Article type : Original Article

Anticoagulant treatment is associated with decreased mortality in severe coronavirus disease 2019 patients with coagulopathy

Ning Tang[1], Huan Bai[1], Xing Chen[1], Jiale Gong[1], Dengju Li[2], Ziyong Sun[1]*

[1] Department of Clinical Laboratory, Tongji Hospital, Tongji Medical College, Huazhong University of Science and Technology, Wuhan, Hubei, China

[2] Department of Hematology, Tongji Hospital, Tongji Medical College, Huazhong University of Science and Technology, Wuhan, Hubei, China

*Corresponding author

Ziyong Sun

Department of Clinical Laboratory, Tongji Hospital, Tongji Medical College, Huazhong University of Science and Technology, Wuhan, Hubei, China

Telephone: 00862783665469; Fax: 00862783663639

Email addresses

First author (Ning Tang): tonyjesus@126.com

Corresponding author (Ziyong Sun): 499388475@qq.com

Word Count (entire manuscript): 2845 words

Number of Figures and Tables (combined): 6

This article has been accepted for publication and undergone full peer review but has not been through the copyediting, typesetting, pagination and proofreading process, which may lead to differences between this version and the Version of Record. Please cite this article as doi: 10.1111/JTH.14817

Conflict of interest statement

The authors declare that they have no conflicts of interest.

Bullet points:

- Heparin treatment has been recommended for COVID-19, however, its efficacy remains to be validated.
- The 28-day mortality between heparin users and nonusers were compared in stratified patients.
- the 28-day mortality of heparin users were lower than nonusers In patients with SIC score $\geqslant 4$ or D-dimer > 3.0 ug/mL.
- Heparin treatment appears to be associated with better prognosis in severe COVID-19 patients with coagulopathy.

Abstract

Background: A relatively high mortality of severe coronavirus disease 2019 (COVID-19) is worrying, the application of heparin in COVID-19 has been recommended by some expert consensus due to the risk of disseminated intravascular coagulation and venous thromboembolism. However, its efficacy remains to be validated.

Methods: Coagulation results, medications and outcomes of consecutive patients being classified as severe COVID-19 in Tongji hospital were retrospectively analysed. The 28-day mortality between heparin users and nonusers were compared, also in different risk of coagulopaphy which was stratified by the sepsis-induced coagulopathy (SIC) score or D-dimer result.

Results: There were 449 patients with severe COVID-19 enrolled into the study, 99 of them received heparin (mainly with low molecular weight heparin, LMWH) for 7 days or longer. The D-dimer, prothrombin time and age were positively, and platelet count was negatively, correlated with 28-day

mortality in multivariate analysis. No difference on 28-day mortality was found between heparin users and nonusers (30.3% vs 29.7%, *P*=0.910). But the 28-day mortality of heparin users were lower than nonusers In patients with SIC score $\geqslant$4 (40.0% vs 64.2%, *P*=0.029), or D-dimer > 6 fold of upper limit of normal (32.8% vs 52.4%, *P*=0.017).

Conclusions: Anticoagulant therapy mainly with LMWH appears to be associated with better prognosis in severe COVID-19 patients meeting SIC criteria or with markedly elevated D-dimer.

Keywords: coronavirus disease 2019, sepsis, coagulopathy, D-dimer, low molecular weight heparin

As recent studies described [1-3], severe coronavirus disease 2019 (COVID-19) is commonly complicated with coagulopathy, disseminated intravascular coagulation (DIC) may exist in the majority of deaths. Due to the evidence of virus infection and respiratory dysfunction, many patients with severe COVID-19 meet the Third International Consensus Definitions for Sepsis (sepsis-3) [4]. In addition, long-term bed rest and likely receiving hormone treatment also increase the risk of venous thromboembolism (VTE) in severe COVID-19. For these reasons, the active application of anticoagulants (such as heparin) for patients with severe COVID-19 has been recommended by some expert consensus in China [5], however, its efficacy remains to be validated.

