Performance of a Qualitative Point-of-Care Strip Test to Detect DOAC Exposure at the Emergency Department: A Cohort-Type Cross-Sectional Diagnostic Accuracy Study

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Abstract

An accurate point-of-care test for detecting effective anticoagulation by direct oral anticoagulants (DOACs) in emergencies is an unmet need. We investigated the accuracy of a urinary qualitative strip test (DOAC Dipstick) to detect relevant DOAC exposure in patients who presented to an emergency department. In this prospective single-center cohort-type cross-sectional study, adults on DOAC treatment were enrolled. We assessed clinical sensitivity and specificity of DOAC Dipstick factor Xa and thrombin inhibitor pads to detect DOAC plasma levels $\geq$30 ng/mL using urine samples as the testing matrix. Liquid chromatography coupled with tandem-mass spectrometry was used as the reference standard method for plasma and urine measurement of DOAC concentrations. Of 293 patients enrolled, 265 patients were included in the analysis, of whom 92 were treated with rivaroxaban, 65 with apixaban, 77 with edoxaban, and 31 with dabigatran. The clinical sensitivity and specificity of the dipstick on urine samples to detect $\geq$30 ng/mL dabigatran plasma levels were 100% (95% confidence interval [CI]: 87–100%) and 98% (95% CI: 95–99%), respectively. The sensitivity and specificity of the dipstick to detect $\geq$30 ng/mL factor Xa inhibitor plasma levels were 97% (95% CI: 94–99%) and 69% (95% CI: 56–79%), respectively. The DOAC Dipstick sensitively identified effective thrombin and factor Xa inhibition in a real-world cohort of patients presenting at an emergency department. Therefore, the dipstick might provide a valuable test to detect relevant DOAC exposure in emergencies, although further studies will be needed to confirm these findings.

Keywords
- cohort study
- anticoagulants
- direct thrombin inhibitor
- direct factor Xa inhibitors
- point-of-care-test

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Introduction

Direct oral anticoagulants (DOACs) represent the main class of oral anticoagulants today. The variable effect of DOACs on routine coagulation tests, however, poses a diagnostic challenge that complicates the acute care of patients in emergencies.[1] Routine coagulation tests like prothrombin time or activated partial thromboplastin time are widely available but require sample processing and have low reliability in detecting relevant DOAC exposure.[2,3] Ecarin-based assays, diluted thrombin time, and chromogenic anti-IIa and anti-Xa assays provide more accurate results but are not ubiquitously available. Liquid chromatography coupled with tandem-mass spectrometry (LC–MS/MS) measurement of DOAC plasma levels is most accurate, although it is complex and unsuitable for routine clinical practice.[4]

A reliable point-of-care test (POCT) may avoid these limitations and accelerate decision-making processes in emergencies. The DOAC Dipstick, a CE-marked qualitative strip test, is intended to detect the presence of DOACs in human urine after 10 minutes.[5,6] Its use may ease decision making in medical conditions that require rapid and reliable knowledge of the presence of effective anticoagulation, including ischemic stroke, life-threatening bleeding, or the need for urgent surgery.

In a cohort of outpatients on stable anticoagulation, the dipstick effectively determined the presence of relevant DOAC plasma levels from urine.[5] However, at present, no data are available on dipstick performance in acutely diseased patients at an emergency department. We assessed the accuracy of the DOAC Dipstick in a series of adults presenting at a European high-volume emergency department.

Methods

Study Design, Setting, and Participants

We conducted a prospective single-center cohort-type cross-sectional study to assess the clinical performance of the DOAC Dipstick at the emergency department of the Medical University of Vienna, Austria, an academic tertiary care facility. The emergency department comprises an outpatient and an inpatient clinic, including an intensive care unit, and treats approximately 90,000 patients annually.[7]

We enrolled adult patients (≥18 years) with direct oral thrombin inhibitor (dabigatran) or factor Xa inhibitor (rivaroxaban, apixaban, edoxaban) intake based on medical history who were able to provide a 10-mL urine sample. Unconscious patients and pregnant or breastfeeding women were excluded from study participation following a statement by the local ethics committee. Oral and written informed consents were obtained before any study-related activity. The study was approved by the Ethics Committee of the Medical University of Vienna and conducted in accordance with the latest version of the Helsinki Declaration.

Study Objective

The objective was the evaluation of the clinical sensitivity and specificity of the DOAC Dipstick thrombin and factor Xa inhibitor pads to detect DOAC plasma levels ≥30 ng/mL using urine samples.

