


Screening platelet function in blood donors

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Abstract

Background: Transfusion of defective platelets could contribute to the inefficiency of platelet transfusion in preventing or stopping bleeding.

Study Design and Methods: This single-center prospective study aimed to determine the prevalence of functional platelet abnormalities in a population of blood donors with a clinical history of bleeding diathesis or with history of hematoma (>4 cm) during blood donation. Donors with positive bleeding screening questionnaire were referred to the reference center for rare platelet diseases at La Timone University Hospital (Marseille) to confirm the bleeding tendency using a more extensive bleeding questionnaire (MCMDMscore) and to assess hemostasis, including a comprehensive platelet analysis.

Results: One hundred and ninety-five donors identified based on a history of hematoma and 2434 blood donors were included in the study. Eighty-eight donors (3.6%) had a bleeding score indicating a potential bleeding disorder. Five donors with a history of hematoma (2.5%) and 15 (17%) donors with a confirmed bleeding score underwent hemostatic analysis, including two men and 18 women with average age of 33.9 years. Minor hemostatic abnormalities were observed in three donors. Two donors exhibited accelerated fibrinolysis with reduced euglobulin lysis time and increased D-dimer levels in serum. Two donors had a platelet granule defect, without identification of genetic abnormality.

Conclusion: The bleeding questionnaire proved to be a valuable tool to screen blood donors for potential platelet defects. Platelet dysfunction was rare in the blood donor population assessed. Additional studies are necessary to understand

Abbreviations: ADP, adenosine diphosphate; Coll, collagen; Epi, epinephrine; Mep, mepacrine; RCPD, Reference Center for Rare Platelet Diseases; Risto, ristocetin; TRAP, thrombin-related activation peptide; vWF, von Willebrand factor.

Marie Christine Alessi and Christophe Picard contributed equally to this study.

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the clinical impact that the transfusion of platelets with qualitative defects has on recipients.

KEYWORDS

blood donor, hemostatic disorder, platelet, prevalence

1 | INTRODUCTION

The prevalence of hemostatic disorders among blood donors is currently unknown. Hemostasis is not assessed in blood donors, which is partly due to the lack of simple automated tests that can provide a comprehensive assessment of hemostatic profile, including platelet function. Indeed, several non-automated tests are required to perform a comprehensive analysis of platelet function.¹

Unlike severe platelet defects that often occur early in life, some less severe disorders that cause only minor bleeding symptoms may persist undetected.² As a result, potential blood donors may be unaware of a moderate platelet disorder, especially if they have not experienced a traumatic injury or undergone surgery. Although the clinical consequences of defective platelets in transfusion recipients remain unknown, platelet function defects may play a role in the hemostasis ineffectiveness of some platelet concentrates. Transfusion of defective platelets from a single donor may be particularly risky, as the defective platelets are concentrated and will not be masked in a pool of platelets derived from several donors.

Very few studies have evaluated platelet function abnormalities in blood donors. One study was performed on whole blood samples using the automated PFA100 platelet function analyzer, which assesses the complex process of primary hemostasis and helps to detect von Willebrand disease and severe platelet dysfunction. Interestingly, approximately 20% of donors displayed prolonged closure time in response to epinephrine and non-steroidal anti-inflammatory drugs (NSAIDs).^{3,4} This finding was confirmed by Curvers et al.⁵ who questioned the donors regarding the recent use of aspirin or NSAIDs. However, these studies did not involve an extensive analysis to detect intrinsic defects in platelet function.

The aim of the current study was to determine the prevalence of qualitative platelet defects in blood donors with a clinical history of bleeding diathesis by performing a comprehensive analysis of donor platelets at the French reference center for constitutional platelet disorders (RCPD, La Timone University Hospital Center, Marseille). Bleeding history information was first collected using a validated screening questionnaire.⁶ Blood donors who developed a hematoma (>4 cm) during a previous blood donation were also included in the study. The

prevalence of non-platelet hemostatic defects was also evaluated. Overall, our findings provide insight to optimize the process to screen potential donors before platelet donation.

