Original Article

Urokinase-type plasminogen activator receptor as a predictor of poor outcome in patients with systemic inflammatory response syndrome

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BACKGROUND: Urokinase-type plasminogen activator (uPA) and urokinase-type plasminogen activator receptor (uPAR) are known as important factors, which mediate a variety of functions in terms of vascular homeostasis, inflammation and tissue repair. However, their role in systemic inflammatory response syndrome (SIRS) has been less well studied. This study aimed to test the hypothesis that the abnormalities of fibrinolysis and degradation of extracellular matrix mediated by uPA and uPAR are directly related to the patients with SIRS. We therefore analyzed their role and clinicopathological significance in patients with SIRS.

METHODS: A case-control study was conducted with 85 patients who were divided into two groups according to the diagnostic criteria of SIRS: SIRS group (n=50) and non-SIRS group (n=35). The SIRS group was divided into MODS group (n=26) and non-MODS group (n=24) by their severity, and survival group (n=35) and non-survival group (n=15) by their prognosis. Another 30 healthy adults served as normal controls. uPA and uPAR in plasma were detected by commercial enzyme-linked immunosorbent assay (ELISA) kits.

RESULTS: The plasma level of uPA was lower in the SIRS group than in the non-SIRS group and controls (P<0.001 and P<0.001). It was lower in sepsis patients and the MODS group than in the non-sepsis patients and the non-MODS patients (all P<0.05). However, there was no difference in uPA level between survivors and non-survivors (P>0.05). The plasma level of uPAR increased in the SIRS group compared with the non-SIRS group and controls (P<0.001 and P<0.001). There was a significant elevation of uPAR in sepsis patients, MODS patients and non-survivors as compared with non-sepsis patients, non-MODS patients and survivors respectively (all P<0.05). Plasma uPAR levels were positively correlated with APACHE II score (r=0.575, P<0.001) and SOFA score (r=0.349, P=0.013). AUCs for the prediction of SIRS mortality were 0.67 and 0.51, respectively, for uPA and uPAR.

CONCLUSION: uPAR could be a predictor of poor outcome in patients with SIRS.

KEY WORDS: Systemic inflammatory response syndrome; Multiple organ dysfunction syndrome; Urokinase-type plasminogen activator; Urokinase-type plasminogen activator receptor

World J Emerg Med 2013;4(3):190–195
DOI: 10.5847/ wjem.j.issn.1920–8642.2013.03.006

INTRODUCTION

Systemic inflammatory response syndrome (SIRS) is a generalized inflammatory process of the organism induced by inflammatory or non-inflammatory factors. SIRS is a common problem in acute medical and surgical practice and an important cause of morbidity and mortality. The systemic release of pro-inflammatory cytokines, chemokines, and lipid and vasoactive mediators in SIRS induces endothelial damage and microvascular thrombosis, potentially culminating in disseminated intravascular coagulation

(DIC), acute respiratory distress syndrome (ARDS), and multiple organ dysfunction syndrome (MODS).[1-3]

The activation of plasminogen to plasmin by the plasminogen activators tissue-plasminogen activator (tPA) and urokinase-plasminogen activator (uPA) is a central step in fibrinolysis. While tPA has a relative specific role in coagulation, uPA has been found to regulate cell migration, cell adhesion and cell proliferation, and also be involved in various inflammatory and immune responses.[4,5] Activation of uPA occurs through binding to its receptor (uPAR, CD87) expressed on endothelium and also on activated T cells, granulocytes and macrophages, which leads to local proteolysis and fibrinolysis.[6,7] The involvement of uPA and uPAR in the pathogenesis of tumors[8] and other non-viral diseases such as pancreatitis,[9] Sjögren syndrome, rheumatoid arthritis,[10] acute myocardial infarction,[11,12] human immunodeficiency virus (HIV) infection,[13,14] and common variable immunodeficiency (CVID)[15] has been recently recognized.