Study Procedures

Demographics, past medical history (comorbidities and concomitant medications), clinical characteristics, and the reason for oral anticoagulation were documented after study enrolment. A 10 mL of urine and 6 mL of citrated whole blood samples were obtained. Urine was sampled into 10-mL vacuum-containing VACUETTE urine tubes (Greiner Bio-One GmbH). The urine samples were used to perform the dipstick test and were afterwards stored at −80°C for LC–MS/MS analysis. Adverse events related to the use of the DOAC Dipstick were documented at the end of the visit.

Whole blood was sampled into two 2-mL vacuum tubes containing 3.8% sodium citrate (VACUETTE, Greiner Bio-One GmbH, Großmünster, Austria). One tube was sent to the hospital's central laboratory for global coagulation studies. The other tube was centrifuged at 2,000 g for 10 minutes, and plasma was stored at −80°C until LC–MS/MS analysis.

Testing Methods

DOAC Dipstick Urinary Point-of-Care Device

The DOAC Dipstick[8] is a CE-marked urine-based POCT, with marketing approval in the European Union.[9] It is intended to qualitatively detect the presence or absence of direct oral thrombin (dabigatran) and factor Xa (rivaroxaban, edoxaban, apixaban) inhibitors by visual color identification. Qualitative test results are available after 10 minutes and can be assessed by the naked eye.

The dipstick consists of four different pads embedded on a reagent strip. In the absence or presence of a thrombin inhibitor the respective pad is orange-brown or pink, as for factor Xa inhibitors the respective pad is yellow or white. In cases of normal or low creatinine levels, the belonging pad is dark-liliac or grey. Pads #4 and #3 determine the presence or absence of a thrombin or factor Xa inhibitor, pad #2 assesses urine color (normal vs. abnormal), and pad #1 determines renal function (normal vs. low creatinine levels in urine). Pad #2 assists in avoiding misinterpretation of pads #3, and #4 results and pad #1 in the case of low urine creatinine. The cut-off of the creatinine pad is 0.25 g/L, corresponding to approximately 30 mL/min creatinine clearance.

During the study initiation, investigators underwent a standardized training program on using the DOAC Dipstick and visual identification of test pad colors. Two investigators independently performed the DOAC Dipstick test according to the manufacturer's instructions. Briefly, after collection of a urine sample, the DOAC Dipstick was immerged into urine for 2 to 3 seconds so that all test pads were completely covered by the urine. Excess urine was wiped off and the dipstick was placed on a flat surface. After 10 minutes of incubation time, the test pads were compared by the naked eye to the corresponding color scales on the label of the tube container. In case of a discordant result, the test was repeated a second time, and mutual agreement was reached. Digital photos were taken after 10-minute incubation of each strip,

and control urine samples were analyzed at the start of patient enrolment and after every 10th patient as internal quality controls. Exclusion from analysis occurred only if Dipstick pads #3 or #4 showed abnormal results, regardless of other parameters such as plasma creatinine or bilirubin levels. This included patients with renal insufficiency, urinary bleeding, or hyperbilirubinemia. Patients with renal insufficiency were excluded only if pad #1 showed the result "low," as this may lead to false-negative thrombin and factor Xa inhibitor pad results.

Measurement of DOAC Concentration in the Urine and Plasma (Reference Standard)

LC–MS/MS was used to measure plasma and urine levels of DOACs. A plasma level threshold of $\geq$30 ng/mL has been previously proposed for safe hemostasis and was used as a threshold discriminating effective anticoagulation.[10–13] The lower limit of quantification of plasma and urine DOAC levels was 3 ng/mL. The chromatographic analysis of plasma and urine samples was performed at the Department of Pharmacy, University of Namur, Belgium.

As we investigated the accuracy of a urine test to determine the predefined DOAC plasma level threshold of $\geq$30 - ng/mL, we measured DOAC levels in plasma and urine to allow for a better interpretation of the DOAC Dipstick pad results.

Performers and readers of the DOAC Dipstick test had no information about the clinical characteristics of the study patients and were not aware of the reference test results. Likewise, clinical information and DOAC Dipstick test results were not available to the assessors of the reference standard. Standard coagulation studies and plasma creatinine levels (reference range: $\leq$1.20 mg/dL) were analyzed at the Department of Laboratory Medicine at the Medical University of Vienna.

