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Changes in platelet levels and prognosis in patients with acute liver failure and late-onset hepatic failure

Manabu Hayashi, MD, PhD[a,*], Masashi Fujita, MD[a], Kazumichi Abe, MD, PhD[a], Atsushi Takahashi, MD, PhD[a], Hiromasa Ohira, MD, PhD[a]

Abstract

The therapeutic strategies for acute liver failure (ALF) and late-onset hepatic failure (LOHF) still have room for improvement. Recent studies have reported an association between platelets and the pathophysiology of ALF. In this study, we investigated changes in platelet levels and clinical findings in ALF and LOHF patients. We retrospectively investigated the clinical data of 62 patients with ALF and LOHF. We analyzed the association between changes in platelet levels for 7 days after admission and the prognosis in patients with ALF and LOHF. The factors associated with changes in platelet levels were also analyzed. The platelet levels on days 1, 3, and 7 were significantly lower in the patients who died or underwent liver transplantation than in the spontaneous survivors. Administration of recombinant thrombomodulin was associated with spontaneous survival. The platelet levels in patients who met the King's College Hospital Criteria or the Japanese scoring system (JSS) for ALF ≥ 4 were significantly decreased 7 days after admission. The area under the receiver operating characteristic curve (AUROC) of a JSS score of 3 for predicting low platelet levels on day 7 was 0.903. Decreased platelet levels were associated with poor prognosis in patients with ALF and LOHF. The patients with low platelet levels and JSS scores on admission showed a high AUROC for predicting low platelet levels on day 7. Decreased platelet levels after admission may be a simple prognostic marker in ALF and LOHF patients.

Abbreviations: ALF = acute liver failure, ALT = alanine aminotransferase, AST = aspartate aminotransferase, DIC = disseminated intravascular coagulation, GGT = gamma-glutamyl transferase, JAAM = the Japanese Association for Acute Medicine, JSS = Japanese scoring system, KCHC = the King's College Hospital Criteria, LT = liver transplantation, LOHF = late-onset hepatic failure, PIs = protease inhibitors, PT = prothrombin time, rTM = recombinant thrombomodulin, SIRS = systemic inflammatory response syndrome, TB = total bilirubin.

Keywords: acute liver failure, late-onset hepatic failure, platelet, recombinant thrombomodulin

1. Introduction

Acute liver failure (ALF) is a clinical syndrome characterized by acute deterioration in liver function due to hepatocyte loss.[1,2] ALF patients develop coagulopathy and hepatic encephalopathy, and the progression of disease causes systemic inflammation and multiorgan failure, resulting in significant morbidity and mortality. Thus, these patients require intensive therapy, including artificial liver support combined with plasma exchange and hemodiafiltration; even with this level of support, mortality is still high. Although liver transplantation (LT) is an effective treatment option for ALF patients, the rate of LT for ALF patients in Japan is not very high. This is due to several reasons, such as a shortage of donors and the high prevalence of comorbidities in ALF patients.[3] Spontaneous survivors of ALF who are treated conservatively have a good prognosis compared with ALF patients who undergo liver transplantation.[4] Improvement in the therapeutic strategy for ALF is therefore needed.

In patients with chronic liver disease, low platelet levels are associated with disease severity. Hypersplenism and splenic pooling, as well as decreased thrombopoietin production, contribute to the low platelet levels observed in patients with advanced chronic liver disease.[5] ALF patients often have thrombocytopenia, but the underlying mechanism is poorly understood. Previous reports have suggested that platelet levels are associated with the mechanism of liver injury in patients with ALF. Local tissue injury and inflammatory responses lead to increased infiltration of inflammatory immune cells in the liver, and activated platelets and the coagulation cascade lead to

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All data generated or analyzed during this study are included in this published article [and its supplementary information files]

The study was approved by the Ethics Committee of the Fukushima Medical University School of Medicine and was performed in accordance with the Declaration of Helsinki. The need to obtain informed consent from the participants was waived by the Ethics Committee of Fukushima Medical University because of the retrospective nature of this study.

[a] Department of Gastroenterology, Fukushima Medical University, Fukushima, Japan.

**Correspondence: Manabu Hayashi, Department of Gastroenterology, Fukushima Medical University School of Medicine, 1 Hikarigaoka, Fukushima City, Fukushima, 960-1295, Japan (e-mail: m884884@fmu.ac.jp).*

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microcirculatory failure and thrombosis.[2,6] Thrombocytopenia is one of the main features of disseminated intravascular coagulation (DIC), and DIC-like coagulopathy can develop in ALF patients.[3] Increased levels of microparticles from platelets have been associated with systemic inflammatory response syndrome (SIRS), complications, and prognosis in ALF patients.[7] ALF patients with poor prognosis have thrombocytopenia after admission.[8] The factors associated with changes in the platelet levels of patients with ALF and LOHF are unknown.

In this study, we analyzed the association between changes in platelet levels for 7 days after admission and prognosis in patients with ALF and LOHF. The factors associated with changes in platelet levels and potential therapeutic options for patients with ALF and LOHF were also analyzed.