The International Society of Thrombosis and Haemostasis (ISTH) has proposed a new category identifying an earlier phase of sepsis-associated DIC, called “sepsis-induced coagulopathy” (SIC) [6], that patients who meet the diagnostic criteria of SIC benefit from anticoagulant therapy have been confirmed [7]. Our study aimed to validate the usefulness of SIC score and other coagulation parameters, in screening out patients who can benefit from anticoagulant through retrospective analysis.

Methods

Consecutive patients with severe COVID-19 admitted to Tongji Hospital of Huazhong University of Science and Technology in Wuhan from January 1 to February 13, 2020, were retrospectively enrolled. Exclusion criteria were a bleeding diathesis, hospital stay <7 days, lack of information about

coagulation parameters and medications, and age <18 years. A retrospective review of the characteristics of these patients was performed through the electronic medical record system of our hospital, the medications and outcomes (28-day mortality) were monitored up to March 13, 2020. This study was approved by the Ethics Committee of Tongji Hospital (Wuhan, China).

The diagnosis of COVID-19 was according to World Health Organization interim guidance [8] and confirmed by RNA detection of the SARS-CoV-2 in clinical laboratory of Tongji hospital. The severe COVID-19 was defined as meeting any one of following items, according to the Diagnosis and Treatment Plan of COVID-19 suggested by National Health Commission of China [9]: Respiratory rate $\geqslant$30 breaths /min; Arterial oxygen saturation$\leqslant$93% at rest; PaO_2/FiO_2 $\leqslant$300 mmHg.

The SIC score system including PT, platelet count and sequential organ failure assessment (SOFA) was described in table 1 [6], in which the SOFA score was developed by an international group of experts to describe the time course of six organ dysfunction using a limited number of routinely measured variables [10]. Meanwhile, in our previous study [3], higher D-dimer and prothrombin time (PT) on admission were associated with poor prognosis in patients with COVID-19. Hence these three parameters were included in this study and the results were recorded at the time the patient meeting the definition of severe COVID-19. Anticoagulant treatment group was defined as receiving unfractionated heparin (UFH) or low molecular weight heparin (LMWH) for 7 days or longer [11], which was the most commonly used anticoagulant therapy for COVID-19 in our hospital.

The coagulation tests, including PT and D-dimer were detected using a STA-R MAX coagulation analyzer and original reagents (Diagnostica Stago, Saint-Denis, France). The platelet counts were analysed by Sysmex XE-2100 haematology analyzer (Sysmex, Kobe, Japan).

Normally and abnormally distributed quantitative variables were compared using the Student's t-test and the Mann–Whitney U test, respectively. Categorical variables were compared using the chi-squared test. The results were given as the mean ± standard deviation, median (interquartile range), or number (percentage), wherever appropriate. Categorical and consecutive variables were evaluated by logistic regression analysis for their ability to predict 28-day mortality. A *P*-value of <0.05 was considered statistically significant. Data were analyzed using SPSS 21.0 for Windows (SPSS Inc., Chicago, IL, USA).

Results

Four hundred and fourty-nine patients (181 females and 268 males) classified as severe COVID-19 were enrolled into the study from consecutive 1786 confirmed cases. The mean age at disease onset was 65.1 ± 12.0 years. Two hundred and seventy-two (60.6%) patients had one or more chronic underlying diseases, mainly including hypertension (n=177, 39.4%), diabetes (n=93, 20.7%), and heart diseases (n=41, 9.1%). Ninety-nine (22.0%) patients received heparin treatment for at least 7 days, in which 94 received LMWH (40-60 mg enoxaparin/day) and 5 received UFH (10000-15000 U/day), no anticoagulants other than heparin had been used for 7 days or longer in our patients. All patients received antiviral and appropriate supportive therapies after admission. Ninety-seven (21.6%) patients met the SIC criteria (total score ⩾4) when they were classified as severe cases. By the end of March 13, 315 (70.2%) patients were still survived and 134 (29.8%) patients had died (Figure 1). No difference on the 28-day mortality was found between heparin users and nonusers (30.3% vs 29.7%, *P*=0.910).

The parameters of coagulation tests and clinical characteristics between survivors and non-survivors were compared (Table 2). Then these parameters were examined in a multivariate logistic regression model to identify independent correlative factor of 28-day mortality of severe COVID-19 (Table 3). The D-dimer, PT and age were positively, and platelet count was negatively, correlated with 28-day mortality.