Statistical Analysis

The sample size was estimated based on precision and derived from previous data of the device. We assumed a prevalence of the reference standard positives to be 60%. According to Harenberg et al, the sensitivity of both pads was expected to be 95%. We chose a sample size where the lower 95% confidence interval (95% CI) band of the sensitivity point estimate did not exceed 5%. A sample size of 250 participants was considered sufficient to assess test accuracy at reasonable precision, practically possible, and accessible in this monocentric study.

Categorical data are presented as absolute numbers and relative frequencies, and continuous data are presented as the mean $\pm$ standard deviation (SD). Plasma and urine levels of DOACs are presented as the median (ng/mL) with 25 to 75% interquartile range and the minimum to maximum range.

Standard measures of test accuracy (sensitivity, specificity, predictive values, accuracy) were calculated according to standard formulas using a dichotomous dipstick as the index test and LC–MS/MS (cut-off $\geq$ 30 ng/mL) as the reference standard, and are presented as mean (%) with 95% CIs. The 95% CIs of the test accuracy measures were calculated based on the Clopper–Pearson exact binomial distribution. The accuracy of the thrombin and the factor Xa inhibitor pad was calculated as the proportion of all samples correctly identified by the test. Dabigatran samples were used as negative controls for the factor Xa inhibitor pad and vice versa. As the performance of the DOAC Dipstick was investigated at an emergency department in a clinical setting and not under laboratory conditions, we use the terms "clinical sensitivity" and "clinical specificity."

Receiver operating characteristic (ROC) curves were used to assess the overall accuracy of the thrombin inhibitor pad and the factor Xa inhibitor pad. As both the DOAC Dipstick test and LC-MS/MS provided dichotomous results, we termed the resulting curve the ROC-type curve. The results are presented as the area under the curve (AUC) with a 95% CI. Kappa coefficients were calculated to assess the strength of agreement between the DOAC Dipstick test and LC-MS/MS and interrater reliability. Spearman's rank-order correlation was used to assess associations between DOAC plasma and urine levels. A p-value of $<$0.05 was considered statistically significant.

Data reporting follows the updated Standards for the Reporting of Diagnostic Accuracy Studies (STARD 2015).[14] Stata Statistical Software, Release 17 (StataCorp 2021, College Station, Texas, United States; StataCorp LLC) was used for statistical analysis.

Results

Participants

Between January 2019 and October 2020, a total of 293 patients were eligible and enrolled in the study (►**Fig. 1**). Twenty-three patients (7.8%) were excluded due to an abnormal urine color pad and/or a low creatinine pad result. Additionally, five patients (1.7%) were excluded due to missing urine samples or dipstick test results. Among the 265 patients (90.4%) in the analysis cohort, 137 were male

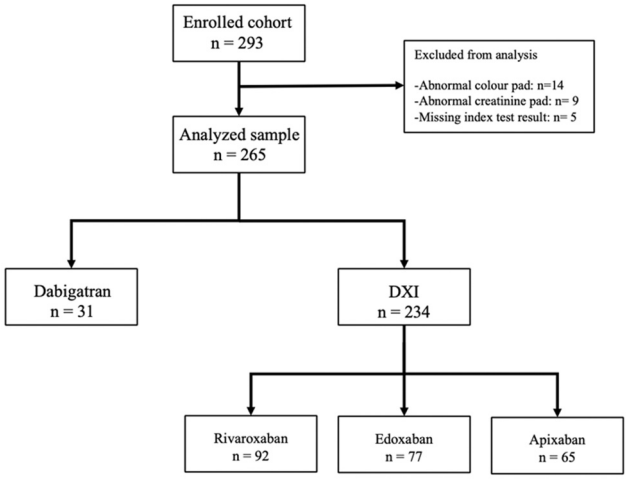

Fig. 1 Flow chart of patients enrolled in the study and included in the analysis. Of 293 patients enrolled in the study between January 2019 and October 2020, 265 patients (90.4%) were analyzed. The remaining 28 patients had to be excluded due to missing, abnormal color, or abnormal creatinine pad results.

(52%), and their mean age was 72 years (SD ± 13 years) (►**Table 1**). Thirty-one patients (11.7%) were on treatment with dabigatran, and 234 patients (88.3%) were on treatment with a direct oral factor Xa inhibitor. Ninety-one patients (34.3%) had elevated plasma creatinine levels >1.2 mg/dL. The proportion of patients with elevated plasma creatinine levels was highest in patients on apixaban (48% [$n = 28$] vs. 37% [$n = 33$], 33% [$n = 10$], and 27% [$n = 20$] in patients on rivaroxaban, dabigatran, and edoxaban). In two patients the dipstick test was repeated due to discrepancies between investigators regarding factor Xa inhibitor pad results. DOAC concentrations in the plasma and urine correlated well, except for apixaban, and are shown in ►**Table 2**.

