

Supplementary Material for
IL-10 plus the EASIX score predict bleeding events after anti-CD19
CAR T-cell therapy

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The PDF file includes:

Supplementary Table 1 to 10;

Supplementary Figure 1 to 7

Supplemental Table 1. Grading of the coagulation parameters

Parameter	Grade 1	Grade 2	Grade 3	Grade 4
PLT	< LLN, ≥ 75	<75, ≥ 50	<50, ≥ 25	<25
FIB	0.75-1 LLN	0.5- 0.75 LLN	0.25- 0.5 LLN	<0.25 LLN
INR	≤ 1.5 ULN	>1.5-2.5 ULN	>2.5 ULN	NA
APTT	≤ 1.5 ULN	>1.5-2.5 ULN	>2.5 ULN	NA
D-dimer	1-3 ULN	>3 ULN	NA	NA
PT	Prolong ≤ 3 s	Prolong > 3s	NA	NA
ATIII	0.75-1 LLN	0.5- 0.75 LLN	0.25- 0.5 LLN	<0.25 LLN
FDP	1-3 ULN	>3 ULN	NA	NA
TT	Abnormal	NA	NA	NA

PLT indicates platelet; FIB, fibrinogen; INR, international normalized ratio; APTT, activated partial thromboplastin time; PT, prothrombin time; ATIII, antithrombin III; FDP, fibrinogen degradation products; TT, thrombin time; LLN, lower limit of normal; ULN, upper limit of normal; NA, not application.

Supplemental Table 2. Grade of APTT prolongation on post-infusion day

Grade	Pre-LD	D0	D7	D14	M1	M3	M6	M9
Grade 0								
(n, %)	49 (87.5)	41 (78.85)	42 (75)	46 (83.64)	34 (94.44)	12 (85.71)	9 (100)	4 (100)
Grade 1								
(n, %)	4 (7.14)	8 (15.38)	6 (10.71)	5 (9.09)	1 (2.78)	1 (7.14)	0 (0)	0 (0)
Grade 2								
(n, %)	3 (5.36)	3 (5.77)	8 (14.29)	4 (7.27)	1 (2.78)	1 (7.14)	0 (0)	0 (0)
Total (n)	56	52	56	55	36	14	9	4

APTT indicates activated partial thromboplastin time; Pre-LD, pre-lymphodepletion; D, day; M, month.

Supplemental Table 3. Grade of TT prolongation on post-infusion day

Grade	Pre-LD	D0	D7	D14	M1	M3	M6	M9
Normal (n, %)	56 (100)	51 (98.08)	54 (96.43)	47 (85.45)	34 (94.44)	14 (100)	9 (100)	4 (100)
Abnormal (n, %)	0 (0)	1 (1.92)	2 (3.58)	8 (14.55)	2 (5.56)	0 (0)	0 (0)	0 (0)
Total (n)	56	52	56	55	36	14	9	4

TT indicates thrombin time; Pre-LD, pre-lymphodepletion; D, day; M, month.

Supplemental Table 4. Grade of ATIII elevation on post-infusion day

Grade	Pre-LD	D0	D7	D14	M1	M3	M6	M9
Grade 0 (n, %)	30 (73.17)	35 (87.5)	36 (80)	32 (80)	15 (83.33)	7 (100)	4 (80)	1 (100)
Grade 1 (n, %)	9 (21.95)	4 (10)	7 (15.56)	7 (17.5)	2 (11.11)	0 (0)	1 (20)	0 (0)
Grade 2 (n, %)	2 (4.88)	0 (0)	2 (4.44)	1 (2.5)	1 (5.56)	0 (0)	0 (0)	0 (0)
Grade 3 (n, %)	0 (0)	1 (2.5)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Total (n)	41	40	45	40	18	7	5	1

ATIII indicates antithrombin III; Pre-LD, pre-lymphodepletion; D, day; M, month.

Supplemental Table 5. Grade of PT prolongation on post-infusion day

Grade	Pre-LD	D0	D7	D14	M1	M3	M6	M9
Grade 0 (n, %)	53 (94.64)	51 (98.08)	51 (91.07)	52 (94.55)	34 (94.44)	13 (100)	9 (100)	4 (100)
Grade 1 (n, %)	3 (5.36)	1 (1.92)	4 (7.14)	3 (5.45)	2 (5.56)	0 (0)	0 (0)	0 (0)
Grade 2 (n, %)	0 (0)	0 (0)	1 (1.79)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Total (n)	56	52	56	55	36	13	9	4

PT indicates prothrombin time; Pre-LD, pre-lymphodepletion; D, day; M, month.

Supplemental Table 6. Grade of INR elevation on post-infusion day

Grade	Pre-LD	D0	D7	D14	M1	M3	M6	M9
Grade 0 (n, %)	53 (94.64)	51 (98.08)	50 (89.29)	51(92.73)	33(91.67)	14(100)	9(100)	4(100)
Grade 1 (n, %)	3(5.36)	1(1.92)	5(8.93)	4(7.27)	3(8.33)	0(0)	0(0)	0(0)
Grade 2 (n, %)	0(0)	0(0)	1(1.79)	0(0)	0(0)	0(0)	0(0)	0(0)
Total (n)	56	52	56	55	36	14	9	4

INR indicates international normalized ratio; Pre-LD, pre-lymphodepletion; D, day; M, month.

Supplemental Table 7. Coagulation, inflammatory, and tumor burden markers by bleeding events

