Characterization and associated risk factors of thrombotic thrombocytopenic purpura in autoimmune related diseases: a retrospective study

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Research Article

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Abstract

Objectives: As a rare but serious disease with high mortality, the features and risk factors of thrombotic thrombocytopenic purpura (TTP) in underlying autoimmune related diseases (AIRDs) are not well characterized.

Methods: We retrospectively reviewed the electronic medical records of TTP patients with AIRDs who were hospitalized in Tongji Hospital from January 2017 to August 2021. 120 AIRDs patients with severe thrombocytopenia were included as controls. Statistical analysis involved Student's t test, Mann-Whitney U test, Chi-squared test, Fisher's exact test, and univariate logistic regression.

Results: The prevalence of TTP in AIRD patients was 1.541‰, with a high mortality of 32.26% in this study. Lower platelet count, higher LDH level, higher cTnI level, more frequent proteinuria and coma on admission were observed in non-survivors when compared with survivors. History of thrombocytopenia, antiphospholipid antibody (aPL) positivity, neurologic symptoms, anti-SSB positivity, proteinuria and hematuria were associated with TTP in AIRD patients who had severe thrombocytopenia. Levels of IBIL, LDH, Hb, cTnI, and D-Dimer had good capabilities to discriminate TTP from severe thrombocytopenia in patients with AIRD.

Conclusion: AIRD-TTP is a rare emergency with a high mortality rate of 32.26%. Organ damages, severities of thrombocytopenia and hemolysis on admission were associated with the outcome of AIRD-TTP patients. Levels of IBIL, LDH, Hb, cTnI, and D-Dimer may provide important information for the diagnosis of AIRD-TTP.

Key Messages

- This study was focused on characteristics of TTP patients with underlying AIRDs.
- Extremely severe thrombocytopenia, high LDH level, proteinuria, cardiac injury and coma on admission were related to early death of AIRD-TTP.
- IBIL, LDH, Hb, cTnI, and D-Dimer were associated factors of TTP in AIRD patients with severe thrombocytopenia.

Introduction

Thrombotic thrombocytopenic purpura (TTP) is a rare but fatal hematologic disorder characterized by severe thrombocytopenia, microangiopathic hemolytic anemia (MAHA), and ischemic end organ injury [1]. TTP is a result of severe deficiency of a metalloprotease named a disintegrin-like and metalloproteinase with thrombospondin type 1 motif, member 13 (ADAMTS13) [2]. Deficiency of ADAMTS13 leads to abnormal accumulation of ultra large von willebrand factor (VWF) multimers and formation of platelet-rich micro-thrombi within small arterioles and capillaries, which subsequently induces wide-spreading microvascular ischemia [3]. Severe ADAMTS13 deficiency can either be caused

by congenital deficiency or acquired immune mediated deficiency. Immune-mediated TTP (iTTP) typically occurs in adults, accounting for 90% of cases, whereas congenital TTP (cTTP) is often detected in the childhood or during pregnancy [4]. iTTP is classified as primary or secondary, depending on whether an obvious underlying disease can be identified or not [5].

Secondary iTTP has been reported to be associated with a variety of autoimmune related diseases (AIRDs), such as systemic lupus erythematosus (SLE), sjögren's syndrome (SS), polymyositis, and other autoimmune diseases [6–10]. In a retrospective analysis, almost 9% of TTP cases had one or more underlying connective tissue diseases [11]. TTP was reported in 2% of SLE patients [12]. The underlying pathophysiology for TTP in AIRDs is not yet clear. It was assumed that autoimmune reaction specifically targeted against ADAMTS13 might be a part of combined autoimmune diseases [13]. TTP is a medical emergency with a mortality of up to 90% if unrecognized, while prompt treatment with plasmapheresis decreased the mortality rate to 10–20% [14]. It is reported that TTP associated with AIRDs had an even higher mortality and poorer response to treatment when compared with primary TTP [15, 16]. Timely identification of TTP in AIRDs is particularly important for prompt treatments and survival benefits. However, clinical diagnosis of TTP might be challenging in patients with AIRDs, because clinical features of acute TTP can be extraordinarily diverse and both TTP and AIRDs can display some or all features of the classic pentad (fever, thrombocytopenia, MAHA, neurological disorders, and renal dysfunction) [3].

Even though there were several studies attempted to summarize the clinical features and prognosis of SLE-TTP, some limitations should be noted. In most previous studies, TTP was diagnosed by unspecific clinical judgement without a confirmatory test of ADAMTS13 deficiency [12, 15, 17]. A study involving ten SLE-TTP patients with severe ADAMTS13 deficiency was of a small sample size and showed insufficient information [18]. In this work, TTP patients with confirmatory ADAMTS13 test were included. We analyzed the clinical features and mortality rate of AIRD-TTP, and identified prognostic factors of AIRD-TTP. Moreover, we explored associated factors of TTP in patients with AIRDs, which may provide useful information for the early diagnosis of AIRD-TTP.

Methods

Patients

We reviewed electronic medical records of patients that were hospitalized in Tongji Hospital between January 2017 and August 2021. The inclusion criteria were: (I) met diagnostic criteria of AIRDs; (II) met diagnostic criteria of TTP, which are specified in the next paragraph; (III) age ≥18 years old. The exclusion criteria were established according to comorbidities that may result in acquired TTP other than AIRDs, including (I) malignancy, (II) pregnancy, (III) sepsis, (IV) organ transplantation, and (V) disseminated intravascular coagulation (DIC) [19]. A total of 31 AIRD-TTP patients were enrolled in this retrospective study. Based on the literature [20], all patients were followed for at least 30 days after admission and divided into survivor and non-survivor groups based on the outcome within 30 days after admission. TTP is characterized by severe thrombocytopenia (generally platelet count of $\leq 30\times10^9$/L); anemia may not be

immediately obvious [5]. Based on the established exclusion criteria, 120 AIRD patients with severe thrombocytopenia (≤30×10^9/L) hospitalized in our institution during the same period were randomly selected as a control named non-TTP group. This study was approved by the Institutional Ethics Committee of Tongji Hospital (TJ-IRB20210853). As data were obtained retrospectively and identifiable personal information was removed, the informed consent was waived.