DOAC Dipstick Test Results

The cross-tabulated DOAC Dipstick test and LC-MS/MS results are shown in ►**Table 3**. ►**Table 4** provides the estimates of test metrics and precision.

The thrombin inhibitor pad identified dabigatran plasma levels ≥30 ng/mL with 100% sensitivity (95% CI: 86.8–100%) and 97.9% (95% CI: 95.2–99.3%) specificity. No false-negative results were observed. The rate of false-positive results was 2.1% (95% CI: 0.7–4.8%). The positive predictive value (PPV) was 83.9% (95% CI 66.3–94.6%), and the negative predictive value (NPV) was 100% (95% CI: 98.5–100%).

The factor Xa inhibitor pad identified ≥30 ng/mL Xa inhibitor plasma levels with a sensitivity of 97.4% (95% CI: 94.1–99.2%) and a specificity of 68.6% (95% CI: 56.4–79.2%). Accordingly, the false-positive and false-negative rates were 31.4% (95% CI: 20.9–43.6%) and 2.6% (95% CI: 0.8–5.9%), respectively. The PPV was 89.6% (95% CI: 84.7–93.4%), and the NPV was 90.6% (95% CI: 79.3–96.9%).

Influence of Plasma Threshold Levels on DOAC Dipstick Accuracy

The sensitivity and specificity values of the thrombin and factor Xa inhibitor pad across DOAC plasma thresholds from 3 to 300 ng/mL are shown in ►**Fig. 2** and ►**Table 5**. The sensitivity for dabigatran was 100% across thresholds. The specificity was constant and ranged from 100 and 98% between 3 and 50 ng/mL and decreased to 89% at 300 ng/mL. The sensitivity for factor Xa inhibitors ranged from 94 to 100% across all thresholds, and the specificity decreased from 100 to 85% at thresholds of 3 to 15 ng/mL and to 25% at 300 ng/mL. ROC-type curves showed an AUC of 0.99 (95%

Table 1 Characteristics of the study cohort

Variable	Direct thrombin inhibitor ($n = 31$)	Direct factor Xa inhibitor ($n = 234$)
Age, y (SD)	72.0 (10.5)	72.4 (13.2)
Male; *n* (%)	22 (71.0)	115 (49.1)
Reason for DOAC intake; *n* (%)		
Atrial fibrillation	28 (90.3)	167 (71.4)
Pulmonary embolism	1 (3.2)	20 (8.5)
Ischemic stroke	2 (6.5)	4 (1.7)
Deep vein thrombosis	0	10 (4.3)
UEVT	0	3 (1.3)
Other	0	22 (9.4)
Unknown	0	8 (3.4)
Dabigatran	31 (100)	0
Apixaban	0	65 (27.8)
Edoxaban	0	77 (32.9)
Rivaroxaban	0	92 (39.3)
Concomitant disease; *n* (%)		
Diabetes mellitus	4 (12.9)	62 (26.5)
Chronic heart failure	9 (29.0)	51 (21.8)
Malignancy	3 (9.7)	22 (9.4)
Coronary artery disease	26 (83.9)	159 (67.9)
Renal insufficiency	3 (9.7)	46 (19.7)
Pulmonary disease	9 (29.0)	57 (24.4)
Reason for admission to ED; *n* (%)		
Chest pain	7 (22.6)	48 (20.5)
Dyspnea	7 (22.6)	44 (18.8)
Palpitations	3 (9.7)	33 (14.1)
Syncope	2 (6.5)	13 (5.5)
Other	12 (38.7)	96 (41.0)
Elevated plasma creatinine; mg/dL (SD)	10 (33.3)	81 (34.6)
APTT; s (SD)	59.7 (24.6)	44.1 (19.8)
PT; % (SD)	67.5 (14.0)	77.3 (25.9)

Abbreviations: APTT, activated partial thromboplastin time; DOAC, direct oral anticoagulant; ED, emergency department; PT, prothrombin time; SD, standard deviation; UEVT, upper extremity vein thrombosis.
Note: Data are mean with standard deviation (SD) or absolute numbers (*n*) and relative frequency (%).