2 | MATERIAL AND METHODS

The study was open to any volunteer, male or female, who was eligible to donate blood and visited the blood donation center. Only donors who agreed to participate in the study and signed informed consent were included in the study. The inclusion criteria included those established by the French blood agency. The patient data used in the study were anonymized, and the institutional review board approved the study (IDRCB: 2016-A00117-44).

To identify donors with bleeding disorders, we used a bleeding questionnaire that was previously used to screen for von Willebrand disease and then improved by Bonhomme et al.⁶ The six questions with yes or no answers represent a simple approach to detect bleeding symptoms (Table 1). One question concerned menstrual bleeding and thus only applied to females. The score was calculated from the total positive responses; therefore, the maximum score was 6 for females and 5 for males.

TABLE 1 Questionnaire used to screen for bleeding disorders

1. Have you ever seen a doctor or received treatment for prolonged or unusual bleeding, such as a nosebleed or a small cut?
2. Do you tend to develop bruises larger than 2 cm in diameter or large hematomas, without shock or trauma or after a minor shock or trauma?
3. Have you seen your dentist for bleeding after a tooth extraction?
4. Have you experienced abnormal bleeding episodes after a surgical procedure (e.g., tonsillectomy or circumcision)?
5. For women: Have you seen a doctor or received treatment for heavy menstrual bleeding (e.g., oral contraception, iron treatment, blood clotting medication such as Exacyl)? Have you experienced abnormal bleeding after childbirth?
6. Is anyone in your immediate family undergoing monitoring for a disorder that causes severe bleeding (e.g., von Willebrand disease, hemophilia)?

For donors with a bleeding score ≥ 2 , we scheduled an inclusion visit at the RCPD to confirm the bleeding diagnosis and perform an in-depth hemostatic analysis. Donors with a bleeding score < 2 were excluded from the study.

We also identified blood donors who had experienced a hematoma > 4 cm in 2015; they were then contacted by phone and invited to visit the RCPD in Marseille for a hemostatic assessment.

At the RCPD, the participants were examined by a specialized physician, and the MCMDM questionnaire was used to establish an extensive bleeding symptom score.⁷ The MCMDM questionnaire was developed to detect von Willebrand disease in patients.⁸ In women suffering from menorrhagia, the MCMDM score can also be applied to assess coagulation and platelet defects.⁹ Close to the MCMDM score, the ISTH-BAT score has been validated in carriers of platelet disorders^{10,11} indicating that platelet disorders are detected with these bleeding scores.

If a bleeding disorder was confirmed using the MCMDM questionnaire, the donor was then invited to undergo a standardized hemostatic assessment. If a hemostatic defect was identified, the donor was informed about the need to perform genetic analyses.

The hemostatic analyses included an assessment of hemoglobin levels and platelet counts (Sysmex, Europe). Coagulation was assessed using a routine analyzer (automated STAR system, Stago) and included global coagulation tests (prothrombin time and activated partial thromboplastin time) fibrinogen levels, factor levels, and cofactor levels (factors II, V, VII, VIII, IX, X, XI, and XIII). von Willebrand factor (vWF) activity (ristocetin cofactor activity) and antigens were analyzed using a coagulation analyzer (ACL AcuStar, Instrumentation Laboratory, and STAR system, Stago).

Fibrinolytic assessment included analyses of D-dimer serum levels, alpha2 antiplasmin activity (STAR system,

