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ORIGINAL RESEARCH

A Prospective Study of the Association of IL6 with the Critical Unit and Their Effect on in-Hospital Mortality in Critically Ill Patients

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Purpose: We previously proposed a new concept, the "critical unit", which covers the structural integrity and function of mitochondria and endothelium. Injury of the critical unit plays a key role in the development of critical illnesses. High levels of inflammation may lead to abnormalities of the critical unit, which is an important mechanism for critical illnesses, and both inflammation and critical unit dysfunction may affect patient prognosis. Here we evaluated the correlation between interleukin-6 (IL6) and the critical unit biomarkers in critically ill patients and the impact of both on prognosis.

Patients and Methods: This study included adult patients admitted to the intensive care unit for various reasons from January 1st to May 31st, 2023. Baseline characteristics, intensive care unit parameters, and laboratory test and outcome data were obtained from the electronic medical records system. Critical unit parameters were measured using polymerase chain reaction and enzyme-linked immunosorbent assay methods. Correlations were examined between IL6, critical unit parameters, and various outcomes.

Results: In critically ill patients, IL6 was closely associated with all the critical unit biomarkers (activated partial thromboplastin time, sphingosine 1-phosphate, mitochondrial DNA, mitochondrial fission 1, and Parkin) and the prognoses of patients. A nomogram was constructed using the critical unit biomarkers to predict the in-hospital mortality of critically ill patients. The area under the curve for the mortality prediction model was 0.708. In sensitivity analyses, the predictive effect was better in the non-surgery and tumor groups compared with the surgery and non-tumor groups, with area under the curve values of 0.885 and 0.891, respectively.

Conclusion: Our study innovatively integrated mitochondrial and endothelial markers in the critical unit to comprehensively evaluate patient prognosis, which may be a trend in the future assessment of critically ill patients. There are few such studies, and ours may promote the progress of related research.

Keywords: critical unit, IL6, critically ill patients, pro-inflammatory response, prognosis

Introduction

Critical diseases are often accompanied by endothelial and mitochondrial damage, which is directly related to the poor prognosis of patients.[1] The interaction between the endothelium and mitochondria plays an important role in the occurrence and progression of critical diseases. On the basis of the inseparable relationship between endothelium and mitochondria and their significant impact on critical diseases, we integrated them into the "critical unit".[2] The critical unit covers the structural and function of mitochondria and endothelium, which is also a key link in critical illness.[2]

Critical diseases are caused by trauma, infection, surgery, and stress, which all induce high inflammation through pathogen-associated molecular patterns (PAMPs) or damage-associated molecular patterns (DAMPs). Studies have confirmed that an excessive inflammatory response is one of the important mechanisms leading to the occurrence and

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progression of critical diseases.[3] Interleukin-6 (IL6) is one of the classical pro-inflammatory factors, and the blood concentration of IL6 reflects the intensity of the pro-inflammatory response of patients.

Pro-inflammatory factors can activate endothelial cells and trigger programmed cell death, leading to regionalization of the local inflammatory response.[4,5] Endothelial dysfunction is not only a key factor in the causes of organ dysfunction in critical diseases such as sepsis but it also seriously affects the prognosis of critically ill patients.[6] Inflammation also affects mitochondrial function in many ways. Pro-inflammatory factors significantly affect the tricarboxylic acid cycle, the core driving force of cell respiration, thus influencing mitochondrial energy metabolism.[7] Pro-inflammatory factors can also lead to the disturbance of mitochondrial dynamics, resulting in damage to respiratory function.[8,9] Mitochondrial respiratory function damage plays a key role in organ dysfunction in critical illness, especially in shock, and is associated with poor prognosis in shock patients.[10,11] The pro-inflammatory response significantly impacts the critical unit (endothelium and mitochondria) and affects the prognosis of critically ill patients. Most studies have only focused on the effect of the pro-inflammatory factor IL6 on the endothelium or mitochondria, and few studies regard the endothelium and mitochondria as a whole, that is, the critical unit. Therefore, it is necessary to explore the specific relationship between IL6 and the critical unit in critically ill patients and the effects on patient prognosis.

In this study, we hypothesized that IL6 is closely related to the critical unit in critically ill patients and that IL6 and critical unit biomarkers can well evaluate the prognosis of patients. We examined the blood concentration of IL6 and critical unit biomarkers (including mitochondrial fission 1 (Fission 1), Parkin RBR E3 ubiquitin-protein ligase (Parkin), mitochondrial DNA (mtDNA), sphingosine 1-phosphate (S1P), and activated partial thromboplastin time (APTT)) in critically ill patients. We also investigated the associations between IL6 and critical unit biomarkers. We then explored the relationship between IL6 and patient prognosis. Finally, a predictive model was constructed based on critical unit biomarkers, and its effectiveness in evaluating the prognosis of critically ill patients was examined. We also performed sensitivity analyses in different subgroups of critically ill patients to evaluate the universality of this approach for a comprehensive assessment of patient prognosis.