Table 2 DOAC levels in plasma and urine as measured by liquid chromatography coupled with tandem-mass spectrometry

DOAC	*n*	Plasma median (ng/mL)	25–75% IQR	Urine median (ng/mL)	25–75% IQR	Spearman's Rho	*p*-Value
Dabigatran	31	93	64–154	3,769	1,239–7,783	0.636	<0.001
Rivaroxaban	92	95	44–226	1,232	434–2,296	0.697	<0.001
Edoxaban	77	120	41–328	13,414	5,954–25,051	0.811	<0.001
Apixaban	65	136	90–207	378	271–518	0.004	0.977

Abbreviations: DOAC, direct oral anticoagulant; IQR, interquartile range.

Table 3 Cross-tabulation of the DOAC Dipstick test results by results of LC-MS/MS

Factor Xa inhibitor pad			
LC-MS/MS measurement in plasma (reference standard)		Dipstick result in urine sample	
		Positive	Negative
Positive (≥30 ng/mL)	195	190	5
Negative (<30 ng/mL)	70	22	48
Sum	265	212	53
Thrombin inhibitor pad			
LC-MS/MS measurement in plasma (reference standard)		Dipstick result in urine sample	
		Positive	Negative
Positive (≥30 ng/mL)	26	26	0
Negative (<30 ng/mL)	239	5	234
Sum	265	31	234

Abbreviations: DOAC, direct oral anticoagulant.
Note: Test results of the thrombin inhibitor pad and the factor Xa inhibitor pad compared with liquid chromatography coupled with tandem-mass spectrometry (LC-MS/MS) results in plasma.

Table 4 Estimates of accuracy metrics for the thrombin and factor Xa inhibitor pads at a threshold of 30 ng/mL

Test accuracy	Thrombin inhibitor pad		Factor Xa inhibitor pad	
	Mean (%)	95% CI	Mean (%)	95% CI
Sensitivity (TPR)	100	86.8–100	97.4	94.1–99.2
Specificity (TNR)	97.9	95.2–99.3	68.6	56.4–79.2
FNR	0	0–13.2	2.6	0.8–5.9
FPR	2.1	0.7–4.8	31.4	20.9–43.6
NPV	100	98.4–100	90.6	79.3–96.9
PPV	83.9	66.3–94.6	89.6	84.7–93.4
Kappa	0.90	0.82–0.99	0.72	0.62–0.82
Accuracy	0.96	0.93–0.98	0.90	0.86–0.93
AUC	0.99	0.98–1.00	0.83	0.76–0.90

Abbreviations: AUC, area under the curve; CI, confidence interval; FNR, false negative rate; FPR, false positive rate; NPV, negative predictive value; PPV, positive predictive value; TNR, true negative rate; TPR, true positive rate.
Note: Accuracy metrics of the DOAC Dipstick pad for individual factor Xa inhibitors are presented in ►**Supplementary Tables S1** and **S2** (available in the online version). Data are mean (%) with 95% CIs. The kappa coefficient represents the strength of agreement between the DOAC Dipstick and LC-MS/MS method. The accuracy of the dipstick was calculated as the proportion of samples correctly identified by the dipstick. The area under the ROC-type curve (AUC) is an aggregate measure of the DOAC Dipstick's accuracy.

CI: 0.98–1.00) for the thrombin inhibitor pad and 0.83 (95% CI: 0.76–0.90) for the factor Xa inhibitor pad (►**Supplementary Fig. S1** [available in the online version]).

Other Findings

The interrater reliability was 1.00 (kappa, $p < 0.001$) for the thrombin inhibitor pad and 0.97 (kappa, $p < 0.001$) for the FXa inhibitor pad. No adverse events occurred from performing the DOAC Dipstick test.

Discussion

We evaluated the clinical accuracy of a qualitative POCT, the DOAC Dipstick, to detect DOAC plasma levels ≥30 ng/mL using urine samples in a series of unscheduled adults seeking acute care at an emergency department. The availability of such a POCT could directly affect emergency department patient care by guiding timely treatment decisions upon ruling out relevant DOAC exposure at the bedside. At present, however, data on the test's diagnostic performance are available only for outpatients on stable DOAC anticoagulation.[15]

In our study cohort of acutely diseased patients, we had to exclude 23 (7.8%) of the 293 eligible patients following the test specifications, and 5 patients (1.7%) were excluded due to missing urine samples or dipstick test results. In the analyzed study cohort, the dipstick had high sensitivity to DOAC plasma levels ≥30 ng/mL (100% for dabigatran and

