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ORIGINAL ARTICLE

Preoperative screening for bleeding disorders: A comprehensive laboratory assessment of clinical practice

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Abstract

Background: Patients with mild bleeding disorders are at risk of perioperative bleeding, but screening for these disorders remains challenging.

Objectives: We aimed to assess the prevalence of hemostatic abnormalities in patients with and without reported bleeding symptoms on a preoperative questionnaire, consisting of guideline-proposed questions, and appraised the diagnostic value of several screening modalities for the identification of patients with hemostatic abnormalities.

Methods: In this observational study, 240 patients with and 95 patients without bleeding symptoms on the preoperative questionnaire were included. Patients with known bleeding disorders, antithrombotic drugs, thrombocytopenia, and anemia were excluded. Preoperatively, all patients underwent elaborate hemostatic testing. Hemostatic abnormalities were defined as coagulation, vWF, or fibrinolysis factor levels below reference range and platelet function defects. Screening modalities included the ISTH Bleeding Assessment Tool (ISTH-BAT), PT, aPTT, TT, Euglobulin Lysis Time (ELT), and Platelet Function Analyser (PFA).

Results: In 21 of 240 (8.8%) patients reporting bleeding symptoms, hemostatic abnormalities were found, including 7 reduced coagulation factor levels, 10 platelet function abnormalities, and 4 reduced vWF levels. In comparison, 10 of 95 (10.5%) patients not reporting bleeding symptoms had abnormalities. The ISTH-BAT could not identify patients with abnormalities, while PT, aPTT, TT, ELT, and PFA had high specificity but low sensitivity to detect abnormalities.

Conclusions: The prevalence of hemostatic abnormalities in both patients with and without reported bleeding symptoms was 9%-10%. This suggests that the guideline-based questionnaire cannot differentiate between patients with and

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without abnormalities, while the discriminative power of the screening modalities is also limited.

KEYWORDS
bleeding disorders, diagnostic techniques, ISTH-Bleeding Assessment Tool, preoperative management

Essentials

- Patients with bleeding disorders are at risk of operative bleeding, but screening for these disorders is challenging.
- Patients with and without bleeding symptoms on a guideline-based screening questionnaire were included and hemostatically phenotyped.
- The questionnaire could not differentiate between patients with and without hemostatic abnormalities.
- The discriminative power of the PT, aPTT, TT, Euglobulin lysis time, PFA, and the ISTH-BAT was also limited.

1 | INTRODUCTION

The term mild bleeding disorders (MBDs) refers to conditions where patients have an increased tendency to skin bruising, menorrhagia, or epistaxis but in whom spontaneous major bleeding episodes generally do not occur. Although, for that matter, many subjects with MBDs remain undiagnosed, there is a clinical relevance in establishing the diagnosis in the preoperative setting, as patients with MBDs are at increased risk of operative bleeding.[1-4] A precise diagnosis of a MBD is important for perioperative hemostatic management, as different disorders require distinct treatment. Merely recognizing a patient as having a "bleeding tendency" limits the types of preventive measures that can be taken.

Accordingly, guidelines developed by the European Society of Anaesthesiology (ESA)[5] and the French Society of Anaesthesiology and Intensive Care (SFAR)[6] advise to screen for bleeding disorders by bleeding questionnaires, and in the case of a positive bleeding history, they suggest consultation of a hematologist. The execution of coagulation tests such as the PT or aPTT is not favored, as these tests were proven insensitive to mild deficiencies.[7-9] In spite of this, a recent survey revealed that these tests are used strikingly often by anesthesiologists.[10] At the same time, the definition of a "positive bleeding history" remains unclear. In clinical practice, most patients reporting bleeding symptoms are not referred to the hematology department; less than 0.01% of preoperative patients are referred for "evaluation of bleeding tendency" in our hospital (1 out of 15 000 patients in 2015; unpublished data). Consequently, most patients who report bleeding symptoms are routinely operated upon, without further hemostatic testing. It is unclear how many of these patients have hemostatic abnormalities that might put them at risk for operative bleeding.

Given remaining uncertainties about current practice, the primary aim was to estimate and compare the prevalence and severity of hemostatic abnormalities in patients with and without reported bleeding symptoms on a guideline-based preoperative questionnaire. Therefore, we extensively tested preoperative patients without known bleeding disorders and not on antithrombotic drugs, using contemporary hemostatic laboratory assays. In addition, we assessed the value of the ISTH-Bleeding Assessment Tool (ISTH-BAT), and laboratory screening tests (Prothrombin Time [PT], activated Partial Prothrombin Time [aPTT], Thrombin Time [TT], Platelet Function Analyser [PFA], Euglobulin Lysis Time [ELT]) for the identification of preoperative patients with hemostatic abnormalities.