Definitions

A diagnosis of TTP was defined according to the following criteria: (I) ADAMTS13 activity <10%; or (II) ADAMTS13 activity ≤20% and ADAMTS13 inhibitor was present [5]. Severe thrombocytopenia was defined when platelet count is ≤30×10^9/L [5]. SLE was diagnosed based on the revised 1997 American College of Rheumatology diagnostic criteria [21]. SS was diagnosed based on the 2002 American-European Consensus Group Criteria [22]. Patients of idiopathic inflammatory myopathies (IIM) fulfilled the diagnostic criteria for DM or PM according to the Bohan and Peter criteria [23]. Patients of systemic sclerosis (SSc) met 2013 ACR/EULAR classification criteria [24]. Other patients who had autoantibodies but did not meet diagnostic criteria established for a specific disease were defined as connective tissue disease (CTD). Proteinuria of ≥2+ and hematuria of ≥2+ were chosen as abnormal cut-off value to avoid the overestimation of proteinuria and hematuria caused by random diurnal sample [25].

Data collection

Data including demographical information, medical history, clinical characteristics, and laboratory results were retrospectively reviewed through the electronic medical records system of Tongji Hospital. The earliest available laboratory values for up to 3 days for patients were collected.

Statistical analysis

Descriptive statistics were given as the Mean ±Standard deviation, median (interquartile range, IQR), or frequencies (percentage). Differences in the mean or median values were analyzed using Student's t test or Mann-Whitney U test. Categorical variables were compared using the Chi-squared test or Fisher’s exact test. Univariate logistic regressions were performed to analyze the potential associated factors for clinical assessment or risk factors of AIRD-TTP mortality. Kaplan-Meier analysis with log-rank test was employed to assess the differences in survival.

The C statistic was calculated from the area under the receiver operator curve (AUC of ROC), and was compared using the Z test. Sensitivity, specificity, and cut-off value, together with their 95% confidence intervals (CI), were calculated. Diagnostic accuracy was assessed at predefined cut-off points. Statistical analyses were performed in GraphPad Prism 6, IBM SPSS Statistics 25. A P-value of ≤0.05 was considered statistically significant.

Results

Clinical features of TTP patients with underlying AIRDs

We summarized the clinical characteristics of TTP patients with AIRDs in Fig. 1 and Table 1. The prevalence rate of TTP among 20117 AIRD patients that were hospitalized in Tongji hospital from January 2017 to Aug 2021 was 1.541‰. There were 16 (51.6%) CTD-related, 11 (35.5%) SLE-related, 2 (6.5%) SS-related, 1 (3.2%) SSc-related, and 1 (3.2%) IIM-related TTP cases. AIRD-TTP occurs most often between the ages of 41 and 50 with a mean age of 45.39 years old.

Table 1
Comparison of characteristics in survivors and non-survivors of AIRD-TTP.

	Survivors, n = 21	non-Survivors, n = 10	P-Value	Univariate analysis
				OR (95%CI)
Gender (female), n (%)	15 (71.4)	7 (70.0)	1.000	-
Age (years), mean (SD)	42.90 ± 12.91	50.60 ± 10.70	0.075	-
Laboratory tests				
PLT ($\times 10^9$/L), median (IQR)	9 (6–12)	4 (3.75-6)	0.001	0.535 (0.323–0.887)
PLT ≤ 6×10^9/L, n (%)	6 (28.6)	9 (90.0)	0.002	22.500 (2.319-218.349)
Hb (g/L), mean (SD)	67 ± 14.32	76.9 ± 14.89	0.086	-
LDH (U/L), median (IQR)	1041.95 ± 397.95	1495.70 ± 351	0.005	1.003 (1.001–1.006)
LDH > 1216 U/L, n (%)	5 (23.8)	9 (90.0)	0.002	28.800 (2.896-286.425)
IBIL (μmol/L), median (IQR)	27.7 (23.05–34.95)	35.3 (17.05–72.73)	0.466	-
Scr (μmol/L), median (IQR)	72 (57-95.5)	84 (69.75-152.25)	0.124	-
cTnI (pg/mL), median (IQR)	8.9 (69.4-107.15)	467.8 (119.3-662.6)	0.001	1.007 (1.002–1.012)
cTnI > 100.3 pg/mL, n (%)	4/17 (23.5)	9/10 (90.0)	0.001	29.250 (2.789-306.811)
cTnI > 250 pg/mL, n (%)	2/17 (11.9)	7/10 (70.0)	0.004	17.500 (2.365-129.506)
D-Dimer (μg/mL), median (IQR)	3.51 (1.73–9.98)	4.91 (2.00-12.56)	0.512	-
Proteinuria ≥ 2+, n (%)	6/18 (33.3)	8/10 (80.0)	0.046	6.789 (1.096–42.065)
Hematuria ≥ 2+, n (%)	13/18 (72.2)	8/10 (80.0)	1.000	-
Complications, n (%)				

Data are shown for survivors (n = 21) and non-survivors (n = 10) unless stated otherwise. Data on cTnI were from 17 survivors and 10 non-survivors; data on D-Dimer were from 19 survivors and 10 non-survivors; data on proteinuria and hematuria were from 18 survivors and 10 non-survivors.

Abbreviation: *PLT* platelet, *Hb* hemoglobin, *LDH* lactate dehydrogenase, *IBIL* indirect bilirubin, *Scr* serum creatinine, *cTnI* cardiac troponin I

	Survivors, n = 21	non-Survivors, n = 10	P-Value	Univariate analysis
				OR (95%CI)
Pulmonary infection	2 (9.5)	4 (40.0)	0.067	-
Coma	1 (4.8)	4 (40.0)	0.027	13.333 (1.242-143.151)
Therapies, n (%)				
Corticosteroid	21 (100.0)	10 (100.0)	1.000	-
Impulse therapy	7 (33.3)	1 (10.0)	0.343	-
Intravenous immunoglobulin	11 (52.4)	7 (70.0)	0.452	-
Plasmapheresis	21 (100.0)	7 (70.0)	0.027	-
Cyclophosphamide	8 (38.1)	1 (10.0)	0.205	-
Rituximab	3 (14.3)	0 (0.0)	0.533	-
Data are shown for survivors (n = 21) and non-survivors (n = 10) unless stated otherwise. Data on cTnI were from 17 survivors and 10 non-survivors; data on D-Dimer were from 19 survivors and 10 non-survivors; data on proteinuria and hematuria were from 18 survivors and 10 non-survivors.				
Abbreviation: *PLT* platelet, *Hb* hemoglobin, *LDH* lactate dehydrogenase, *IBIL* indirect bilirubin, *Scr* serum creatinine, *cTnI* cardiac troponin I				