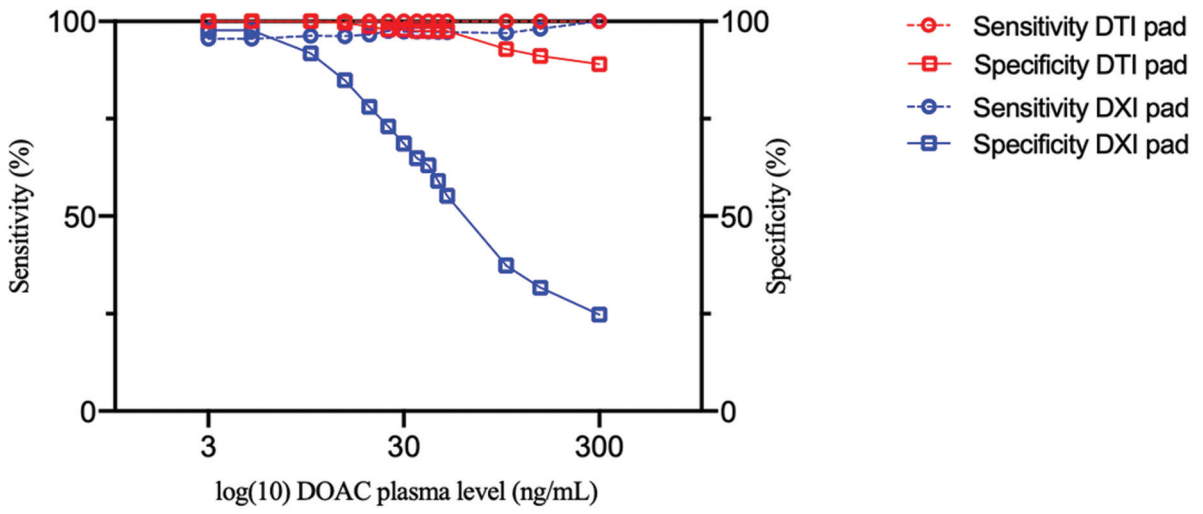

Fig. 2 Clinical sensitivity and specificity of the dipstick according to DOAC plasma thresholds from 1 to 300 ng/mL. *X*-axis: DOAC plasma level (ng/mL, log (10) scale). Left *Y*-axis: sensitivity (%). Right *Y*-axis: specificity (%). The sensitivity (true positive rate, *dashed line with circles*) and the specificity (true negative rate, *solid line with squares*) of the thrombin inhibitor pad (*red*) and the factor Xa inhibitor pad (*blue*) are plotted against selected plasma thresholds for positivity from 3 ng/mL (LLOQ) to 300 ng/mL. DOAC, direct oral anticoagulant; LLOQ, lower limit of quantitation.

Table 5 Clinical sensitivity and specificity values for selected plasma thresholds (from 3 to 300 ng/mL) including the generally accepted threshold of 30 ng/mL, for which, however, there is little evidence

Threshold (ng/mL)	Clinical sensitivity DTI	Clinical specificity DTI	Clinical sensitivity DXI	Clinical specificity DXI
3	100	100	95.5	97.7
5	100	100	95.5	97.7
10	100	100	96.3	91.8
15	100	99.6	96.2	84.9
20	100	98.7	96.6	78.0
25	100	97.9	97.5	73.0
30	**100**	**97.9**	**97.4**	**68.6**
35	100	97.5	97.4	64.9
40	100	97.5	97.4	63.0
45	100	97.5	97.3	59.0
50	100	97.5	97.2	55.2
100	100	92.9	97.0	37.4
150	100	91.1	98.1	31.7
300	100	89.0	100	24.8

Abbreviations: DTI, direct thrombin inhibitor pad; DXI, direct factor Xa inhibitor pad.

97.4% for factor Xa inhibitors) as well as a high specificity for dabigatran (97.9%), but limited specificity for direct factor Xa inhibitors (68.6%) with good interrater agreement. The specificity of the factor Xa inhibitor pad increased by decreasing the plasma threshold.

The need for critical decision making under time pressure at the emergency department requires rapid and accurate information on the presence or absence of relevant anticoagulation. This requirement is impeded by the unreliability and probably long turnaround times of conventional coagulation tests. Furthermore, conditions commonly present in emergency patients, including cognitive disorders, language discordance, unconsciousness, unreliable drug adherence, and ignorance of drug intake, may contribute to a critical lack of information about a patient's coagulation status.[16] In addition, the increasing availability of expensive

DOAC antidotes potentially requires diagnostic tools to facilitate timely and rationale decision-making on their use in emergencies, as the time of last dose intake may be unknown in emergency patients.