2 | METHODS

2.1 | Study population and design

In this explorative observational study, the study population was recruited from consecutive patients who were scheduled for any kind of elective surgery in the Maastricht University Medical Centre (MUMC) in the period from September 2013 to January 2016. During this time frame, 35 000 patients were scheduled for surgery. Subjects eligible for inclusion were ≥18 years old, did not have known bleeding disorders, did not use antithrombotic or nonsteroidal anti-inflammatory drugs. Patients with thrombocytopenia (<100 × 10^9/L) or anemia (men: hemoglobin level <13.2 g/dL, women: <11.8 g/dL) and pregnant women were excluded.

All patients completed a preoperative anesthesiology bleeding questionnaire by themselves, which is part of the routine screening protocol in the hospital. Although a standardized questionnaire is not available, the MUMC questionnaire contains all bleeding questions recommended by the French Society of Anaesthesiology and Intensive Care (SFAR).[6] All preoperative patients who did not object to screening of their medical record by the study team were screened for the presence of bleeding symptoms on this questionnaire, and other in- and exclusion criteria. We primarily aimed to include *all* patients who reported ≥1 bleeding symptoms on this questionnaire ("positive questionnaire"), and a *sample* of patients who did not report bleeding symptoms ("negative questionnaire") (Table 1, Figure 1). In general, patients with a scheduled surgical intervention in the nearest future were approached first. During the study visit, patients completed the ISTH-BAT and blood was drawn

TABLE 1 Self-reported bleeding symptoms on the anaesthesiology bleeding questionnaire

Questions	Patients reporting bleeding symptom(s) (n = 240) n (%) yes	Patients not reporting bleeding symptoms (n = 95) n (%) yes
Prolonged bleeding after pulling teeth/molars or after an operation or after delivery?	48 (20)	0
Spontaneous gum bleeds?	28 (12)	0
Spontaneous large hematomas?	51 (21)	0
Spontaneous nosebleeds?	57 (24)	0
Prolonged bleeding after small wounds (for instance after shaving)?	41 (17)	0
Heavy blood loss during menstruation?	102 (59% of women)	0
Do you have any family relatives with blood clotting problems? (Not due to blood thinning medication)	20 (8.3)	0

Patients could answer "yes" or "no" to each bleeding question present in the anesthesiology questionnaire. Patients completed the questionnaire without supervision of an anesthesiologist. The number of patients answering "yes" to the questions is depicted here. When patients self-reported one or more bleeding symptoms to the questionnaire, they were eligible for inclusion in the "patients reporting bleeding symptom(s)" group.

for hemostatic screening and confirmatory tests (Table 2). A hemostatic abnormality was defined as the presence of a positive result in the confirmatory assays.

The health status of all patients was recorded according to the American Society of Anaesthesiologists physical status classification system.[11]

This study complied with the Declaration of Helsinki. Definite ethical approval for this study was provided by the local Medical Ethical Committee. Written informed consent was obtained from all patients.

2.2 | Blood collection, preparation, and storage

Patients were asked to avoid fat-containing food 4 hours before venepuncture, as light transmission aggregometry (LTA) may be hindered by a lipemic sample.[12] Venous blood was drawn between 9:00-14:30 hours and collected using vacuum tubes. For the preparation of platelet-rich plasma (PRP), citrated-blood was centrifuged at 170 *g* for 10 min at 18°C. Platelet free plasma (PFP) was obtained by centrifugation of citrated blood at 2500 *g* for 5 min and then at 10 000 *g* for 10 min at 18°C (as validated in our laboratory). For all hemostatic tests, citrated blood was used (3.2% sodium citrate, Greiner bio-one; ELT: Trinilyze Stabilyte).

Blood count, blood type (EDTA 7.2 mg, Plymouth), aPTT, PT, TT, fibrinogen, and LTA measurements were performed within 2 hours of blood collection; the PFA 1 hour after blood collection. All other tests were performed in stored PFP (−80°C), frozen within 2 hours of blood collection.

2.3 | Hemostatic confirmatory tests

2.3.1 | Coagulation, von Willebrand Factor (vWF), and fibrinolysis

FII, FV, FVII, FX (Neoplastine R), FVIII, FIX, FXI, FXII (CK Prest) activity levels were measured by clotting assays using specific factor-depleted plasma as substrate on Sta-R (Stago, Paris, France). Fibrinogen level (Clauss method, Thrombin Reagent; Siemens, Marburg, Germany), FXIII activity and vWF antigen and activity (FXIIIact/subs, vWF Reag and vWF Ac Reagens; Siemens) measurements were performed on a Sysmex CS2100i. α2-antiplasmin level was measured using a chromogenic assay (Stachrom; Stago), tissue plasminogen activator (tPA) and plasminogen activator inhibitor 1 (PAI-1) activity were measured using bio-immunoassays (Zymutest, Hyphen-BioMed, Neuville-sur-Oise, France).