Materials and Methods

Study Design and Research Population

This study was approved by the local ethics committee of Peking Union Medical College Hospital (PUMCH) in China (I-22PJ1072). Adult patients who were admitted to the intensive care unit (ICU) with a diagnosis of infection-related diseases from January 1 to May 31, 2023 were included in our study. All patients or their relatives provided written informed consent. The exclusion criteria included minors and patients that did not provide written informed consent. The study procedure is shown in Figure 1. The research data are available on reasonable request from the corresponding author following the data sharing rules made by PUMCH.

Data Collection

Electronic Data

Most of the clinical data were obtained from the electronic medical records system in PUMCH, which is a mature database with well preserved data. The following clinical data were collected: baseline characteristics including age, gender, tumor information, and surgery information; ICU parameters on admission, which mainly included heart rate (HR), mean atrial pressure (MAP), lactate, Sequential Organ Failure Assessment (SOFA) score, Acute Physiology and Chronic Health Evaluation (APACHE) II score, and dosage of norepinephrine (NE); and endpoints. The primary endpoints included in-hospital mortality and the secondary outcomes included mechanical ventilation time (MVt), ICU days, and length of in-hospital stay (LOS). Data on inflammation biomarkers (IL6) were collected from the system. While APTT is a functional index for coagulation, it reflects endothelial injury to some extent. Therefore, we use it as an indirect index for the critical unit.[2] Wang G and Lian H extracted the data independently and cross-checked the extracted data. Data were confirmed by Wang X and Zhang H.

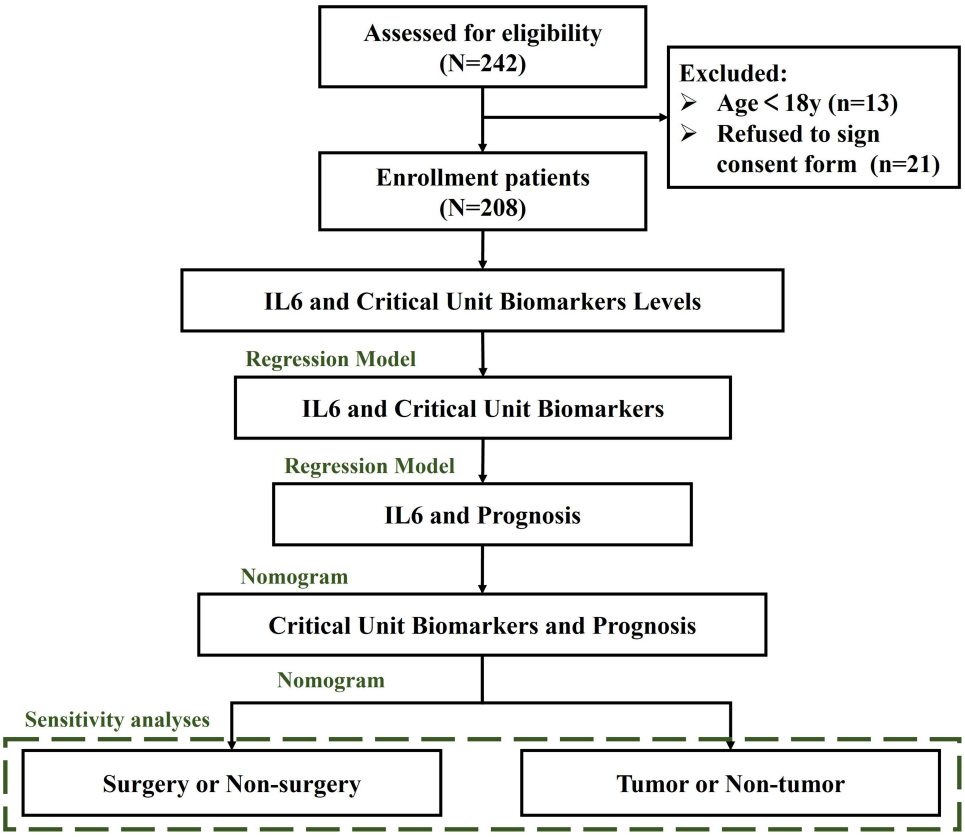

Figure 1 Flowchart of the study.
Abbreviation: IL6, Interleukin-6.

Other Critical Unit Biomarkers

Peripheral blood samples were collected within 24 h after ICU admission and immediately centrifuged. Serum was obtained and stored at −80°C. Plasma mtDNA was extracted using a commercially available kit (DP 318, Tiangen Biochemistry, Beijing, China). Polymerase chain reaction (PCR) was performed following previously published methods.[12] Gene expression was calculated by the $2^{-\Delta\Delta Cq}$ method using standards in the kit. The primers for the mtDNA gene were forward 5′-ATA CCCATGGCCAACCTCCT-3′ and reverse 5′-GGGCCT TTGCGTAGTTGTAT-3′. Other serum proteins, such as Fission 1, Parkin, and S1P, were determined using commercially available enzyme-linked immunosorbent assay (ELISA) kits following manufacturers' instructions (Fission 1: Abbexa, abx151559, Cambridge, UK; Parkin: Abcam, ab212159, Shanghai, China; S1P: MSKbio, kt99298, Wuhan, China). The ELISA assays were specific for native proteins, with no significant cross-reactivity with known analogs. Each well of the ELISA plate was loaded with 100 µL undiluted blood sample, and protein concentration was not measured before loading the plate.