Laboratory findings	Total	Non-bleeding	Bleeding	P value (non-bleeding vs bleeding)
PLT ($\times 10^9$/L)				
Pre-LD	115.50 (67.75–197.75)	131.00 (82.00–210.00)	82.00 (29.5–142.5)	0.022
Minimum	49.0 (16.2–105.5)	50.50 (17.75–114.75)	22.50 (10.75–83.25)	0.056
D-dimer (mg/L)				
Pre-LD	0.66 (0.37–1.22)	0.64 (0.36–1.06)	0.67 (0.38–2.45)	0.373
Peak	1.59 (0.81–4.62)	0.96 (0.71–4.96)	2.42 (1.16–11.97)	0.040
APTT (s)				
Pre-LD	37.00 (34.10–40.90)	37.05 (34.38–40.60)	37.00 (33.70–42.65)	0.827
Peak	43.70 (37.26–50.50)	46.40 (37.35–51.28)	43.45 (40.60–60.33)	0.993
FIB (g/l)				
Pre-LD	3.22 (2.52–4.24)	3.15 (2.30–4.16)	3.43 (2.77–4.85)	0.348
Minimum	2.32 (1.47–3.30)	2.29 (1.65–3.00)	2.31 (1.11–3.43)	0.599
Peak	4.00 (3.21–4.90)	4.03 (3.12–4.72)	4.13 (3.46–5.52)	0.859
FDP (ug/ml)				
Pre-LD	4.00 (4.00–5.10)	4.00 (4.00–4.55)	4.00 (2.50–7.80)	0.738
Peak	5.50 (4.08–17.25)	4.65 (4.00–15.75)	7.25 (4.75–50.35)	0.103
TT (s)				
Pre-LD	17.00 (15.80–18.00)	17.00 (16.05–18.00)	16.50 (15.70–18.15)	0.964
Peak	18.60 (17.30–21.75)	18.58 (16.73–21.73)	18.45 (17.48–26.65)	0.556
ATIII (%)				
Pre-LD	89.00 (77.50–96.00)	92.00 (79.00–96.00)	81.00(65.5–94.5)	0.120
Peak	104.00 (94.88–110.50)	103.50 (94.75–110.00)	103.50(94.00–112.50)	0.712
PT (s)				
Pre-LD	13.30 (13.00–13.80)	13.20 (12.80–13.73)	13.70 (13.25–14.55)	0.014
Peak	14.07 (13.38–15.55)	13.95 (13.18–15.35)	14.50 (13.58–17.23)	0.076
Monocyte ($\times 10^9$/L)				

Pre-LD	0.30 (0.16–0.48)	0.27 (0.13–0.44)	0.30 (0.21–0.68)	0.123
Minimum	0.02 (0.0–0.06)	0.02 (0.00–0.06)	0.00 (0.00–0.02)	0.017
Peak	0.41 (0.24–0.63)	0.44 (0.24–0.60)	0.29 (0.17–0.50)	0.212
Ferritin (ng/ml)				
Pre-LD	654.55 (234.88–1310.88)	552.35 (174.27–1168.57)	865.55 (445.98–2293.48)	0.204
Peak	1420.80 (431.23–5558.35)	1470.90 (372.50–10528.88)	1721.80 (578.08–5798.55)	0.795
LDH (U/L)				
Pre-LD	242.00 (194.25–494.50)	224.00 (181.00–327.00)	304.00 (209.00–719.00)	0.105
Peak	312.00 (225.50–1050.50)	279.50 (221.25–853.00)	607.50 (238.50–2259.25)	0.073
Tumor burden (%)				
Pre-LD	32.25 (4.00–96.50)	21.50 (5.54–91.00)	66.50 (4.00–96.50)	0.329
CRP (mg/L)				
Pre-LD	10.99 (1.97–24.14)	10.23 (1.54–19.13)	12.16 (4.44–61.21)	0.309
Peak	51.36 (15.12–101.20)	62.32 (16.54–103.99)	63.28 (30.81–118.18)	0.493
IL-2 (pg/ml)				
Pre-LD	2.06 (1.59–2.68)	2.11 (1.67–2.61)	1.81 (1.41–4.12)	0.670
Peak	3.03 (2.52–4.49)	2.76 (2.31–4.62)	3.50 (2.81–5.12)	0.047
IL-4 (pg/ml)				
Pre-LD	1.71 (1.45–2.99)	1.69 (1.41–2.83)	1.79 (1.53–3.12)	0.585
Peak	2.55 (2.23–2.97)	2.39 (2.21–2.99)	2.72 (2.24–3.55)	0.199
IL-6 (pg/ml)				
Pre-LD	9.33 (5.20–23.37)	6.37 (4.52–12.00)	18.09 (8.39–28.25)	0.025
Peak	50.74 (21.35–740.29)	38.35 (15.78–321.35)	55.23 (33.58–1244.67)	0.082
IL-10 (pg/ml)				
Pre-LD	4.66 (3.36–10.68)	3.71 (3.08–5.44)	9.13 (4.56–17.09)	0.006
Peak	23.86 (12.31–101.01)	20.80 (11.17–70.01)	81.36 (16.86–147.45)	0.017
IFNγ (pg/ml)				
Pre-LD	1.89 (1.57–2.80)	1.73 (1.57–2.63)	2.62 (1.45–3.05)	0.403
Peak	4.47 (2.88–11.00)	4.51 (2.90–13.27)	5.94 (3.24–16.04)	0.363
TNFα (pg/ml)				
Pre-LD	1.56 (1.25–2.88)	1.54 (1.26–2.45)	2.01 (0.92–3.04)	0.946
Peak	2.76 (2.16–4.08)	2.67 (2.03–3.77)	2.72 (2.18–3.41)	0.688

PLT indicates platelet; Pre-LD, pre-lymphodepletion; APTT, activated partial thromboplastin time;

FIB, fibrinogen; FDP, fibrinogen degradation products; TT, thrombin time; ATIII, antithrombin III; PT, prothrombin time; LDH, dehydrogenase; CRP, C-reactive protein; IL, interleukin; IFN γ , interferon γ ; TNF α , tumor necrosis factor α .

Supplemental Table 8. Correlation matrix showing the correlation among coagulation markers, inflammation markers and tumor burden markers