2.3.2 | Platelet function

For the LTA (Hart Biologicals, Hartlepool, UK), platelet count in PRP was unadjusted if $<600 \times 10^9$/L; higher counts were adjusted to 500×10^9/L.[12] Platelets were stimulated using arachidonic acid (AA; Bio/Data) 1 mmol/L, thrombin receptor activating peptide (TRAP; Boom H8105) 15 µmol/L, Collagen (Chrono-par Ref385) 1 and 4 µg/mL, Ristocetin (Chronolog Stago Ref396) 1.5 mg/mL, Epinephrine 10 µmol/L (chronolog CH393), ADP (chronology CH384) 5 and 10 µmol/L. Maximal aggregation was recorded and curves were visually assessed. A curve was considered abnormal in case of maximal aggregation <60%, reversibility to <50% and if maximal aggregation was delayed (not reached within 3 minutes; delayed aggregation to epinephrine was considered normal). In case of a deviant curve, aggregation with the concerning agonist was repeated (with the same PRP).

2.4 | Hemostatic screening tests and tools

2.4.1 | Coagulation

aPTT (Actine FSL; Siemens), PT (Innovin Pt; Siemens), and TT (Thromboclotin; Siemens) were performed on a Sysmex CS2100i.

2.4.2 | Fibrinolysis

The ELT was performed similar to Kowalski et al.[13] Differently, the precipitate was dissolved in 1 mL Veronal buffer and the