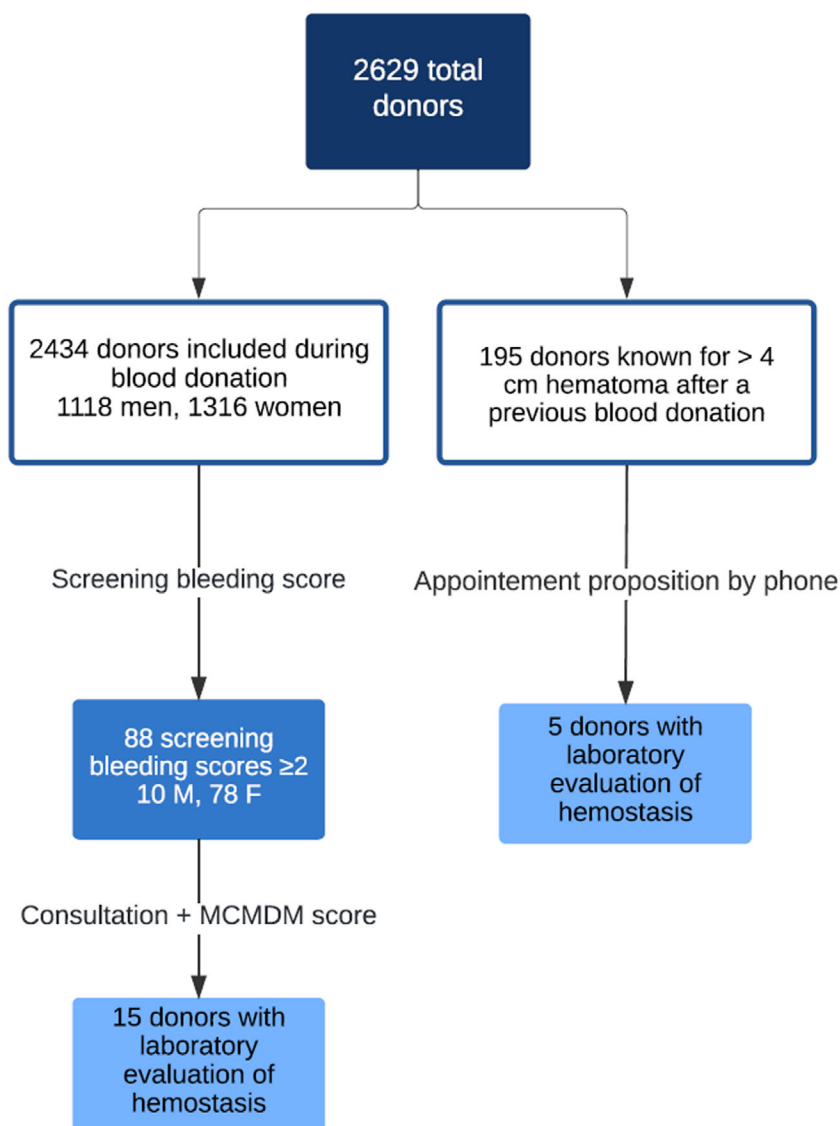


FIGURE 1 Study flowchart [Color figure can be viewed at wileyonlinelibrary.com]

TABLE 2 Clinical results for the 20 donors identified using the bleeding questionnaire and hematoma screening

Donor	Gender	Age (years)	Bleeding score	MCMDM score	Bleeding history	Bleeding family history
Donor 1	F	27	3	6	Easy bruising Epistaxis Menorrhagia	None
Donor 2	F	48	4	8	Intraoperative bleeding (abdominoplasty) Post-partum hemorrhage (third pregnancy only)	None
Donor 3	F	30	2	7	Easy bruising, epistaxis	None
Donor 4	F	25	2	9	Menorrhagia, epistaxis, gingivorrhagia	None
Donor 5	F	19	3	6	Menorrhagia Epistaxis	None
Donor 6	M	23	Donor with hematoma	1	None	None
Donor 7	F	41	Donor with hematoma	0	None	None
Donor 8	M	49	2	6	Easy bruising Bleeding post-tonsillectomy with transfusion. One episode of digestive bleeding	None
Donor 9	F	22	3	5	Wisdom tooth extraction bleeding Cut bleeding	None
Donor 10	F	27	3	9	Hematoma, menorrhagia	Undocumented familial thrombocytopenia
Donor 11	F	36	2	5	Menorrhagia	Hemophilic paternal uncle (father healthy)
Donor 12	F	49	2	4	Menorrhagia	None
Donor 13	F	19	2	4	Buccal bleeding	None
Donor 14	F	40	2	4	Wisdom tooth extraction bleeding	None
Donor 15	F	60	2	-2	Easy bruising	None
Donor 16	F	30	Donor with hematoma	7	Hematoma (road traffic accident)	None
Donor 17	F	28	3	5	Menorrhagia, epistaxis, gingivorrhagia	Father with perioperative bleeding
Donor 18	F	20	Donor with hematoma	1	None	None
Donor 19	F	25	Donor with hematoma	1	None	None
Donor 20	F	60	5	16	Easy bruising Menorrhagia Bleeding during dental care Post-surgery hematoma	Son with easy bleeding

