Elevated D-Dimer levels correlate with the development of hepatorenal syndrome and a poor outcome in patients with cirrhosis

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ABSTRACT

Objectives: Whether hemostatic status was correlated with the diverse types of acute kidney injury in cirrhotic patients is unclear. The present study aimed to investigate the relationship between hemostatic markers and the diverse types of acute kidney injury (AKI) in liver cirrhosis.

Patients and methods: Cirrhotic patients with consecutive treatment at the First Affiliated Hospital of Qianjiang University, Hangzhou, China were included in this study. Their demographic and clinical data, biochemical parameters and hemostatic markers were assessed to identify risk factors for the development and prognosis of AKI.

Results: A total of 773 cirrhotic patients were included in this cohort. Patients with hepatorenal syndrome (HRS) had significantly higher D-Dimer than those with the other types of AKI. In univariate COX regression, APTT, TT, INR, D-Dimer and Fib were correlated with the development of AKI and HRS in multivariate COX regression. The area under the ROC curve of D-Dimer was 0.755 (95% CI, 0.718–0.793) in predicting the development of AKI, 0.879 (95% CI, 0.791–0.967) in predicting the development of HRS, respectively. D-Dimer was used for diagnosis of HRS with a sensitivity of 87.3% and specificity of 72.9% at the cutoff of 3.7 (mg/L FEU). Survival rates differed significantly between groups by D-Dimer level.

Conclusions: Hemostatic markers were significantly associated with the diverse types of AKI. D-Dimer was an independent risk factor for HRS and correlated with a poor outcome in cirrhotic patients.

Introduction

Liver cirrhosis (LC) is the end stage for a variety of liver diseases, and it often leads to hemostatic alterations, which are very complex and involve every aspect of the hemostatic system, including prothrombotic and antithrombotic changes [1]. It is well known that hemostasis is the result of a tightly controlled balance between prothrombotic and antithrombotic factors, involving primary hemostasis, coagulation, and fibrinolysis [2]. Indeed, owing to the central role of the liver in the hemostatic process, changes in hemostasis are involved in the overall balance in cirrhotic patients, and they are highly individual and depend on LC cause and severity [3]. Hemostatic alterations and their individual imbalances will determine the patients’ thrombotic or hemorrhagic risk, and they are related to severe complications among patients with cirrhosis [4].

Acute kidney injury (AKI) is one of the most frequent complications in patients with cirrhosis. 25%-50% of cirrhotic patients suffering from AKI, and most of them may display poor consequences [5,6]. AKI is classified into three types of prerenal azotemia (PRA), hepatorenal syndrome (HRS) and acute tubular necrosis (ATN) according to the common causes of kidney injury in cirrhotic patients [7]. HRS has the worst prognosis because it arises during severe portal hypertension and circulatory dysfunction. Cirrhotic patients with these conditions have increased procoagulant or bleeding risks [8,9]. Disturbance of the hemostatic system may result in intravascular coagulation which could cause the reduction of renal blood flow [10]. A large number of studies have investigated the relationship between hemostatic disorder and renal failure, and some studies have reported the role of coagulopathy in the development of AKI in surgery or critically ill patients [11–13]. However, few studies have focused on the relationship of hemostatic disorder with the diverse types of AKI seconded to advanced cirrhosis. Different types of AKI have different causes and knowledge regarding the roles of hemostatic markers in the development of a...
different type of AKI in cirrhotic patients is limited [14]. It is hypothesized that hemostatic disorders are involved in the development of AKI and hemostatic markers may respond differently to diverse causes of AKI. The present study investigated the roles of hemostatic markers in the development and prognosis of AKI induced by different causes in advanced cirrhotic patients.

Patients and methods

Study design and patients

This retrospective observational cohort analysis used the database of the First Affiliated Hospital of Medicine School, Zhejiang University. We extracted data derived from the electronic records of hospitalized cases between January 2018 and March 2021, and any patients who had been diagnosed with liver cirrhosis were pooled into a single cohort. Liver cirrhosis was diagnosed based on histology, imaging, laboratory data and clinical assessment [15]. Patients aged 18–80 years who had received consecutive treatment at this hospital were eligible. Exclusion criteria included: recently received anticoagulation and/or antithrombotic therapy or contrast agent administration; previously treated with liver transplantation or with other surgery; liver neoplasms or chronic extrahepatic diseases. Patients with less than three months of follow-up or high data miss rates were also excluded. The Ethics Research Committee of the First Affiliated Hospital of College of Medicine, Zhejiang University, approved this study.

Diagnosis and classification of AKI

The diagnostic criteria and classification of AKI have evolved over the past decades. AKI was defined as an increase in serum creatinine (sCr) at least 0.3 mg/dL (26.5 μmol/L) within 48 h or a percentage increase in sCr at least 50% from baseline according to the International Club of Ascites (ICA) current criteria [16].