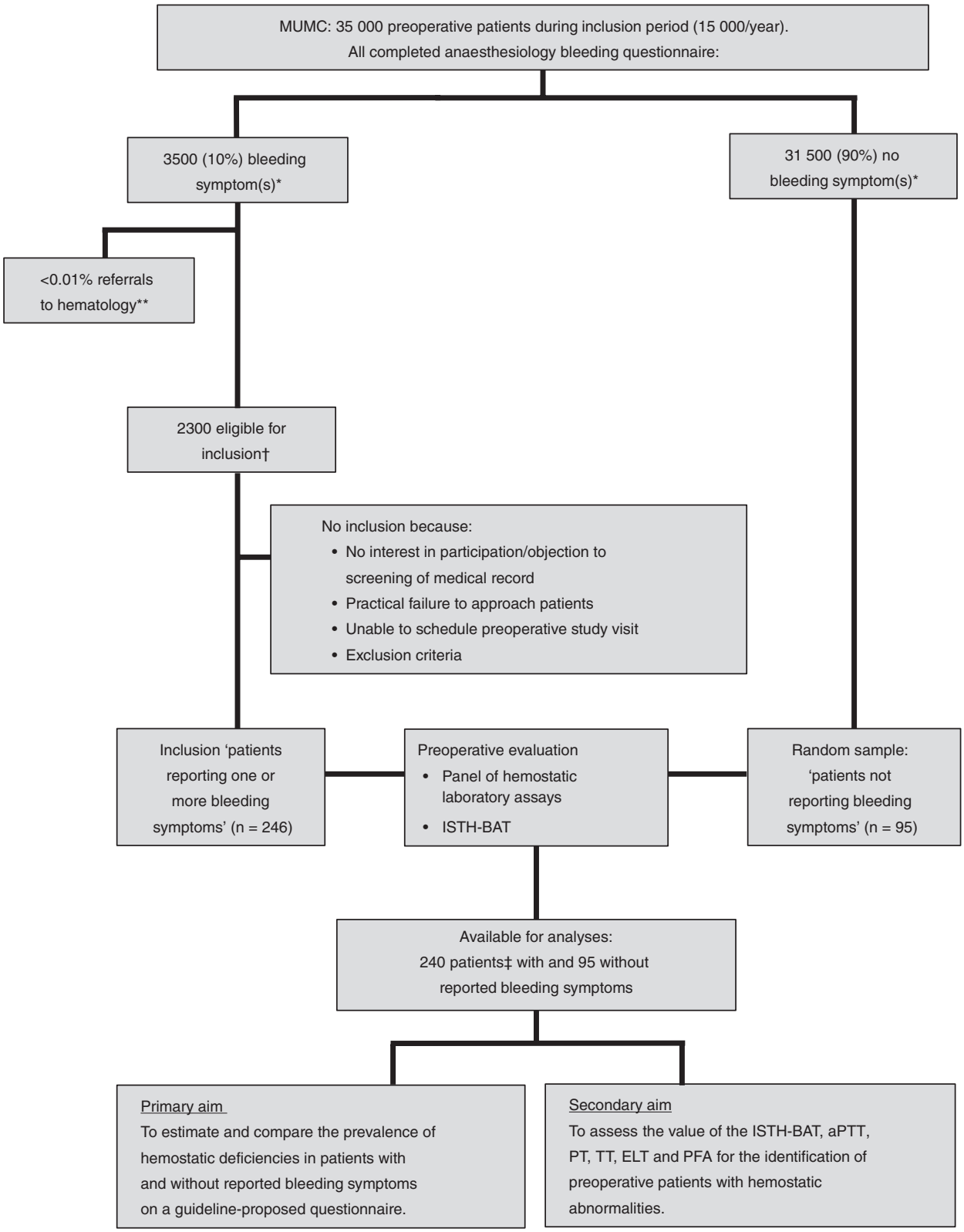

FIGURE 1 Flow chart of the study. *Based on unpublished data from our hospital. **In 2015, only one patient was referred for hemostatic consultation, but this patient was not diagnosed with or treated for a bleeding disorder. †About two-thirds of patients reporting bleeding symptoms were eligible as they met in- and exclusion criteria; most importantly these patients did not use antithrombotic drugs. As many patients as possible were included. ‡Exclusion: 6 patients without any hemostatic test results due to failure of blood withdrawal. aPTT, activated Partial Prothrombin Time; ELT, Euglobulin Lysis Time; MUMC, Maastricht University Medical Centre; ISTH-BAT, International Society of Thrombosis and Haemostasis – Bleeding Assessment Tool; PFA, platelet function analyser; PT, prothrombin time; TT, thrombin time

TABLE 2 Hemostatic tests

	Coagulation	vWF	Platelet function	Fibrinolysis
Confirmatory tests	Fibrinogen, factor II, V, VII, VIII, IX, X, XI, XII, XIII activity	Antigen and activity	LTA with AA, TRAP, Collagen, Epinephrine, Ristocetin, ADP	tPA, PAI, α2-antiplasmin activity
Screening tests	aPTT, PT, TT	PFA	PFA	ELT

Overview of confirmatory and screening laboratory tests. AA, Arachidonic Acid; ADP, Adenosine Diposphate; aPTT, activated Partial Prothrombin Time; ELT, Euglobulin Lysis Time; LTA, Light Transmission Aggregometry; PAI, Plasminogen Activator Inhibitor; PFA, Platelet Function Analyser; PT, Prothrombin Time; tPA, tissue Plasminogen Activator; TT, thrombin time; TRAP, Thrombin Receptor Activating Peptide; vWF, von Willebrand Factor.

reaction was started by adding 25 μL thrombin 100 U/mL (Thrombin Reagent; Siemens). Every 10 to 15 minutes, clot resolution was assessed visually, and every 5 minutes when clot dissolution was almost reached.

2.4.3 | Platelet function and vWF

Collagen-ADP (C-ADP) and collagen-epinephrine (C-epi) cartridges were used to measure closure times (CT; seconds) on the PFA-200 (Siemens).

2.4.4 | ISTH-BAT

Patients completed the ISTH-BAT together with trained medical research personnel, but not with a hematologist or anesthesiologist. We participated in the ISTH-BAT Repository maintained by the Rockefeller University Laboratory of Blood, Vascular Biology, and Informatics Core. The ISTH-BAT consists of 14 bleeding items and the maximal score is 4 per item (latest version, updated July 19, 2011).[14] Information was administered to the Rockefeller website to retrieve bleeding scores.[15]

2.5 | Definitions of hemostatic abnormalities

Patients with positive test results in the confirmatory tests were considered to have a hemostatic abnormality. Positive test results were defined as coagulation or fibrinolysis factor activity level below the reference range, or ≥2 abnormal LTA agonists (or single abnormality in 4 μg/mL Collagen or Ristocetin[16]). Hospital reference ranges were applied (Table 4). Only low FXII and PAI-1 activity were not defined as abnormalities, as low FXII activity does not cause bleeding[17] and PAI-1 activity of 0 is frequently found in healthy individuals.[18]

Local reference ranges for tPA and ELT were not available. For these we generated reference values using 40 healthy volunteers (previously described[19]) per Clinical Laboratory Standards Institute guidelines, preparing the samples and performing the tests exactly as described above.

2.6 | Clinical bleeding outcomes

The proportion of patients requiring perioperative red blood cell transfusions was chosen as bleeding outcome, as bleedings requiring transfusion were considered most relevant and were well-documented.

2.7 | Study policy regarding screening assay abnormalities

Whole blood count and most screening assays (PT, aPTT, TT, PFA) were immediately available, and therefore blinding of all study members for these results was impossible. For ethical reasons, abnormal results in these assays were discussed with a hematologist and a clinical chemist. When the abnormalities *and* the bleeding history were reason for concern, our study team referred patients to the hematologist.

2.8 | Statistical analysis

Continuous variables are expressed as mean and standard deviation (SD) for normally distributed variables; categorical variables are expressed as counts and percentages. Continuous variables were compared using Student's *t* test for normally distributed variables. Categorical variables were compared using the Chi squared-test or Fisher's exact test when expected frequencies were <5.

Two separate binary logistic regression analysis models were applied using the presence vs absence of hemostatic abnormalities as dependent variable, and the results of the anesthesiology bleeding questionnaire (positive/negative) and the ISTH-BAT score as independent variables, respectively. These models allowed evaluation of the ability of these questionnaires to discriminate between patients with and without hemostatic abnormalities while adjusting for differences in baseline variables.

Diagnostic parameters of laboratory screening assays were evaluated by using confirmatory test results as reference. Diagnostic performance was quantified by sensitivity, specificity, positive and negative predictive value (PPV, NPV).