	D-dimer	PLT	FIB	PT	APTT	ATIII	TT	FDP	CRP	Ferritin	LDH	Bone-marrow blast	IL-6	IL-10
D-dimer	1.0	-0.676, P<0.001	-0.519, P<0.001	0.664, P<0.001	0.473, P<0.001	0.170, P=0.244	0.508, P<0.001	0.924, P<0.001	0.360, P=0.007	0.669, P<0.001	0.745, P<0.001	0.488, P=0.057	0.609, P<0.001	0.568, P<0.001
PLT	-0.676, P<0.001	1.0	0.495, P<0.001	-0.536, P<0.001	-0.412, P=0.002	-0.108, P=0.462	-0.455, P<0.001	-0.696, P<0.001	-0.269, P=0.045	-0.662, P<0.001	-0.722, P<0.001	-0.734, P=0.002	-0.412, P=0.002	-0.558, P<0.001
FIB	-0.519, P<0.001	0.495, P<0.001	1.0	-0.502, P<0.001	-0.360, P=0.006	-0.037, P=0.801	-0.596, P<0.001	-0.516, P<0.001	0.032, P=0.813	-0.399, P=0.003	-0.545, P<0.001	-0.250, P=0.349	-0.340, P=0.010	-0.558, P<0.001
PT	0.664, P<0.001	-0.536, P<0.001	-0.502, P<0.001	1.0	0.644, P<0.001	-0.026, P=0.857	0.478, P<0.001	0.598, P<0.001	0.295, P=0.027	0.548, P<0.001	0.622, P<0.001	0.631, P=0.010	0.569, P<0.001	0.595, P<0.001
APTT	0.473, P<0.001	-0.412, P=0.002	-0.360, P=0.006	0.644, P<0.001	1.0	-0.075, P=0.607	0.456, P<0.001	0.532, P<0.001	0.539, P<0.001	0.582, P<0.001	0.450, P<0.001	0.174, P=0.519	0.578, P<0.001	0.497, P<0.001
ATIII	0.170, P=0.244	-0.108, P=0.462	-0.037, P=0.801	-0.026, P=0.857	-0.075, P=0.607	1.0	0.141, P=0.334	0.157 P=0.283 1	0.045 P=0.757	0.168 P=0.253	0.154 P=0.291	0.515, P=0.044	-0.099 P=0.499	0.155 P=0.287
TT	0.508, P<0.001	-0.455, P<0.001	-0.596, P<0.001	0.478, P<0.001	0.456, P<0.001	0.141, P=0.334	1.0	0.495 P<0.001	0.105 P=0.441	0.501 P<0.001	0.427 P=0.001	0.474, P=0.065	0.479 P<0.001	0.315 P=0.018
FDP	0.924, P<0.001	-0.696, P<0.001	-0.516, P<0.001	0.598, P<0.001	0.532, P<0.001	0.157 P=0.283	0.495 P<0.001	1.0	0.395 P=0.004	0.696 P<0.001	0.744 P<0.001	0.447, P=0.084	0.728 P<0.001	0.628 P<0.001
CRP	0.360, P=0.007	-0.269, P=0.045	0.032, P=0.813	0.295, P=0.027	0.539, P<0.001	0.045 P=0.757	0.105 P=0.441	0.395 P=0.004	1.0	0.442 P=0.001	0.355 P=0.007	-0.065, P=0.814	0.448 P=0.001	0.456 P<0.001
Ferritin	0.669, P<0.001	-0.662, P<0.001	-0.399, P=0.003	0.548, P<0.001	0.582, P<0.001	0.168 P=0.253	0.501 P<0.001	0.696 P<0.001	0.442 P=0.001	1.0	0.632 P<0.001	0.688, P=0.004	0.429 P=0.001	0.611 P<0.001
LDH	0.745, P<0.001	-0.722, P<0.001	-0.545, P<0.001	0.622, P<0.001	0.450, P<0.001	0.154 P=0.291	0.427 P=0.001	0.744 P<0.001	0.355 P=0.007	0.632 P<0.001	1.0	0.494, P=0.005	0.510 P<0.001	0.585 P<0.001
Bone-marrow blast	0.4882, P=0.057	-0.734, P=0.002	-0.250, P=0.349	0.631, P=0.010	0.174, P=0.519	0.515, P=0.044	0.474, P=0.065	0.447, P=0.084	-0.065, P=0.814	0.688, P=0.004	0.494, P=0.005	1.0	0.274, P=0.054	0.679, P=0.304
IL-6	0.609, P<0.001	-0.412, P=0.002	-0.340, P=0.010	0.569, P<0.001	0.578, P<0.001	-0.099 P=0.499	0.479 P<0.001	0.728 P<0.001	0.448 P=0.001	0.429 P=0.001	0.510 P<0.001	0.274, P=0.054	1.0	0.483 P<0.001
IL-10	0.568, P<0.001	-0.558, P<0.001	-0.321, P=0.016	0.595, P<0.001	0.497, P<0.001	0.155 P=0.287	0.315 P=0.018	0.628 P<0.001	0.456 P<0.001	0.611 P<0.001	0.585 P<0.001	0.679, P=0.304	0.483 P<0.001	1.0

PLT indicates platelet; FIB, fibrinogen; PT, prothrombin time; APTT, activated partial thromboplastin time; ATIII, antithrombin III; TT, thrombin time; FDP, fibrinogen degradation products; CRP, C-reactive protein; LDH, dehydrogenase; IL, interleukin.

Supplemental Table 9. Risk stratification according to IL-10 level and the EASIX score on bleeding events

Risk group	Parameters		n*	Events, n	Cumulative Inc, %	HR	95% CI	P value
	IL-10 (pg/mL)	EASIX						
High risk	>7.98	> 7.649	7	7	100.00	14.47	2.78–75.29	< 0.0001
Intermediate risk	>7.98	≤ 7.649	13	5	38.46	3.68	0.82 –16.67	0.089
	≤7.98	> 7.649						
Low risk	≤7.98	≤ 7.649	17	2	11.76	1 (reference)	-	-

IL indicates interleukin; EASIX, endothelial activation and stress index; Cumulative Inc, cumulative incidence; HR, hazard ratio; CI, confidence interval.

High risk vs intermediate risk: HR, 5.495; 95% CI, 1.346–22.43; $P = 0.0003$.

