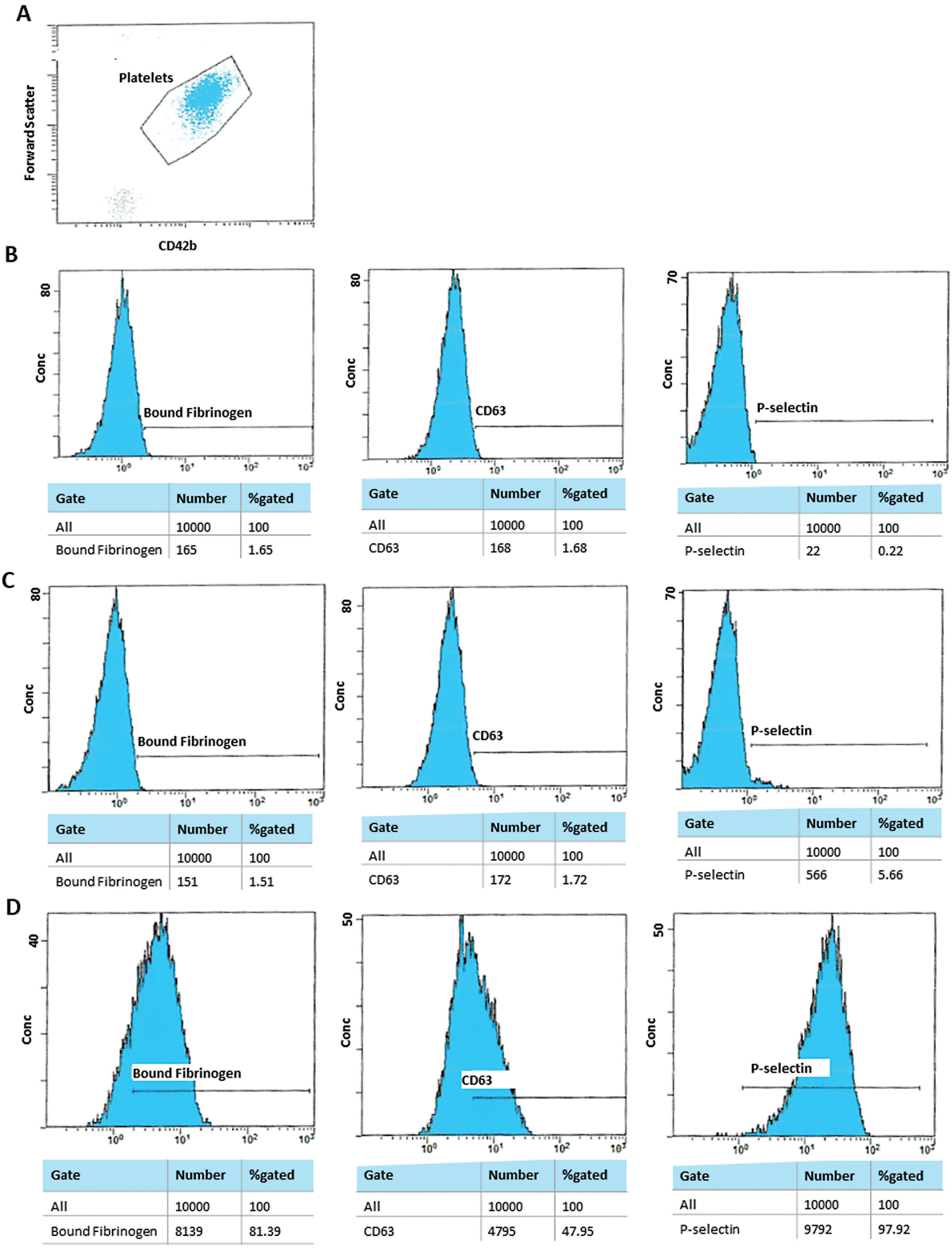


Gate	Number	%gated
All	10000	100
Bound Fibrinogen	165	1.65

Gate	Number	%gated
All	10000	100
CD63	168	1.68

Gate	Number	%gated
All	10000	100
P-selectin	22	0.22

Gate	Number	%gated
All	10000	100
Bound Fibrinogen	151	1.51

Gate	Number	%gated
All	10000	100
CD63	172	1.72

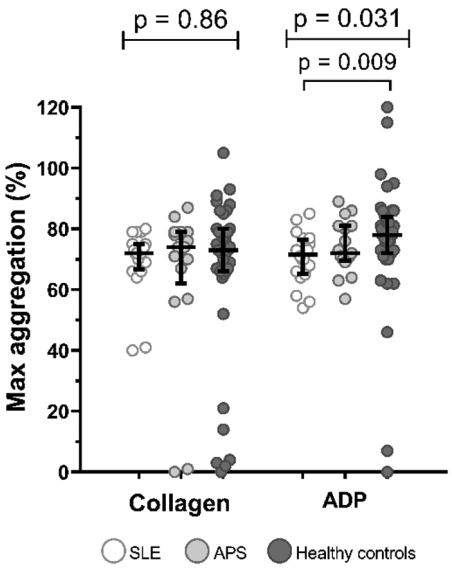
Gate	Number	%gated
All	10000	100
P-selectin	566	5.66

Gate	Number	%gated
All	10000	100
Bound Fibrinogen	8139	81.39

Gate	Number	%gated
All	10000	100
CD63	4795	47.95

Gate	Number	%gated
All	10000	100
P-selectin	9792	97.92

Supplementary Fig. S1 (**A**) Gating strategy for identification of platelets based on the surface expression of CD42b and forward scatter intensity. Platelet – platelet aggregates were excluded based on forward scatter intensity and forward scatter peak. (**B**) The gating strategy for all three receptors in the negative control sample (EDTA). Bound fibrinogen and CD63 gates were adjusted to include 1.5 to 2%-positive platelets. For P-selectin, gates were adjusted to include below 0.1%-positive platelets. (**C**) The gating strategy applied to the HEPES sample. Gates for bound fibrinogen and CD63 were adjusted if %-positive values for bound fibrinogen and CD63 were below 1.5%. All values above 1.5% were accepted. The %-positive platelets for P-selectin in the HEPES buffer sample equals the preactivation. (**D**) Bound fibrinogen, CD63, and P-selectin expression in a sample stimulated with ADP, applying the gates set in the negative controls. Additional settings, including compensation, analyses, and signal characteristics, followed the MIFlowCyt guideline.[1] ADP, adenosine diphosphate.