All these 31 TTP patients had severe thrombocytopenia. None of the patients had a previous history of TTP. There were 2 cases (6.5%) reported a history of thrombocytopenia. The symptoms of AIRD-TTP patients included neurologic symptoms, fatigue, purpura, hemorrhage and fever. Neurologic symptoms were the most common (87.1%), followed by fatigue (38.7%). Purpura, hemorrhage, and fever were reported in 35.5%, 25.8%, and 19.4% patients. Among the AIRD-TTP patients, ANA, anti-SSA, and anti-Ro-52 were observed most frequently (87.1%, 64.5% and 51.6%).

Risk factors associated with early mortality in AIRD-TTP

During the observational period of 30 days after the admission of TTP patients, 10 patients died resulting in a total mortality of 32.26%. Characteristics of these TTP-AIRD patients with different clinical outcomes were summarized in Table 1. ADAMTS13 activity, subcategory of AIRDs, autoantibodies, and symptoms showed no significant differences between these two groups (Supplementary Table 1). Compared with survivors, deceased patients had lower platelet counts [4 (3.75-6) vs. 9 (6–12) $\times 10^9$/L, P = 0.001], higher serum LDH level (1495.70 ± 351 vs. 1041.95 ± 397.95 U/L, P = 0.005), and higher cTnI level [467.8 (119.3-662.6) vs. 8.9 (69.4-107.15) pg/mL, P = 0.001]. Based on ROC analysis, cTnI, PLT, and LDH had the potential to predict early mortality in TTP-AIRD patients with an AUC > 0.8. No significant difference was found among these three ROC curves when compared using Z test. Optimal cut-off points of cTnI, PLT,

and LDH to predict early death by ROC analysis were of > 100.3pg/mL (sensitivity: 90%, specificity: 76.19%), ≤ 6×10^9/L (sensitivity: 90%, specificity: 71.43%), and > 1216 U/L (sensitivity: 90%, specificity: 76.19%) (Fig. 2). Proteinuria ≥ 2 + was discovered in 80% of non-survivors and in 33.3% of survivors (P = 0.046). Pulmonary infection defined by computed tomography was reported in 40% of non-survivors and 9.5% of survivors, but the difference was not statistically significant (P = 0.067). One of the survivors and four of the deaths were in coma on admission (P = 0.027).

Plasmapheresis was performed in 90.32% of AIRD-TTP patients. All of the three patients without plasmapheresis died in hospital. Two out of these three patients died within 24 hours of admission and the last one died eight days after the admission. All the three patients who received rituximab treatment survived. Corticosteroid impulse therapy (≥ 500 mg/d), intravenous immunoglobulin (≥ 0.4 g/kg), and cyclophosphamide showed no significant differences between the two groups.

Univariate analysis indicated that PLT, LDH, cTnI, proteinuria, and coma were possible risk factors for early death (Table 1) and Kaplan-Meier survival curves of these factors were shown in Fig. 3.

Factors associated with TTP in AIRD patients

A high mortality of 32.26% in AIRD-TTP may be largely due to delayed diagnosis of TTP. Thus, we compared 31 AIRD-TTP patients with 120 AIRD patients with severe thrombocytopenia to determine the associated factors of TTP in patients with AIRDs and severe thrombocytopenia. The comparisons of TTP group and non-TTP group are shown in Table 3. There were no statistically significant differences between the two groups in age, sex, and category of underlying AIRDs. History of thrombocytopenia was significantly less common in TTP group than that in non-TTP group (P 0.001). Neurologic symptoms were more prevalent in TTP group than non-TTP group (87.1% vs. 10.8%, P 0.001). Antiphospholipid antibody (aPL) was found in 31.9% of non-TTP patients but only in 3.8% of TTP patients (P = 0.004). Conversely, anti-SSB positivity was observed more frequently in TTP group than non-TTP group (P = 0.049). Even though all patients enrolled in this study had platelet count of 30×10^9/L, the absolute platelet counts were lower in TTP patients than that in the controls [7(4–11) ×10^9/L vs. 19(9–26) ×10^9/L, P 0.001]. The value of hemoglobin (Hb) was significantly lower in TTP group than non-TTP group (70.19 ± 15.01 vs. 103.7 ± 25.12 g/L, P 0.001). Patients with TTP had higher levels of serum lactic dehydrogenase (LDH) and indirect bilirubin (IBIL) when compared with non-TTP patients (P 0.001 and P 0.001). Elevated levels of serum creatinine (Scr) and blood urea nitrogen (BUN) were more prevalent in TTP group than non-TTP group (P = 0.004 and P 0.001). Positive Coombs tests were observed in 17.2% (5/29) of TTP patients and 53% (44/83) of non-TTP patients (P 0.001). The incidences of proteinuria and hematuria in AIRD-TTP patients were more frequent than those in non-TTP patients (P 0.001 and P 0.001). Cardiac troponin I (cTnI) level was significantly higher in TTP patients than that in non-TTP patients [100.3 (15.50–357.0) vs. 4.0 (1.9–9.15) pg/mL, P 0.001]. 74.1% (20/27) of TTP patients showed abnormity of cTnI. Levels of D-Dimer and erythrocyte sedimentation rate (ESR) were significantly higher in TTP group than non-TTP group (P 0.001 and P = 0.001).

Nine serum markers showed significant differences between these two groups (Fig. 4A). ROC analyses of these continuous variables were performed (Fig. 4B-C). Good diagnostic accuracy (AUC ≥ 0.8) was observed in the ROC analysis of Hb, LDH, IBIL, cTnI, and D-Dimer. The ability of IBIL and LDH to discriminate between patients with and without TTP was more excellent than the others, as indicated by an AUC above 0.97. Z test showed that there is no significant difference between these two variables. IBIL provided a sensitivity of 96.77% and a specificity of 88.33% with a cut-off value of 12.8 pg/mL. A sensitivity of 96.77% and a relatively high specificity of 93.33% were observed for LDH at the optimal cut-off point of 554 U/L.