The classification of AKI was as follows: PRA was considered when patients experienced fluid losses in the preceding days (because of diarrhea, diuretics, or other causes) together with the absence of other causes of kidney injury, and resolution as indicated by a return of sCr to within 25% of baseline within 48 h after fluid resuscitation [7,17]; HRS was defined as worsening kidney function in cirrhotic patients with ascites that meets the ICA-AKI criteria [18]; No sustained improvement in kidney function after volume expansion with albumin and diuretic withdrawal; the absence of recent exposure to nephrotoxic agents (such as aminoglycosides, nonsteroidal anti-inflammatories, or contrast media); and no evidence of shock or signs of structural kidney disease (proteinuria, hematuria and abnormal renal ultrasonographic findings) [16,19]; ATN is characterized by a sharp decline in glomerular filtration rate accompanied with fluid and electrolyte disturbances [18,20]. There is no gold standard, the recognition of ATN rely on synthesis data from clinical and laboratory studies, including a 50% decrease in estimated glomerular filtration rate (eGFR) or a 0.5 mg/dL (40 μmol/L) increase in sCr from baseline, together with granular casts in urine sediment and Urine–plasma creatinine ratio < 20 or FENa > 2% without diuretics [19,21].

Data collection

We collected data on demographics, cause of liver cirrhosis, medical history and laboratory data from electronic medical records. All data were collected through a manual review of the medical records. In the patient with an available sCr value before admission, the most recent stable value of sCr within the last 3 months before admission was considered as baseline; In the patient without an available sCr before hospitalisation, the sCr on admission was used as baseline [19]. Patients were followed up by clinical record review until death, liver transplant (LT) or the end of follow-up. The occurrence and classification of AKI were monitored based on their clinical and laboratory data. All assays for serum biochemical parameters were routinely performed at the central clinical laboratory of this hospital. Plasma D-Dimer was analyzed on the SYSMEX CS 5100 using the INNOVANCE D-Dimer assay (Siemens Healthcare, Marburg, Germany). D-Dimer values at baseline ranged from 0.19 to 4.4 mg/l FEU. Automated sample redilution extends the measuring range up to 35.2 mg/l FEU. The coefficients of variation of D-dimer determination were 3.2%(within-run) and 5.5%(between-run). The eGFR was calculated based on the Modification of Diet in Renal Disease (MDRD) [22]. The equation of MDRD is as follow:

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eGFR = \frac{186 \times (sCr^{-1.154} \times (age^{-0.203} \times 0.742) \text{ female})}{C3}
\]

Data are expressed as the mean ± SD, median (interquartile range, IQR) or number (%). Student’s t-test and Mann-Whitney U-test were used to comparing continuous variables, and the Chi-Squared test was used to compare categorical variables. A Cox proportion hazard regression was performed to analyze hemostatic markers associated with the development of AKI. In order to avoid multicollinearity among variables (for example, eGFR vs sCr; MELD vs INR and TBil), the potentially most powerful parameters were identified using variance inflation factor statistics. Time-dependent ROC curve analysis was performed to evaluate the accuracy of the hemostatic markers for predicting the development of HRS. The diagnostic performance of the hemostatic markers at the cutoff point based on the Youden index was assessed. Survival rates were calculated by Kaplan-Meier plots, and differences between groups were assessed by the log-rank test. All data analyses were performed by SPSS version 22 and RStudio version 3.6. P-values were two-sided, and values <.05 were considered statistically.
Results

Patient cohort

A total of 1282 patients were preliminarily selected for this study. 368 of them were excluded due to previous treatment with liver or kidney transplantation, liver neoplasms or complications of chronic extrahepatic diseases. 63 patients were discharged from the hospital and could not be contacted for further follow-up. 78 patients had incomplete coagulation function tests. The remaining 773 patients were enrolled in the study. Of these, 108 patients had AKI at admission and 194 of the 665 patients without AKI developed it during follow-up. The overall incidence of AKI was 23.6%. Of the total of 302 patients with AKI, 181 were diagnosed with PRA, 70 with HRS and 51 with ATN (Figure 1).

Characteristics of the patients at baseline

Baseline characteristics of patients with or without AKI are shown in Table 1. There was no significant difference in the proportion of men or liver disease etiology between the non-AKI and AKI groups. Compared to patients without AKI, patients with AKI at admission had significantly higher age, incidences of ascites and hepatic encephalopathy (HE), white blood cell (WBC), C-reactive protein (CRP), aspartate transaminase (AST), total bilirubin (TBil), sCr, Blood urea nitrogen (BUN), Uric Acid (UA), activated partial thromboplastin time (APTT), prothrombin time (PT), international normalized ratio (INR), D-Dimer and MELD score, but lower albumin (Alb), eGFR and platelet (PLT) baseline levels. There was no significant difference in baseline ALT, Fibrinogen (Fib) between patients with AKI at admission and non-AKI patients; similarly, patients with AKI during follow-up had significantly higher age, incidences of ascites and HE, WBC, CRP, AST, alanine transaminase (ALT), TBil, sCr, BUN, UA, APTT, PT, INR, D-Dimer and MELD score, but lower Alb, eGFR, Fib and PLT baseline levels.