The association between heparin treatment and outcome in stratified patients according to SIC score or D-dimer result were evaluated (Table 4 and Figure 2). The heparin treat was associated with lower mortality in patients with SIC score ⩾4 (40.0% vs 64.2%, *P*=0.029), but not in those with SIC score <4 (29.0% vs 22.6%, *P*=0.419). As patients were stratified by D-dimer result, the mortality in heparin users basically maintained at same level, but in nonusers, the mortality rised with the rising D-dimer. When D-dimer exceeding 3.0 ug/mL (6 fold of upper limit of normal, 6 ULN), approximate 20% reduction in mortality with heparin treat was found (32.8% vs 52.4%, *P*=0.017).

Discussion

The dysfunction of endothelial cells induced by infection results in excess thrombin generation and fibrinolysis shutdown, which indicated a hypercoagulable state in patient with infection [12, 13], such as COVID-19. In addition, the hypoxia found in severe COVID-19 can stimulate thrombosis through not only increasing blood viscosity, but also a hypoxia-inducible transcription factor-dependent signaling pathway [14]. As evidence, occlusion and microthrombosis formation in pulmonary small vessels of critical patient with COVID-19 has been reported from a recent lung organ dissection [15]. Hence, early application of anticoagulant therapy in severe COVID-19 was suggested in China for improving outcome [5], however, no specific inclusion or exclusion criteria has been pointed out so far. As anticoagulant was seldom used in early stage due to lack of understanding of this disease, and increasingly used later during this outbreak of COVID-19, we could retrospectively include enough cases to analyze the difference on outcomes between patients with and without receiving anticoagulant.

LMWH was the most commonly used anticoagulant in our hospital for preventing DIC and VTE in patients, also because of its anti-inflammatory effect [16]. Another reason is that other anticoagulants, such as recombinant soluble thrombomodulin or antithrombin, is unavailable in China yet. The prophylactic dose of LMWH was used in most of our heparin users, bleeding complications were unusual and commonly mild, it is not known if higher doses would have been better. Due to the evidence suggests that the prevalence and genetic risk factors of VTE vary significantly among ethnic populations, and the incidence of VTE in Asian populations (21-29 cases per 100,000 individuals per year) is lowest [17, 18], a higher dose of LMWH could be considered in non-Asian patients with severe COVID-19. However, the effectiveness of anticoagulant therapy for sepsis-associated DIC is still controversial [19, 20], even the Japanese guideline for management of sepsis has against the use of heparin or heparin analogs as a standard treatment in sepsis-associated DIC [21], some studies suggested that septic patients might just benefit from early recognition and specific treatment [22, 23]. As platelet count decline and PT prolongation are correlated with increased mortality, and hypofibrinogenemia is not common in sepsis, the ISTH developed the SIC criteria to guide anticoagulant therapy, the usefulness of this simple score has been validated previously [7].

As organ dysfunction is mainly limited in lung and virus is the main pathogen, the coagulation feature of severe COVID-19 might not be identical with sepsis in general. Perhaps due to the reactively increased thrombopoietin following pulmonary inflammation [24], platelet count may not be a sensitive marker for coagulopathy of COVID-19, in current study, only 21.6% of patients with severe COVID-19 met the SIC criteria, that suggested limited patients needing anticoagulant treat. However, as an indirect marker of coagulation activation, markedly elevated D-dimer (>6 ULN) also suggested benefit from heparin treat, in a larger groups of severe patients (161 of 449, 35.9%).

As the activation of coagulation also contributes to compartmentalization of pathogens and reduces their invasion [25], therefore, anticoagulant treatment in patients without significant coagulopathy has potential risk. This may explain the relatively higher mortality of heparin users comparing to nonusers in patients with D-dimer$\leqslant 1$ ULN, although the difference was not statistically significant (*P*=0.260).

