

Changes of histidine-rich glycoprotein levels in critically ill septic patients

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CHANGES OF HISTIDINE-RICH GLYCOPROTEIN LEVELS IN CRITICALLY ILL SEPTIC PATIENTS

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ABSTRACT—Background: Histidine-rich glycoprotein (HRG), a potential prognostic factor in sepsis, lacks clarity regarding its relevance in septic-induced shock, disseminated intravascular coagulation (DIC), and acute respiratory distress syndrome (ARDS) pathogenesis. This study investigated the association between HRG concentrations and these critical conditions. **Methods:** Blood samples were collected from 53 critically ill patients on days 1, 3, 5, and 7 after ICU admission at the Kyushu University Hospital. Daily clinical and laboratory data were recorded, and patient survival was assessed 28 days after ICU admission. **Results:** Serum HRG concentrations were significantly reduced on days 3, 5, and 7 in patients with septic shock and DIC but not in those with ARDS. While initial HRG levels on day one were not correlated with survival, nonsurvivors displayed decreased HRG levels, notably on days 3, 5, and 7 post-ICU admissions. The HRG levels remained stable in survivors. A progressive decrease was associated with higher mortality rates, particularly on days 5 and 7. On day 5, an HRG level with a cutoff of 25.5 µg/mL showed a sensitivity of 0.77 and a specificity of 0.75, indicating significantly lower survival rates (log-rank test, $P < 0.05$). **Conclusion:** HRG presents a potential intervention for critically ill sepsis patients, providing a novel strategy to enhance outcomes. Further research is needed to explore the therapeutic potential of HRG in sepsis management.

KEYWORDS—Sepsis; septic shock; disseminated intravascular coagulation; acute respiratory distress syndrome

INTRODUCTION

Sepsis is a leading cause of global fatalities and critical illnesses (1), contributing to over 20% of worldwide deaths (2). Despite an increase in sepsis cases and deaths, Japan has seen a decrease in sepsis patient mortality over the past 8 years, mirroring a global trend (3). In 2017 alone, sepsis affected over 48.9 million people, causing over 11 million deaths globally (2). Mortality risks associated with septic shock (4), acute respiratory distress syndrome (ARDS) (5), and disseminated intravascular coagulation (DIC) (6) underscore the severity of sepsis-related complications.

Septic shock has a hospital mortality risk of 40–60% (3), while ARDS has a 30–40% mortality rate (5). In Japan, sepsis-induced DIC patients have a mortality rate of 24.8% compared with 17.5% in non-DIC patients (6). Therefore, various therapeutic modalities, such as catecholamines and steroids, have been used to improve the prognosis of patients with septic shock (6). Recombinant human thrombomodulin and plasma-derived or recombinant antithrombin

drugs have been used for DIC and they improve the prognosis of these patients, especially in Japan (7).

Recent research has revealed the critical role of histidine-rich glycoproteins (HRG) during immune and inflammatory responses. This 75 kDa plasma glycoprotein, primarily synthesized in the liver, exerts a pivotal influence on various physiological functions, including coagulation, fibrinolysis, immunity, and inflammation (8,9). With plasma concentrations typically falling between 60 to 150 µg/mL (8,10), HRG emerges as a crucial player in maintaining neutrophil shape, enhancing their ability to engulf bacteria (11), and mitigating neutrophil extracellular traps (NETs) formation (12). Additionally, HRG has been found to regulate vascular endothelial cell activity and restrain the release of HMGB1 into the extracellular space (13,14). Importantly, studies have indicated a correlation between reduced HRG levels in septic conditions and patient survival (15–17), underscoring its urgent clinical relevance in critical illness.

However, the association between HRG concentration and sepsis-related DIC, shock, and ARDS remains unclear. Given the prognostic impact of these conditions on sepsis (3,5,6), our study aimed to investigate changes in HRG concentrations and their relationship with concurrent DIC, ARDS, and septic shock.