Supplementary Fig. S2 Platelet aggregation, measured using LTA, in response to agonist stimulation with collagen or ADP in patients with SLE ($n = 20$), patients with APS ($n = 17$), and healthy controls ($n = 39$). Bars indicate median with interquartile range. Capped lines indicate the *p*-value across all three groups, calculated with ANOVA or Kruskal–Wallis. When significant, the tick-down lines indicate the *p*-values between two groups, calculated with unpaired *t*-tests or Mann–Whitney *U*-tests. Three data points are above 100% max aggregation, as illustrated in ►**Supplementary Fig. S2**. This is theoretically not possible, but happens due to the calibration of the aggregometer. These data points were censored at 100% in the statistical analyses. ADP, adenosine diphosphate; APS, antiphospholipid syndrome; LTA, light transmission aggregometry; SLE, systemic lupus erythematosus.

Supplementary Table S1 Disease-specific characteristics for SLE patients

ACR criteria (cumulative)	SLE
Malar rash (ACR1)	11 (55)
Discoid lupus (ACR2)	4 (20)
Photosensitivity (ACR3)	10 (50)
Oral/nasal ulcers (ACR4)	9 (45)
Arthritis (ACR5)	16 (80)
Serositis (ACR6)	9 (45)
Nephritis (ACR7)	2 (10)
CNS (ACR8)	0 (0)
Hematological (ACR9)	19 (95)
Immunological (ACR10)	17 (85)
ANA (ACR11)	20 (100)
Number of ACR criteria	6 [5; 7]
Biochemical and clinical data on the day of inclusion	
SLEDAI at inclusion	4.0 [3.0; 7.5]
SLICC	0 [0; 1]
dsDNA antibodies $>10^3$ int. units/L	6 (30)
Urine albumin/creatinine ratio >200 mg/g	1 (5)
Medical treatment on the day of inclusion	
Hydroxychloroquine	17 (85)
Prednisolone	8 (40)
Other immunosuppressives[a]	11 (55)
Pain relief (NSAID/paracetamol)	3 (15)