2. Materials and methods

2.1. Subjects

In this retrospective single-center study, we analyzed ALF patients who were treated between 2003 and 2020 at the Department of Gastroenterology at Fukushima Medical University Hospital. We retrospectively collected and reviewed the patients' clinical data. ALF and LOHF were diagnosed based on the guidelines of the 2011 Japanese Acute Hepatic Failure Study Group.[9] Briefly, patients showing prothrombin time (PT) activity ≤ 40% or PT-INRs ≥ 1.5 due to severe liver damage within 8 weeks of the onset of disease symptoms were diagnosed as ALF. Patients showing PT activity ≤ 40% or PT-INRs ≥ 1.5 and grade II or more severe hepatic coma between 8 and 24 weeks of the onset of disease symptoms were diagnosed as LOHF. Treatments for ALF and LOHF, such as glucocorticoid administration and artificial liver support (plasma exchange and/or hemodiafiltration), were also analyzed. The King's College Hospital Criteria (KCHC),[10] Japanese scoring system (JSS) for ALF,[11] and MELD score[12] on admission were used. We also investigated prognosis, spontaneous survivors, survivors with liver transplantation, and deaths. Spontaneous survivors were defined as survivors who did not undergo liver transplantation. The platelet levels on admission and on days 3 and 7 were investigated. We investigated DIC-like coagulopathy in ALF and LOHF patients. We defined "DIC-like coagulopathy" as coagulopathy fulfilling the Japanese Association for Acute Medicine (JAAM) criteria on admission in this study.[13] We investigated treatment with protease inhibitors (PIs) and recombinant thrombomodulin (rTM) for DIC-like coagulopathy in the ALF and LOHF patients. Gabexate mesylate, nafamostat mesylate, and ulinastatin therapy was regarded as PI therapy. The median platelet level was used as the cutoff value for distinguishing high and low platelet levels in this study.

The study was approved by the Ethics Committee of the Fukushima Medical University School of Medicine and was performed in accordance with the Declaration of Helsinki. The need to obtain informed consent from the participants was waived by the Ethics Committee of Fukushima Medical University because of the retrospective nature of this study.

2.2. Statistical analysis

Quantitative variables are presented as the median and interquartile range. Differences in the quantitative variables between the groups were compared by the Mann–Whitney *U* test. Differences in the categorical variables were determined using Fisher's exact test or the chi-squared test. Correlations between platelet levels and laboratory data were assessed by Spearman's rank correlation test. The association between platelet levels on day 7 after admission and clinical data was analyzed using multivariate linear regression analysis. Multivariate logistic regression analyses were used to assess the predictors of prognosis in patients with ALF and LOHF. Survival curves were constructed using the Kaplan–Meier method, and the prognosis of patients with ALF and LOHF was analyzed using the log–rank test. Differences were considered statistically significant at $P < .05$. Data analyses were carried out using GraphPad Prism 7.0 software (GraphPad Software, La Jolla, CA) and EZR (Saitama Medical Center, Jichi Medical University, Saitama, Japan),[14] a graphical user interface for R software (The R Foundation for Statistical Computing, Vienna, Austria).

3. Results

3.1. Baseline characteristics of patients with ALF and LOHF

The baseline characteristics of the 62 ALF or LOHF patients at the time of admission are shown in Table 1. The median age was 53 years, and 42% of the patients were male. Hepatitis B virus infection (29%) and drug-induced liver injury (29%) were the leading causes of ALF or LOHF, and 16% of the patients had autoimmune hepatitis. There were 30 (48%) ALF patients without coma, 10 ALF patients with acute-type coma, 15 patients with subacute-type coma, and 7 patients with LOHF. The median DIC score based on the JAAM criteria was 3, and 24 patients (38%) were diagnosed at the time of admission. In 62 patients, 31 patients (50%) received PI or rTM therapy during the hospital stay (gabexate mesylate; n = 21. Nafamostat mesylate; n = 3. Ulinastatin; n = 1. rTM; n = 6). There were 32 patients who were spontaneous survivors, 8 patients who underwent liver transplantation, and 22 patients who died. During the hospital stay, 4 patients (6%) developed bleeding symptoms (3 had gastrointestinal bleeding, and 1 had alveolar hemorrhage), 12 patients (19%) developed infection, 11 patients (17%) developed renal failure, and 5 patients (8%) developed heart failure. The median platelet level on day 1 was 13.2 [8.2–16.1] × 10^4/μL.

Seven days after admission, 8 patients died (n = 4) or underwent liver transplantation (n = 4). Among 54 patients, the median platelet level on day 7 was 10.8 (6.3–20.2) × 10^4/μL. We compared the baseline characteristics according to the platelet levels on day 7 (Table 1). There were significant differences in aspartate aminotransferase (AST), alanine aminotransferase (ALT), gamma-glutamyl transferase (GGT), and total bilirubin (TB) levels, platelet count, KCHC, JSS for ALF, MELD score, and the rate of patients with coma grade ≥ II.