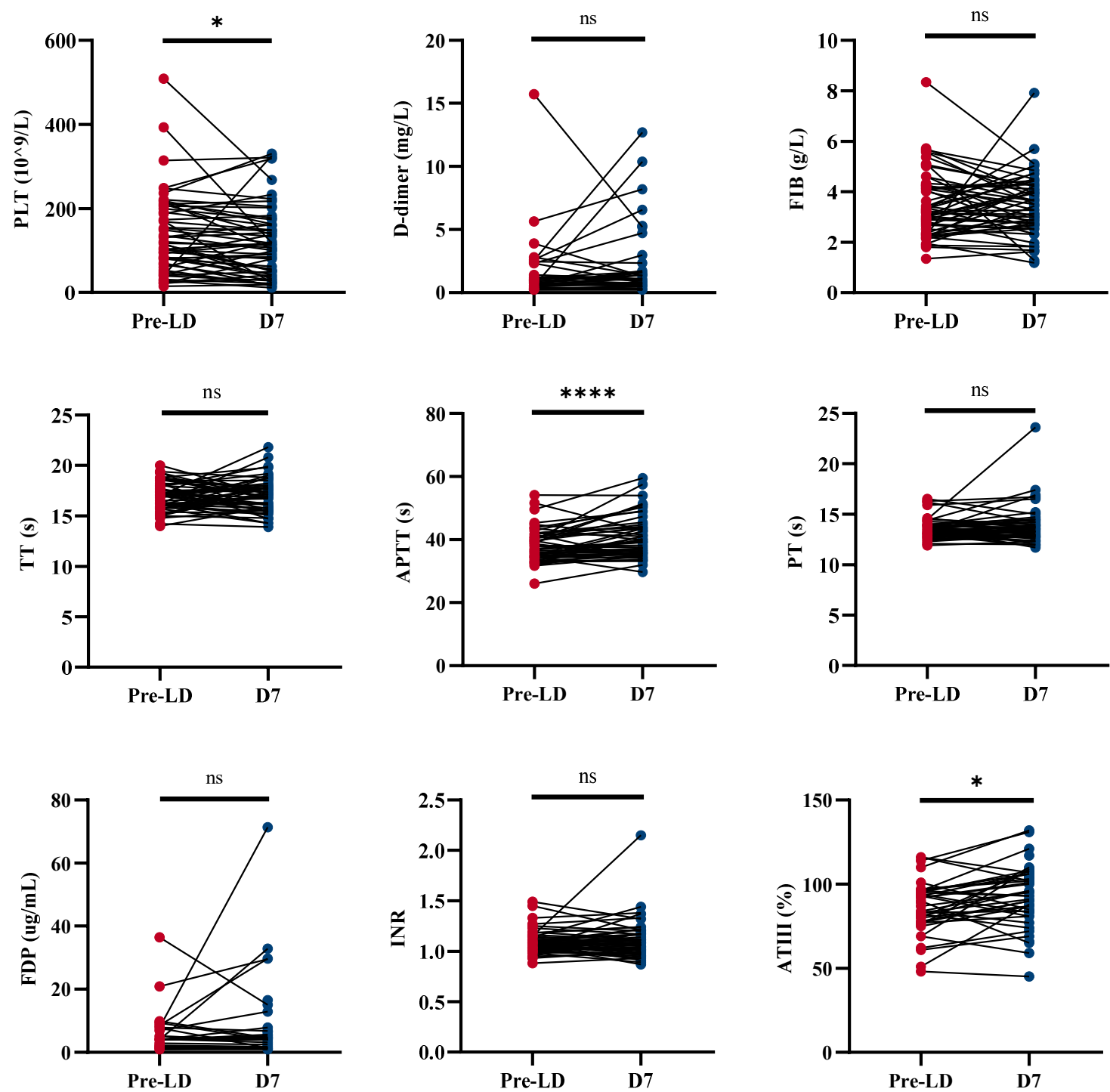
*Only patients with available IL-10 levels were eligible for this analysis.

Supplemental Table 10. Univariate and multivariate logistic regression analysis of grade 2–3 bleeding events

Marker	Subgroup	No. case	No. Non-case	Unadjusted OR (95% CI)	P value	Adjusted OR (95% CI)	P value
Pre-LD D-dimer (mg/L)	≤0.58	0	21	1 (reference)	0.998		
	> 0.58	6	20	484642457.48 (0–)			
Pre-LD CRP (mg/L)	≤10.39	1	24	1 (reference)	0.101		
	>10.39	6	23	6.26 (0.70–56.10)			
Pre-LD IL-6 (pg/ml)	≤7.49	0	16	1 (reference)	0.998		
	>7.49	7	13	869871062.59 (0–)			
Pre-LD IL-10 (pg/ml)	≤7.98	0	24	1 (reference)	0.998		
	>7.98	6	5	1938569843.51 (0–)			
Pre-LD Ferritin (ng/ml)	≥96.3	4	32	1 (reference)	0.038		
	<96.3	2	1	16.00 (1.17–218.86)			
Pre-LD EASIX	≤8.81	2	41	1 (reference)	0.002	1 (reference)	0.002
	>8.81	4	3	27.33 (3.47–215.07)		27.33 (3.47–215.07)	
Pre-LD PLT (10⁹/L)	≥102.5	1	30	1 (reference)	0.045		
	<102.5	6	19	9.47 (1.06–84.96)			
Pre-LD TNFα (pg/ml)	≤2.93	3	24	1 (reference)	0.100		
	>2.93	3	5	4.80 (0.74–31.08)			
CRS	≤0.5	2	24	1 (reference)	0.301		
	>0.5	5	24	2.50 (0.44–14.17)			