Abbreviations: ACR, American College of Rheumatology; ANA, antinuclear antibodies; aPL, antiphospholipid antibodies; CNS, central nervous system; IQR, interquartile range; RI, reference interval; NSAID, non-steroidal anti-inflammatory drug; SLE, systemic lupus erythematosus; SLEDAI, systemic lupus erythematosus disease activity index, SLICC; systemic lupus international collaborating clinics.
Note: Values are written as median [IQR] or *n* (%).
[a]Includes rituximab, belimumab, methotrexate, mycophenolate, azathioprine, and colchicine.

Supplementary Table S2 Lectin pathway protein concentrations, flow cytometry measurements, and LTA measurements in patients with SLE, APS, and healthy controls

Serum concentration of lectin pathway proteins ((ug/mL) unless otherwise stated)					
Protein		*SLE, n = 20*	*APS, n = 17*	*Healthy controls, n = 39*	*p*
MBL		1.623 (0.461–2.175)	2.466 (0.712–3.814)	2.030 (0.693–2.975)	0.32
CL-L1		0.636 (0.554–0.678)	0.578 (0.546–0.596)	0.588 (0.532–0.631)	0.18
CL-K1		0.409 (0.359–0.448)	0.379 (0.338–0.418)	0.386 (0.356–0.416)	0.28
M-ficolin		3.656 (2.165–4.406)	4.560 (3.980–5.810)	4.151 (3.477–5.249)	0.01
L-ficolin		2.932 (2.139–3.650)	3.885 (3.526–4.198)	3.196 (2.433–3.888)	0.03
H-ficolin		35.54 (30.26–41.77)	36.55 (31.61–40.51)	28.45 (25.06–34.13)	0.01
MASP-1		14.03 (11.64–15.77)	13.09 (11.81–15.42)	13.70 (12.26–16.06)	0.95
MASP-3		8.665 (7.115–9.579)	7.955 (6.983–9.895)	6.977 (6.274–8.819)	0.28
MAp44		2.466 (2.104–3.207)	2.664 (2.317–3.000)	2.590 (2.192–3.026)	0.67
MASP-2		0.522 (0.355–0.735)	0.530 (0.454–0.766)	0.453 (0.331–0.562)	0.10
MAp19		0.652 (0.555–0.708)	0.789 (0.633–0.845)	0.572 (0.518–0.703)	< 0.001
C3dg (mUnits/mL)		36.3 (28.0–67.8)	35.6 (30.1–45.5)	33.0 (26.8–39.4)	0.21
Flow cytometry (%-positive platelets)					
Surface marker	*Agonist*	*SLE, n = 20*	*APS, n = 17*	*Healthy controls, n = 39*	*p*
Bound fibrinogen	Collagen	78.5 (65.5–90.6)	74.9 (68.0–81.2)	73.5 (60.4–87.3)	0.53
	ADP	81.2 (75.2–85.1)	79.8 (75.7–87.6)	83.2 (75.2–85.9)	0.86
	TRAP	86.5 (77.3–90.1)	89.0 (83.5–93.1)	88.3 (84.5–91.0)	0.21
	AA	98.2 (95.5–98.8)	96.8 (92.4–97.9)	92.2 (82.2–96.7)	0.01
CD63	Collagen	64.5 (49.0–74.9)	51.5 (41.9–64.6)	57.5 (30.0–70.9)	0.26
	ADP	42.3 (34.3–50.1)	37.7 (30.8–48.2)	38.6 (29.3–46.6)	0.45
	TRAP	87.4 (78.8–89.8)	86.8 (81.0–89.1)	86.8 (83.1–89.7)	0.94
	AA	65.5 (58.6–73.4)	55.0 (46.6–68.6)	50.7 (37.0–61.9)	0.02
P-selectin	Collagen	92.7 (88.3–97.2)	87.8 (82.8–95.0)	90.6 (78.3–95.6)	0.19
	ADP	97.0 (94.6–97.7)	95.7 (94.0–97.3)	95.1 (92.9–96.9)	0.28
	TRAP	99.4 (98.7–99.6)	99.3 (99.0–99.4)	99.1 (98.8–99.4)	0.39
	AA	98.6 (98.2–98.8)	97.7 (96.4–98.5)	96.8 (94.6–98.0)	0.01
Flow cytometry (MFI)					
Surface marker	*Agonist*	*SLE, n = 20*	*APS, n = 17*	*Healthy controls, n = 39*	*p*
Bound fibrinogen	Collagen	5.8 (4.4–6.8)	5.5 (4.4–6.9)	7.2 (5.2–8.1)	0.05
	ADP	5.0 (4.1–5.7)	4.9 (3.8–6.5)	6.3 (5.0–7.8)	0.01
	TRAP	4.6 (3.9–5.1)	5.1 (4.4–6.0)	5.6 (4.8–7.2)	< 0.001
	AA	12.4 (10.0–15.9)	12.9 (8.6–14.4)	11.0 (9.1–14.5)	0.82
CD63	Collagen	11.7 (10.3–12.8)	12.6 (10.3–14.1)	13.2 (11.7–15.8)	0.28
	ADP	8.5 (7.9–9.7)	9.6 (8.8–10.2)	9.7 (8.7–11.5)	0.04
	TRAP	14.4 (13.4–18.0)	16.5 (15.8–17.1)	17.1 (15.8–18.9)	0.02
	AA	10.3 (9.4–11.6)	11.3 (10.7–12.3)	12.4 (10.7–14.0)	0.01
P-selectin	Collagen	19.6 (14.2–25.9)	17.3 (13.5–21.4)	20.1 (11.0–26.8)	0.68
	ADP	15.7 (12.1–17.4)	12.8 (12.3–16.5)	14.9 (12.7–17.9)	0.83
	TRAP	37.4 (32.0–40.2)	38.8 (32.7–42.0)	36.1 (33.0–42.0)	0.95
	AA	23.5 (19.8–26.8)	20.5 (16.8–25.4)	19.7 (15.9–24.7)	0.19