MATERIALS AND METHODS

Patients and data collection

This was a single-center retrospective analysis. From June 2019 to October 2021, 65 critically ill patients with sepsis were consecutively admitted to the emergency ICU of Kyushu University Hospital. We excluded patients younger than 20 years of age, patients with cancer in the terminal stages, patients with liver cirrhosis, and individuals who died within 3 days of ICU stay. Ultimately, 53 patients were included, with the primary reasons for ICU admission being septic shock requiring adrenalin >0.1 µg/kg or ARDS necessitating mechanical ventilation. There were 29 patients with septic shock, 25 with DIC, and 33 with ARDS, with some showing overlap between these conditions.

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Septic shock

Sepsis and septic shock were diagnosed according to the criteria outlined in the Third International Consensus Definition for Sepsis and Septic Shock (Sepsis-3) (1). This definition characterizes sepsis as life-threatening organ dysfunction arising from dysregulated host responses to infection (1). Septic shock, a subset of sepsis, is characterized by profound abnormalities in circulatory, cellular, and metabolic functions, leading to a substantial increase in the risk of mortality (1).

DIC

Disseminated intravascular coagulation (DIC) is an acquired syndrome resulting from both infectious and noninfectious triggers, marked by widespread activation of coagulation within the blood vessels (18,19). DIC invariably arises as a complication of one or more underlying clinical conditions. The leading cause of DIC is infection, with other common causes including malignancies, obstetrical complications, and trauma (20). DIC was diagnosed using the Japanese Association for Acute Medicine DIC score (Table Supplementary 1, <http://links.lww.com/SHK/B964>) (21). The Japanese Association for Acute Medicine DIC score consists of platelet count, prothrombin time, fibrin degradation products (FDP) or D-dimer, and systemic inflammatory response syndrome (21). This score was developed for early diagnosis of DIC and has been used in Japan.

ARDS

ARDS was diagnosed according to the Berlin definition. The Berlin definition of ARDS is the presence of symptoms or worsening of the respiratory system within 1 week, with diffuse bilateral opacities on chest radiographs, which are not attributed to congestive heart failure or intravascular volume overload. The Berlin definition categorizes ARDS as mild ($200\text{ mmHg} < \text{PaO}_2/\text{FiO}_2 \leq 300\text{ mmHg}$), moderate ($100\text{ mmHg} < \text{PaO}_2/\text{FiO}_2 \leq 200\text{ mmHg}$), and severe ($\text{PaO}_2/\text{FiO}_2 \leq 100\text{ mmHg}$) (22). In the present study, COVID-19 was prevalent; therefore, ARDS in most patients was caused by COVID-19. The diagnosis of COVID-19 was based on positive polymerase chain reaction (PCR) results.

Other definitions

The Sequential Organ Failure Assessment and Acute Physiology and Chronic Evaluation II scores were calculated from the patients' daily clinical and laboratory data obtained throughout their ICU stay. On day 28 postadmission, patients' conditions were assessed to ascertain their survival status.

Sample collection

Blood samples were collected from patients on the first, third, fifth, and seventh days after admission to the ICU at Kyushu University Hospital. Blood samples were obtained from two healthy volunteers for comparative analysis. Samples were collected in both serum and EDTA plasma tubes. Plasma tubes were slowly shaken and centrifuged at $1000 \times g$ for 15 min, whereas serum tubes were gently inverted twice and incubated at room temperature for 45 min to complete the coagulation

process. Subsequently, the same centrifugation conditions as those used for plasma samples were applied. The resulting supernatant was collected and stored at -80°C .

Measurement of HRG

Serum and plasma HRG concentrations were assessed following the manufacturer's protocol using a human enzyme-linked immunosorbent assay (ELISA) kit (Abcam, Cambridge, UK). Diluted blood samples and standards were added to each well and the wells were shaken gently for 2.5 h at room temperature. After four washes with $1 \times$ wash buffer, $1 \times$ biotinylated anti-human HRG detection antibodies were added to each well, and the wells were gently shaken for 1 h at room temperature. Following four additional washes, the HRP-streptavidin concentrate was added to each well and gently shaken for 45 min at room temperature. The wells were then rewashed four times, and TMB Substrate Reagents were added, followed by gentle shaking for 30 min in the dark at room temperature. Stop Solutions were added to each well, and the plates were immediately read at 450 nm using an ELISA microplate reader (Tecan, Männedorf, Switzerland).