Statistical Analysis

Continuous variables are presented as quantile percentile after normality test. Categorical variables are presented as number (percentage). χ^2 tests were used for comparisons of categorical variables. Generalized Estimating Equations (GEE) was used to conduct single-factor and multi-factor analyses. We adjusted age, gender, and APACHE II in the multi-factor model. Nomograms were used to build the prediction model. The receiver operating characteristic (ROC) curve was used to assess the accuracy of the predicting model. P<0.05 indicated statistical significance. All statistical analyses and figures were conducted using R software, version 4.2.3 (http://www.R-project.org/). Figure 1 was created in WPS.

Results

A total of 208 patients were included in the study. Among the 208 patients, 110 were female and 98 were male. Most patients (85.1%) underwent surgery before entering the ICU and 47.1% were diagnosed with a tumor. The median level of NE was 0.1 μg/(kg/min), and the median SOFA and APACHE II scores were 7 and 14, respectively. Of the 208 patients, 10 died in the hospital. The median ICU stay was 3 days. The median in-hospital length of stay was 15 days, and the median ventilation time was 13.5 h (Table 1). IL6 concentration ranged from 0 to 423016 pg/mL, with a median level of 157.9 pg/mL (Table 2). Most patients had a shortened APTT. The levels of S1P, mtDNA, Fission 1, and Parkin in the study population are listed in the table; information on normal levels for these markers has not been established.

Table 3 shows the regression results from the GEE model. IL6 showed markedly correlations with mitochondrial indicators, such as Fission 1, Parkin, and mtDNA. IL6 was positively correlated with Fission 1 and Parkin and negatively correlated with mtDNA. Fission 1 had a stronger correlation with IL6 than Parkin. The β value and 95% CI were 0.489 (0.358, 0.620) and 0.070 (0.030, 0.110) in a multi-factor model. While mtDNA was negatively correlated with IL6, the correlation coefficient was −0.497 in both single-factor and multi-factor models. Regarding endothelial biomarkers, IL6 only correlated with APTT, an indirect indicator for endothelial injury. The β value and 95% CI were 0.059 (95% CI: 0.055, 0.063). There was a negative trend between IL6 and S1P, although without statistical significance.

We next examined IL6 and patient outcomes (Table 4). We found no significant results in terms of primary outcomes (in-hospital mortality) in both the single-factor and the multi-factor model. However, IL6 showed a negative correlation in terms of secondary outcomes. An increase in IL6 was associated with a shorter LOS, ICU stay, and MVt in both single-factor and multi-factor models. The β value and 95% CI were −0.043 (−0.053, −0.034) and −0.017 (−0.021,

Table 1 Clinical Characteristics of the Study Population

Continuous Variable	N=208					Categorical Variable	N=208
	Minimum	25th	Medium	75th	Maximum		N(%)
Age (years)	25	50	60	69	95	**Gender**	
HR (bpm)	57	78	91	104	145	Female	110(52.9%)
MAP (mmHg)	0	86	97	106	228	Male	98(47.1%)
NE (μg/kg/min)	0.0	0.0	0.1	0.3	4.0	**Non-Surgery**	31(14.9%)
Lac (mmol/l)	0.4	1.0	1.7	3.5	18	**Surgery**	177(85.1%)
SOFA	0	4	7	10	21	**Non-Tumor**	110(52.9%)
APACHE II	6	10	14	17	46	**Tumor**	98(47.1%)
LOS (days)	2	10	15	22	119	**In-hospital mortality**	
ICU stay (days)	1	2	3	5	55	No	198(95.2%)
MVt (hours)	0	3	13.5	46.5	1137	Yes	10(4.8%)

Abbreviations: HR, Heart Rate; MAP, Mean Arterial Pressure; NE, Norepinephrine; Lac, Lactate; SOFA, Sequential Organ Failure Assessment; APACHE II, Acute Physiology And Chronic Health Evaluation II; LOS, Length Of Stay; ICU, Intensive Care Unit; MVt, Mechanical Ventilation time; bpm, beats per minute.

Table 2 IL6 and the Critical Unit Markers of the Study Population

	N=208				
	Minimum	25th	Medium	75th	Maximum
IL6 (pg/mL)	0	73.3	157.9	460.5	423016
APTT (s)	18.6	25.8	28.4	32.8	131.4
S1P (nmol/l)	311.6	953.8	1607.6	2189.4	2953.8
MtDNA (copies/μL)	1.5	49.6	158.2	309.9	872.6
Fission 1 (pg/mL)	295.5	488.7	663.8	807.6	991.8
Parkin (pg/mL)	82.0	133.8	183.9	231.1	317.5

Abbreviations: IL6, Interleukin-6; APTT, Activated Partial Thromboplastin Time; S1P, Sphingosine 1-Phosphate; MtDNA, Mitochondrial DNA; Fission 1, Mitochondrial Fission 1; Parkin, Parkin RBR E3 ubiquitin-protein ligase.