3.2. Changes in platelet levels and prognosis in patients with ALF and LOHF

We analyzed the association between the platelet levels on day 1 and day 7. There were no significant differences between the median platelet levels on day 1 and day 7 in patients with ALF and LOHF ($P = .325$) (Fig. 1A). The platelet levels on day 1 were positively correlated with the platelet levels on day 7 (Fig. 1B). There were no significant differences between the changes in platelet levels within 7 days in the patients with low platelet levels and those of the patients with high platelet levels on day 1 (high platelet on day 1: -1.1 [-8.8-6.7] × 10^4/μL, low platelet on day 1: -1.1 [-4.0-1.9] × 10^4/μL, $P = .325$) (Fig. 1C). In 32 patients with low platelet levels on day 1, 6 patients died or underwent liver transplantation (18%), 18 patients had low platelet levels on day 7 (56%), and 7 patients had high platelet levels on day 7 (21%). In 32 patients with high platelet levels on day 1, 2 patients died or underwent liver transplantation (6%), 9 patients had low platelet levels on day 7 (28%), and 20 patients had high platelet levels on day 7 (62%) ($P = .003$) (Fig. 1D).

We analyzed the platelet levels and prognosis in patients with ALF and LOHF. The platelet levels on days 1, 3, and 7 were

Table 1

Baseline characteristics of the ALF/LOHF patients.

Variables	All patients (n = 62)	Survival for more than 7 days (n = 54)	High platelet on d 7 (n = 27)	Low platelet on d 7 (n = 27)	*P*
Age, yrs	54 (37–65)	54 (37–65)	50 (35–65)	56 (49–65)	.216
Sex, male; n (%)	26 (42%)	24 (44%)	14 (51%)	10 (37%)	.412
HAV/HBV/AIH/ DILI/Others	5/18/10/18/11	5/18/10/18/11	5/4/5/7/6	0/9/4/10/4	.093
AST, U/L	1531 (386–5627)	1392 (365–3725)	1898 (1233–6245)	519 (188–1994)	.003
ALT, U/L	1726 (537–3946)	1568 (497–3710)	2349 (1516–4478)	787 (352–2048)	.001
ALP, U/L	480 (346–654)	474 (347–651)	468 (355–536)	480 (325–666)	.653
GGT, U/L	174 (78–305)	178 (79–324)	238 (145–392)	129 (74–204)	.007
Total bilirubin, mg/dL	10.2 (4.3–19.2)	10.6 (4.3–19.9)	7.2 (3.3–15.2)	15.3 (8.9–21.4)	.043
D/T ratio	0.66 (0.59–0.72)	0.66 (0.59–0.71)	0.66 (0.61–0.74)	0.66 (0.56–0.70)	.197
Albumin, g/dL	3.3 (2.8–3.5)	3.2 (2.8–3.5)	3.2 (2.8–3.5)	3.2 (2.8–3.4)	.879
Prothrombin time, %	35.1 (25.5–40.7)	37 (29–41)	39 (34–41)	32 (22–40)	.096
Prothrombin time, INR	1.81 (81.61–2.48)	1.77 (1.59–2.24)	1.71 (1.58–1.91)	1.94 (1.61–2.69)	.158
Platelets, $\times 10^4/\mu L$	13.2 (8.2–16.1)	13.5 (9.3–16.8)	15.6 (13.0–19.0)	10.0 (6.7–14.2)	<.001
FDP, μg/dL (N = 52)	11.3 (4.3–20.4)	11.2 (3.7–20.4)	11.7 (5.1–17.9)	10.1 (3.7–20.4)	.826
Fibrinogen, mg/dL (N = 51)	153 (108–219)	152 (113–211)	180 (127–239)	132 (94–158)	.004
Creatinine, mg/dL	0.76 (0.55–1.18)	0.75 (0.51–1.06)	0.75 (0.53–0.85)	0.76 (0.49–1.25)	.467
Without coma/Acute type /Subacute type/LOHF	30/10/15/7	30/4/13/7	24/0/2/1	6/4/11/6	<.001
JAAM score ≥ 4, n (%)	24 (38%)	18 (33%)	6 (22%)	12 (44%)	.148
KCHC met, n (%)	22 (35%)	18 (33%)	3 (12%)	15 (62%)	.001
JSS for ALF ≥ 4, n (%)	29 (46%)	26 (48%)	4 (16%)	22 (91%)	<.001
MELD score	21 (15-26)	20 (15-24)	17 (14-21)	22 (17-28)	.012
Treatment					
Glucocorticoid	49 (79%)	45 (83%)	23 (85%)	22 (81%)	1
Artificial liver support	35 (56%)	28 (51%)	11	17	.173
Treatment for DIC	31 (50%)	26 (48%)	9	17	.056
PI/rTM	25/6	21/5	8/1	13/4	

Quantitative variables are presented as the median and interquartile range.

