐	3.3
9.1.3 Covariates and other characteristics?	X	☐	☐	3.7
9.2 Does the protocol describe the information available from the data source(s) on:				
9.2.1 Exposure? (e.g. date of dispensing, drug quantity, dose, number of days of supply prescription, daily dosage, prescriber)	X	☐	☐	3.3
9.2.2 Outcomes? (e.g. date of occurrence, multiple event, severity measures related to event)	X	☐	☐	3.3
9.2.3 Covariates and other characteristics? (e.g. age, sex, clinical and drug use history, co-morbidity, co-medications, lifestyle)	X	☐	☐	3.3
9.3 Is a coding system described for:				
9.3.1 Exposure? (e.g. WHO Drug Dictionary, Anatomical Therapeutic Chemical (ATC) Classification System)	X	☐	☐	Appendix
9.3.2 Outcomes? (e.g. International Classification of Diseases (ICD), Medical Dictionary for Regulatory Activities (MedDRA))	X	☐	☐	Appendix
9.3.3 Covariates and other characteristics?	X	☐	☐	Appendix
9.4 Is a linkage method between data sources described? (e.g. based on a unique identifier or other)	X	☐	☐	3.3

Comments:

Section 10: Analysis plan	**Yes**	**No**	**N/A**	**Section Number**
10.1 Are the statistical methods and the reason for their choice described?	X	☐	☐	3.6
10.2 Is study size and/or statistical precision estimated?	☐	X	☐	
10.3 Are descriptive analyses included?	X	☐	☐	3.6
10.4 Are stratified analyses included?	X	☐	☐	3.6
10.5 Does the plan describe methods for analytic control of confounding?	X	☐	☐	3.7
10.6 Does the plan describe methods for analytic control of outcome misclassification?	☐	X	☐	
10.7 Does the plan describe methods for handling missing data?	☐	X	☐	
10.8 Are relevant sensitivity analyses described?	X	☐	☐	3.6.2, 3.6.3

Comments:

Section 11: Data management and quality control	**Yes**	**No**	**N/A**	**Section Number**
11.1 Does the protocol provide information on data storage? (e.g. software and IT environment, database maintenance and anti-fraud protection, archiving)	X	☐	☐	5
11.2 Are methods of quality assurance described?	X	☐	☐	5
11.3 Is there a system in place for independent review of study results?	☐	X	☐	5

Comments:

Section 12: Limitations	**Yes**	**No**	**N/A**	**Section Number**
12.1 Does the protocol discuss the impact on the study results of:				
12.1.1 Selection bias?	X	☐	☐	3.7.2
12.1.2 Information bias?	☐	X	☐	
12.1.3 Residual/unmeasured confounding? (e.g. anticipated direction and magnitude of such biases, validation sub-study, use of validation and external data, analytical methods).	X	☐	☐	3.7
12.2 Does the protocol discuss study feasibility? (e.g. study size, anticipated exposure uptake, duration of follow-up in a cohort study, patient recruitment, precision of the estimates)	X	☐	☐	3.3

Comments:

Section 13: Ethical/data protection issues	**Yes**	**No**	**N/A**	**Section Number**
13.1 Have requirements of Ethics Committee/ Institutional Review Board been described?	X	☐	☐	3.9
13.2 Has any outcome of an ethical review procedure been addressed?	☐	X	☐	
13.3 Have data protection requirements been described?	X	☐	☐	3.9

Comments:

Section 14: Amendments and deviations	**Yes**	**No**	**N/A**	**Section Number**
14.1 Does the protocol include a section to document amendments and deviations?	X	☐	☐	7

Comments:

Section 15: Plans for communication of study results	**Yes**	**No**	**N/A**	**Section Number**
15.1 Are plans described for communicating study results (e.g. to regulatory authorities)?	X	☐	☐	6
15.2 Are plans described for disseminating study results externally, including publication?	X	☐	☐	6

Comments:

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Date: 16/November/2020

Signature: Ditte Bork Iversen