There were several limitations in current study. First, potential selection bias exists in this retrospective study, for instance, LMWH might tend to be used in patients with targeted symptom or medical history, which we have not controlled. Second, due to insufficient medical resource at early stage of the COVID-19 outbreak in Wuhan, China, the severity and mortality of the included patients might not be representative. Third, the influence of other therapies on these patients has not been evaluated, in addition, as we enrolled patients over a 6 week period, It is possible that some non-pharmacological change has taken place in the management of patients as the doctors learned more about this disease over the period. Nonetheless, this study included a large critical patient population, and due to lack of specific drugs against the infection of SARS-CoV-2 up to now [26], the majority of patients with severe COVID-19 should have received similar supportive treatment after admission. Hence, we believe that the results of current study still have certain clinical significance.

In conclusion, a relatively high mortality of severe COVID-19 is worrying, our study suggests that anticoagulant may not benefit to the unselected patients, instead, only the patients meeting SIC criteria or with markedly elevated D-dimer may benefit from anticoagulant therapy mainly with LMWH. Further prospective studies are needed to confirm this result.

Author Contributions

N.Tang drafted the manuscript, N.Tang, H.Bai, X.Chen and J.Gong collected and analysed the data, D.Li interpreted the data, N.Tang and Z.Sun designed the study.

Funding

National Mega Project on Major infectious Disease Prevention of China. (No.2017ZX10103005-007)

Conflict of interest statement

The authors declare that they have no conflicts of interest.

References

1. Chen N, Zhou M, Dong X, et al. Epidemiological and clinical characteristics of 99 cases of 2019 novel coronavirus pneumonia in Wuhan, China: a descriptive study. *Lancet*. 2020; 395(10223): 507-513.

2. Huang C, Wang Y, Li X, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. *Lancet*. 2020; 395(10223): 497-506.

3. Tang N, Li D, Wang X, Sun Z. Abnormal Coagulation parameters are associated with poor prognosis in patients with novel coronavirus pneumonia. *J Thromb Haemost*. 2020 [published online ahead of print].

4. Singer M, Deutschman CS, Seymour CW, et al. The Third International Consensus Definitions for Sepsis and Septic Shock (Sepsis-3). *JAMA*. 2016; 315(8):801–810.

5. Shanghai Clinical Treatment Expert Group for COVID-19. Comprehensive treatment and management of coronavirus disease 2019: expert consensus statement from Shanghai (in Chinese). *Chin J Infect*. 2020, 38 [published online ahead of print].

6. Iba T, Levy JH, Warkentin TE, et al. Diagnosis and management of sepsis-induced coagulopathy and disseminated intravascular coagulation. *J Thromb Haemost*. 2019; 17(11):1989–1994.

7. Iba T, Nisio MD, Levy JH, Kitamura N, Thachil J. New criteria for sepsis-induced coagulopathy (SIC) following the revised sepsis definition: a retrospective analysis of a nationwide survey. *BMJ Open*.

2017; 7(9):e017046.

8. World Health Organization. Clinical management of severe acute respiratory infection when novel coronavirus (nCoV) infection is suspected: interim guidance. Published January 28, 2020. Accessed January 31, 2020. https://www.who.int/publications-detai l/clinical-management-of-severe-acute-respiratory-infection-whennovel-coronavirus-(ncov)-infection -is-suspected

9. National Health Commission of China. The diagnosis and treatment plan for the novel coronavirus disease (the seventh edition). 2020.

10. Vincent JL, Moreno R, Takala J, et al. The SOFA (Sepsis-related Organ Failure Assessment) score to describe organ dysfunction/failure. On behalf of the Working Group on Sepsis-Related Problems of the European Society of Intensive Care Medicine. *Intensive Care Med*. 1996; 22(7):707–710.

11. Venclauskas L, Llau JV, Jenny JY, Kjaersgaard-Andersen P, Jans Ø; ESA VTE Guidelines Task Force. European guidelines on perioperative venous thromboembolism prophylaxis: Day surgery and fast-track surgery. *Eur J Anaesthesiol*. 2018; 35(2):134–138.

12. Levi M, van der Poll T. Coagulation and sepsis. *Thromb Res*. 2017; 149:38–44.

13. Schmitt FCF, Manolov V, Morgenstern J, et al. Acute fibrinolysis shutdown occurs early in septic shock and is associated with increased morbidity and mortality: results of an observational pilot study. *Ann Intensive Care*. 2019; 9(1):19.