ALP = alkaline phosphatase, AST = aspartate aminotransferase, DIC = disseminated intravascular coagulation, DILI = drug-induced liver injury, D/T ratio = direct/indirect bilirubin ratio, FDP = fibrin/fibrinogen degradation products, GGT = γ-glutamyl transpeptidase, HAV = hepatitis A virus, HBV = hepatitis B virus, INR = international normalized ratio, JAAM = Japanese association for acute medicine, JSS = Japanese scoring system, KCHC = King's College Hospital Criteria, LOHF = late-onset hepatic failure, MELD = Model for End-Stage Liver Disease, PI = protease inhibitor, rTM = recombinant thrombomodulin.

significantly lower in the patients who died or underwent liver transplantation than in the spontaneous survivors (Fig. 1E). The rate of spontaneous survival in the patients with low platelet levels on day 1 and day 7 was significantly lower than that in the other patients (Fig. 1F and G). We analyzed the rate of spontaneous survival in the 4 groups according to the platelet levels on day 1 and day 7 (A: High platelet levels on day 1 and day 7. B: Low platelet levels on day 1 and high platelet levels on day 7. C: High platelet levels on day 1 and low platelet levels on day 7. D: Low platelet levels on day 1 and day 7). This analysis revealed that the patients with high platelet levels on day 7 (Group A and B) showed a high rate of spontaneous survival even though their platelet levels were low on day 1 (Group C and D) ($P = .003$) (Fig. 1H). In multivariate logistic analysis including potential therapeutic options for ALF, administration of recombinant thrombomodulin was associated with spontaneous survival independent of the development of hepatic coma and low platelet levels on day 7 (odds ratio: 0.003, 95% CI: 0.00004–0.23, $P = .008$) (Table 2).

3.3. Factors associated with platelet levels on day 7 in patients with ALF and LOHF

We analyzed the association between the platelet levels and laboratory data (Fig. 2). PT activity on day 1 was positively correlated with the platelet levels on day 1 ($R = 0.28$, $P = .024$), but TB on day 1 ($R = 0.15$, $P = .22$) and AST on day 1 ($R = -0.01$, $P = .93$) did not show a significant correlation with the platelet levels on day 1. TB ($R = -0.39$, $P = .003$) and AST ($R = 0.47$, $P < .001$) on day 1 were positively correlated with the platelet levels on day 7, but the PT activity on day 1 did not show a significant correlation with the platelet levels on day 7 ($R = 0.26$, $P = .05$).

In multivariate linear regression analysis, TB on day 1 (coefficient: −0.41, $P = .002$), the platelet levels on day 1 (coefficient: 0.58, $P < .001$), and development of coma (coefficient: −5.74, $P = .004$) showed a significant association with the platelet levels on day 7 (Table 3).

We analyzed the association between DIC criteria and the platelet levels in patients with ALF and LOHF (Fig. 3A). The patients with DIC-like coagulopathy on day 1 had lower platelet counts on day 1 (without DIC-like coagulopathy: $14.4 \times 10^4/\mu L$, with DIC-like coagulopathy: $7.5 \times 10^4/\mu L$, $P < .001$) and day 7 (without DIC-like coagulopathy: $12.0 \times 10^4/\mu L$, with DIC-like coagulopathy: $6.6 \times 10^4/\mu L$, $P = .021$) than those of the patients without DIC-like coagulopathy. However, there was no significant association between the presence of DIC-like coagulopathy on day 1 and changes in platelet levels for 7 days (without DIC-like coagulopathy: $-1.7 \times 10^4/\mu L$, with DIC-like coagulopathy: $-0.6 \times 10^4/\mu L$, $P = .539$). Next, we analyzed the association between the prognostic criteria of ALF and the platelet levels in patients with ALF and LOHF. The MELD score on day 1 did not show a significant correlation with the platelet levels on day 1, but the MELD score on day 1 showed a significant correlation with the platelet levels on day 7 and changes in platelet levels (Fig. 3B). There were no significant differences between the platelet levels in the patients who met the KCHC on day 1 and those of the patients who did not meet the KCHC on day 1 (KCHC−: $13.7 \times 10^4/\mu L$, KCHC+: $12.4 \times 10^4/\mu L$, $P = .167$). However, the patients who met the KCHC on day 1 showed significantly lower platelet levels (KCHC−: $14.6 \times 10^4/\mu L$,