As there is no consensus on cut-off values for the ISTH-BAT, the diagnostic performance was visualized by Receiver-Operating-Characteristics curves (ROC). The area under the curve (AUC) with 95% confidence intervals (CI) was calculated. To demonstrate the diagnostic performance of the ISTH-BAT when using previously proposed cut-off values,[20] we calculated diagnostic parameters with the cut-offs of >5 for women and >3 for men.

The primary aim was to estimate the prevalence of hemostatic abnormalities in patients with a positive anesthesiology

questionnaire (reporting ≥1 bleeding symptoms). A sample size of at least 196 patients with a positive result was required for estimation of an expected prevalence of 15% with a maximal width of the 95% CI of ±5%. To enable comparison of the observed prevalence with patients having a negative questionnaire, a random sample of these patients was taken to prevent selection bias. The inclusion rate of patients with a negative score on the questionnaire was based on maximal feasibility; we primarily included patients reporting bleeding symptoms and patients *not* reporting bleeding symptoms were included whenever this was practically possible.

Statistical analyses were performed with IBM SPSS statistics version 24.0 (International Business Machines Corp., Armonk, NY, USA); significance was assumed at $P < .05$.

3 | RESULTS

The flow diagram in Figure 1 shows that 240 patients reporting ≥1 bleeding symptoms on the anaesthesiology questionnaire and 95 patients who did not report bleeding symptoms were included (Tables 1-3).

3.1 | Hemostatic abnormalities in patients reporting bleeding symptoms

Twenty-one of 240 patients (8.8%) with a positive anaesthesiology bleeding questionnaire had a hemostatic abnormality as detected by the confirmatory tests (Table 4).

One patient had combined mildly reduced FII and FVII activities, 5 patients had decreased FVII activity, and 1 patient had decreased FXI activity. Four patients had reduced vWF levels; in 3 patients vWF antigen or activity were just below 50%, while one patient had levels of 27% and 17%, respectively. No patients showed hyperfibrinolytic abnormalities. In 10 patients, abnormal platelet function was detected.

Three patients required perioperative blood transfusion with packed cells. These patients underwent major abdominal or thoracic surgery. Two patients had no hemostatic abnormalities, while one patient showed signs of a platelet function abnormality.

Based on an abnormal screening test (PFA: prolonged CTs) and the bleeding history, one patient who later also appeared to have a hemostatic abnormality in our confirmatory assays (vWF antigen/activity of 27%/17%), was sent to the hematologist *preoperatively by our study team* (for ethical reasons). This patient received perioperative FVIII/vWF concentrates and therefore potential bleeding may have been prevented.

TABLE 3 Baseline characteristics

Patient and laboratory characteristics	Patients reporting bleeding symptom(s) (n = 240) Mean (SD) or n (%)	Patients not reporting bleeding symptoms (n = 95) Mean (SD) or n (%)	*P* values
Female	173 (72)	43 (45)	<.001
Age (years)	50 (14.8)	56 (13.4)	.001
BMI (kg/m^2)	26.5 (5.2)	26.6 (4)	.86
Renal dysfunction (eGFR <60 mL/min)	15 (6.3)	7 (7.5)	.7
Liver dysfunction	0	0	1
Blood type 0	112 (46.7)	28 (29.5)	.004
ASA 1 ASA 2 ASA 3 ASA 4 or 5	98 (40.1) 134 (55.8) 8 (3.3) 0	42 (44.2) 52 (54.7) 1 (1.1) 0	.47
Orthopedic surgery General surgery Gynecological surgery Oral surgery Ear nose throat surgery Urological surgery Neurosurgery Other	59 (24.6) 55 (22.4) 35 (14.6) 8 (3.3) 16 (6.5) 11 (4.5) 22 (8.9) 34 (13.8)	32 (33.7) 21 (22.1) 4 (4.2) 2 (2.1) 9 (9.5) 5 (5.3) 5 (5.3) 17 (17.9)	.12
Hemoglobin (g/dL) Women Men	 13.7 (1.1) 15.3 (1)	 13.9 (1) 15.0 (1.2)	 .38 .29
MCV (fL)	90.2 (4.6)	90.6 (3.6)	.41
Thrombocytes (10^9/L)	270 (60)	271 (75)	.93
MPV (fL)	10.5 (0.87)	10.5 (0.88)	.42

Baseline characteristics. ASA, American Society of Anesthesiologists; BMI, body mass index; eGFR, estimated glomerular filtration ratio; MCV, mean cell volume; MPV, mean platelet volume; SD, standard deviation.