14. Gupta N, Zhao YY, Evans CE. The stimulation of thrombosis by hypoxia. *Thromb Res*. 2019; 181:77–83.

15. Luo W, Yu H, Gou J, et al. Clinical Pathology of Critical Patient with Novel Coronavirus Pneumonia (COVID-19). *Preprints.* 2020, 2020020407.

16. Poterucha TJ, Libby P, Goldhaber SZ. More than an anticoagulant: Do heparins have direct anti-inflammatory effects?. *Thromb Haemost*. 2017; 117(3):437–444.

17. Stein PD, Kayali F, Olson RE, Milford CE. Pulmonary thromboembolism in Asians/Pacific Islanders in the United States: analysis of data from the National Hospital Discharge Survey and the United States Bureau of the Census. *Am J Med*. 2004; 116(7):435–442.

18. Zakai NA, McClure LA. Racial differences in venous thromboembolism. *J Thromb Haemost*. 2011;

9(10):1877–1882.

19. Aikawa N, Shimazaki S, Yamamoto Y, et al. Thrombomodulin alfa in the treatment of infectious patients complicated by disseminated intravascular coagulation: subanalysis from the phase 3 trial. *Shock*. 2011; 35(4):349–354.

20. Liu XL, Wang XZ, Liu XX, et al. Low-dose heparin as treatment for early disseminated intravascular coagulation during sepsis: A prospective clinical study. *Exp Ther Med*. 2014; 7(3):604–608.

21. Nishida O, Ogura H, Egi M, et al. The Japanese Clinical Practice Guidelines for Management of Sepsis and Septic Shock 2016 (J-SSCG 2016). *Acute Med Surg*. 2018; 5(1):3–89.

22. Umemura Y, Yamakawa K, Ogura H, Yuhara H, Fujimi S. Efficacy and safety of anticoagulant therapy in three specific populations with sepsis: a meta-analysis of randomized controlled trials. *J Thromb Haemost*. 2016; 14(3):518–530.

23. Iba T, Gando S, Thachil J. Anticoagulant therapy for sepsis-associated disseminated intravascular coagulation: the view from Japan. *J Thromb Haemost*. 2014; 12(7):1010–1019.

24. Menter DG, Kopetz S, Hawk E, et al. Platelet "first responders" in wound response, cancer, and metastasis. *Cancer Metastasis Rev*. 2017; 36(2):199–213.

25. Sun H, Wang X, Degen JL, Ginsburg D. Reduced thrombin generation increases host susceptibility to group A streptococcal infection. *Blood*. 2009; 113(6):1358–1364.

26. Dong L, Hu S, Gao J. Discovering drugs to treat coronavirus disease 2019 (COVID-19). *Drug Discov Ther*. 2020; 14(1):58–60.

Table 1 ISTH SIC scoring system

Item	Score	Range
Platelet count ($\times 10^9$/L)	1	100-150
	2	<100
PT -INR	1	1.2-1.4
	2	>1.4
SOFA score	1	1
	2	⩾2
Total score for SIC	⩾4	

INR, International Normalized Ratio; SOFA, sequential organ failure assessment.

Table 2 Clinical and coagulation characteristics of patients being classified as severe COVID-19

Parameters	Normal range	Total (n=449)	Survivors (n=315)	Non-survivors (n=134)	*P* values
Age (years)		65.1±12.0	63.7±12.2	68.7±11.4	<0.001
Sex ratio (male/female)		268/181	178/137	90/44	0.036
With underlying diseases		272 (60.6%)	181 (57.5%)	91 (67.9%)	0.136
Receiving heparin		99 (22.0%)	69 (21.9%)	30 (22.4%)	0.910
Meeting SIC criteria		97 (21.6%)	42 (13.3%)	55 (41.0%)	<0.001
Coagulation parameters					
PT (sec)	11.5-14.5	15.2±5.0	14.6±2.1	16.5±8.4	<0.001
Platelet count (×109/L)	125-350	215±100	231±99	178±92	<0.001