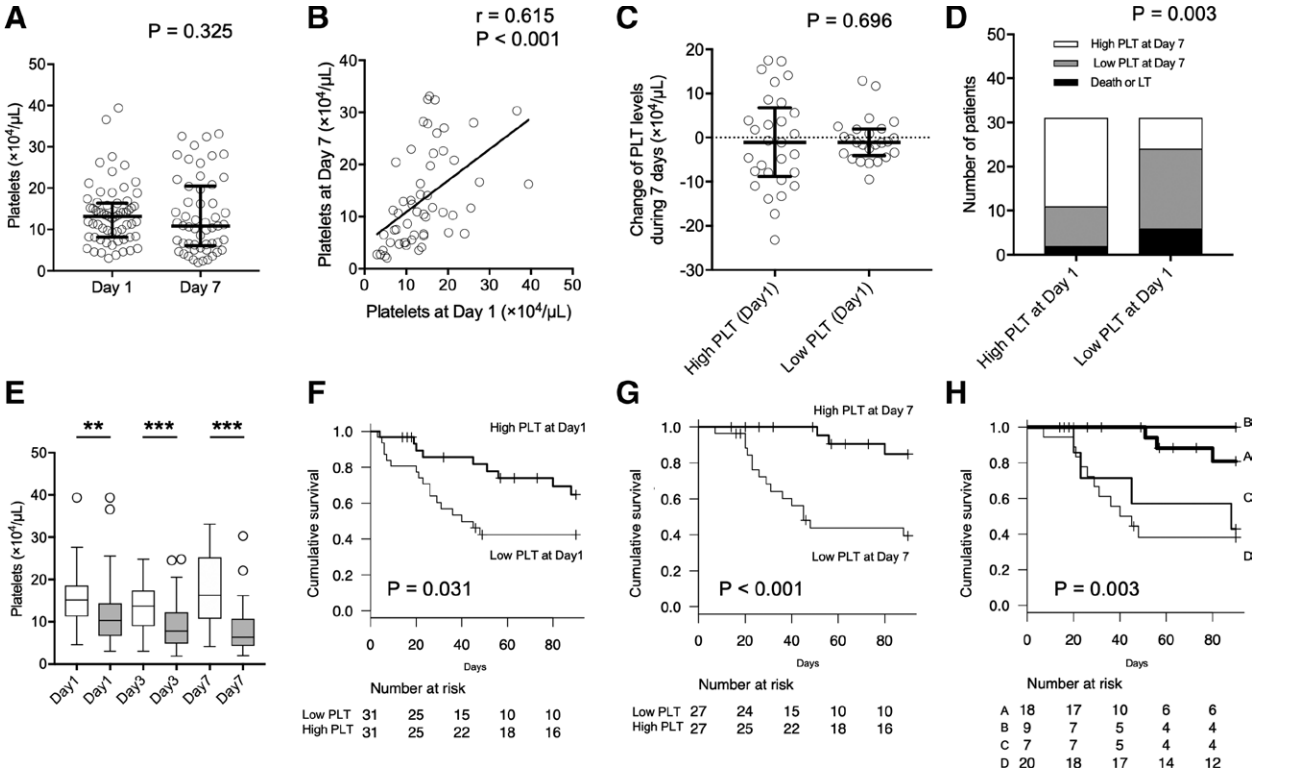

Figure 1. Platelet levels on day 1 and day 7 in patients with ALF and LOHF. (A) Comparison between the platelet levels on day 1 and day 7 (Mann–Whitney *U* test). (B) Correlation between the platelet levels on day 1 and day 7 (Spearman's rank correlation test). (C) Changes in the platelet levels for 7 days according to the platelet levels on admission (Mann–Whitney *U* test). (D) Rate of low platelet levels on day 7 according to the platelet levels on admission (Chi-squared test). (E) Comparison between the platelet levels in patients with spontaneous survival and those with poor prognosis. (White box: spontaneous survivor. Gray box: death or liver transplantation) (Mann–Whitney *U* test). (F) Survival rate according to the platelet levels on admission (log-rank test). (G) Survival rate according to the platelet levels on day 7 (log-rank test). (H) Survival rate according to the platelet levels at days 1 and 7. (A: High platelet levels on day 1 and day 7. B: Low platelet levels on day 1 and high platelet levels on day 7. C: High platelet levels on day 1 and low platelet levels on day 7. D: Low platelet levels on day 1 and day 7) (log-rank test). ALF = acute liver failure, LOHF = late-onset hepatic failure.

Table 2

Therapeutic factors associated with prognosis during the hospital stay.

Variables	Odds ratio	95% CI	P
Development of coma	226	10.4–4930	<.001
High platelet levels on d 7*	0.059	0.003–0.91	.042
Artificial liver support	0.283	0.011–7.03	.441
Glucocorticoid	0.699	0.024–19.8	.833
No DIC treatment	1		
Protease inhibitor	0.629	0.021–18.1	.787
Recombinant thrombomodulin	0.003	0.00004–0.23	.008

CI = confidence interval, DIC = disseminated intravascular coagulation.
*The cutoff value was 10.8 × 10⁴/μL.

KCHC+: 5.2 × 10⁴/μL, *P* < .001) and decreased platelet levels on day 7 (KCHC–: 1.8 × 10⁴/μL, KCHC+: –4.3 × 10⁴/μL, *P* = .008) (Fig. 3C). The patients with JSS for ALF ≥ 4 showed significantly lower platelet levels on day 1 (JSS < 4: 14.2 × 10⁴/μL, JSS ≥ 4: 11.3 × 10⁴/μL, *P* = .043) and day 7 (JSS < 4: 18.1 × 10⁴/μL, JSS ≥ 4: 5.9 × 10⁴/μL, *P* < .001) and decreased platelet levels on day 7 (JSS < 4: 2.8 × 10⁴/μL, JSS ≥ 4: -4.65 × 10⁴/μL, *P* < .001) (Fig. 3D).

3.4. Early predictor of low platelet levels on day 7 in patients with ALF and LOHF

We evaluated clinical parameters on day 1 as early predictors of low platelet levels on day 7 in patients with ALF and LOHF. The AUROCs of the platelet levels on day 1, TB on day 1, MELD score on day 1, and JSS for ALF for predicting low platelet levels on day 7 were 0.768, 0.661, 0.700, and 0.903, respectively (Fig. 4).