Hemostatic markers at baseline according to the type of AKI

Of the 108 patients with AKI at admission, 64 (59%) were diagnosed with PRA, 19 (18%) with HRS, and 25 (23%) with ATN. Median values for APTT, PT, INR and D-Dimer were significantly lower in patients without AKI than those with AKI. APTT did not differ significantly between patients with PRA and those with HRS ($p = 0.058$) or ATN ($p = 0.280$) and between patients with HRS and those with ATN ($p = 0.488$) (Figure 2A). PT did not differ significantly between patients with PRA and those with HRS ($p = 0.062$) or ATN ($p = 0.211$) and between patients with HRS and those with ATN ($p = 0.721$) (Figure 2B). INR did not differ significantly between patients with PRA and those with HRS ($p = 0.062$) or ATN ($p = 0.211$) and between patients with HRS and those with ATN ($p = 0.721$) (Figure 2B). INR did not differ significantly between patients with PRA and those with HRS ($p = 0.087$) or ATN ($p = 0.141$) and between patients with HRS and those with ATN ($p = 0.917$) (Figure 2C).

Figure 1. Flow diagram of the follow-up and outcomes at the first affiliated hospital, school of medicine, Zhejiang University.
D-Dimer did not differ significantly between patients with PRA and those with ATN (p = .799), but it was significantly higher in patients with HRS than in those with PRA (p = .001) and ATN (p = .005) (Figure 2D). TT, Fib and PLT did not differ significantly between the three groups (PRA, HRS and ATN).

Hemostatic markers and the development of AKI during follow-up

Of 665 cirrhotic patients without AKI at baseline, 194 patients developed AKI (PRA = 117, HRS = 51, ATN = 26) during 90 days of follow-up. For the further Cox regression, the patients were classified as AKI (PRA, HRS and ATN) versus non-AKI, or HRS versus PRA and not AKI, or ATN versus non-ATN (HRS, PRA and not-AKI). The analysis of multicollinearity was completed before Cox proportional hazards analysis. eGFR was collinear with serum creatinine and MELD was collinear with INR and TBil. Since their VIF were both greater than 5, so they were excluded from the next analysis (Supplementary Table 1). In univariate analysis, the development of AKI was significantly associated with APTT (HR = 1.033, p < .001), PT (HR = 1.109, p < .001), INR (HR = 1.122, p < .001) and D-Dimer (HR = 1.070, p < .001). Adjusted for the significant covariates of ascites, WBC, CRP, AST, TBil, sCr, BUN, and UA in univariate analysis, the development of AKI was only correlated with D-Dimer (HR = 1.039, p < .001). Similarly, the development of HRS was significantly correlated with APTT (HR = 1.046, p < .001), PT (HR = 1.138, p < .001), INR (HR = 1.156, p < .001), D-Dimer (HR = 1.079, p < .001) and Fib (HR = 0.503, p < .05). Adjusted for the significant covariates of ascites, WBC, CRP, TBil, Alb and sCr in univariate analysis, the development of HRS was only correlated with D-Dimer (HR = 1.055, p < .05). However, the development of ATN was not correlated with any hemostatic markers adjusted for the significant covariates of ascites, WBC, CRP, TBil and sCr, although it was significantly associated with APTT (HR = 1.130, p < .05), PT (HR = 1.048, p < .05), INR (HR = 1.153, p < .05), D-Dimer (HR = 1.060, p < .05) and Fib (HR = 0.443, p < .05) in univariate Cox regression (Table 2).

Prediction of the development of AKI and HRS by D-Dimer

A time-dependent ROC curve analysis was applied to estimate the prediction performance of D-Dimer in the development of AKI and HRS. Meanwhile, D-Dimer was compared to Cr. For predicting the development of AKI within 30 days, the
area under the ROC curve was 0.755 (95% CI, 0.718–0.793) for D-Dimer, and 0.676 (95% CI, 0.628–0.723) for Cr, respectively (Figure 3A); For predicting the development of HRS within 30 days, the area under the ROC curve was 0.879 (95% CI, 0.791–0.967) for D-Dimer, and 0.669 (95% CI, 0.559–0.778) for Cr, respectively (Figure 3B). D-Dimer significantly improved the prediction performance compared with sCr. In addition, D-Dimer exhibited a higher AUC in the prediction of HRS than in the prediction of AKI. Findings were similar for 60- and 90-days follow-ups (Table 3).