D-dimer (ug/mL)	<0.5	1.94 (0.90-9.44)	1.47 (0.78-4.16)	4.70 (1.42-21.00)	<0.001

Table 3 Multivariate correlative factors of 28-day mortality in severe COVID-19

	Multivariate analysis	
	Odds ratio (95% CI)	*P* value
Age	1.033 (1.013-1.055)	0.002
Sex ratio	0.677 (0.425-1.078)	0.100
With underlying diseases	0.861 (0.538-1.379)	0.534
Treating with heparin	1.647 (0.929-2.921)	0.088
Prothrombin time	1.107 (1.008-1.215)	0.033
Platelet count	0.996 (0.993-0.998)	0.001
D-dimer	1.058 (1.028-1.090)	<0.001

Table 4 The association between heparin treatment and outcomes in stratified patients

Patients with	28-day mortality		Univariate analysis	
	Treating with Heparin	Non-treating with heparin	Odds ratio (95% CI)	*P* value
SIC socre⩾4 (n=97)	40.0%	64.2%	0.372 (0.154-0.901)	0.029
SIC score⩽4 (n=352)	29.0%	22.6%	1.284 (0.700-2.358)	0.419
D-dimer⩽1 ULN (n=34)	33.3%	9.7%	4.667 (0.320-68.03)	0.260

D-dimer >1 ULN (n=415)	30.2%	32.7%	0.934 (0.569-1.533)	0.788
D-dimer >2 ULN (n=317)	32.1%	36.9%	0.810 (0.477-1.375)	0.435
D-dimer >3 ULN (n=253)	31.1%	42.5%	0.611 (0.344-1.086)	0.093
D-dimer >4 ULN (n=224)	33.3%	44.5%	0.623 (0.345-1.127)	0.118
D-dimer >5 ULN (n=190)	34.9%	48.8%	0.563 (0.301-1.050)	0.071
D-dimer >6 ULN (n=161)	32.8%	52.4%	0.442 (0.226-0.865)	0.017
D-dimer >8 ULN (n=150)	33.3%	54.8%	0.412 (0.207-0.817)	0.011

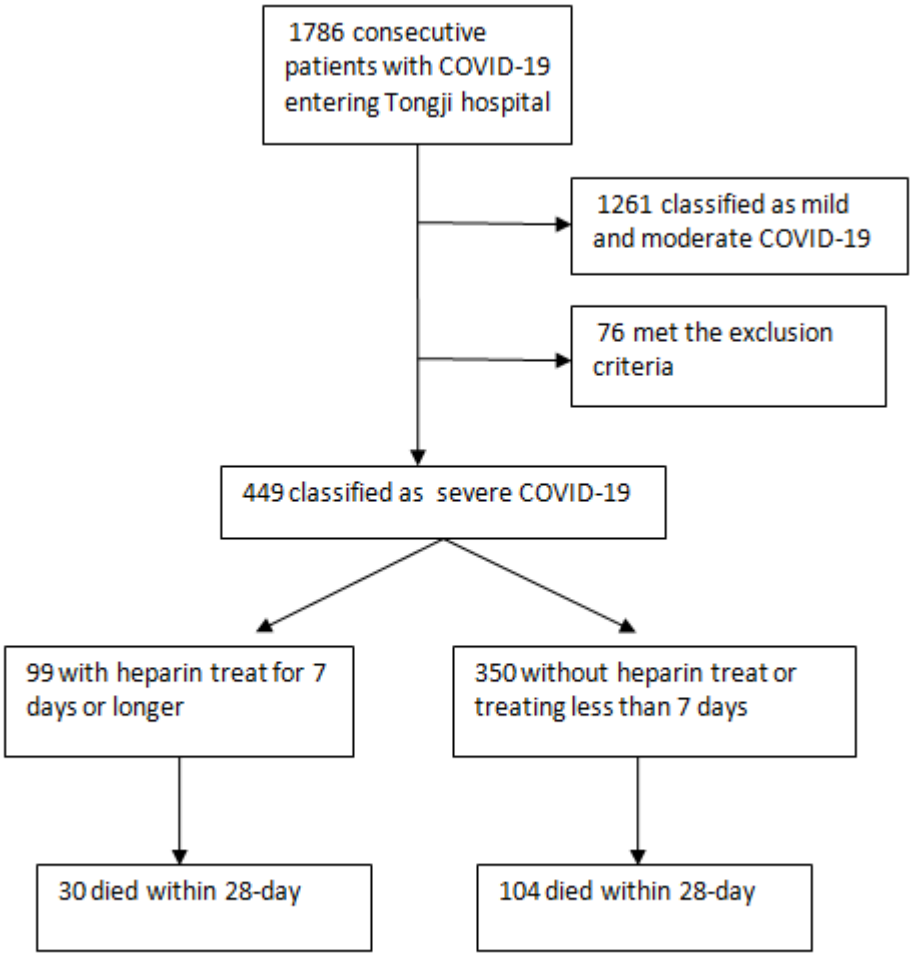
ULN: upper limit of normal, 0.5ug/mL for D-dimer.