4. Discussion

The platelet count was included in the JSS for ALF as a prognostic factor,[11] and low baseline platelet levels have been associated with poor prognosis in patients with hepatitis E virus-related ALF.[15] In this study, low platelet levels on day 7 were associated with poor prognosis in patients with ALF and LOHF. The platelet levels on day 7 were positively correlated with the platelet levels on day 1. However, in the patients with high platelet

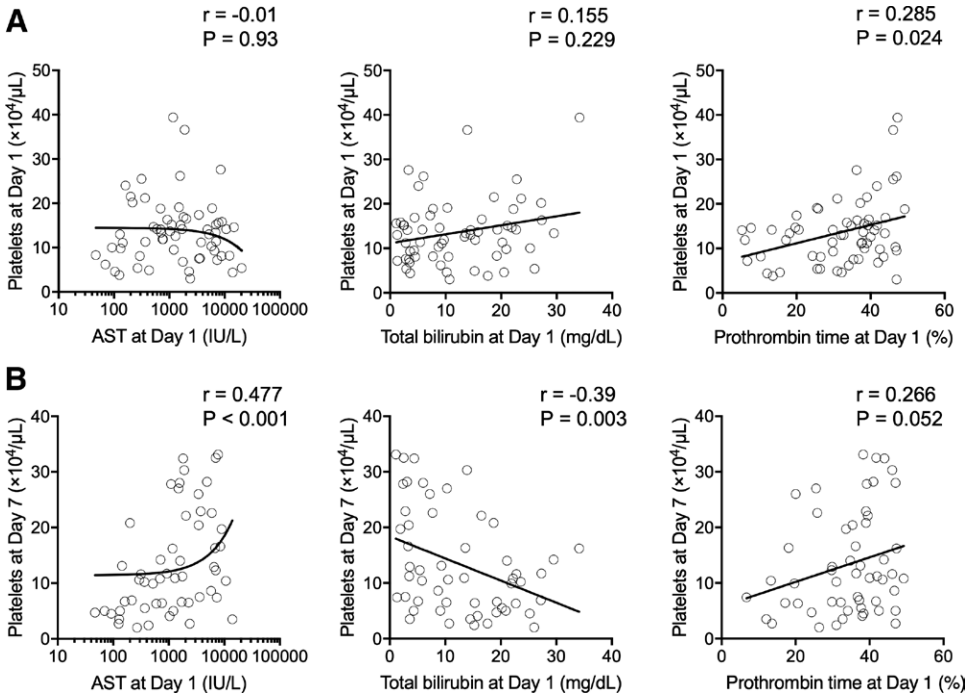

Figure 2. Correlation between platelet levels and laboratory data on admission. (A) Correlation between the platelet levels on day 1 and AST, TB, and PT on day 1. (B) Correlation between the platelet levels on day 7 and AST, TB, and PT on day 1. Spearman's rank correlation test was used. AST = aspartate aminotransferase, PT = prothrombin time, TB = total bilirubin.

Table 3

Factors associated with platelet levels on day 7 after admission.

Variables	Coefficient	95% CI	SE	*P*
Intercept	12.53	2.86–22.19	4.80	.012
AST, 1000 U/L	0.057	–0.69 to 0.80	0.37	.877
Prothrombin time, %	0.038	–0.17 to 0.24	0.10	.715
Total bilirubin, mg/dL	–0.41	–0.67 to –0.15	0.131	.002
Serum creatinine, mg/dL	–0.96	–2.84 to 0.92	0.93	.309
Platelets, ×10⁴/μL	0.58	0.31–0.84	0.13	<.001
Development of coma	–5.74	–9.64 to –1.84	1.93	.004

AST = aspartate aminotransferase, CI = confidence interval, SE = standard error.

levels on day 1, 28% of these patients had low platelet levels on day 7, and these patients had a low survival rate. In other words, patients with low platelet levels on admission showed high 90-day mortality, but patients with low platelet levels on admission and high platelet levels on day 7 showed low 90-day mortality. Our results suggested that a change in the platelet level after admission is a simple prognostic marker in ALF and LOHF patients.

Our results showed that there were no significant differences between the changes in the platelet levels in the patients with low platelet levels on admission and the other patients. High TB levels on day 1 and the development of coma were significantly associated with low platelet levels on day 7. Prognostic criteria and scores for ALF, such as the MELD score, KCHC, and JSS for ALF on admission, were associated with low platelet levels on day 7. In particular, a high JSS score for ALF on admission showed a high AUROC for predicting low platelet levels on day 7. Even if ALF and LOHF patients showed high platelet levels on admission, patients with a high JSS score for ALF showed decreased platelet levels and poor prognosis. The presence of DIC-like coagulopathy was not associated with changes in platelet levels after admission. These results suggested that platelet levels were closely associated with liver dysfunction compared with DIC-like coagulopathy on admission. On the other hand, the association between the platelet levels on day 1 and prognostic criteria on day 1 was not significant or was weak in this study. Decreases in platelet levels may reflect liver dysfunction a few days earlier.