When a cutoff of 2.8 (mg/L FEU) was used for diagnosis of AKI, the sensitivity of D-Dimer for predicting AKI within 30 days was 74.1%, and specificity 71.1%, the positive predictive value in this cohort was 53.4% and the negative predictive value was 86.0% with a diagnostic DOR of 6.5. Similarly, a cutoff of 3.7 (mg/L FEU) was used for diagnosis of HRS, the sensitivity of D-Dimer for predicting HRS within 30 days was 87.3%, and specificity 72.9%, the positive predictive value in this cohort was 24.5% and negative predictive value was 98.3% with a diagnostic DOR of 16.0 (Table 4).
Survival analysis according to D-Dimer levels

During follow-up, 184 patients died. The overall survival rates at 30, 60 and 90 days were 79.3%, 75.8% and 74.1%, respectively. The patients were categorized into a high D-Dimer group and a low D-Dimer group based on the median D-Dimer (2.465 mg/L FEU). Kaplan-Meier curve analysis indicated that the high D-Dimer group had significantly higher mortality than the low D-Dimer group (log-rank test, \( p < .001 \)). The survival rates at 30, 60 and 90 days were 86.9%, 84.9% and 83.5%, respectively, in the low D-Dimer group; 61.1%, 53.8% and 51.4%, respectively, in the high D-Dimer group. In multivariate Cox regression analysis, Higher baseline levels of D-dimer (quartile 4 versus 1) were associated with significantly higher mortality after adjustment for age and sex and remained a risk factor for mortality after adjustment for other significant risk factors: ascites, HE, WBC, CRP, Cr, BUN, UA, ALT, AST, TBil, Alb and MELD Score (Table 5).

Discussion

In the present study, we found that D-Dimer was significantly higher in patients with HRS than in those with the other type of AKI. Interestingly, D-Dimer was correlated with HRS, but not with ATN after adjusting for the significant covariates. D-Dimer could be an important risk factor for the development of HRS in patients with cirrhosis. Moreover, D-Dimer was correlated with an increased risk of mortality in patients with cirrhosis.
D-Dimer is a degradation product of cross-linked fibrin, which is a well-known marker for clotting disorders, such as disseminated intravascular coagulation, deep vein thrombosis and pulmonary embolism. Recent studies have provided evidence that elevated D-Dimer Levels are correlated with AKI and a poor outcome in different disease groups [28–30]. Our study revealed that elevated D-Dimer was independently associated with HRS in patients with cirrhosis. Although it has been noted that D-Dimer is strongly correlated with AKI, the role of D-Dimer in the development of HRS remains elusive. D-Dimer may not only be a marker of hypercoagulability and prothrombotic state but may also be a risk factor for HRS. Increased D-Dimer may be deposited in the renal vasculature and increase the risk of microthrombi in visceral organs [31]. Moreover, there is an extensive cross-talk between inflammation and coagulation, whereby inflammation contributes to the activation of coagulation, and coagulation also considerably affects inflammatory activity, which damages the vascular endothelium [32,33]. D-Dimer is closely associated with inflammatory activity and endothelial dysfunction and may contribute to reducing the renal blood flow and impairing renal function [34,35]. It is now recognized that HRS not only involves circulatory dysfunction but also systemic inflammation [36,37].

HRS is believed to be a reversible cause of AKI. Early diagnosis and treatment of HRS are crucial for determining the outcome [21]. Recent studies showed several non-invasive methods and Zheng Luyan for her critical reading and feedback on our manuscript.

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**Table 5.** Associations between D-Dimer levels by quartile and mortality.

| D-Dimer Level (mg/L, FEU) | adjusted $I^*$ HR (95% CI) | $P$ value | adjusted $II^*$ HR (95% CI) | $P$ value |
|---------------------------|---------------------------|-----------|----------------------------|-----------|
| <0.958                    | 1                         | <.001     | 1                          | .036      |
| 0.958–2.465               | 2.003 (0.970–4.136)       |           | 1.568 (0.741–3.316)        |           |
| 2.465–5.815               | 3.697 (1.877–7.280)       |           | 1.799 (0.902–3.588)        |           |
| ≥5.815                    | 9.321 (4.852–17.908)      |           | 2.132 (1.049–4.331)        |           |

*Data are expressed as hazard ratios (95% CIs). $I^*$ is adjusted for D-Dimer quartile, age and gender; $II^*$ is adjusted for D-Dimer quartile, ascites, HE, WBC, CRP, Cr, BUN, UA, ALT, AST, TBil, Alb and MELD Score.*
Disclosure statement
There are no conflicts of interest reported by the authors.

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