Figure 1. The enrollment of patients with severe COVID-19

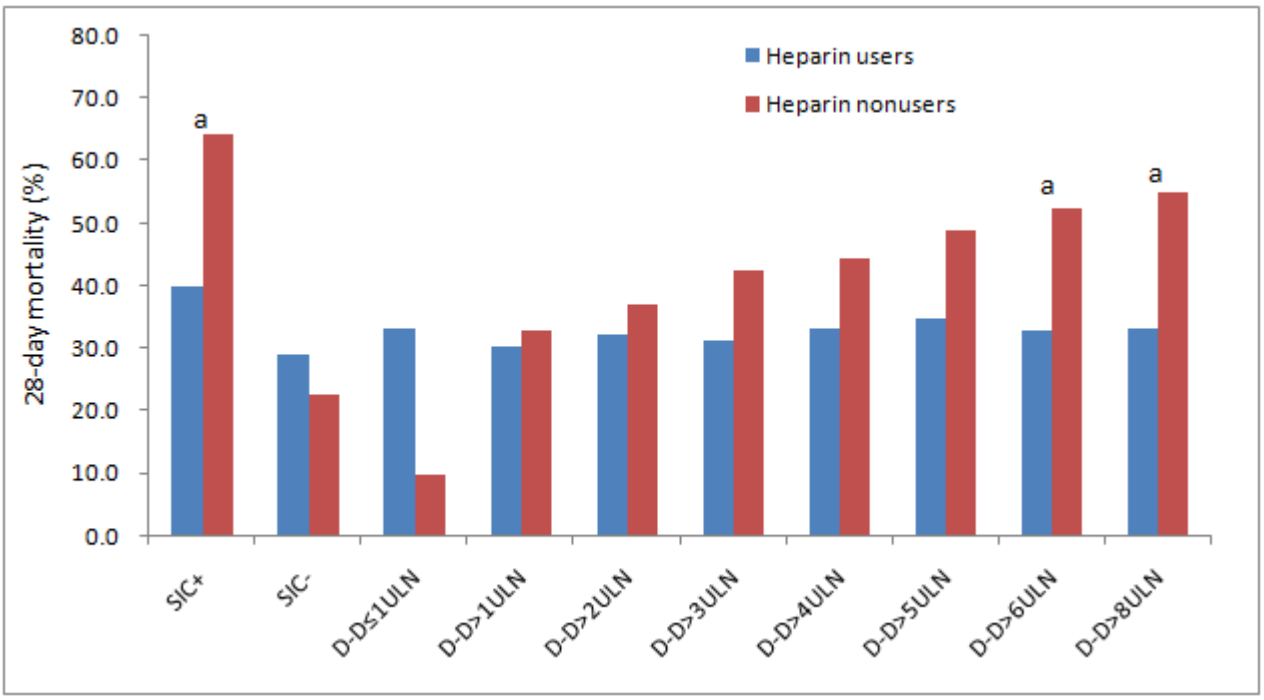
.

Figure 2. A paired bar chart showing the mortality between heparin users and nonusers in stratified patients

SIC+, SIC score⩾4; SIC-, SIC score<4; D-D, D-dimer; ULN, upper limit of normal (0.5ug/mL); a, *P*<0.05 between heparin users and nonusers.



jth_14817_f1.png



jth_14817_f2.png