TABLE 4 Hemostatic abnormalities (confirmatory tests)

Coagulation, platelet function and fibrinolysis tests (reference range)	Patients reporting bleeding symptom(s) (n = 240) Abnormalities n (%)	Range abnormalities	Patients not reporting bleeding symptoms (n = 95) Abnormalities n (%)	Range abnormalities	*P* values
Fibrinogen (<1.7 g/L)	0	*NA*	0	*NA*	*NA*
FII (<60%)	1 (0.4)	59	0	*NA*	1
FV (<60%)	0	*NA*	0	*NA*	*N*
FVII (<60%)	6 (2.5)	47-59	1 (1.1)	57	.68
FVIII (<50%)	0	*NA*	0	*NA*	*NA*
FIX (<60%)	0	*NA*	0	*NA*	*NA*
FX (<60%)	0	*NA*	1 (1.1)	56	.28
FXI (<60%)	1 (0.4)	57	0	*NA*	1
FXII (<60%)[a]	12 (5)	38-59	2 (2.1)	58-59	.37
FXIII (<70%)	0	*NA*	1 (1.1)	14	.28
vWF antigen (<50%)	3 (1.2)	26.6-48.9	0	*NA*	.56
vWF activity (<50%)	4 (1.7)	16.8-49.8	0	*NA*	.58
LTA AA 1 mmol/L (<60%)	2 (0.8)	15.4-19.3	0	*NA*	1
LTA TRAP 15 µmol/L (<60%)	4 (1.7)	17.7-33.9	2 (2.1)	32.2-37.9	1
LTA collagen 4 µg/mL (<60%)	0	*NA*	0	*NA*	*NA*
LTA collagen 1 µg/mL (<60%)	6 (2.5)	5.3-54.1	2 (2.1)	15.7-43	1
LTA ristocetin 1.5 mg/mL (<60%)	0	*NA*	0	*NA*	*NA*
LTA epinephrine 10 µmol/L (<60%)	4 (1.7)	4.2-37.5	0	*NA*	.58
LTA ADP 5 µmol/L (<60%)	3 (1.3)	51.0-57.2	2 (2.1)	55.6-59.2	.63
LTA ADP 10 µmol/L (<60%)	0	*NA*	0	*NA*	*NA*
Abnormal platelet function (≥2 agonists deviated, or collagen 4/ristocetin abnormalities)	10 (4.2)	2-4	6 (6.3)	2-3	.41
tPA activity (>2.23 IU/mL)[b]	0	*NA*	1 (0.6)	3.16	.29
α2-antiplasmin (<80%)	0	*NA*	0	*NA*	*NA*
PAI-1 (0 ng/mL)[a,c]	88 (36.7)	0	20 (21.1)	0	.006
Any kind of abnormality	21 (8.8%)		10 (10.5%)		.61

AA, arachidonic acid; Aggr, aggregation; ADP, adenosine diphosphate; LTA, light transmission aggregometry; PAI-1, plasminogen activator inhibitor-1; tPA, tissue plasminogen activator; TRAP, thrombin receptor activating peptide; vWF, von Willebrand factor. Reference ranges were adapted from our hospital and established in accordance with the Clinical Laboratory Standards Institute guidelines.

Hemostatic abnormalities as detected by the confirmatory assays, including the range of detected abnormalities.

[a]Low levels of FXII or PAI are not considered to be hemostatic abnormalities.

[b]Reference ranges were established using EP-evaluator; nonparametric Index Method.

[c]As the reference range of PAI-activity includes 0 ng/mL, we can only assess whether PAI-activity of 0 is more frequent in patients with a high score.

3.2 | Hemostatic abnormalities in patients not reporting bleeding symptoms

In comparison, 10 of 95 patients (10.5%) who scored negatively on the anaesthesiology questionnaire showed hemostatic abnormalities (Table 4). In 2 patients FVII or FX activity were decreased and one patient had FXIII activity of 14%. A possible platelet function defect was detected in 6 patients. One patient showed an increased tPA activity.

None of these 95 patients needed perioperative blood transfusion, nor was referral to the hematologist by the study team necessary.

3.3 | Discriminative power of the anaesthesiology bleeding questionnaire

There was no difference in the prevalence of hemostatic abnormalities between patients with a positive (8.8%) or negative (10.5%) score on the anesthesiology bleeding questionnaire (Table 4). Binary logistic regression analysis revealed that after adjustment for differences in age, gender, and blood type, patients with a positive anesthesiology questionnaire did not have a higher chance of having a hemostatic abnormality than patients with a negative questionnaire (positive questionnaire: OR 0.73, 95% CI 0.31-1.73, $P = .46$. Supplementary Table S1A). We adjusted for difference in blood type,

TABLE 5 Diagnostic performance of screening tests regarding detection of hemostatic abnormalities