Stago), and plasma PAI-1 antigen levels. Euglobulin lysis time tests were performed to measure overall fibrinolysis as previously described.¹²

Platelet assessments were performed in a department dedicated to platelet analysis at the RCPD. We measured

platelet aggregation in response to ADP (2.5 and 10 μ M), arachidonic acid (1 mM), epinephrine (7.5 μ M), collagen (2.5 μ g/ml), and ristocetin (0.5 and 1.25 mg/ml). Flow cytometry was performed to rule out platelet adhesion and aggregation receptor deficiency. Residual factor II

TABLE 3 Hemostasis results for blood donors with bleeding tendency

Gender		Coagulation												Von Willebrand factor					Fibrinolysis																		
		PC		PT		APTT		II		V		X		VII		VIII		IX		XIII		Fib.		Act.		Ag		Act/Ag		AZAP		ELT		PAI1p		DD	
		Hb	Hg	g/L	>150	>110	>1.2	>70	>70	>70	>70	>70	>70	>70	>70	>70	>70	>70	>70	>70	>70	>70	g/L	%	%	%	%	%	%	%	%	%	min	ng/ml	µg/ml		
Normal range		F: 115-146 M: 130-160		>150	>110	<1.2	>70	>70	>70	>70	>70	>70	>70	>70	>70	>70	>70	>70	>70	>70	>70	1.8-4	50-150	50-160	>0.7	70-130	90-240	0-25	<2								
1	F	111	237	114	0.9	98	74	132	204	140	118	3.13	150	167	150	1.11	96	90	2	0.93																	
2	F	116	271	90	1	71	106	99	195	114	100	3.14	139	144	139	1.04	104	240	4	0.79																	
3	F	138	277	100	0.9	92	134	88	155	150	146	2.79	114	92	114	0.81	106	270	11	2.29																	
4	F	125	209	97	1	84	113	100	133	118	86	2.15	101	91	101	0.9	107	240	7	0.38																	
5	F	122	331	94	1	99	105	123	141	127	86	3.31	13	97	13	0.79	120	180	5	1.11																	
6	M	149	206	81	1	73	68	115	113	101	89	2.68	137	135	137	0.99	81	240	9	0.94																	
7	F	132	288	113	1	91	114	146	103	95	84	3.04	102	102	95	1.07	101	180	3	0.82																	
8	M	149	308	94	1	85	90	101	137	87	116	2.03	106	98	106	0.93	113	120	2	1.12																	
9	F	131	306	75	1	107	99	106	106	128	96	2.87	94	94	114	0.83	79	60	3	>4.00																	
10	F	133	147	107	1	95	94	111	110	113	79	2.84	73	65	73	0.89	95	70	1	2.94																	
11	F	128	224	117	1	107	119	116	114	138	115	3.58	102	96	102	0.94	101	270	12	0.48																	
12	F	142	269	120	1	106	121	132	146	115	108	3.24	113	98	113	0.87	117	180	3	1.51																	
13	F	103	241	106	1	109	78	130	156	133	114	3.18	102	97	102	0.95	109	240	20	0.86																	
14	F	123	228	106	0.9	89	100	128	169	103	112	2.95	144	140	144	0.97	125	260	7	0.76																	
15	F	126	343	93	1.1	83	89	94	91	92	125	4.89	86	64	86	0.74	105	240	6	0.4																	
16	F	127	320	113	1	96	90	123	140	133	97	3.6	158	143	158	0.91	114	330	16	0.35																	
17	F	130	303	127	0.9	91	138	117	145	150	104	2.82	101	96	101	0.95	101	300	5	0.38																	
18	F	139	239	98	1.1	92	87	94	101	113	98	2.78	85	91	85	1.07	100	300	9	0.48																	
19	F	123	213	107	0.9	104	102	138	292	183	134	4.45	241	241	267	0.9	110	120	7	>4.00																	
20	F	121	156	94	1.1	ND	ND	ND	157	ND	132	3.57	132	110	132	0.8	91	90	1	ND																	