In the univariate analysis, history of thrombocytopenia, neurologic symptoms, anti-SSB, aPL, PLT count, serum level of Hb, LDH, IBIL, cTnI, and D-Dimer, proteinuria and hematuria were significantly associated with TTP in AIRDs (Table 2).

Table 2
Comparisons of characteristics between AIRD patients with TTP and without TTP.

	Without TTP, n = 120	With TTP, n = 31	P-Value	Univariate analysis
				OR (95%CI)
Gender (female), n (%)	100 (83.3)	22 (71.0)	0.119	-
Age (years), mean (SD)	44.72 ± 15.90	45.39 ± 11.27	0.826	-
Underlying AIRDs, n (%)				
CTD	51 (42.5)	16 (51.6)	0.363	-
SLE	51 (42.5)	11 (35.5)	0.479	-
SS	17 (14.2)	2 (6.5)	0.248	-
IIM	3 (2.5)	1 (3.2)	0.822	-
SSc	2 (1.7)	1 (3.2)	0.579	-
History of thrombocytopenia (≥ 3months), n (%)	58 (48.3)	2 (6.5)	0.001	0.468 (0.301–0.727)
Autoantibodies, n (%)				
ANA	115 (95.8)	22 (87.1)	0.067	-
Anti-SSA	60 (50.0)	20 (64.5)	0.149	-
Anti-Ro52	48 (40.0)	16 (51.6)	0.243	-
Anti-SSB	15 (12.5)	9 (29.0)	0.049	2.864 (1.112–7.372)
Anti-Smith	19 (15.8)	4 (12.9)	0.686	-
Anti-dsDNA	12 (10.0)	1 (3.2)	0.231	-
aPL	29/91 (31.9)	1/26 (3.8)	0.004	0.157 (0.025-.973)

Data are shown for TTP patients (n = 31) and non-TTP patients (n = 120) unless stated otherwise. Data on aPL were from 26 TTP patients and 91 non-TTP patients; data on cTnI were from 27 TTP patients and 82 non-TTP patients; data on D-Dimer were from 29 TTP patients and 115 non-TTP patients; data on Coomb's were from 29 TTP patients and 83 non-TTP patients; data of ESR were from 16 TTP patients and 89 non-TTP patients; data on proteinuria and hematuria were from 28 TTP patients and 111 non-TTP patients.

Abbreviation: *TTP* thrombotic thrombocytopenic purpura, *CTD* connective tissue disease, *SLE* systemic lupus erythematosus, *SS* sjögren's syndrome, *IIM* idiopathic inflammatory myopathies, *SSc* systemic sclerosis, *ANA* antinuclear antibodies, *aPL* antiphospholipid antibodies, *PLT* platelet, *Hb* hemoglobin, *LDH* lactate dehydrogenase, *IBIL* indirect bilirubin, *Scr* serum creatinine, *BUN* blood urea nitrogen, *cTnI* cardiac troponin I, *ESR* erythrocyte sedimentation rate.

	Without TTP, n = 120	With TTP, n = 31	P-Value	Univariate analysis
				OR (95%CI)
Symptoms, n (%)				
Neurologic symptoms	13 (10.8)	27 (87.1)	<0.001	60.750 (18.159-203.239)
Fatigue	30 (25.0)	12 (38.7)	0.129	-
Purpura	38 (31.7)	11 (35.5)	0.686	-
Hemorrhage	32 (26.7)	8 (25.8)	0.923	-
Fever	21 (17.5)	6 (19.4)	0.81	-
Laboratory tests				
PLT ($\times 10^9$/L), median (IQR)	19 (9–26)	7 (4–11)	<0.001	0.866 (0.812–0.923)
$\leq 15\times 10^9 L^{-1}$, n (%)	43 (35.8)	29 (93.5)	<0.001	48.261 (6.363-366.018)
Hb (g/L), mean (SD)	103.7 ± 25.12	70.19 ± 15.01	<0.001	0.934 (0.910–0.959)
≤ 90 g/L, n (%)	35 (29.2)	30 (96.8)	<0.001	72.857 (9.560-555.227)
LDH (U/L), median (IQR)	301.75 (202.5-489.5)	1551 (1203–1867)	<0.001	1.005 (1.003–1.007)
LDH > 214 U/L, n (%)	50 (41.7)	31 (100.0)	<0.001	-
IBIL (μmol/L), median (IQR)	4.8 (3.33–8.35)	27.7 (19.4–38.4)	<0.001	1.498 (1.263–1.777)
IBIL > 12.9 μmol/L, n (%)	11 (9.2)	29 (95.3)	<0.001	143.682 (30.153-684.646)
Src (μmol/L), median (IQR)	60 (49-74.75)	76 (57–108)	0.003	-

Data are shown for TTP patients (n = 31) and non-TTP patients (n = 120) unless stated otherwise. Data on aPL were from 26 TTP patients and 91 non-TTP patients; data on cTnI were from 27 TTP patients and 82 non-TTP patients; data on D-Dimer were from 29 TTP patients and 115 non-TTP patients; data on Coomb's were from 29 TTP patients and 83 non-TTP patients; data of ESR were from 16 TTP patients and 89 non-TTP patients; data on proteinuria and hematuria were from 28 TTP patients and 111 non-TTP patients.