Table 3 Regression Models Between IL6 and Critical Unit Biomarkers of the Study Population

Critical Unit Biomarkers	Single-Factor Model	Multi-Factor Model
	β value and 95% CI	
Fission 1	**0.451(0.330, 0.573)**	**0.489(0.358, 0.620)**
Parkin	**0.068(0.031, 0.105)**	**0.070(0.030, 0.110)**
MtDNA	**−0.497(−0.610, −0.384)**	**−0.497(−0.616, −0.379)**
S1P	−0.129(−0.610, 0.353)	−0.088(−0.57, 0.394)
APTT	**0.060(0.056, 0.064)**	**0.059(0.055, 0.063)**

Abbreviations: IL6, Interleukin-6; Fission 1, Mitochondrial Fission 1; Parkin, Parkin RBR E3 ubiquitin-protein ligase; MtDNA, Mitochondrial DNA; S1P, Sphingosine 1-Phosphate; APTT, Activated Partial Thromboplastin Time.

Table 4 Regression Models Between IL6 and Outcomes of the Study Population

Outcomes	Single-Factor Model	Multi-Factor Model
	OR/β value and 95% CI	
Mortality	0.996(0.990,1.003)	0.996(0.988, 1.004)
LOS	**−0.041(−0.049, −0.033)**	**−0.043(−0.053, −0.034)**
ICU stay	**−0.016(−0.021, −0.011)**	**−0.017(−0.021, −0.012)**
MVt	**−0.132(−0.225, −0.039)**	**−0.146(−0.229, −0.063)**

Abbreviations: IL6, Interleukin-6; LOS, Length of Stay; ICU, Intensive Care Unit; MVt, Mechanical Ventilation time.

−0.012) for LOS and ICU stay, respectively, indicating the coefficient was larger with LOS than ICU stay. The same trends were found in the multi-factor model. Elevated levels of IL6 correlated with MVt; the β values and 95% CIs were −0.132 (−0.225, −0.039) and −0.146 (−0.229, −0.063) in single-factor and multi-factor models.

After confirming the correlation between IL6 and the outcomes or critical unit biomarkers, we built a predictive model to explore the correlation between critical unit biomarkers and the outcomes (Figure 2). The predictive model was logit(mortality) = −4.42920 −0.00434*Parkin +0.00108*Fission 1+0.00050*mtDNA+0.00035*S1P+0.02507*APTT. The area under ROC curve (AUC) was 0.708 (Figure 3). The sensitivity and specificity were 80.0% and 65.3%, respectively. We conducted sensitivity analyses to further explore the effect of the critical unit on the in-hospital mortality. In the non-surgery group, the predictive model was logit(mortality) = −1.27371 −0.02806*Parkin+0.00201*Fission 1 +0.00309*mtDNA +0.00178*S1P-0.01422*APTT. The AUC was 0.885 (Figure 4A). For patients who underwent surgery, the predictive model was logit(mortality) = −6.49573 −0.00495*Parkin-0.00123*Fission 1+0.00013*mtDNA-0.00017*S1P+0.14413*APTT, with an AUC of 0.794 (Figure 4A). Nearly half of the study population was diagnosed with a tumor, which could be an influencing factor for both the critical unit and the outcomes. Thus, we also conducted sensitivity analyses in tumor and non-tumor groups (Figure 4B). The predictive models were logit(mortality) = −3.13891 −0.00279*Parkin-0.00077*Fission 1 −0.00296*mtDNA+0.00097*S1P+0.00492*APTT for non-tumor groups and logit-(mortality) = −10.68360 −0.01405*Parkin+0.00444*Fission 1+0.00120*mtDNA-0.00027*S1P+0.21603* APTT for the tumor group. The AUCs were 0.891 and 0.698, respectively.

Discussion

For this study we conceptualized mitochondria and endothelial cells as a whole, defined as the "critical unit", and explored the association between IL6 and the critical unit, as well as the effect of both on in-hospital mortality in critically ill patients. We found that IL6 was closely related to the critical unit, positively correlated with Fission 1, Parkin, and APTT, and negatively correlated with mtDNA. Additionally, IL6 was partially related to the prognosis of critically ill patients and showed negative correlations with LOS, ICU stay, and MVt. We also built a model using critical