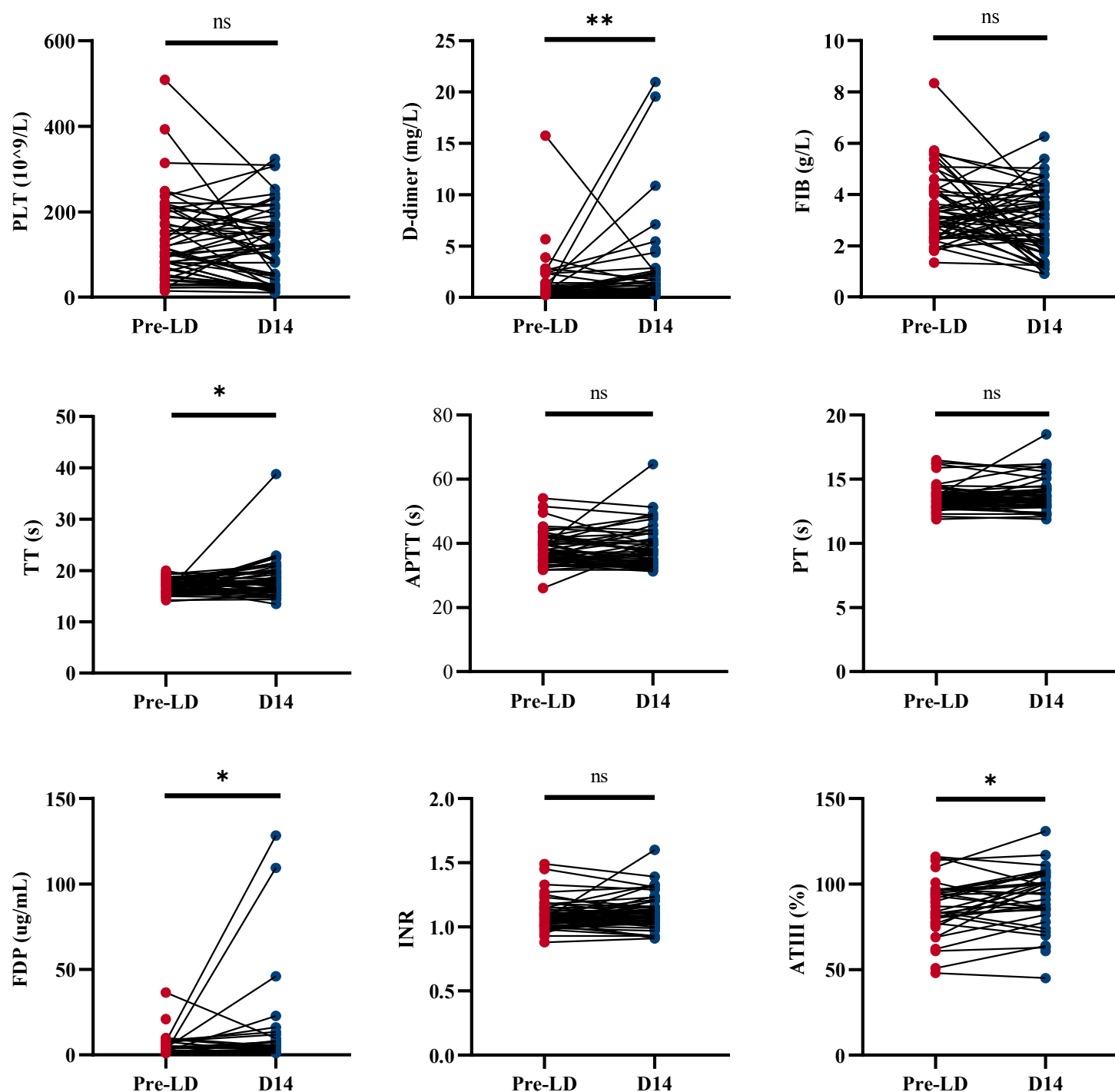
OR, odds ratio; LD, lymphodepletion; CRP, C-reactive protein; IL, interleukin; EASIX, endothelial activation and stress index; PLT, platelet; TNFα, tumor necrosis factor α; CRS, cytokine release syndrome.

Supplemental Figure 1.



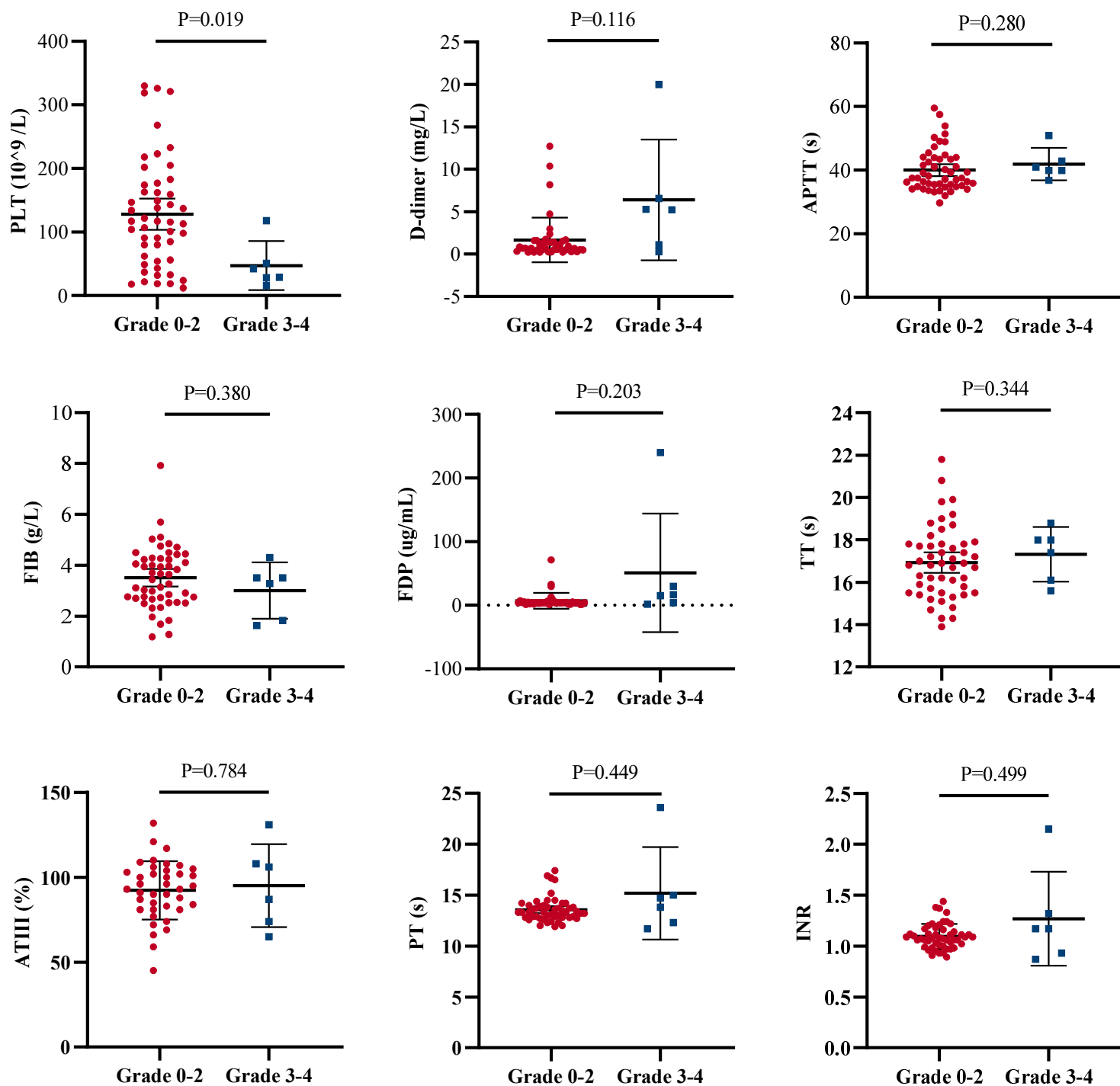
Supplemental Figure 1. Coagulation parameters changes from pre-LD to day 7. Changes of PLT, D-dimer, FIB, TT, APTT, PT, FDP, INR, and ATIII from pre-LD to day 7. * $P < .05$. **** $P < .0001$. ns, not significant. Pre-LD, pre-lymphodepletion; PLT, platelet; FIB, fibrinogen; TT, thrombin time; APTT, activated partial thromboplastin time; PT, prothrombin time; FDP, fibrinogen degradation products; INR, international normalized ratio; ATIII, antithrombin III.