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	Without TTP, n = 120	With TTP, n = 31	P-Value	Univariate analysis
				OR (95%CI)
Src > 84 μmol/L, n (%)	9 (7.5)	8 (25.8)	0.004	-
BUN (mmol/L), median (IQR)	5.7 (4.27–7.72)	8.20 (5.29–9.60)	0.002	-
BUN > 7.5 mmol/L	26 (21.7)	17 (54.8)	＜0.001	-
cTnI (pg/mL), median (IQR)	4.0 (1.9–9.15)	100.30 (15.50–357.0)	＜0.001	1.008 (1.002–1.014)
cTnI > 15.6 pg/mL, n (%)	13/82 (15.9)	20/27 (74.1)	＜0.001	7.269 (1.359–38.883)
D-Dimer (μg/mL), median (IQR)	0.64 (0.31–2.10)	4.42 (1.82-10)	＜0.001	1.148 (1.047–1.260)
D-Dimer > 0.5 μg/mL, n (%)	63/115 (54.8)	28/29 (96.6)	＜0.001	13.943 (1.692-114.918)
Positive Coombs test, n (%)	44/83 (53.0)	5/29 (17.2)	＜0.001	-
ESR (mm/h), median (IQR)	19 (9–43)	54 (41.5–88)	0.001	-
ESR > 20 mm/h	44/89 (49.4)	13/16 (81.3)	0.019	-
Proteinuria ≥ 2+, n (%)	10/111 (9.0)	14/28 (50.0)	＜0.001	10.386 (3.985–27.069)
Hematuria ≥ 2+, n (%)	21/111 (18.9)	21/28 (75.0)	＜0.001	10.648 (3.934–28.818)

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Discussion

Studies focused on TTP-AIRD, a subgroup of TTP, were rare. We analyzed autoimmune rheumatic disease associated TTP patients who were admitted in our hospital from 2017–2021. TTP was rarely considered the first manifestation of AIRDs. In AIRD-associated TTP patients, only 15% of patients were diagnosed with TTP and SLE simultaneously in a study from US [26]. In a recent Chinese study, however, 60% of

patients were diagnosed with SLE and TTP concurrently [17]. Similarly, TTP as the first presentation of AIRDs was common in our research. 29% of AIRD-TTP cases had preceding AIRDs, whereas 71% had new-onset AIRDs. This finding indicates that detection of autoantibody profile is essential for TTP patients to uncover the underlying AIRDs. SLE was the most frequently reported AIRD associated with TTP [27]. In this study, SLE coexisted with TTP in 35.5% cases, but CTD did in 51.6%. These patients who were defined as CTD had autoantibodies but did not meet diagnostic criteria of a specific disease. There have been conflicting reports regarding to the clinical outcomes of AIRDs associated TTP. Letchumanan et al reported that the mortality of eight SLE-TTP patients was as high as 62.5%, despite the timely and aggressive therapies [15]. However, in the report of SLE-TTP by Cai et al., all of the ten patients with SLE-TTP survived [18]. A high mortality rate of 32.3% was observed in the 31 AIRD-TTP patients from our data. A mortality of 36.36% was revealed in SLE-TTP subtype. These controversial results may be attributed to the heterogeneity of study population and the bias of small sample size.

Benhamou et al reported that old age was a strong predictor for death in iTTP [28]. In Kwok' study, infection is the only independent risk factor for the mortality of SLE-TTP patients [12]. Our data demonstrated that older age and pulmonary infection were more frequent in non-survivors than survivors, although no statistical significances were found between the two groups. Previously, no significant differences were found in platelet counts and LDH levels between survivors and non-survivors of TTP [28]. Our study revealed that platelet counts were significantly lower and extremely high serum LDH levels were more prevalent in the deceased patients, suggesting that more severe thrombocytopenia and hemolysis were related to the death of AIRD-TTP. A cTnI level of > 250 pg/mL had been reported as a predictor of early death in acquired TTP (sensitivity: 64%, specificity: 66%) [28]. In this study, a cut-off point of 250pg/mL also showed good discrimination between non-survivors and survivors (sensitivity: 70%, specificity: 88.24%). What is more, the optimal cut-off value of cTnI determined by ROC curve was 100.3 pg/mL, possessing a better sensitivity for predicting early death of AIRD-TTP patients (sensitivity: 90%, specificity: 76.5%). Coma on admission is another event significantly related to early death in this study. These observations are similar to previous findings; elevated cTnI level and neurological involvement were reported to be poor prognostic markers in patients with TTP [1, 20]. Severe proteinuria might predict worse prognosis of AIRD-TTP patients. Thus, organ involvement including cardiac injury, proteinuria and coma in AIRD-TTP suggested adverse outcomes, and aggressive management should be initiated to reduce the risk of death.

Although some innovative drugs, such as anti-CD20 monoclonal antibody, caplacizumab and recombinant ADAMTS13, have emerged in recent years as novel treatments for TTP [4], plasmapheresis remains the cornerstone of current management of iTTP. In this study, three patients did not receive plasmapheresis treatment all died. 90.3% (28/31) of patients underwent plasmapheresis; of them, 25% (7/28) still succumbed to their illness. TTP patients with AIRDs were recommended to receive appropriate treatments for underlying AIRDs, in addition to standard TTP therapies [4]. Immune suppression therapy in AIRD-TTP patients not only targets antibody production to promote the recovery of ADAMTS13 level, but also treats underlying AIRDs. All TTP patients were treated with corticosteroid in this study. Corticosteroid pulse therapy and cyclophosphamide were administered more frequently in survivors

(33.3% vs 10%, 38.1% vs 10%), but differences were not statistically significant, which is likely due to the limited sample size. Rituximab is a monoclonal antibody against CD20, which had been confirmed in several researches to improve the outcome of TTP, especially in refractory cases [29, 30]. In this study, three TTP patients who received rituximab intravenous injection all survived.

Although ADAMTS13 activity defines TTP ultimately, it is a consensus that timely clinical assessment and prompt treatment are critical for TTP patients as the mortality before the era of plasmapheresis reached 90%. Severe thrombocytopenia is a prominent and persistent sign of TTP [13]. In the setting of underlying autoimmune disease, overlapping features may relax a physician's vigilance for TTP diagnosis. For instance, severe thrombocytopenia in SLE is a not a rare manifestation, which was strongly associated with other manifestations such as neurologic disorder, hemolysis, and renal injury. TTP in AIRD patients may be wrongly attributed to AIRDs flare and delay the prompt treatment [11].