DOACs were introduced to clinical practice more than 10 years ago. A simple and reliable test for the timely detection of relevant DOAC exposure is still a diagnostic need, which the DOAC Dipstick is intended to address. The test has previously been shown to perform well in patients on stable anticoagulation but has not yet been tested in emergency settings, where POCT may offer the most significant benefit.

In the current study, we used the ≥30 ng/mL plasma level threshold to indicate effective anticoagulation, as was done in a previous study evaluating the dipstick's accuracy in outpatients on stable anticoagulation.[5] At this threshold, the sensitivity of the thrombin and factor Xa inhibitor pads was comparably high. False-negative pad results were observed only in apixaban samples, likely due to its reduced renal clearance. Accordingly, urine levels of apixaban were lower compared with rivaroxaban and edoxaban and did not correlate with plasma levels. Furthermore, in our study cohort of emergency patients, apixaban urine levels were three times lower on average than in a cohort of stable patients recruited from nonemergency departments of a Croatian University hospital, as recently reported by Margetić and colleagues.[17] The difference is probably explained by the substantial proportion of patients with impaired renal function in our study cohort and may have contributed to the number of false-negative DOAC Dipstick results with apixaban samples.

The thrombin inhibitor pad's specificity was similar to that previously reported by Harenberg et al.[5] The specificity of the Xa inhibitor pad, in contrast, was lower at ≥ 30 ng/mL, where false-positive rates were higher in edoxaban than in rivaroxaban- and apixaban-containing samples, possibly due to its more predominant renal clearance.

We decided to sum the results of the factor Xa pad of patients with dabigatran (n = 31) and with factor Xa inhibitor (n = 234, ►**Table 1**) according to the literature and vice versa for the thrombin inhibitor pad (n = 265, ►**Table 3**).[5,15,17] The same procedure was followed for the evaluations of the rivaroxaban, apixaban, and edoxaban subgroups. For the evaluations of the subgroups with rivaroxaban, apixaban, and edoxaban, we further decided to use the factor Xa inhibitor pad of the dabigatran patients, which resulted in a higher number of results in ►**Supplementary Table S2** (available in the online version) by $n = 31$ compared with the patients in ►**Table 2** who were actually treated with a factor Xa inhibitor. Specificity for the subgroups (values ranging from 77% to 94%) are therefore higher than for the overall factor Xa inhibitor group (68.6%) and in a comparable range to the literature.[5,15,17] In this study, sensitivity, NPV, and PPV of the total and subgroups of factor Xa inhibitors and all values for dabigatran are less affected by the imbalance in the number of patients treated with thrombin and factor Xa inhibitors.

The ≥30 ng/mL plasma threshold has been proposed as the safe-for-treatment threshold for thrombolysis, invasive procedures, surgery, and DOAC reversal.[12,18,19] However, the evidence for clinical importance supporting this threshold is limited, and the clinical value of a qualitative test crucially depends on the discrimination threshold set by the quantitative reference test. Whether the ≥30 ng/mL plasma level is suitable for guiding clinical decisions at the emergency department is unclear and may depend on the individual situation.[20] We, therefore, included accuracy calculations for plasma thresholds ranging from 3 to 300 ng/mL. The sensitivity and specificity of the thrombin inhibitor pad were largely stable between 3 and 300 ng/mL plasma levels. In contrast, the false-positive rate of the factor Xa inhibitor pad substantially increased with increasing plasma levels, resulting in a specificity of 69% at the 30 ng/mL threshold. A 3 ng/mL threshold in plasma samples yielded a sensitivity of >93% and a specificity of 100% for both pads. It is debatable whether these values justify critical treatment decisions but it supports the concept that a sensitive detection of DOAC in urine provides and interesting way for estimating relevant DOAC levels in plasma.

POCTs can be valuable tools in emergencies by reducing turnaround times and hastening patient care and triage. Conditions requiring rapid rule out of DOAC exposure, including the need for emergency surgery, urgent interventional procedures, thrombolysis, or life-threatening bleeding, might be reasonable areas of application for the DOAC Dipstick at emergency departments.[12,18,19] Given the nearly 100% sensitivity and NPV of the DOAC Dipstick observed in the current and previous studies, a negative test result may support immediate clinical decision making. The 100% sensitivity of the direct thrombin inhibitor pad across plasma thresholds from 3 to 300 ng/mL may be suitable to safely rule out effective anticoagulation with dabigatran, for example, in ischemic stroke before intravenous lysis treatment or in cases of severe bleeding or acute surgery before specific antidote administration. However, the number of dabigatran samples in the study cohort was low, and the finding may have limited validity but is comparable to previously published data.[17]

The sensitivity of the factor Xa pad was likewise high across plasma thresholds. Yet, the wrongful administration of thrombolysis or withholding of antidote administration in 3% of patients due to false-negative pad results might not be acceptable. However, the predictive value of a test depends on the prevalence of exposure in a given population, and the NPV of a test increases with decreasing prevalence (e.g., in emergency patients with unknown DOAC exposure).[15] If the DOAC Dipstick is positive, in contrast, decisions may be made based only on the individual clinical situation and additional quantitative laboratory test results.