Screening tests (reference range)	All patients (n = 335), Abnormal screening test results n (%)	Range of screening test abnormalities (sec, min, score)	Sensitivity % (95% CI)	Specificity % (95% CI)	PPV % (95% CI)	NPV % (95% CI)
aPTT (>32 s)	3 (0.9)	34	5.9 (0.3-15)	99 (99-100)	33 (1.8-87)	95 (95-96)
PT (>11.5 s)	5 (1.5)	11.8-12.2	63 (32-63)	100 (99-100)	100 (50-100)	99 (98-99)
TT (>21 s)	0 (0)	—	*NA*	100	*NA*	100
PFA-epi (CT>160 s)	19 (5.7)	161-300	10 (1.8-30)	93 (93-95)	8.7 (1.5-2.7)	94 (94-95)
PFA-ADP (CT>118 s)	23 (6.9)	119-300	5.0 (0.3-24)	94 (94-96)	5.3 (0.3-25)	94 (94-95)
ELT (<40 min)[a]	1 (0.3)	15	0 (0-91)	99.7 (99-100)	0 (0-91)	99.7 (99-100)
ISTH-BAT females (>5), n = 216 ISTH-BAT males (>3), n = 119	62 (29) 21 (18)	0-16 0-14	31 (12-58) 27 (9.2-52)	72 (70-74) 84 (81-87)	8.1 (3.2-15) 19 (6.6-37)	93 (91-96) 89 (86-93)

CI; confidence interval. CT; Closure Time. NPV; negative predictive value. PPV; positive predictive value.
Diagnostic performance of the prothrombin time (PT) for detecting reduced FII, V, VII, and X activity levels, activated partial prothrombin time (aPTT) for detecting reduced FII, V, VIII, IX, X, XI, and XII activity levels, thrombin time (TT) for reduced fibrinogen levels (and dysfunction), the platelet function analyser (PFA) for detecting reduced von Willebrand factor antigen/activity or platelet function defects, the euglobulin lysis time for high tPA levels, and the ISTH-BAT for any kind of hemostatic abnormality. See Supplemental Digital Content Table S2 for cross tables. Hospital reference ranges were applied, except for ELT.
[a]Reference ranges were established using EP-evaluator; nonparametric Index Method.

as patients with blood type 0 have 20% and 30% lower vWF levels due to increased clearance from the circulation.[21]

3.4 | Diagnostic performance of screening tests and ISTH-BAT

Last, we appraised the diagnostic performances of the hemostatic screening tests and the ISTH-BAT in all subjects, to see if these tests could discriminate between patients with and without hemostatic abnormalities (eg, instead of the anaesthesiology bleeding questionnaire).

The aPTT, PT, PFA, ELT and the ISTH-BAT (using proposed BAT cut-off values[20]) had high specificity but low sensitivity, indicating that these screening modalities cannot exclude mild coagulation factor, vWF or platelet function, and hyperfibrinolytic abnormalities, respectively (Table 5 and Supplementary Table S2). The PT performed best, with a sensitivity of 63% and NPV of 99%. ROC-curve analysis revealed that the ISTH-BAT had poor ability to identify patients with hemostatic abnormalities using any cut-off level (women: AUC 0.51, 95% CI 0.34-0.67, $P = .94$. men: AUC 0.55, 95% CI 0.39-0.72, $P = .51$. Supplementary Figure S1). Also, when correcting for age, gender and blood type, a higher ISTH-BAT score was not associated with a higher chance of having a hemostatic abnormality (OR 1.05, 95% CI 0.93-1.20, $P = .41$. Supplementary Table S1B).

4 | DISCUSSION

Patients with MBDs are at risk of perioperative bleeding, which was shown to have a major impact on duration of hospital stay, morbidity and mortality.[1-3,22] The prevalence of MBDs and how to screen for these disorders in the preoperative setting remains uncertain.

In this explorative observational study, we performed an elaborate hemostatic laboratory workup in preoperative patients with and without reported bleeding symptom(s) on the guideline-based bleeding questionnaire, and found a prevalence of hemostatic abnormalities of 9% and 10%. Most hemostatic abnormalities were mild and would not prompt treatment in clinical practice.[9,23] Moreover, patients reporting ≥1 bleeding symptoms on the anaesthesiology questionnaire did not have more abnormalities than those who did not report bleeding symptoms, nor were these more severe. The observation that anesthesiologists refer practically no patients to the hematology department (one patient in 2015) indicates that the screening procedure does not rightly recognize patients as having an undiagnosed MBD. Altogether, our results suggest that screening with a guideline-based bleeding questionnaire is not adequate to identify patients with MBDs.

Yet, we cannot merely advise to stop screening for MBDs with a bleeding questionnaire. Although the low diagnostic yield of the questionnaire is in accordance with some previous studies,[24,25] it contrasts others.[26,27] Discrepancies between these studies are most likely explained by methodological differences. In line with our study, the former studies[24,25] conducted hemostatic tests in all patients and related a bleeding questionnaire to these tests or clinical outcomes, while the latter studies performed more hemostatic tests in those patients reporting bleeding symptoms on a questionnaire, and thereby introduced detection bias.[26,27]

An important secondary finding of this study was that the ISTH-BAT, the PFA, ELT, PT, and aPTT also seemed unable to discriminate between patients with and without hemostatic abnormalities, which indicates that they should not be used as preoperative screening tools. The value of the PFA was assessed as it is often used to screen for platelet function disorders (PFDs),[28] even though previous studies demonstrated low sensitivity for mild PFDs.[29,30] The aPTT is quite

often prolonged due to the presence of lupus anticoagulant activity, which can lead to an extensive but unnecessary coagulation work-up.[31] Our findings underpin the advice of the ESA[5] and the SFAR[6] guidelines that in case of a positive bleeding history, hematologist consultation is preferred over measurement of PT or aPTT. However, these tests are still often used by anesthesiologists.[10] We fear that measuring PT and aPTT may lead to a false sense of security in case of normal test results, and believe that hematologist consultation for the performance and interpretation of hemostatic confirmatory assays is indeed the most appropriate diagnostic step when a MBD is suspected.