Gender		Maximal intensity platelet aggregation																															
		ADP		ADP		ADP		Coll.		TRAP		TRAP		AA		Epi		Risto0.5		Risto1.2		Mep 0 µM		Mep 1 µM uptake		Mep 2 µM uptake		Mep 1 µM decrease		Mep 2 µM decrease		PAI Ag in platelets	
		2.5 µM	10 µM	ADP	10 µM	ADP	10 µM	Coll.	3 µg/ml	TRAP	50 µM	TRAP	10 µM	AA	1 mM	Epi	7.5 µM	Risto0.5	mg/ml	Risto1.2	mg/ml	Mep 0 µM	AU	Mep 1 µM uptake	AU	Mep 2 µM uptake	AU	Mep 1 µM decrease	>40%	Mep 2 µM decrease	>40%	PAI Ag in platelets	µg × 10 ⁹ platelets
Normal range		83-93	84-95	84-95	84-95	79-83	79-83	86-97	86-97	76-91	73-89	0	78-94	0.11-0.18	0.50-1.14	0.61-1.28	0.33-1.07																
1	F	86	88	88	89	89	86	88	88	87	87	0	90	0.12	0.74	0.88	44	1.17															
2	F	91	96	96	95	95	99	93	93	94	96	0	98	0.12	0.51	0.65	54	0.65															
3	F	81	83	83	87	87	85	85	85	87	81	0	89	0.12	0.74	0.85	47	0.61															
4	F	92	88	88	90	90	85	90	90	91	83	0	93	0.12	0.79	0.92	45	1.04															
5	F	87	89	89	89	89	88	90	90	90	87	0	91	0.14	0.54	0.65	62	0.92															
6	M	98	93	93	97	97	95	96	96	97	97	0	91	0.19	0.69	0.82	49	0.86															
7	F	89	92	92	92	92	87	88	88	90	89	0	91	0.12	0.57	0.68	53	1.1															
8	M	83	79	79	91	91	90	ND	ND	87	90	0	94	0.12	0.6	0.69	18	0.83															
9	F	80	81	81	84	84	83	81	81	84	73	0	83	0.12	0.58	0.68	67	1.15															

(Continues)

TABLE 3 (Continued)

B. Results for the platelet function assessment

Gender	Maximal intensity platelet aggregation														
	ADP 2.5 μM %	ADP 10 μM %	Coll. 3 μg/ml %	TRAP 10 μM %	TRAP 50 μM %	AA 1 mM %	Epi 7.5 μM %	Risto0.5 mg/ml %	Risto1.2 mg/ml %	Mep 0 μM AU	Mep 1 μM uptake AU	Mep 2 μM uptake AU	Mep 1 μM % decrease >40%	Mep 2 μM % decrease >40%	PAI Ag in platelets μg × 10 ⁹ platelets
10	F	87	89	90	91	90	90	85	0	93	0.12	0.92	48	48	1.18
11	F	73	84	89	88	91	94	84	0	93	0.12	0.62	63	65	0.78
12	F	87	88	90	91	93	90	97	0	96	0.14	0.66	52	55	0.82
13	F	89	87	90	88	90	90	86	0	93	0.12	0.75	52	51	1.1
14	F	82	85	88	84	84	88	86	0	94	0.13	0.58	40	40	0.74
15	F	92	94	92	93	95	94	89	0	97	0.15	0.63	58	57	0.78
16	F	87	93	93	93	95	93	92	0	94	0.13	0.7	52	49	0.81
17	F	85	90	85	89	91	91	86	0	92	0.12	0.66	60	56	0.78
18	F	92	90	95	92	94	99	93	0	99	0.13	0.62	49	47	0.74
19	F	85	86	90	89	91	91	86	0	92	0.13	0.68	57	53	1.01
20	F	81	ND	67	34	82	65	77	18	88	0.14	0.35	8	6	0.23