Ethics statement

This study was conducted in accordance with the ethical guidelines of the Declaration of Helsinki and International Conference on Harmonization Guidelines for Good Clinical Practice. The study was approved by the Kyushu University Hospital Institutional Review Board (approval number: 21117-00), and informed consent was obtained before data collection.

Statistical analysis

Data are presented as mean $\pm$ standard deviation. The correlation between serum and plasma HRG concentrations was analyzed using the Pearson's correlation matrix and simple linear regression. Pearson's r values >0.70 were considered indicative of a strong correlation. Differences between two groups were assessed using the unpaired Student's t test, while two-way analysis of variance with Dunnett's multiple comparisons test was applied for three or more groups. The Fisher's exact probability test was used to compare the ratios of the two categorical groups. Receiver operating characteristic (ROC) curve analysis was conducted to evaluate HRG concentrations on days 1, 3, 5, and 7 for prognostic prediction. Kaplan-Meier survival curves and log-rank tests were performed by categorizing patients into two groups using cutoff levels obtained from the ROC curve analysis. All two-tailed tests were considered statistically significant at $P < 0.05$. Statistical analyses were conducted using the GraphPad Prism software (GraphPad Prism 9.5.1; GraphPad Software, San Diego, CA).

RESULTS

Patients' characteristics

As outlined in Table 1, the patients had a mean age of 63.0 ± 14.8 years, with 60.4% being men (32 men and 21 women).

TABLE 1. Patients' characteristics

Factor	All, N = 53	Survivor, n = 41	Nonsurvivor, n = 12	P
Age, years	63.6 $\pm$ 14.8	61.9 $\pm$ 15.4	69.4 $\pm$ 10.7	0.125
Male/female, (%)	32/21 (60.4/39.6)	24/17 (58.5/41.5)	8/4 (66.7/33.3)	0.612
Site of infection	53			
- Lung	30	24	6	-
- Abdomen	15	11	4	-
- Urinary tract	3	3	0	-
- Soft tissues	5	3	2	-
SOFA	9.7 $\pm$ 3.9	9.2 $\pm$ 3.7	11.4 $\pm$ 3.9	0.08
APACHE II	27.7 $\pm$ 7.6	26 $\pm$ 7.4	33.5 $\pm$ 4.9	0.002*
Acute DIC score	2.6 $\pm$ 2.5	2.6 $\pm$ 2.7	2.7 $\pm$ 1.7	0.9
Platelet ($\times 10^3$)	167.5 $\pm$ 90.8	171.3 $\pm$ 87.1	154.3 $\pm$ 105.3	0.573
ICU stays, d	14.6 $\pm$ 13.3	15.3 $\pm$ 14.6	12.5 $\pm$ 7.7	0.529
Diagnosis				
- Only septic shock	5	4	1	-
- Septic shock with DIC	15	13	2	-
- Septic shock with DIC and ARDS	9	2	7	-
- ARDS with DIC	1	1	0	-
- Only ARDS	23	21	2	-

Data are reported as the mean $\pm$ SD.

APACHE II, Acute Physiology and Chronic Health Evaluation; ARDS, acute respiratory distress syndrome; DIC, disseminated intravascular coagulation; SOFA, Sequential Organ Failure Assessment.

On day one after admission to the ICU, the mean APACHE II, Sequential Organ Failure Assessment, and acute DIC scores were 27 ± 7.6 , 9 ± 3.9 , and 2 ± 2.5 , respectively. Among these patients, 29 (54.7%) had septic shock, 34 (64.2%) had ARDS, and 25 (47.2%) had DIC. Of the 29 patients with septic shock, 24 had concomitant DIC. Twenty patients with ARDS were diagnosed with coronavirus disease 2019 (COVID-19). In total, 12 patients (22.6%) died within 28 days.

Association between serum and plasma HRG concentrations

We analyzed the plasma and serum HRG concentrations in blood samples from patients and volunteers. Plasma and serum HRG concentrations showed a positive correlation, with a coefficient (r) of 0.9486. ($Y = 0.911X + 4.57$, $P < 0.0001$) (Fig. 1).