PLASMIC score [31] and French score [20] have been derived to help with rapid clinical diagnosis of TTP. Notably, verification of MAHA is a prerequisite for these two diagnostic models. Fragmented red blood cells on peripheral smear (schistocytes) and negative Coombs test are key points of MAHA [32]. However, schistocytes on peripheral smear is not often readily available and remains subjective [32]. Positive Coombs test was reported in 22.5% of cases of TTP [16]. Similarly, Coombs test positivity was found in 17.2% of AIRD-TTP patients, supporting that MAHA should not be excluded by positive Coombs test completely. Hence, ambiguity of MAHA in AIRD-TTP limits the application of PLASMIC score or French score, and more evidence is still required to determine the predictors which can rapidly assess TTP in AIRD patients with severe thrombocytopenia. Li et al. suggested that when new kidney and neurological symptoms appeared in children with SLE, assessment of TTP should be initiated [17]. For adult patients with SLE, renal involvement increased the risk for TTP [12]. However, TTP patients in these studies were diagnosed based on clinical judgment, lacking of confirmatory tests of ADAMTS13 activity. In our study, AIRD-TTP patients suffered from more severe thrombocytopenia and anemia than non-TTP group. Neurologic disorders ranging from dizziness to coma were still prominent symptoms for AIRD-TTP patients. Isolated proteinuria/hematuria was the most frequent renal disorder in TTP [33]. We also found that proteinuria and hematuria were prevalent in AIRD-TTP (50% and 75%), whereas elevated serum creatinine was observed only in a quarter of these cases. It had been raised that thrombocytopenia and elevated D-Dimers require exclusion of DIC from TTP [34]. However, raised D-Dimer was seen in 96.6% of AIRD-TTP patients without typical signs of DIC. Abundant micro-thrombosis formation may contribute to increased D-Dimer concentrations. In summary, clinical suspicion of TTP should be aroused in AIRD patients with new-onset severe thrombocytopenia, neurologic symptoms, anti-SSB positivity, aPL negativity, proteinuria or hematuria. IBIL and LDH were excellent predictors for a rapid assessment of the risks of TTP in AIRD patients with severe thrombocytopenia. Hb, cTnI and D-Dimer level also had a potential value to distinguish TTP from AIRDs. Repeat blood smear and ADAMTS13 measurement should be performed to confirm the suspicion of TTP. More importantly, empiric therapy for TTP should be initiated immediately to reduce the risk of death.

The strength of our study is that all TTP cases were confirmed by severe ADAMTS13 deficiency. Nonetheless, there are several limitations. The first one is the retrospective study design, which was an inevitable issue because of the rarity of AIRD-TTP. Second, our hospital is a tertiary referral center in central China, receiving critically patients referred from many other hospitals. Thus, mortality rate of AIRD-TTP in current study might be overestimated. Third, small number of AIRD-TTP patients did not allow determination of independent factors linked to early death, as well as independent factors related to rapid identification of TTP in AIRDs.

In conclusion, AIRD-TTP is a rare emergency with a high mortality rate. Extremely severe thrombocytopenia, higher LDH level, proteinuria, cardiac injury, and coma on admission were related to early death of AIRD-TTP, and more intensive therapies should be considered. Clinical suspicion of TTP should be aroused in AIRD patients with new-onset severe thrombocytopenia, neurologic symptoms, anti-SSB positivity, aPL negativity, proteinuria or hematuria. IBIL, LDH, Hb, cTnI, and D-Dimer were predictors of TTP development in adult AIRD patients with severe thrombocytopenia, which may improve the accuracy of clinical assessment before ADAMTS13 activity testing.

Abbreviations

TTP: Thrombotic thrombocytopenic purpura; AIRD: Underlying autoimmune related disease; MAHA: Microangiopathic hemolytic anemia; ADAMTS13: A disintegrin-like and metalloproteinase with thrombospondin type 1 motif, member 13; VWF: Von willebrand factor; SLE: Systemic lupus erythematosus; SS: Sjögren's syndrome; IIM: Idiopathic inflammatory myopathies; SSc: Systemic sclerosis; DIC: Disseminated intravascular coagulation; CTD: Connective tissue disease; IQR: Interquartile range; ROC: Receiver operating characteristic; AUC: Area under the curve; CI: Confidence intervals; ANA: Antinuclear antibodies; aPL: Antiphospholipid antibodies; PLT: Platelet; Hb: Hemoglobin; LDH: Lactate dehydrogenase; IBIL: Indirect bilirubin; Scr: Serum creatinine; BUN: Blood urea nitrogen; cTnI: Cardiac troponin I; ESR: Erythrocyte sedimentation rate.

Declarations

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Not applicable.

Authors’ contributions

LD and JZ conceived and designed the study. LZ, GS and LL contributed to the data collection. LZ and GS analyzed the data and drafted the paper. LZ, GS, JZ and LD edited and revised the paper. All authors approved the final manuscript.

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Availability of data and materials

The datasets supporting the conclusions of this article are included within the article and its additional file.

Ethics approval and consent to participate

This study was approved by the Institutional Ethics Committee of Tongji Hospital (TJ-IRB20210853). Written informed consent was exempted due to the retrospective nature of this study.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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Figures