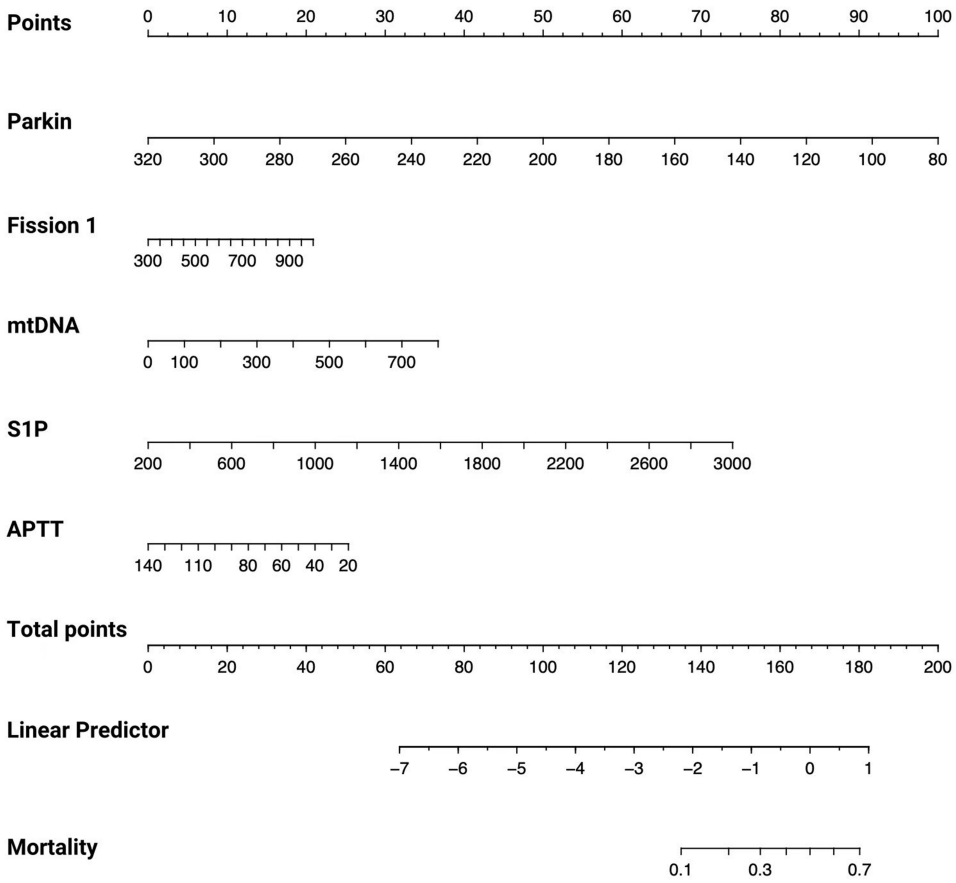

Figure 2 Nomogram of the critical unit biomarkers predicting mortality. Predictive model: logit(mortality) = −4.42920 −0.00434*Parkin +0.00108*Fission1 +0.00050*mtDNA +0.00035*S1P +0.02507*APTT.

Abbreviations: Parkin, Parkin RBR E3 ubiquitin-protein ligase; Fission 1, Mitochondrial Fission 1; MtDNA, Mitochondrial DNA; S1P, Sphingosine 1-Phosphate; APTT, Activated Partial Thromboplastin Time.

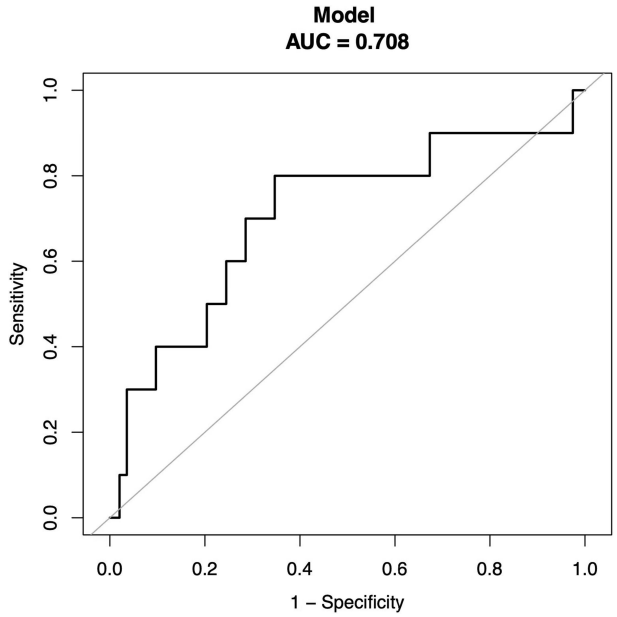

Figure 3 ROC curve of the mortality-predicting model in the study population.

Abbreviations: ROC, Receiver Operating Characteristic; AUC, Area Under ROC Curve.