Changes in serum HRG concentrations in survivors and nonsurvivors

In the survivors, the average HRG concentrations were 32.5, 28.6, 30.3, and 32.6 $\mu\text{g/mL}$ on days 1, 3, 5, and 7, respectively (Fig. 2). Conversely, in nonsurvivors, the average HRG concentrations were 36.3, 23.3, 19.7, and 20.3 $\mu\text{g/mL}$ on days 1, 3, 5, and 7, respectively. Notably, HRG concentrations in nonsurvivors were significantly lower on days 3 ($P < 0.05$), 5 ($P < 0.01$), and 7 ($P < 0.05$) than on day 1. Furthermore, the HRG concentrations on days 5 and 7 were significantly lower in nonsurvivors than in survivors.

Changes in HRG concentrations in patients with septic shock, ARDS, and DIC

Mean HRG concentrations on days 1, 3, 5, and 7 were 35.6, 25.2, 24.7, and 25 $\mu\text{g/mL}$ in patients with septic shock and 31, 230, 32.6, and 33.1 $\mu\text{g/mL}$ in patients with ARDS. Patients experiencing septic shock exhibited a significant decline in HRG concentrations on days 3, 5, and 7 compared to day 1 (all $P < 0.01$). Moreover, HRG concentrations were significantly lower in patients with septic shock than in those with ARDS on days 5. No significant changes in HRG concentrations were found in patients with ARDS (Fig. 3A).

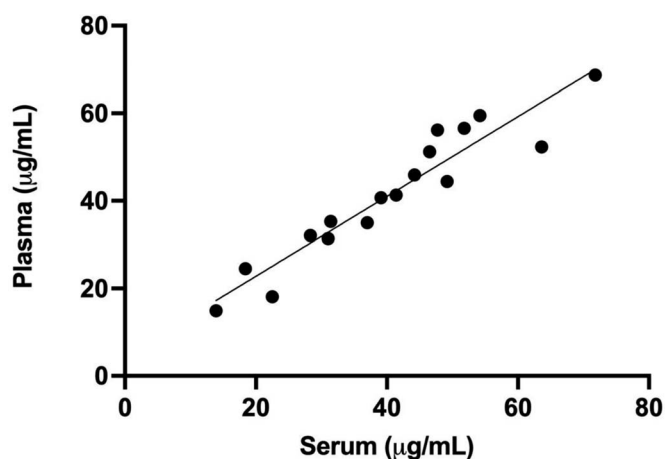


FIG. 1. Correlation between serum HRG concentrations and plasma HRG concentrations. In preliminary experiments, serum and plasma HRG concentrations showed a high positive correlation ($n = 17$, $r = 0.9486$, $P < 0.0001$) in blood samples from two voluntary donors and 15 critically ill patients. HRG, histidine-rich glycoprotein.

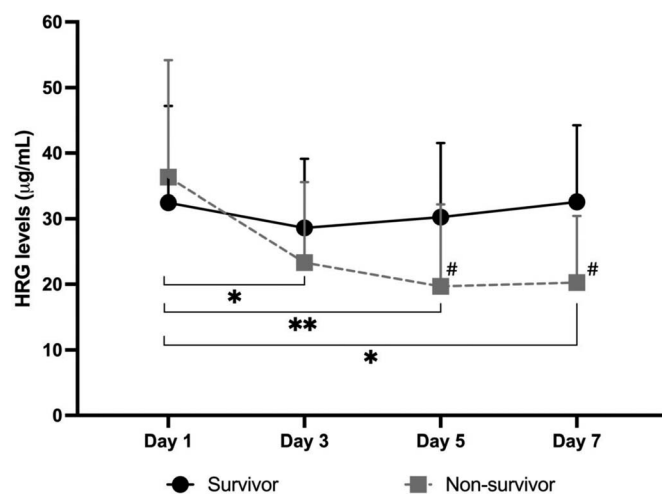


FIG. 2. Differences in HRG concentrations between survivors and nonsurvivors in critically ill patients. In nonsurvivors, the HRG concentrations were significantly lower on days 3, 5, and 7 than on day 1. HRG concentrations were also significantly lower in nonsurvivors ($n = 12$) than in survivors ($n = 41$) on days 5 and 7. Data are presented as mean and standard deviation. * $P < 0.05$ vs. nonsurvivors on days 3 and 7; ** $P < 0.01$ vs. nonsurvivors on day 5; # $P < 0.05$ vs. survivors on days 5 and 7. HRG, histidine-rich glycoprotein.