The mechanism leading to the decrease in platelet levels in ALF and LOHF is not fully understood. Low platelet levels were observed in an animal model of ALF with sinusoidal fibrin deposition.[16] This result suggests that the platelet levels decreased due to consumption secondary to coagulation in the liver. Recent reports have suggested that platelet and platelet-derived factors are associated with liver injury. Platelet depletion using an anti-CD41 antibody was found to reduce APAP-induced liver injury in rats.[17] Platelet activation due to C-type lectin-like receptor (CLEC)-2 by podoplanin has been shown to contribute to the sterile inflammatory response involving neutrophils and macrophages.[18] Deficient platelet CLEC-2 or deletion of podoplanin in hematopoietic cells reduces liver injury by APAP or CCl4 in mice.[19] Levels of procoagulant microparticles from platelets have been associated with SIRS and prognosis in patients with ALF.[8] Furthermore, thrombocytopenia has been found to exacerbate cholestasis-induced liver disease via hepatocyte growth factor-Met signaling in mice.[20]

Our analysis suggested that administration of rTM may have a potential therapeutic effect independent of low

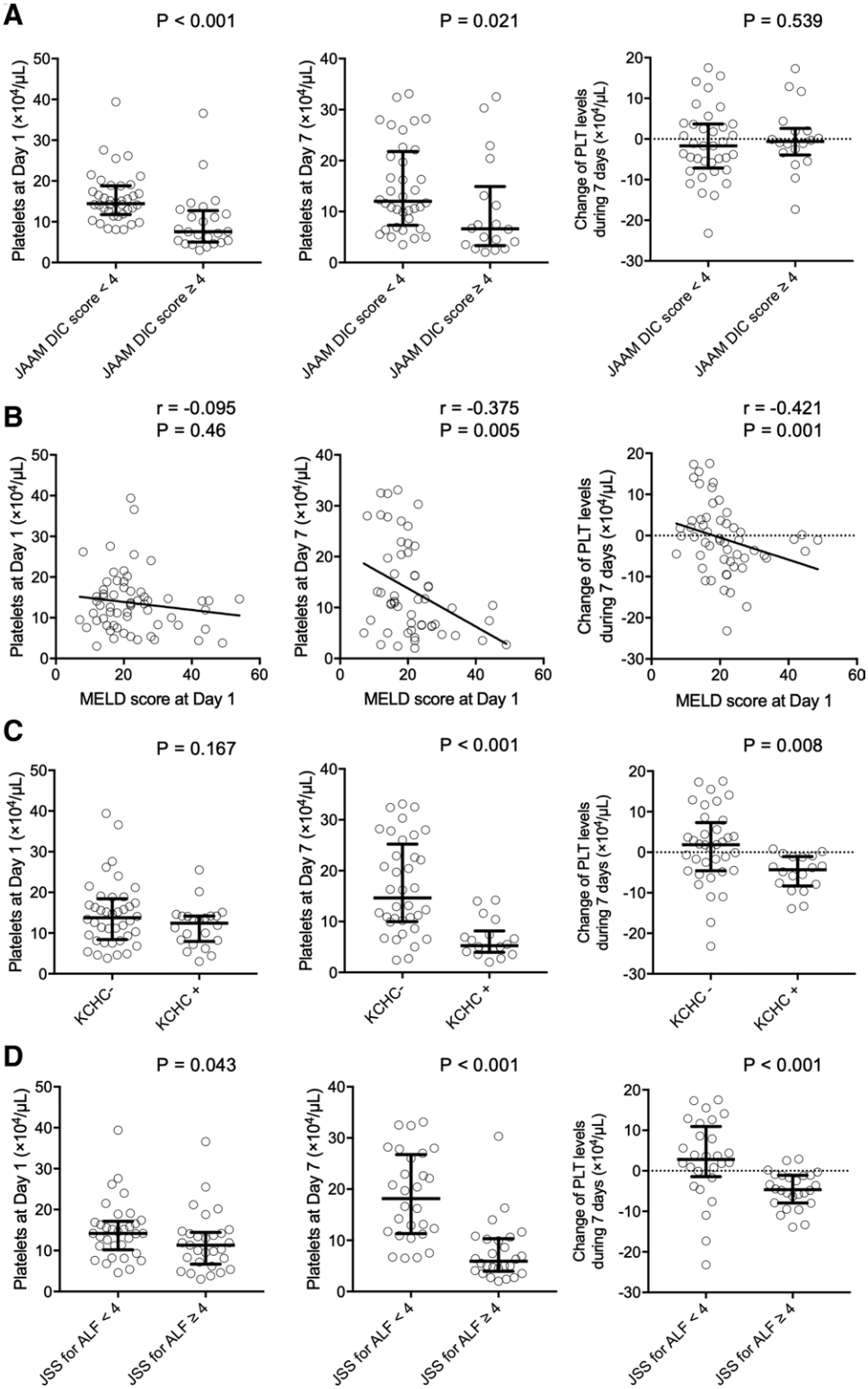

Figure 3. Relationship between platelet levels on day 7, DIC criteria, and prognostic criteria for ALF. Association between platelet levels and (A) disseminated intravascular coagulation-like coagulopathy, (B) model for End-Stage Liver Disease, (C) the King's College Hospital criteria, and (D) the Japanese scoring system for ALF. The Mann–Whitney *U* test and Spearman's rank correlation test were used. ALF = acute liver failure, DIC = disseminated intravascular coagulation.