Which plasma level threshold might be most suitable for use in the emergency department setting and whether this threshold may differ between individual DOACs and according to the individual clinical situation need further investigation.[20]

Limitations

The study was designed as a single-center prospective and controlled cohort study. We included a selected cohort of conscious adults on DOAC treatment who could provide a 10 mL urine sample. In this study, in 7% of enrolled patients, the thrombin and factor Xa inhibitor pads could not be interpreted due to abnormal urine color or creatinine levels and the associated risk of misinterpretation. In this context, it should be noted that our study was conducted in patients with known DOAC exposure, the vast majority of whom gave urine voluntarily. Thus, our findings need to be investigated in additional intended populations. In this regard, it should also be investigated whether the method of urine collection may affect the performance of the dipstick, taking into account the time of the last bladder emptying. As we were interested in the performance of the DOAC Dipstick under "real-life" emergency conditions, the time since the last bladder emptying was not recorded. This information is likely to be unknown in most cases of dipstick use in emergency practice anyway, reflecting the pragmatism of the study design, but may be addressed as part of a follow-up study.

Furthermore, the distribution of DOACs observed in our study is center-specific and may not be representative of other institutions and settings. This is particularly important because the observed sensitivity and specificity values of the factor Xa inhibitor pad for a $\geq$30 ng/mL plasma threshold may vary depending on the individual factor Xa inhibitor. In this context, it must be noted that the study was primarily not designed to evaluate the accuracy of the DOAC Dipstick factor Xa inhibitor pad for individual factor Xa inhibitors. Additionally, the number of patients on dabigatran was small. Thus, appropriate caution should be exercised in interpreting the accuracy estimates observed in the study.

Finally, the clinical value of a POCT depends not solely on the test accuracy, but also on its practicability and ergonomics, which need to be determined in future studies. These should include medical personnel intending to perform the test in the clinical routine (mainly nurses). In this context, a point-of-care reflectance photometer (the Doasense Reader) has been recently developed, which provides an automatic readout with an objective and user-independent test interpretation and might further facilitate the use of the DOAC Dipstick in stressful emergency situations.[17] However, further studies are needed to determine the agreement between visual assessment and Doasense reader result.

Conclusion

This study investigated the clinical accuracy of a POCT to detect relevant DOAC exposure at the bedside in a real-world cohort of adults presenting at an emergency department. In this cohort, the DOAC Dipstick sensitively excluded effective thrombin and factor Xa inhibition at a threshold of $\geq$30 - ng/mL. Further studies may investigate the optimal DOAC plasma level threshold for use in different emergency populations and evaluate whether using the dipstick can directly affect emergency department patient care.

What is known about this topic?

- Direct oral anticoagulants (DOACs) have a variable effect on routine coagulation tests and form a diagnostic challenge that complicates the acute care of patients in emergencies.
- Routine and more specific coagulation tests and chromogenic assays have low reliability in detecting relevant DOAC exposure or are more accurate but are not ubiquitously available. Liquid chromatography coupled with tandem-mass spectrometry (LC–MS/MS) measurement of DOAC plasma levels is most accurate but not suitable for routine clinical practice.
- A reliable point-of-care test (POCT) may avoid these limitations and accelerate decision-making processes in emergencies.

What does this paper add?

- In this study the recently developed POCT strip test on patients' urine samples was first used in real-life emergency settings and was compared with plasma threshold concentration of $\geq$30 ng/mL determined by LC-MS/MS.
- This research provides novel and valuable data for clinical practice concerning timely detection of DOAC exposure and potential therapeutical consequences in adults presenting at an emergency department based on the sensitivity and specificity analysis.
- An accurate point-of-care test may assist physicians to rapidly detect DOAC anticoagulation in patients in an acute care setting and could expedite acute decision-making.

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Conflict of Interest
J.H. is the CEO and founder of Doasense GmbH. J.D. is CEO of Qualiblood SA. All other authors declare no conflicts of interest.

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