The mean HRG concentrations were 31.1, 23.3, 22.9, and 22.8 $\mu\text{g/mL}$ in patients with DIC and 35.2, 31.1, 33.3, and 36.1 $\mu\text{g/mL}$ in patients without DIC on days 1, 3, 5, and 7, respectively. Patients with DIC did not exhibit significant changes in HRG concentrations on days 3, 5, and 7 compared with those on day 1. However, patients with DIC consistently showed significantly lower HRG concentrations than those without DIC on days 3, 5, and 7 (Fig. 3B).

HRG concentrations between survivors and nonsurvivors in patients with septic shock, DIC, and ARDS

Mean HRG concentrations were 35.3, 28.0, 27.4, and 29.6 $\mu\text{g/mL}$ in septic shock survivors, while nonsurvivors had concentrations of 35.4, 19.0, 16.6, and 16.4 $\mu\text{g/mL}$ on days 1, 3, 5, and 7, respectively. In nonsurvivors of septic shock, HRG concentrations were notably lower on days 3, 5, and 7 than on day 1. Additionally, there were significant differences in HRG concentrations between nonsurvivors and survivors on days 5 and 7 ($P < 0.05$; Fig. 4A).

Survivors with DIC displayed mean HRG concentrations of 29.5, 26.0, 26.4, and 27.0 $\mu\text{g/mL}$, whereas nonsurvivors displayed concentrations of 28.1, 14.5, 16.6, and 16.4 $\mu\text{g/mL}$ on days 1, 3, 5, and 7, respectively. Although HRG concentrations in nonsurvivors with DIC were lower than those in survivors, significant differences were observed only on days 3 and 5 ($P < 0.05$, Fig. 4B).

In survivors with ARDS, the mean HRG concentrations were 29.3, 28.9, 32.5, and 34.3 $\mu\text{g/mL}$, while nonsurvivors had concentrations of 36.3, 24.2, 20.6, and 20.3 $\mu\text{g/mL}$ on days 1, 3, 5, and 7, respectively. Notably, HRG concentrations were significantly lower on days 5 and 7 in nonsurvivors of ARDS, with no significant differences observed between the time points in survivors of ARDS (Fig. 4C). Additionally, HRG concentrations were significantly lower in nonsurvivors than in survivors on days 5 and 7.

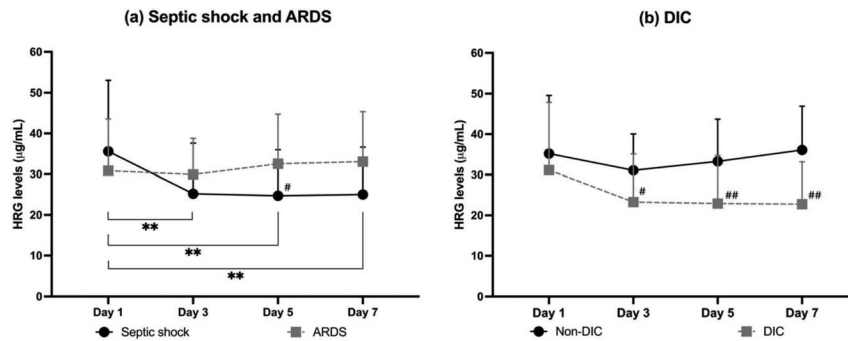


FIG. 3. **Changes in serum HRG concentrations in critically ill patients with Septic shock, ARDS, and DIC.** A, HRG concentrations in patients with septic shock were significantly lower on days 3, 5, and 7 than on day 1 after ICU admission. Patients with septic shock ($n = 27$) had significantly lower HRG concentrations than those with ARDS ($n = 24$) on days 5. No significant difference in HRG concentrations was found in patients with ARDS. B, Patients with DIC ($n = 23$) had lower HRG concentrations than those without DIC ($n = 28$) on days 3, 5, and 7. Data are presented as the mean and standard deviation. ** $P < 0.01$ vs. septic shock on days 3, 5, and 7; # $P < 0.05$ and ## $P < 0.01$ vs. the other group. ARDS, acute respiratory distress syndrome; DIC, disseminated intravascular coagulation; HRG, histidine-rich glycoprotein; ICU, intensive care unit.