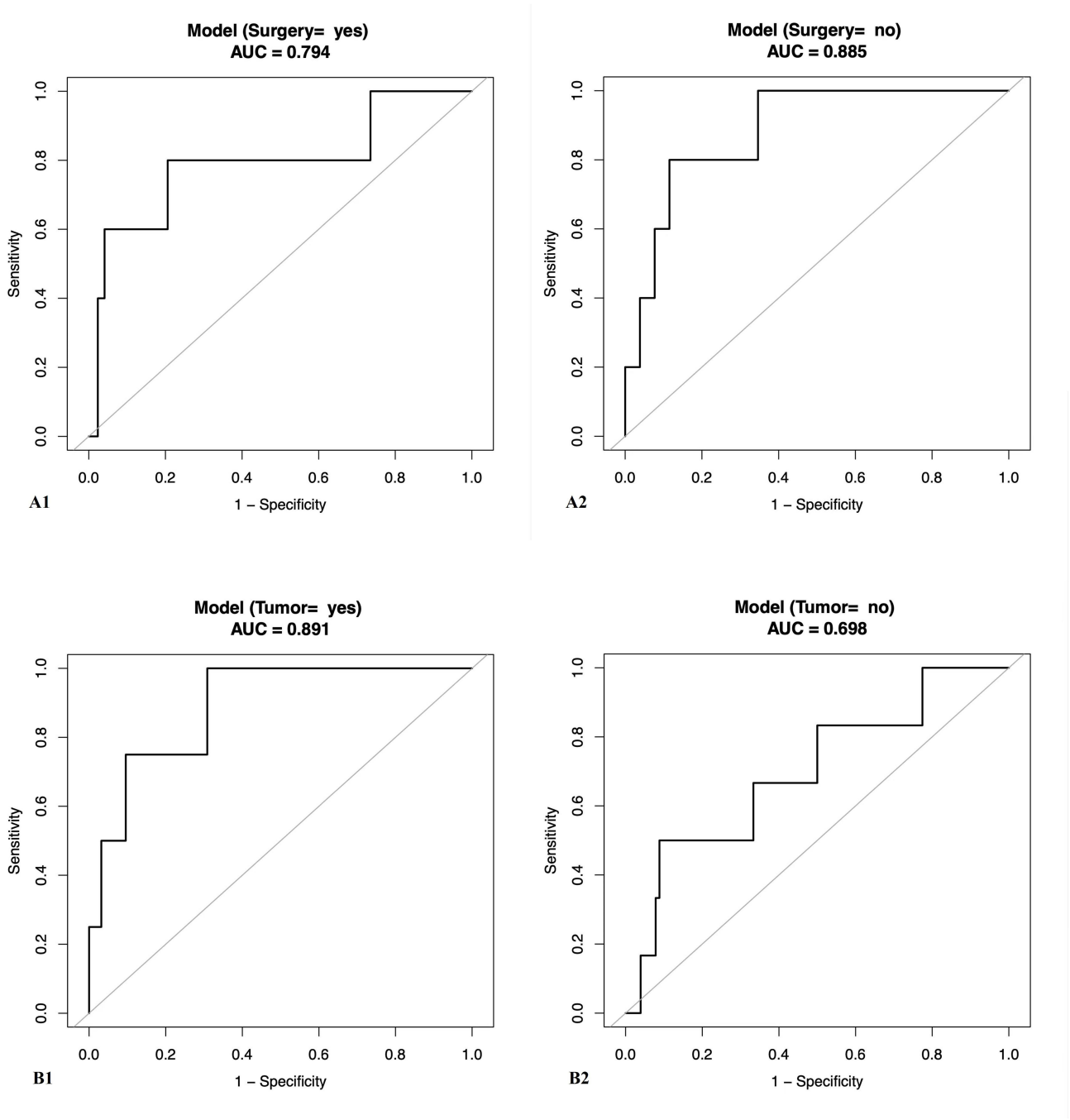

Figure 4 Sensitivity analyses for the mortality-predicting model. (**A**) Surgery (A1) or Non-surgery (A2). (**B**) Tumor (B1) and Non-tumor (B2).
Abbreviation: AUC, Area Under ROC Curve.

unit biomarkers to predict the in-hospital mortality of critically ill patients. Sensitivity analysis in different groups of critically ill patients found that the model built with critical unit biomarkers had good performance in predicting in-hospital mortality in surgery and non-surgery patients as well as tumor and non-tumor patients. This study confirms the feasibility of evaluating patient prognosis on the basis of the critical unit, thus providing a strong complement to the field.

In this study, we focused on the correlation between the pro-inflammatory response and the critical unit. A high inflammation status in critically ill patients is one of the important mechanisms leading to the onset and progression of critical diseases.[3] The results of our study shows that the serum concentration of IL6 was generally increased in critically ill patients, and the median concentration level was 157.9 pg/mL, which revealed that high inflammation status was

common in critically ill patients. The critical unit covers the structural integrity and function of mitochondria and endothelial cells and plays an important role in critical diseases.[2] Mitochondrial fission and mitophagy can eliminate damaged mitochondria.[13] Fission 1 and Parkin are the signature proteins of mitochondrial fission and mitophagy, respectively. Mitochondrial damage can result in the release of mtDNA.[14] Therefore, Fission 1, Parkin, and mtDNA can together comprehensively reflect mitochondrial damage. S1P maintains the integrity of endothelial cells and plays a key role in improving endothelial cell function.[15,16] Endothelial cell damage often leads to abnormal coagulation function.[17] While APTT indicates coagulation function, it also represents the degree of endothelial damage to a certain extent. Therefore, we evaluated the condition of critical units by detecting Fission 1, Parkin, mtDNA, S1P, and APTT.