Supplemental Figure 2.



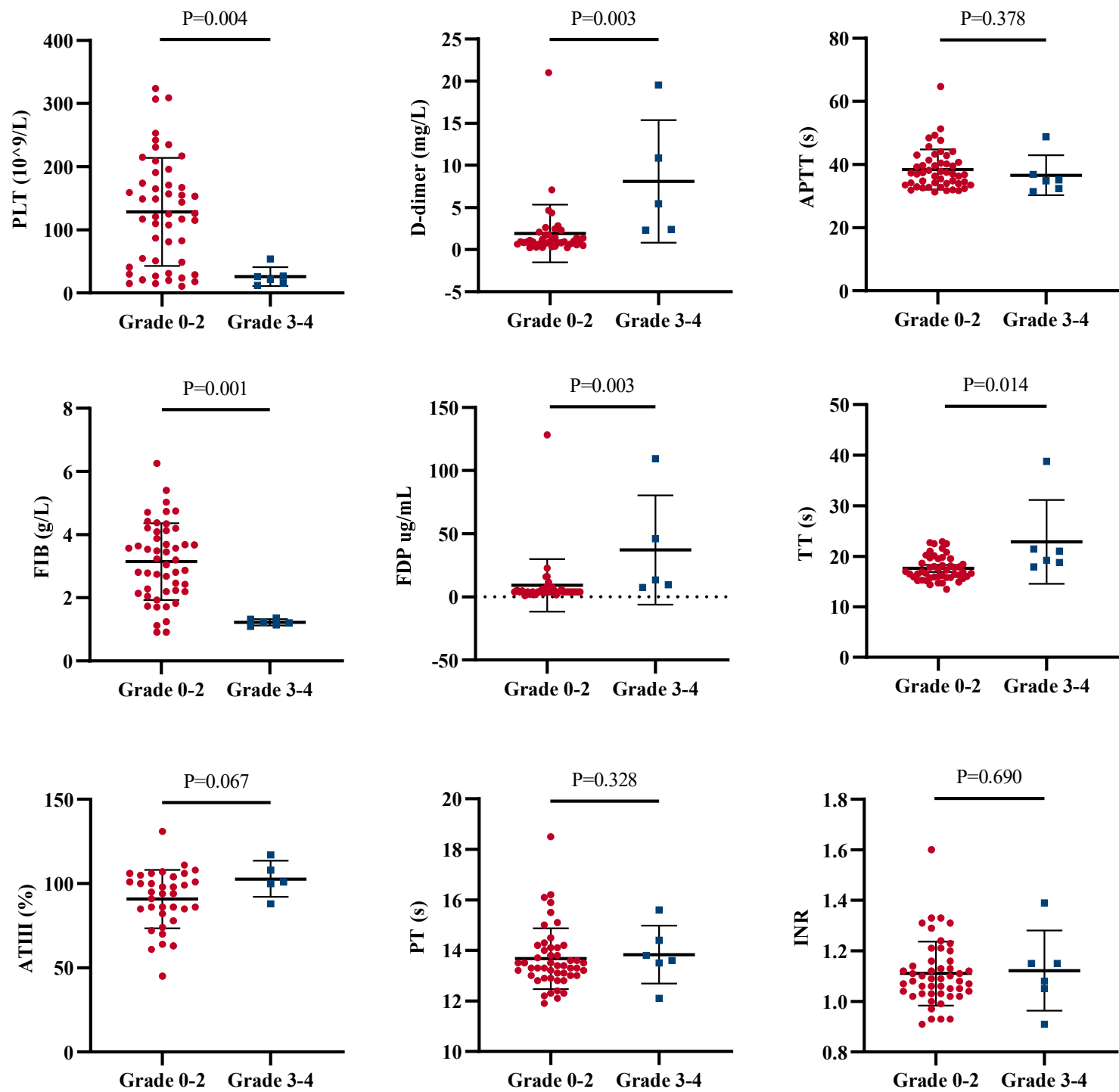
Supplemental Figure 2. Coagulation parameters changes from pre-LD to day 14. Changes of PLT, D-dimer, FIB, TT, APTT, PT, FDP, INR, and ATIII from pre-LD to day 14. * $P < .05$. ** $P < .01$. ns, not significant. Pre-LD, pre-lymphodepletion; PLT, platelet; FIB, fibrinogen; TT, thrombin time; APTT, activated partial thromboplastin time; PT, prothrombin time; FDP, fibrinogen degradation products; INR, international normalized ratio; ATIII, antithrombin III.

Supplemental Figure 3.