Receiver operator characteristic analysis to predict the prognosis of critically ill patients with sepsis

We assessed the predictive value of HRG concentration using ROC curves (Fig. 5A). The areas under the curve for HRG concentrations were 0.566, 0.651, 0.764, and 0.798 on days 1, 3, 5, and 7, respectively. On day 5, HRG concentrations demonstrated a sensitivity of 0.77 and, specificity of 0.75, with a 25.5 $\mu\text{g/mL}$ cutoff level. Similarly, on day 7, the sensitivity and specificity were 0.75 and 0.81, respectively, with a 25.7 $\mu\text{g/mL}$ cutoff level. Since these cutoff levels were very close, Kaplan-Meier analysis was performed using HRG concentrations of 25.5 $\mu\text{g/mL}$ on day 5. Patients with HRG concentrations $<25.5 \mu\text{g/mL}$ exhibited a significantly lower survival curve (log-rank test, $P < 0.05$) (Fig. 5B).

DISCUSSION

In this single-center observational study, we demonstrated alterations in serum HRG concentrations in critically ill patients with sepsis. To our knowledge, only one multicenter observational study has reported changes in plasma HRG concentrations (17). Previous studies on sepsis have primarily assessed HRG concentrations in the plasma (15–17). Our preliminary experiments indicated a robust positive correlation between the serum and plasma HRG concentrations. Given the greater stability and preservability of serum samples than plasma, we conducted measurements using serum samples in this study.

The primary discovery of our study was the changes and differences in HRG levels between survivors and nonsurvivors

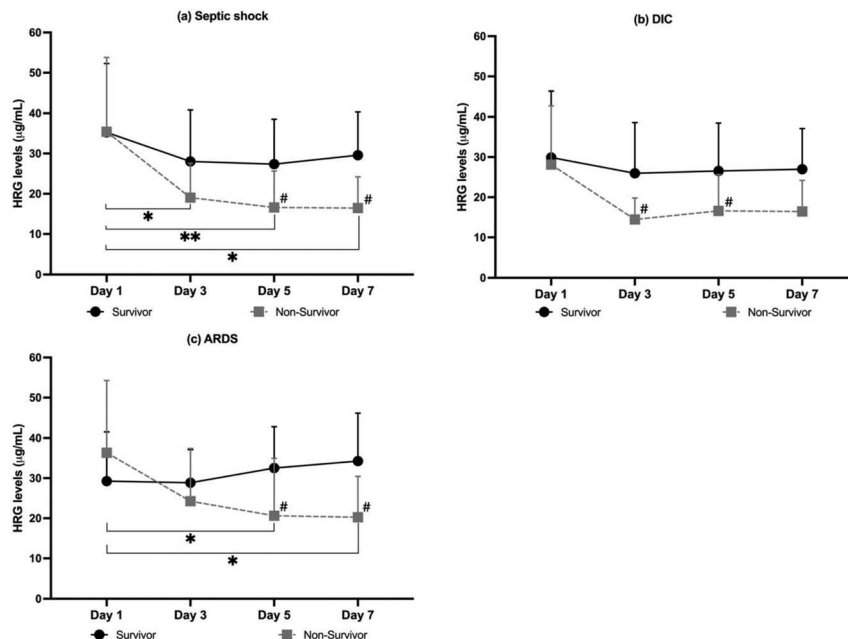


FIG. 4. **Comparison of HRG concentrations between survivors and nonsurvivors in patients with septic shock, DIC, or ARDS.** A, In nonsurvivors with septic shock, HRG concentrations were significantly lower on days 3, 5, and 7 than on day 1 of ICU admission. In patients with septic shock, HRG concentrations were significantly lower in nonsurvivors ($n = 10$) than in survivors ($n = 19$) on days 5 and 7. B, In patients with DIC, nonsurvivors ($n = 6$) had significantly lower HRG concentrations than survivors ($n = 18$) on days 3 and 5. C, In patients with ARDS, HRG concentrations were significantly lower in nonsurvivors ($n = 9$) on days 5 and 7 than on day 1 of ICU admission, while no significant difference was found in survivors ($n = 25$). Data are presented as the mean and standard deviation. * $P < 0.05$ vs. septic shock nonsurvivor on days 3 and 7; * $P < 0.05$ vs. ARDS nonsurvivors on days 5 and 7; ** $P < 0.01$ vs. septic shock nonsurvivors on day 5; # $P < 0.05$ vs. the other group. ARDS, acute respiratory distress syndrome; DIC, disseminated intravascular coagulation; HRG, histidine-rich glycoprotein; ICU, intensive care unit.