Note: The shaded numbers indicate abnormal values. Abbreviations: AZAP, alpha2antiplasmin; Act, activity; ADP, adenosine diphosphate; Ag, antigen; APTT, activated partial thromboplastin time; Coll, collagen; DDs, D-dimers in serum; Epi, epinephrine; ELT, Eupoglobulin lysis test; Fib, fibrinogen; Mep, mepacrine; min, minutes; ND, not determined; PAI-1p, plasminogen activator inhibitor 1 in plasma; PT, prothrombin time; Risto, ristocetin; TRAP, thrombin-related activation peptide.

levels were evaluated to assess platelet procoagulant capacity. PAI-1 antigen levels in serum were determined to rule out platelet alpha granule deficiency. Several methods were used to identify platelet dense granules. We assessed dense granules for mepacrine incorporation and release upon stimulation using flow cytometry. In case of dense granule quantitative abnormalities, whole-mount electron microscopy and intraplatelet serotonin analysis were performed using high-performance liquid chromatography. In case of platelet abnormalities, we sequenced a panel of 80 genes associated with congenital platelet disorders.

3 | RESULTS

3.1 | Cohort results

A total of 2629 donors were recruited from July 2017 to April 2019, including 1179 men (44.8%) and 1450 women (55.2%). The mean age was 33.6 years, and the median age was 34.0 years. The study flowchart is presented in Figure 1.

A total of 2434 donors were included at the time of blood donation (46% male and 54% female). Eighty-eight (3.6%) of the donors had a bleeding score ≥2; the majority of these donors were female (89%). Fifteen donors (17%) underwent hemostatic assessment (one male and 14 females). Donors with a history of hematoma ≥4 cm during a previous blood donation (n = 195) were contacted by phone. Five of these donors were screened for hemostatic defects (one male and four females). A total of 20 blood donors were screened for hemostatic abnormalities, including two men and 18 women, with a mean age of 35 years and a median age of 32 years.

3.2 | Hemostatic abnormalities in the hematoma group

Four donors with hematoma had a low MCMDM score (score: 0–1), indicating no risk of bleeding. None of the donors in the hematoma group had a family history of bleeding or significant hemostatic abnormalities (Tables 2 and 3). One donor with hematoma (Donor 16) exhibited a high MCMDM score (score: 7) and suffered major hematomas following a motor vehicle accident. Donor 16 did not exhibit hemostatic defects.

3.3 | Hemostatic abnormalities in the blood donor group

The MCMDM score confirmed the bleeding questionnaire results for all donors but one (Donor 15, Table 2).

Donor 20 with the highest bleeding score (score: 5) also had the highest MCMDM score (score: 16).

The most common bleeding symptoms were menorrhagia (eight cases), epistaxis (five cases), gingivorragia (two cases), easy bruising (two cases), and bleeding associated with dental treatment (three cases). Only two donors reported a family history of bleeding, and one donor reported bleeding symptoms in offspring. None of the repeat donors (>3 donations) reported hematoma after whole blood donation.

Despite the prevalence of moderate forms of von Willebrand disease in the general population around 1%,^{13,14} none of the donors showed decreased vWF levels. Unexpectedly, six donors presented with elevated levels of factor VIII (155%–292%), vWF (1.67 and 2.41 U/ml), and/or fibrinogen (4.45 and 4.89 g/L). Two donors displayed reduced levels of factor VII (48%, Donor 9) and factor V (68%, Donor 6) (Table 3). We observed two cases with accelerated fibrinolysis and reduced euglobulin lysis time associated with increased D-dimer levels in serum. Elevated D-dimer levels were also observed in a third donor without abnormal lysis time and elevated levels of vWF, factor VIII, and fibrinogen, thus suggesting an inflammatory condition.