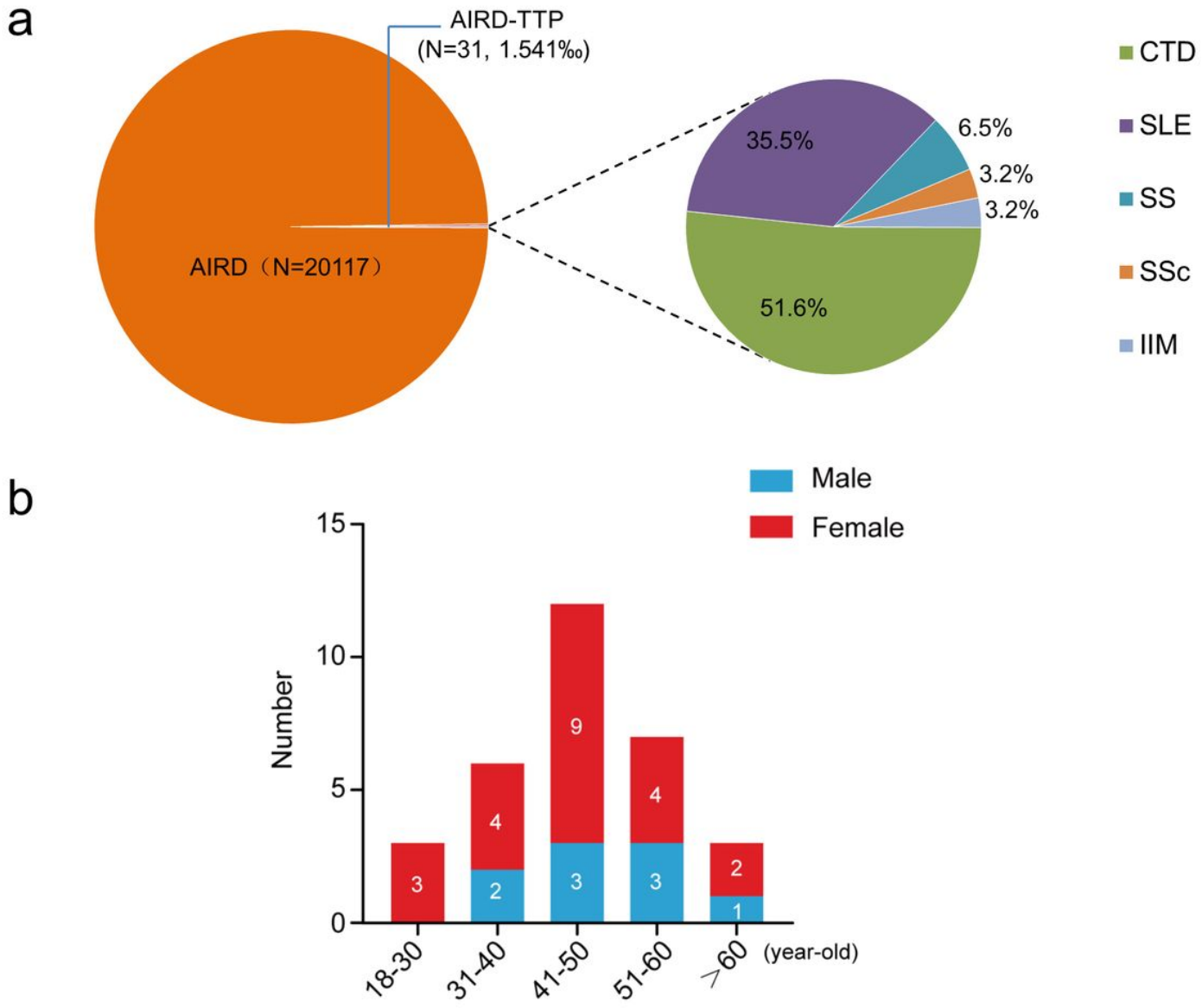
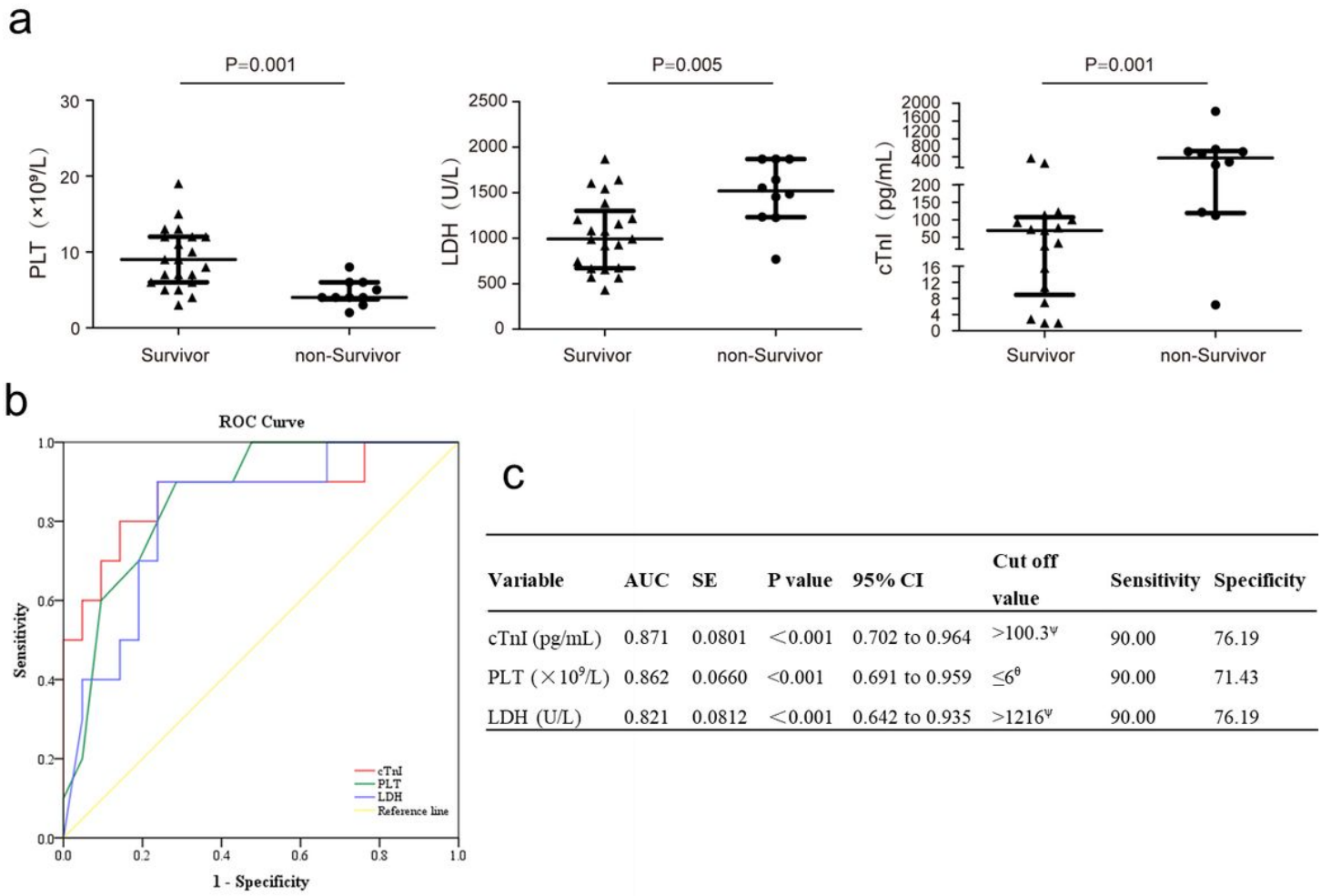