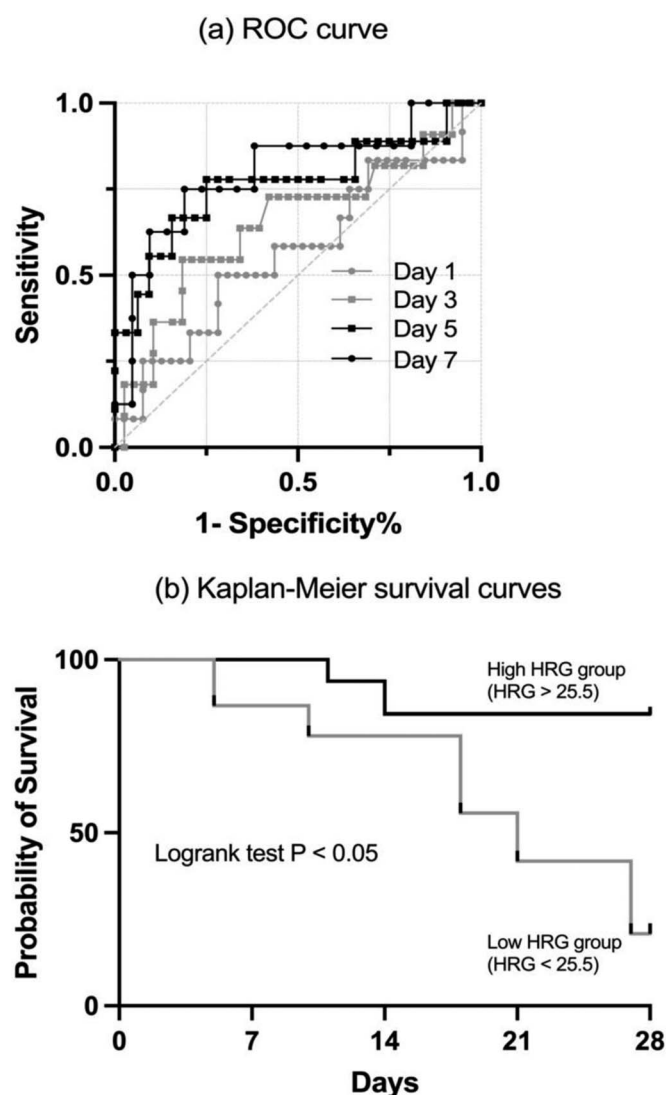


FIG. 5. Receiver operating characteristic curves and Kaplan-Meier survival curves. A, HRG levels at day 1, day 3, day 5, and day 7. The area under the curve is 0.566, 0.651, 0.764, and 0.798, respectively. B, Patients were categorized as low HRG ($n = 16$) or high HRG ($n = 28$) using a 25.5 g/mL cutoff level. There was a significant difference between the high and low HRG groups. HRG, histidine-rich glycoprotein; ROC, receiver operating characteristic.

among critically ill patients with sepsis. In contrast to earlier studies showing significantly lower first-day HRG levels in nonsurvivors than in survivors (16,17), our study revealed slightly higher HRG levels in nonsurvivors on the first day, although the difference was not statistically significant. However, we observed a significant decrease in HRG levels from days 3 to 7, reaching the lowest average on day 5 in the nonsurvivors. Compared to survivors, the HRG levels in nonsurvivors were considerably lower, particularly on days 5 and 7. These findings align with prior research, indicating that HRG levels are consistently lower in nonsurvivors than in survivors (17).