Platelet analysis revealed minor thrombocytopenia in one individual (Donor 10) (Table 3) without other platelet abnormalities. None of the donors had relevant defects in platelet aggregation (Table 3) or platelet procoagulant activity (residual prothrombin levels 2%–9%; normal value <20%). Interestingly, two donors, Donors 8 and 21, had marked granule defects. For Donor 21, dense granule deficiency in this donor was associated with an alpha granule defect as indicated by low platelet PAI-1 content (Table 3). Electronic microscopy confirmed that Donor 21 had a severe deficiency in both dense granules and alpha granules. Immature alpha granules were observed, thus indicating a defect in alpha granule biogenesis and/or granule trafficking. Assuming a genetic disorder, we conducted gene sequencing on samples derived from Donor 21 using a panel of 80 genes associated with congenital platelet disorders; however, this assessment failed to shed light on the genetic etiology of the condition.

4 | DISCUSSION

This study aimed to determine the prevalence of qualitative platelet abnormalities in blood donors using a simple questionnaire to screen donors for bleeding symptoms. The frequency of qualitative platelet defects in blood donors was rare, as approximately one of 2000 donors (0.05%) displayed a characterized platelet granule defect.

Overall, the bleeding questionnaire was a useful tool to screen blood donors for potential bleeding disorders.

The bleeding questionnaire used in this study consists of six questions with binary yes or no answers. The questionnaire is simple to use and takes approximately 5 min to complete. This questionnaire was initially developed and validated to screen individuals for von Willebrand disease and has shown good specificity (98%) and sensitivity (89%) to detect congenital hemostatic disorders. The questionnaire has also recently been tested in a case-control study by Bonhomme et al.⁶ to assess bleeding risk in donors referred for hemostatic analysis. In our study, this questionnaire also proved to be effective, as all but one donor had a high MCMDM score. To assess bleeding tendency, the questionnaire is based on a –1 to +4 scoring system for each bleeding symptom. Normal scores range from –3 to +3.¹⁵ Negative values are used to adjust for the lack of bleeding in response to major hemostatic events such as surgery, childbirth, or dental extraction. In our study, the 15 blood donors displayed a mean MCMDM score of 6.1 ± 3.9 . Dietary factors may cause platelet dysfunction when donors are encouraged to eat before donating blood.¹⁶ Thus, using the questionnaire, our study excluded any transient hemostatic disorders associated with diet before donation.

Our study is the first to extensively assess the prevalence of platelet defects in a population of blood donors. Previous studies of blood donors have used closure time measurement (PFA-100) to detect primary hemostatic disorders, including platelet function defects. Jilma-Stohlawetz et al.³ and Harrison et al.⁴ have observed prolonged collagen-epinephrine closure times in up to 20% and 16% of platelet donors, respectively. In both studies, this observation was consistent with the use of NSAIDs. As we did not observe a reduced response to arachidonic acid, which is used to assess aspirin/NSAID therapy, donors undergoing antiplatelet therapy were likely not included in our study.¹⁷

A retrospective analysis by Benlakhel et al.¹⁸ has suggested using a factor VII procoagulant activity level threshold of 10% to administer recombinant factor VIIa for patients undergoing major surgery. Bauer et al.¹⁹ have demonstrated that restoring factor VII levels to 15%–20% that of normal levels is usually sufficient to control bleeding. Similarly, factor V levels <20% are associated with bleeding. When factor V levels are above 20%, hemorrhagic signs are rarer and most often moderate.^{20–22} Thus, the slight decrease in factor V and factor VII levels observed in two donors in our study is unlikely to be involved in the bleeding disorder. Nevertheless, Saes et al.²³ have concluded that previously determined factor activity thresholds are inaccurate to ensure that bleeding is avoided in patients with rare bleeding disorders. In their study, 48% of patients had more severe bleeding than expected based on their baseline activity levels,

which was particularly true for factor V and factor VII deficiencies.

Von Willebrand disease is a common deficiency, which is often diagnosed late in mild forms.^{13,14} The most common form of vWD is type 1 marked by decreased antigen and activity. None of the donors had a vWF activity/antigen ratio below 0.7. This ratio would represent an uncommon type 2 variant. Thus, this highlights the fact that blood donors are not representative of the general population. People with VWD or known platelet dysfunction, even with mild bleeding symptoms, are very unlikely to become blood donors. Unexpectedly, we observed increased levels of factor VIII and vWF in some blood donors, which suggests a possible inflammatory condition. Many extrinsic patient-related parameters may influence factor VIII and vWF levels, such as age, body weight, ABO antigen status, diet, smoking, ethnicity, and physical activity.²⁴ However, increases in these hemostatic factors have also been described in many chronic conditions associated with inflammation, thus indicating that some blood products may contain inflammatory and/or prothrombotic mediators.