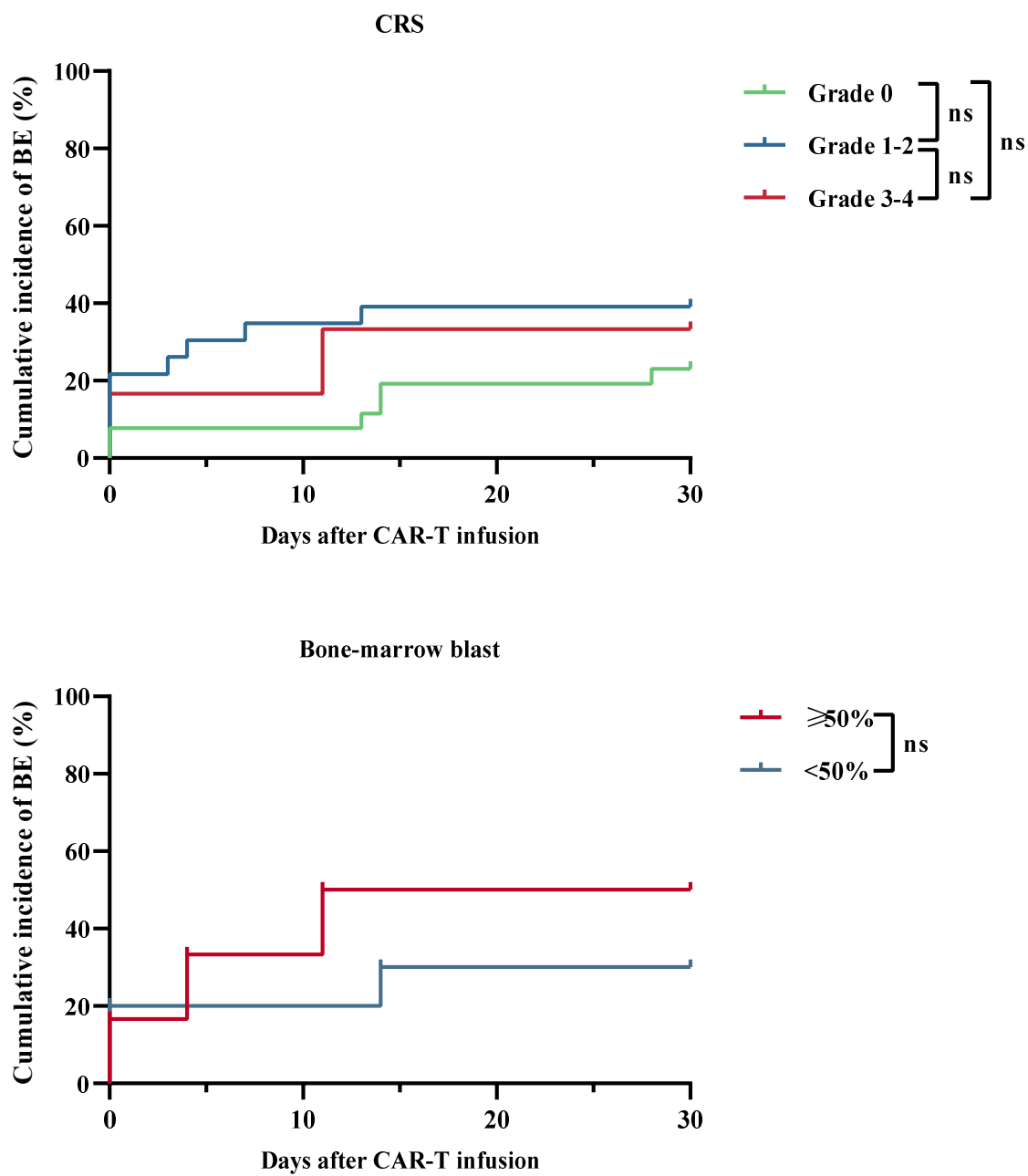
Supplemental Figure 3. Coagulation parameters by the severity of CRS on day 7. PLT, platelet; FIB, fibrinogen; TT, thrombin time; APTT, activated partial thromboplastin time; PT, prothrombin time; FDP, fibrinogen degradation products; INR, international normalized ratio; ATIII, antithrombin III.

Supplemental Figure 4.



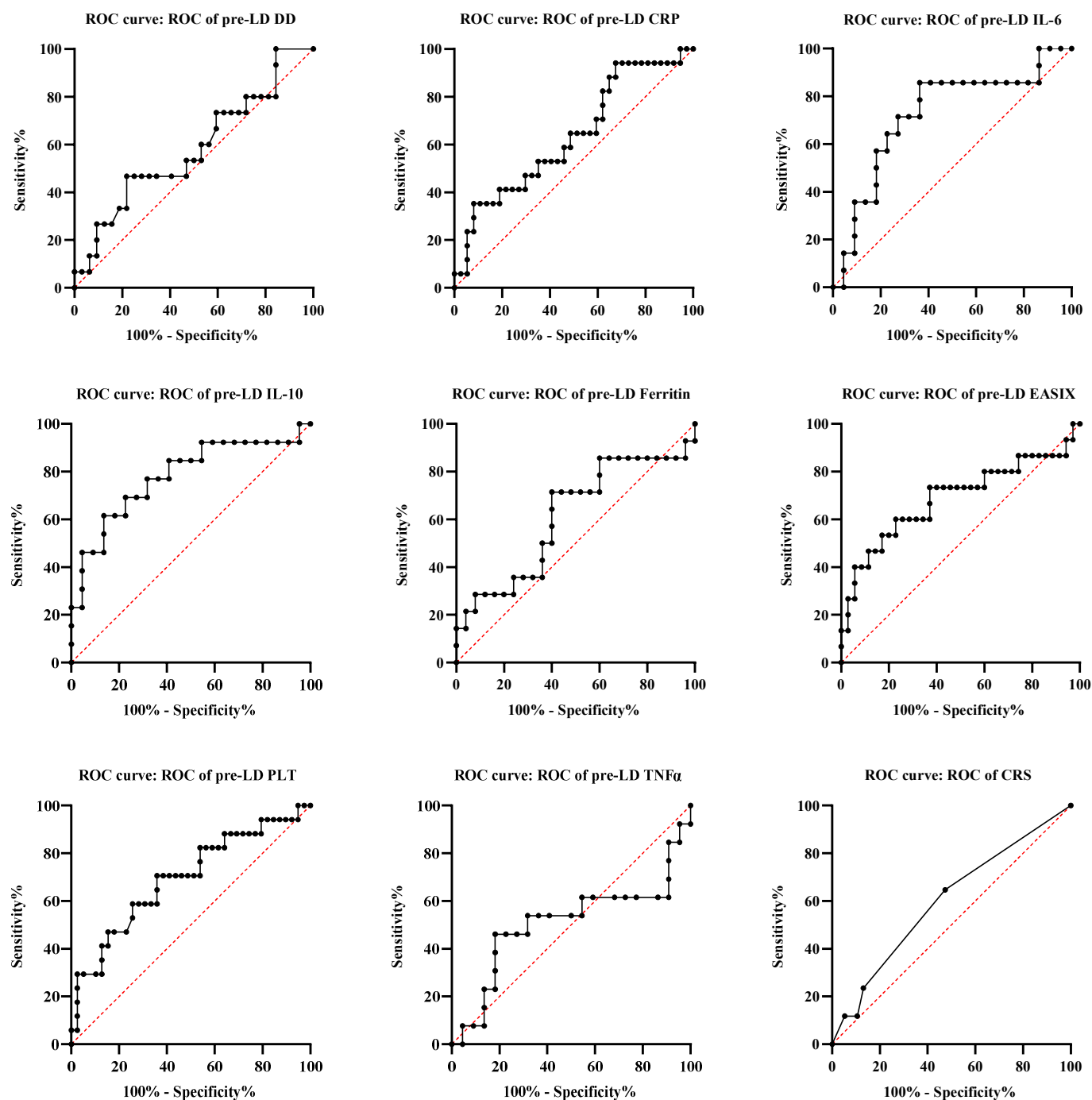
Supplemental Figure 4. Coagulation parameters by the severity of CRS on day 14. PLT, platelet; FIB, fibrinogen; TT, thrombin time; APTT, activated partial thromboplastin time; PT, prothrombin time; FDP, fibrinogen degradation products; INR, international normalized ratio; ATIII, antithrombin III.