Supplementary Table S2 (*Continued*)

LTA (maximum aggregation)				
Agonist	*SLE, n = 20*	*APS, n = 17*	*Healthy controls, n = 39*	*p*
Collagen	72 (68–75)	74 (67–79)	73 (66–80)	0.86
ADP	72 (66–76)	72 (70–81)	78 (72–84)	0.03

Abbreviations: AA, arachidonic acid; ADP, adenosine diphosphate; APS, antiphospholipid syndrome; CL-K1, collectin kidney 1; CL-L1, collectin liver 1; Collagen, collagen-related peptide; IQR, interquartile range; LTA, light transmission aggregometry; MAp19/44, MBL-associated protein of 19/44 kDa; MASP, MBL-associated protease; MBL, mannose-binding lectin; MFI, median fluorescence intensity; SLE, systemic lupus erythematosus; TRAP, thrombin receptor activating peptide-6.
Note: *p*-Values across all groups were calculated using one-way ANOVA or Kruskal–Wallis test for data following and not following Gaussian distribution respectfully. Values are written as median (IQR).

Supplementary Table S3 Correlations between MASP-2 concentrations and flow cytometry analyses of bound fibrinogen to the surface of platelets after stimulation with agonists, measured as both MFI and %positive platelets

		SLE, $n = 20$		APS, $n = 17$		Healthy controls, $n = 39$	
Measurement	**Agonist**	***r***	***p***	***r***	***p***	***r***	***p***
MFI	Collagen	− 0.23	0.34	− 0.51	0.046	0.16	0.34
	ADP	0.03	0.91	− 0.83	< 0.001	0.07	0.65
	TRAP	0.22	0.35	− 0.77	< 0.001	0.03	0.87
	AA	− 0.13	0.60	− 0.69	0.002	− 0.06	0.73
%-positive platelets	Collagen	− 0.17	0.49	0.01	0.97	0.17	0.29
	ADP	0.20	0.39	− 0.74	0.001	0.25	0.13
	TRAP	0.38	0.10	− 0.34	0.18	0.21	0.21
	AA	− 0.35	0.13	− 0.31	0.22	0.06	0.73