The relationship between IL6 and mitochondria is one of the important manifestations of the close relationship between the pro-inflammatory response and the critical unit. We found that IL6 was significantly positively correlated with Fission 1 and Parkin, suggesting that mitochondrial fission and mitophagy increased when the pro-inflammatory response was enhanced. This result is also similar to those of other studies. For example, Lee et al[18] reported that treatment of ovarian cancer cells with IL6 caused an increase in mitochondrial division, while Tyrrell et al[19] found that IL6 induced an increase in mitophagy in the cerebral vasculature. As the only organelle with independent DNA besides the nucleus, mitochondria are highly dynamic and maintain homeostasis through continuous fission/fusion and mitophagy.[20,21] Mitochondrial fission and subsequent mitophagy regulate the clearance of damaged mitochondria.[22] This may also explain why an increase in IL6 is closely related to a decrease in mtDNA. Increased IL6 leads to increased mitochondrial fission, which may lead to the differentiation and removal of mutated mtDNA.[23] In contrast, the absence of mitochondrial fission leads to mtDNA accumulation.[24,25] Increased mitochondrial fission selectively removes and degrades damaged mitochondria through Parkin-mediated mitophagy.[13,26] The increase in IL6 level is related to the decrease of mitochondrial transcription factor A (TFAM) protein production, and TFAM plays an important role in mtDNA replication, which means IL6 itself may affect mtDNA level, thus partly explaining our results.[27] Studies have shown that increased mitochondrial fission can induce mtDNA leakage into the cytoplasm, resulting in increased mtDNA levels.[28,29] The above results are not completely contradictory, which may be related to the increase degree in mitochondrial fission. Mitochondrial fission is necessary to eliminate mutated mtDNA, and a certain degree of increase in mitochondrial fission will lead to the decrease of mtDNA; however, an excessive increase in mitochondrial fission will disrupt mitochondrial function and cause an abnormal release of mtDNA.

We also investigated the association of IL6 with endothelial cell damage to further clarify the relationship between the pro-inflammatory response and the critical unit. We found that the increase of IL6 was significantly correlated with the prolonged APTT. While IL6 was not significantly correlated with S1P, it tended to be negatively correlated. Abnormal inflammatory responses in critically ill patients often lead to endothelial cell death and barrier damage. For example, in sepsis, exogenous PAMPs and DAMPs both cause endothelial cell activation and may damage endothelial cell structure and function.[30,31] Coagulation begins with endothelial damage. In theory, endothelial cell damage may lead to the exposure of negatively charged subendothelial collagen, activated platelets and various coagulation factors, and eventually enhanced coagulation, which may be related to our result of the slight shortening of APTT in most critically ill patients.[32] Therefore, APTT, a classic coagulation indicator, is inextricably related to endothelial damage. Another reason to include APTT in the evaluation of endothelial damage is the results of the recent study by Tang et al,[33] who found a significant positive correlation between APTT and endothelial damage in patients infected with severe fever with thrombocytopenia syndrome virus, which may be from heparin-like structures (for example, heparin sulfate) in the endothelium that degrade and release into the blood during sepsis.[34] The pharmacological activity of heparin sulfate in vitro is negligible. However, it is an effective antithrombotic agent in vivo, and its release into the blood can lead to prolonged coagulation time.[35] This result partly explains the increase in IL6 and the prolonged APTT identified in the current study. Another explanation is that endothelial damage promotes the process of thrombosis, and coagulation factors are consumed in large quantities, resulting in abnormal coagulation function. Although IL6 did not have a significant correlation with S1P, there was a negative correlation trend. This downward trend in S1P, an important component of endothelial integrity and function, suggests that increased IL6 may cause endothelial damage. The increase of IL6 is highly correlated with the elimination of damaged mitochondria and the damage of endothelial cells, thus

confirming the close association of IL6 with the critical unit. Few studies regard mitochondria and endothelial cells as a whole and explore the relationship between IL6 and the critical unit. Our study is a contribution to this study field.