Supplemental Figure 5.



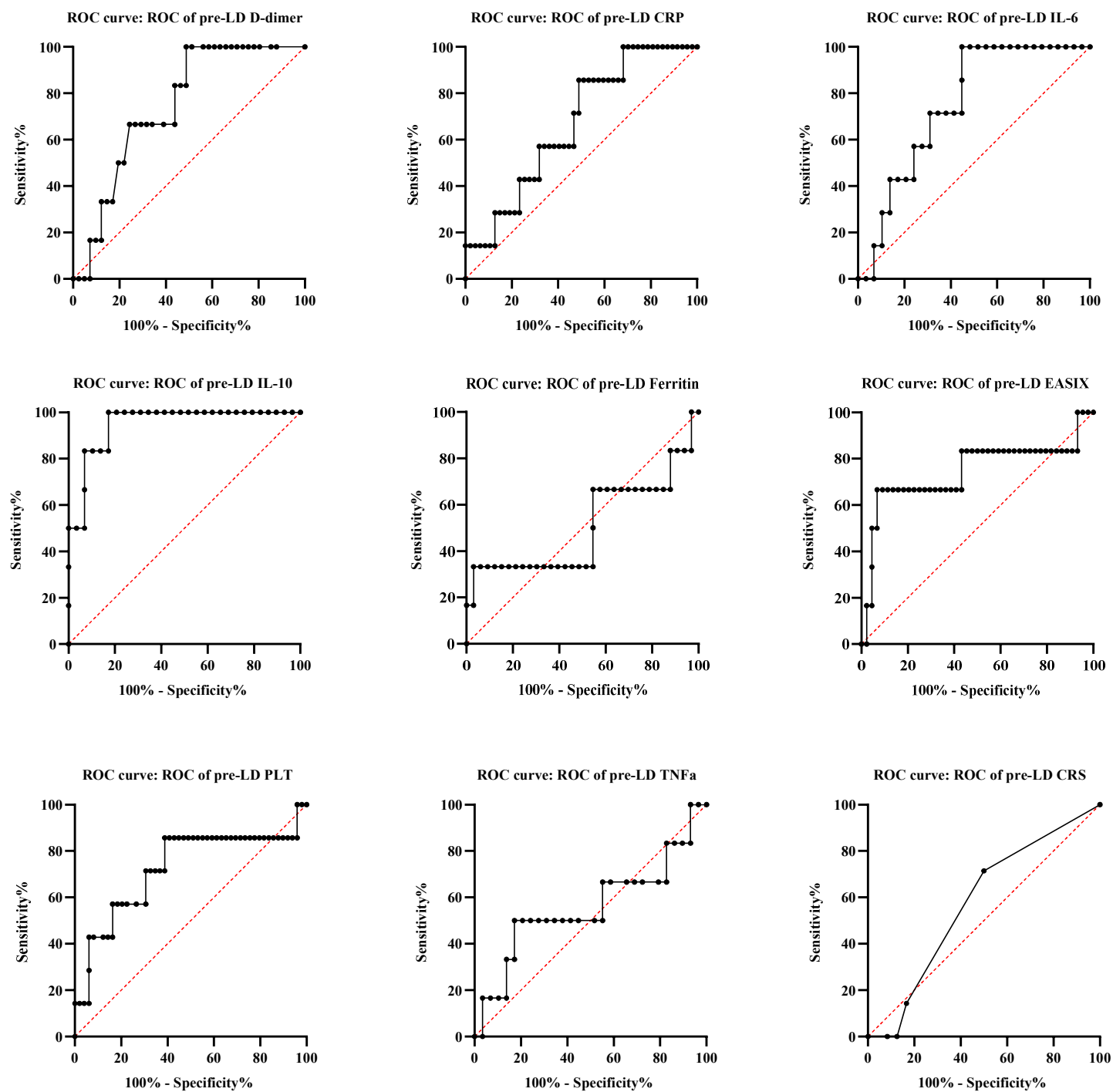
Supplemental Figure 5. Cumulative incidence of bleeding events by grade of CRS and bone-marrow tumor burden. BE, bleeding events; CRS, cytokine release syndrome.

Supplemental Figure 6.



Supplemental Figure 6. ROC curve analysis of D-dimer, CRP, IL-6, IL-10, ferritin, EASIX score, PLT, TNF α , and CRS pre-lymphodepletion for bleeding events. ROC, receiver operating characteristic; CRP, C-reactive protein; IL, interleukin; EASIX, endothelial activation and stress index; PLT, platelet; TNF, tumor necrosis factor; CRS, cytokine release syndrome.

Supplemental Figure 7.



Supplemental Figure 7. ROC curve analysis of D-dimer, CRP, IL-6, IL-10, ferritin, EASIX score, PLT, TNF α , and CRS pre-lymphodepletion for grade 2-3 bleeding events. ROC, receiver operating characteristic; CRP, C-reactive protein; IL, interleukin; EASIX, endothelial activation and stress index; PLT, platelet; TNF, tumor necrosis factor; CRS, cytokine release syndrome.