Abbreviations: AA, arachidonic acid; ADP, adenosine diphosphate; APS, antiphospholipid syndrome; Collagen, collagen-related peptide; MASP, MBL-associated protease; MFI, median fluorescence intensity; SLE, systemic lupus erythematosus; TRAP, thrombin receptor activating peptide-6.
Note: Correlation analyses were performed, calculating Spearman's rank correlation.

Supplementary Table S4 Correlations between C3dg concentrations and flow cytometry analyses of platelet activation after stimulation with agonists

C3dg concentrations vs. MFI of platelet surface markers							
		SLE, n = 20		*APS, n = 17*		*Healthy controls, n = 39*	
Surface marker	*Agonist*	*r*	*p*	*r*	*p*	*r*	*p*
Bound fibrinogen	Collagen	0.02	0.94	− 0.16	0.54	0.02	0.88
	ADP	− 0.10	0.68	− 0.27	0.30	0.05	0.74
	TRAP	− 0.23	0.32	− 0.24	0.36	0.07	0.66
	AA	0.01	0.97	− 0.40	0.12	− 0.16	0.36
CD63	Collagen	0.07	0.78	0.01	0.96	0.05	0.77
	ADP	− 0.10	0.69	− 0.05	0.86	0.01	0.97
	TRAP	− 0.17	0.48	− 0.09	0.72	0.28	0.08
	AA	0.10	0.67	− 0.01	0.96	− 0.09	0.58
P-selectin	Collagen	− 0.08	0.74	− 0.38	0.152	0.11	0.52
	ADP	− 0.28	0.23	− 0.47	0.056	0.11	0.50
	TRAP	− 0.22	0.36	− 0.82	<0.001	0.23	0.16
	AA	0.36	0.12	− 0.49	0.045	− 0.06	0.73
C3dg concentrations vs. %-platelets positive for platelet surface markers							
		SLE, n = 20		*APS, n = 17*		*Healthy controls, n = 39*	
Surface marker	*Agonist*	*r*	*p*	*r*	*p*	*r*	*p*
Bound fibrinogen	Collagen	− 0.10	0.68	− 0.28	0.29	0.05	0.74
	ADP	− 0.08	0.74	− 0.25	0.32	0.06	0.74
	TRAP	− 0.18	0.45	− 0.21	0.43	0.11	0.49
	AA	− 0.07	0.77	− 0.48	0.05	− 0.17	0.32
CD63	Collagen	− 0.19	0.43	− 0.30	0.26	0.01	0.96
	ADP	− 0.41	0.07	− 0.22	0.39	− 0.21	0.20
	TRAP	− 0.30	0.20	− 0.21	0.41	0.05	0.77
	AA	− 0.11	0.65	− 0.35	0.17	− 0.24	0.16
P-selectin	Collagen	− 0.11	0.65	− 0.17	0.53	0.07	0.67
	ADP	− 0.32	0.17	− 0.44	0.07	0.02	0.92
	TRAP	− 0.26	0.27	− 0.20	0.44	0.12	0.48
	AA	− 0.02	0.94	− 0.62	0.008	− 0.23	0.18

Abbreviations: AA, arachidonic acid; ADP, adenosine diphosphate; APS, antiphospholipid syndrome; Collagen, collagen-related peptide; LTA, light transmission aggregometry; MFI, median fluorescence intensity; SLE, systemic lupus erythematosus; TRAP, thrombin receptor activating peptide-6.
Note: Platelet activation was measured as both median fluorescence intensity and %positive platelets for all three platelet surface markers. Correlation analyses were performed using Spearman's rank correlation.

Reference

1 Lee JA, Spidlen J, Boyce K, et al. MIFlowCyt: the minimum information about a Flow Cytometry Experiment. Cytometry A 2008;73(10):926–930