Fibrinolysis is barely assessed in rare bleeding disorders. Furthermore, rare fibrinolytic disorders are difficult to diagnose, as diagnostic protocols are largely lacking and the methods used are challenging. In a study by Saes et al.,²³ fibrinolytic disorders were relatively common, accounting for 21% of patients with rare bleeding disorders, of which 27% of patients displayed reductions in lysis time. In our study, two donors exhibited accelerated fibrinolysis with both reduced lysis time and increased D-dimer levels in serum. In the rare Quebec platelet syndrome due to elevated urokinase levels in platelets, D-dimer levels in serum are markedly elevated.²⁵ This observation indicates that elevated D-dimer levels in serum may be an indicator of increased plasmin generation. Further analysis is required to understand the mechanism and clinical relevance of the reduced euglobulin lysis time in platelet-poor plasma associated with elevated D-dimer levels.

In our study, donors with platelet disorders were quite uncommon. In Donor 10, it is unlikely that the isolated case of moderate thrombocytopenia without functional defects was the underlying cause of the bleeding tendency. Indeed, when bleeding is associated with moderate thrombocytopenia, a platelet dysfunction is often observed, as we previously described in some cases of constitutional thrombocytopenia.²⁶

The prevalence of dense granule deficiency in patients with rare hemostatic disorders is unknown. Although it is rare, some groups have suggested that the prevalence is probably underestimated because the diagnostic process is complex and requires expertise.^{27,28} Interestingly, two donors displayed a significant defect in dense granules. While Donor 8 showed reduced mepacrine release upon

platelet stimulation, Donor 20 had a severe quantitative abnormality in dense granules and alpha granules. Donor 8 may harbor a signaling defect that impairs dense granule release rather than a true dense granule defect. The deficiency in dense and alpha granules detected in Donor 21 was observed in several tests. This donor had the highest bleeding scores, thus confirming the clinical impact of this deficiency. Few genes have been identified to play a role in concomitant decreases in dense granules and alpha granules. As our genetic sequencing assessment failed to identify candidate genes, further investigation is required to understand the etiology of this condition.

The clinical consequences of transfusing recipients with defective platelets are poorly understood.²⁹ In our study, none of the individuals donated platelets via apheresis. As the platelets were distributed in platelet pools, it was not possible to evaluate the impact on recovery and efficacy in the recipients.

A few study limitations should be noted. Only 15 donors out of 88 took part in the hemostatic analysis phase of the study. This may be due to the moderately high bleeding score threshold applied (≥ 2). It is possible that donors with low bleeding risk were not motivated to continue the study. A more restrictive threshold of 3 may be better suited to identify donors at risk.

Overall, the blood donor population assessed in our study displayed very few qualitative platelet defects. Only one donor displayed a confirmed severe deficiency in dense granules and alpha granules of unknown etiology. The bleeding questionnaire proved to be a valuable tool to screen blood donors for potential bleeding disorders. This questionnaire could be useful to avoid transfusing defective platelets, which is especially critical when transfusing single-donor units to patients who may be prone to bleeding.

ACKNOWLEDGMENTS

This work was supported by the French Blood Agency (grant no. 2016-27-CHIARONI-AM). The authors would like to thank P. Suchon (MD) for their contribution to data extraction. The authors would like to thank all blood donors.

CONFLICT OF INTEREST

The authors have disclosed no conflicts of interest.

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How to cite this article: Pedini P, Baudey J-B, Pouymayou K, Falaise C, Ibrahim-Kosta M, Véliér M, et al. Screening platelet function in blood donors. *Transfusion.* 2022;62(8):1643–51. <https://doi.org/10.1111/trf.16990>