This study was the first to analyze HRG concentrations in patients with septic shock, DIC, and ARDS. As depicted in Figure 3A, HRG levels in septic shock patients demonstrated a significant decrease over time, notably declining compared to HRG levels in nonseptic shock patients on days 5. To determine the HRG levels in patients with sepsis-associated DIC, which is

a major cause of multiple organ failure and an independent predictor of mortality, we analyzed septic shock and DIC separately. As shown in Table 1, most patients with septic shock progressed to DIC, whereas only one patient with initial ARDS developed DIC. This indicates that the results from DIC patients primarily reflect the outcomes of those with septic shock.

Our analysis revealed that patients who developed DIC had significantly lower HRG levels on days 3, 5, and 7 compared to those who did not develop DIC (Fig. 3B). Furthermore, HRG levels were also lower in DIC patients who did not survive (Fig. 4B). These findings suggest a potential association between reduced HRG levels and the progression of septic shock to DIC and poorer outcomes in DIC patients. HRG plays a critical role as an endogenous regulator of blood clotting and has been identified as an anticoagulant that inhibits Factor XIIa, thus preventing its self-activation and subsequent activation of FXI (23). Recent studies in rabbits have shown that HRG depletion increases the risk of catheter thrombosis. These findings suggest that replenishing HRG during conditions like sepsis or cancer might reduce the risk of catheter thrombosis (24,25). However, this potential therapeutic approach requires further investigation, as limited clinical evidence supports it. Furthermore, hereditary thrombophilia resulting from congenital HRG deficiency has been documented as a familial condition (26), warranting additional research into HRG's role in coagulation disorders.

Unlike in patients with septic shock, we did not find a significant decrease in HRG concentration in patients with ARDS (see Fig. 3A). Among the 34 patients diagnosed with ARDS, 20 (58.8%) had COVID-19 as the underlying cause. Of these COVID-19 patients, 18 had only ARDS. One patient had both ARDS and septic shock, while another developed ARDS, septic shock, and DIC. COVID-19 is associated with high coagulopathy, and although coagulopathy may appear similar to DIC, noticeable differences exist (27). Therefore, this result may have been influenced by the bias introduced by the COVID-19 patients. However, there was a notable decline in HRG levels among the nonsurviving ARDS patients (Fig. 4C). Several studies have suggested that a lower HRG concentration is associated with a worse prognosis in COVID-19 (28), severe community-acquired pneumonia (29), and ventilator-associated pneumonia patients (30). In our extended analysis, we compared patients who were extubated within 10 days with those who were not extubated. All nonextubated patients underwent tracheotomy during their ICU stay. Importantly, there was a significant difference between the two groups on days 5 and 7 (Fig. S1, <http://links.lww.com/SHK/B965>, and S2, <http://links.lww.com/SHK/B966>).

HRG has been shown to have several beneficial effects on patients with sepsis. Notably, HRG demonstrates antibacterial activity against both gram-positive bacteria (such as *Enterococcus faecalis* and *Staphylococcus aureus*) and gram-negative bacteria (such as *Escherichia coli* and *Pseudomonas aeruginosa*) (31), as well as systemic *Candida* infections (32). There is also evidence suggesting that HRG may offer protective effects against HIV-1 infection (33).

Additionally, recent research has revealed that HRG plays a crucial role in maintaining the round shape of neutrophils, enhancing phagocytic activity against bacteria (11), and decreasing the formation of neutrophil extracellular traps (NETs) (12). HRG also regulates vascular endothelial cell activity and inhibits the

release of HMGB1 into the extracellular space (13,14). Therefore, maintaining adequate HRG levels is beneficial in patients with sepsis. Given its diverse effects in mitigating the severity of sepsis through various mechanisms, therapeutic supplementation with HRG may be advantageous. To our knowledge, no clinical studies have yet explored the administration of HRG in human patients. However, a number of experimental studies, including those involving animal models, suggest that HRG use may yield promising outcomes in the treatment of sepsis (10–14). Further research is needed to refine supplementation protocols and fully harness HRG's therapeutic potential of HRG in sepsis management.

In summary, the correlation between lower HRG concentrations and adverse outcomes in patients with sepsis suggests their relevance in prognosis. Moreover, because HRG has various effects on mitigating the severity of sepsis, it may become a predictive biomarker and therapeutic drug.

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