The low diagnostic value of the ISTH-BAT might be explained by several factors. First, like all bleeding questionnaires, the BAT is susceptible to the subjective interpretation of the severity of bleeding symptoms by both the patient and the professional administering the BAT.[32] Second, healthy subjects are known to report bleeding symptoms.[33,34] Third, some hemostatic abnormalities present in our study population might not be expected to cause spontaneous bleeding symptoms (but may hinder operative hemostasis). As such, these abnormalities are less likely to be detected by the BAT.

The major strength and distinctive quality of this study concerns the extent of hemostatic laboratory testing. This is the first study to perform most (clinically) available coagulation, platelet function and fibrinolysis tests, in many preoperative patients with positive answers to guideline-proposed bleeding questions, to gain insight into the prevalence and severity of hemostatic abnormalities in these patients. In addition, we are the first to study the validity of the ISTH-BAT for detecting hemostatic abnormalities in a preoperative patient population. Furthermore, we directly linked the laboratory screening tests to the confirmatory tests that are used to diagnose bleeding disorders, whereas previous studies in the field related the screening tests to surgical outcomes. Although the latter is clinically relevant, it is also an indirect and difficult assessment, merely because perioperative bleeding is largely dependent on other factors such as type and success of the intervention.

This study had several limitations. As it was not designed to relate hemostatic traits and the bleeding questionnaire to the outcome bleeding, the perioperative significance of (mild) hemostatic abnormalities and the diagnostic value of a positive bleeding questionnaire regarding the identification of patients who bleed during surgery remain unknown. However, to optimize perioperative preventive measures, establishing a diagnosis of a hemostatic deficiency is preferred over recognizing a patient as having a "positive bleeding history," as measures to prevent bleeding differ per disorder.[35-37] Other limitations concern the study design that led to an imbalance between the two patient groups and the relative low number of hemostatic abnormalities that was found. More patients with a positive than with a negative bleeding questionnaire were included (n = 240 vs n = 95) and baseline characteristics differed between the groups. The small number of hemostatic abnormalities limits the interpretation of the diagnostic performance of the screening assays. Moreover, the fact that only part of the patients eligible for this study consented to participate, may have induced selection bias. Additional limitations include the extent of our laboratory package and definitions of hemostatic abnormalities. The laboratory package was extensive but not exhaustive, meaning that some abnormalities might have been missed. These include platelet storage pool diseases,[29,38] and some vWD types.[39] We did not perform repeated measures, which is usually performed in the diagnostic work-up of bleeding disorders, as many variables such as stress and circadian variation influence hemostatic measurements.[7,40,41] Furthermore, the definition of a "hemostatic abnormality" is subject to debate. Some experts might say that hemostatic levels just below our study cut-off values do not cause a bleeding tendency, but minimal functional levels of coagulation factors to prevent perioperative bleeding are not established.[3,42]

In summary, this explorative observational study showed that the prevalence of hemostatic abnormalities in both patients with and without reported bleeding symptom(s) was 9%-10%, which implies that the guideline-based questionnaire cannot differentiate between patients with and without abnormalities. The value of the screening assays aPTT, PT, PFA, ELT, and ISTH-BAT was also limited, suggesting that these should not be used to identify preoperative patients with hemostatic abnormalities. However, most abnormalities were mild and would currently not prompt hemostatic treatment. Regarding future research, the connection between hemostatic traits and operative bleeding outcome is the most relevant question remaining in the field. A large multi-center study including many patients undergoing major surgical procedures that have a comparable risk of (major) bleeding, in which hemostatic tests are performed and standardized clinical outcomes are recorded, would be required to establish minimal surgical hemostatic requirements and the relevance of mild abnormalities (eg, as detected in our study). How to screen for these relevant hemostatic abnormalities in the preoperative setting, would be a subsequent research step.

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AUTHOR CONTRIBUTIONS

MJ Vries wrote the manuscript and was coordinator of the study. PE van der Meijden participated in the description of the results and discussion session of this paper. GJ Kuiper and MD Lancé participated in the design of the study and the ethical approval, they critically checked the manuscript. PJ Nelemans participated and advised in all the statistical calculations and the design of the results section. RJ Wetzels and RG van Oerle performed the laboratory experiments and checked the information in the Methods section of the manuscript. H ten Cate and YM Henskens are the principal investigators of the study and were responsible for the study design and advised and checked the preparation of the manuscript in detail.

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.

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