Figure 1

Prevalence and demographic characteristics of AIRD patients with TTP. a The prevalence rate of TTP among 20117 hospitalized AIRD patients. **b** Age and sex distributions of TTP-AIRD.



c

Variable	AUC	SE	P value	95% CI	Cut off value	Sensitivity	Specificity
cTnI (pg/mL)	0.871	0.0801	<0.001	0.702 to 0.964	>100.3$^{\Psi}$	90.00	76.19
PLT (×10^9/L)	0.862	0.0660	<0.001	0.691 to 0.959	≤6$^{\theta}$	90.00	71.43
LDH (U/L)	0.821	0.0812	<0.001	0.642 to 0.935	>1216$^{\Psi}$	90.00	76.19

Figure 2

Predictors for early death in AIRD-TTP patients. a Scatter plots of levels of LDH, levels of cTnI and platelet counts. Data of PLT and LDH were from 21 survivors and 10 non-survivors. Data of cTnI were obtained from 10 survivors and 17 non-survivors. **b-c** ROC of predictors for early death. $^{\Psi}$ larger values than the cut-off indicated stronger evidence for death; $^{\theta}$ lower values than the cut-off indicated stronger evidence for death.

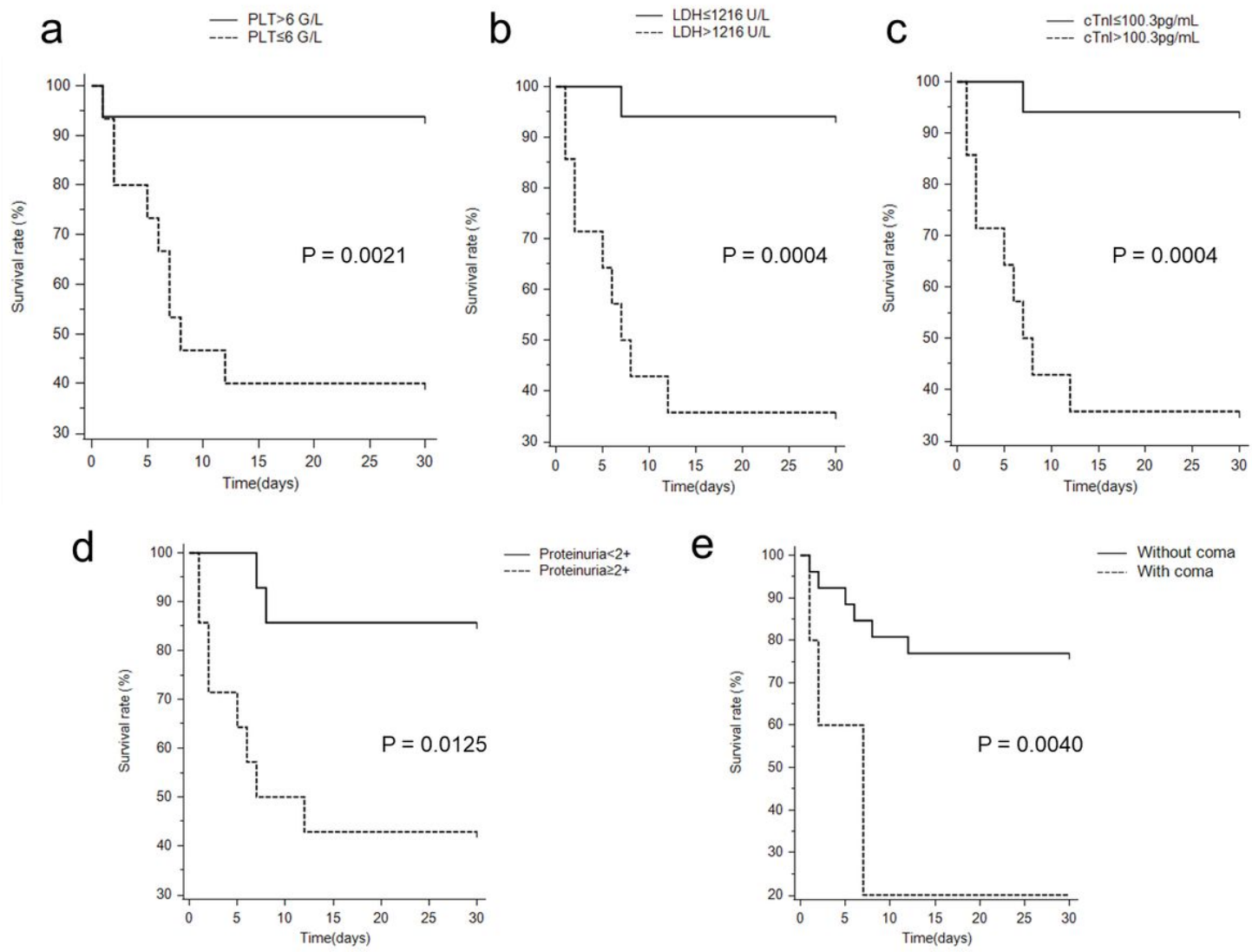


Figure 3

Kaplan-Meier survival curves for death within 30 days in AIRD-TTP patients with different clinical characteristics. a 30-day mortality in AIRD-TTP patients with and without PLT $\leq 6\times10^9$/L. **b** 30-day mortality in AIRD-TTP patients with and without LDH >1216 U/L. **c** 30-day mortality in AIRD-TTP patients with and without cTnI >100.3 pg/mL. **d** 30-day mortality in AIRD-TTP patients with and without proteinuria ≥2+. **e** 30-day mortality in AIRD-TTP patients with and without coma on admission.

Figure 4

Predictors for TTP occurrence in AIRD patients. **a** Scatter plots of laboratory tests. Data are shown for patients with TTP (n=31) and non-TTP patients (n=120) unless otherwise stated. Data of ESR were obtained from 17 TTP patients and 89 non-TTP patients; data on D-Dimer were from 29 TTP patients and 115 non-TTP patients; data on cTnI were from 27 TTP patients and 82 non-TTP patients. **b-c** ROC of

laboratory tests. $^{\Psi}$ larger values than the cut-off indicated stronger evidence for TTP; $^{\theta}$ lower values than the cut-off indicated stronger evidence for TTP.

Supplementary Files

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- SupplementaryTable1.docx