The pro-inflammatory response may also have an important effect on the prognosis of critically ill patients. We found that higher levels of IL6 correlated with patient outcome. IL6 is one of the classical pro-inflammatory factors, and its plasma concentration is correlated with the severity of critically ill patients.[36] The inflammatory response is an important feature of critical diseases such as sepsis and is consistent throughout the disease.[3] Excessive release of multiple inflammatory factors leads to inflammatory storms, which play a key role in the occurrence and progression of critical diseases.[3] Numerous studies have been conducted on the predictive value of cytokines for clinical outcomes. For example, a study of critically ill patients with sepsis found that IL6 was more effective than procalcitonin, presepsin, and C-reactive protein in predicting the 30-day mortality in patients.[37] Takahashi et al[38] found that the prognostic value of serum IL6 levels increased with time, up to 7 days. Therefore, serum IL6 levels are a useful prognostic biomarker for critically ill patients. Our study found a certain correlation between IL6 and the prognosis of critically ill patients, mainly showing that the increase of IL6 was significantly negatively correlated with LOS, ICU stay, and MVt. While IL6 was not significantly associated with in-hospital mortality, there was a protective trend against in-hospital mortality. This result is at odds with other studies. One reason for this phenomenon is that inflammation itself is a protective response of the host to infection and tissue damage, which can prevent the spread of pathogens or promote tissue repair, so a certain degree of inflammation may contribute to the recovery of the disease.[39,40] However, an excessive inflammatory response has more negative effects and ultimately leads to poor prognosis. A recent study of sepsis patients by Zeng et al[41] partially confirmed this view, finding that patients with a high inflammatory phenotype were indeed given higher doses of vasoactive drugs and had a higher mortality than patients with a low inflammatory phenotype. Notably, the sample size of patients included in this study was relatively small, so the relationship between IL6 and patient prognosis could not be fully explored. Future studies with a larger sample size and even multi-center cohort studies are required for further exploration. Another possible reason is that higher levels of IL6 indicate a higher severity degree of critical disease, which can draw more attention from clinicians and make patients' families more pessimistic about treatment expectations. These factors may influence how to treat and whether to pursue further treatment. For a complicated reason, the shorter LOS, ICU stay, and MVt appeared.

Given the confirmation of the correlation between IL6 and the prognosis of critically ill patients and considering the close relationship between IL6 and the critical unit, it is necessary to further explore the impact of the critical unit on the prognosis of critically ill patients. Mitochondria and endothelium play an independent role in the occurrence and progression of critical diseases and influence each other. For example, endothelial damage can lead to an enhanced inflammatory response, which affects mitochondrial function. Correspondingly, when mitochondria are damaged, DAMPs, such as mtDNA, are released into endothelial cells, triggering endothelial inflammation.[42] Abnormal release of mtDNA can also inhibit endothelial cell proliferation, and hindering the recognition of mtDNA can improve endothelial cell proliferation.[43] Therefore, it is valuable to integrate mitochondria and endothelium, the critical unit, for evaluation. Some studies are often based on a single aspect of mitochondrial or endothelial indicators to evaluate the prognosis of critically ill patients, and a more comprehensive analysis is required. Therefore, we constructed a predictive model based on critical unit biomarkers to explore the correlation between the critical unit and the prognosis of critically ill patients. The AUC of the model was 0.708, and the sensitivity and specificity were 80% and 65.3%, respectively. This model is only suitable for adult critically ill patients and helpful in comprehensively evaluating the prognosis of such patients. However, more studies are needed to validate the predictive model. Considering that many surgery and tumor patients were included in the study, we conducted a sensitivity analysis to further explore the predictive efficacy of the critical unit for in-hospital mortality in different groups of critically ill patients. We constructed predictive models in critically ill patients with surgery or non-surgery, and patients with tumors or without tumors. We found that the predictive models based on the critical unit biomarkers had good predictive efficacy in these critically ill patients.

Limitations

This study has several limitations. First, this is a single-center prospective study and the sample size was relatively small. Thus, the results need to be verified in a multicenter cohort study with a large sample size. Second, the results of our

study differ from the results of other studies, which may result from the small sample size. We will continue to include more such patients to provide higher-quality evidence for our conclusions. Additionally, IL6 concentrations vary in different stages of the disease. However, our study only included measurements at a single point in time, and further dynamic studies are needed to clarify the relationship between IL6 and the critical unit and between IL6 and prognosis in critically ill patients. In the future, we will dynamically incorporate the test results and relevant data of patients and build a mixed-effect model to analyze the repeated measurement data dynamically. We will also clarify the influence of the disease course and clinical treatment on our study results.

Conclusion

Our study confirmed a significant association between IL6 and the critical unit in critically ill patients, with increased IL6 associated with mitochondrial fission, enhanced mitophagy, and decreased mtDNA. While IL6 was not significantly associated with in-hospital mortality, increased IL6 was associated with shorter LOS, ICU stay, and MVt. The predictive model based on critical unit biomarkers comprehensively evaluated the in-hospital mortality of critically ill patients with good efficacy. Additionally, sensitivity analyses in different critically ill patient populations further confirmed that it is feasible to evaluate patient prognosis using critical unit biomarkers. Our study innovatively integrates mitochondrial and endothelial markers in the form of the critical unit to comprehensively evaluate patient prognosis, which may be a trend in the future assessment of critically ill patients. There are few such studies, and our study may promote the progress of related research.

Data Sharing Statement

All datasets used and analyzed during the current study are available from the corresponding author on reasonable request.

Ethics Approval and Consent to Participate

This study was approved by the ethics committee of Peking Union Medical College Hospital, Beijing, China (Approval No. I-22PJ1072). Written informed consent was obtained from the next of kin of each patient. Our study complies with the Declaration of Helsinki.

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Author Contributions

All authors made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis and interpretation, or in all these areas; took part in drafting, revising or critically reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

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Disclosure

The authors declare that they have no competing interests in this work.

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