platelet levels after admission in patients with ALF and LOHF. Thrombomodulin is a receptor of thrombin that is expressed on vascular endothelial cells.[21] Thrombin bound to thrombomodulin impairs platelet aggregation. The administration of rTM activated protein C is the same for the administration of native thrombomodulin. Activated protein C inactivates factor Va and factor VIIIa, resulting in the regulation of thrombin generation and coagulation. In DIC model rats using lipopolysaccharide (LPS) or tissue factor, the peripheral platelet count was decreased, and administration of rTM attenuated impairments

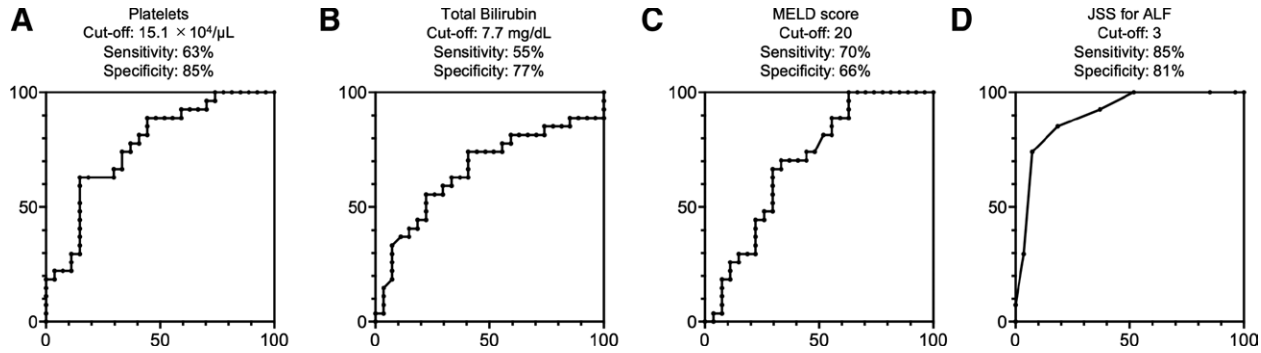

Figure 4. Area under the receiver operating characteristic (AUROC) of potential predictors on admission for predicting low platelet levels on day 7. AUROC of (A) platelet levels on day 1, (B) total bilirubin on day 1, (C) Model for End-Stage Liver Disease score on day 1, and (D) Japanese scoring system for ALF on day 1. ALF = acute liver failure.

in platelet aggregation.[22] Thrombomodulin is associated with the inflammatory response. The C-type lectin domain of thrombomodulin can bind to high-mobility group protein B1, leading to suppression of the lethal inflammatory response due to LPS.[23] The EGF domain of thrombomodulin can recognize CD14 on macrophages and inhibit the CD14-mediated inflammatory response.[24] Administration of rTM can attenuate the inflammatory response and liver injury in a model of acute liver injury.[25,26] Interestingly, suppression of the inflammatory response by thrombomodulin occurs independently of coagulation inhibition.[24] The administration of rTM to ALF patients may suppress the inflammatory pathway by modulating platelet and monocyte activation in the liver.

Our study had several limitations. First, the sample size was small, especially because the number of patients receiving rTM therapy was small. In addition, this was a retrospective and single-center analysis. Treatment initiation and type of therapy were decided by each physician. A large number of prospective studies are needed to elucidate the treatment effect of therapeutic options for patients with ALF. We evaluated DIC-like coagulopathy using JAAM criteria in this study, because we aimed to evaluate the association between coagulopathy diagnosed using JAAM DIC criteria and decreased platelet levels after admission in ALF and LOHF patients. However, ALF and LOHF patients often fulfilled the JAAM criteria, but the diagnosis of DIC in ALF patients is still controversial. Further research is needed for diagnosis of DIC or DIC-like coagulopathy in ALF patients.

In conclusion, changes in platelet levels were associated with prognosis in patients with ALF and LOHF. Poor prognostic criteria for ALF on day 1 were associated with a decrease in platelet levels for 7 days. The JSS score for ALF on day 1 showed a high AUROC for low platelet levels on day 7. Administration of recombinant thrombomodulin may be a therapeutic option for ALF and LOHF patients. Further research is needed to elucidate the association between platelets and the pathophysiology of ALF.

Author contributions

M.H. and H.O. designed the study. M.H. performed the research, wrote the manuscript and prepared the figures. M.H., M.F., K.A., and A.T. acquired and analyzed the data. All authors reviewed the manuscript.

Conceptualization: Manabu Hayashi, Hiromasa Ohira.
Data curation: Manabu Hayashi, Masashi Fujita, Kazumichi Abe, Atsushi Takahashi.
Formal analysis: Manabu Hayashi, Kazumichi Abe.
Investigation: Manabu Hayashi.
Writing – original draft: Manabu Hayashi.
Writing – review & editing: Manabu Hayashi, Hiromasa Ohira.

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