Based on their role in inflammation and fibrinolysis, we hypothesized a role for uPA/uPAR in SIRS. In the present study, we examined the plasma levels of uPA/uPAR in different subgroups of SIRS and healthy controls, trying to relate the levels of these parameters to distinct clinical features of SIRS.

METHODS

Patients

Fifty patients with SIRS admitted to the intensive care unit (ICU), Wuhan Central Hospital, Tongji Medical College, Huazhong University of Science and Technology, were included in this study (Table 1). SIRS was defined according to the criteria of the American College of Chest Physicians and the Society of Critical Care Medicine.[16] Patients with hematological systemic disorder, cardiopulmonary resuscitation, malignant tumors or immunodeficiency disease were excluded from the study. The study group comprised 26 men and 24 women with a mean age of 68.2±8.7 years (range, 41–86years). Acute physiology and chronic health evaluation II (APACHE II) score (17.2±5.2) and sequential organ failure assessment (SOFA) score (7.1±2.0) were assessed when the patients were diagnosed with SIRS. There were 27 patients with sepsis, including pneumonia 8, necrotizing fasciitis 3, enterocolitis 4, necrotizing pancreatitis 8, pyelonephritis 2, and vasculitis 2. Twenty patients had organ failures diagnosed according to the criteria of MODS by Bone et al.[17] Seventeen patients died during the observation period in our department. All the other patients survived. In addition, 35 patients from internal medicine without infection or SIRS severed as non-SIRS group. Meanwhile, 30 age-matched and gender-matched healthy individuals were enrolled as healthy controls. The study was conducted according to the ethical guidelines of the hospital, which are consistent with the *Helsinki declaration.*

Table 1. Characteristics of patients with SIRS

Variables	Value
Number of patients	50
Average age (yr)	68.2±8.7
Age range (yr)	41–86
Sex (male/female)	26/24
APACHE II score	17.2±5.2
SOFA score	7.1±2.0
Origins of SIRS	
Trauma	10
Pneumonia	8
Necrotizing fascitis	3
Cerebral infarction	9
Enterocolitis	4
Necrotizing pancreatitis	8
Pyelonephritis	2
Unstable angina	4
Vasculitis	2

Experimental procedure

Venous blood samples were collected via routine venipuncture. All samples were placed in tubes containing tri-sodium citrate, immediately centrifuged, and stored at −80 °C. The plasma levels of uPA and uPAR were measured by ELISA (ADI, America). Arterial blood gasses (ABG) were only measured by the attending physician when necessary.

Statistical analysis

All data were expressed as means±SD. The two groups were compared using the independent-samples *t* test. Pearson's product-moment correlation coefficient analysis was used to evaluate the relationship between the APACHE II score, SOFA score and the levels of uPA/uPAR. Receiver operating characteristic (ROC) curves were drawn and the area under the curve (AUC) was calculated to visualize and compare the performance of uPA and uPAR. All *P* values less than 0.05 were considered statistically significant. The data were analyzed using SPSS version 17.0 (SSPS Inc., Chicago, IL). The figures were drawn using GraphPad Prism version 5.0 (GraphPad Software, San Diego, CA).

RESULTS

The plasma level of uPA was found to be significantly decreased in patients with SIRS compared with the non-SIRS group and controls ($P<0.001$ and $P<0.001$; Table

2, Figure 1), and the patients with SIRS had significantly higher plasma uPAR values than the non-SIRS patients and controls (P<0.001 and P<0.001; Table 2, Figure 1).

The SIRS patients were divided into sepsis group (n=27) and non-sepsis group (n=23). The plasma levels of uPA were significantly decreased in the sepsis group as compared with the non-sepsis group (P=0.001, Table 2, Figure 2). In addition, the levels of uPAR observed in the sepsis group were higher than those in the non-sepsis group (P=0.020, Table 2, Figure 2).

Changes in plasma uPA and uPAR levels in the MODS patients (n=20) and non-MODS patients (n=30) are shown in Table 2 and Figure 3. The plasma level of uPA decreased more markedly in the MODS patients than in the non-MODS patients (0.446±0.208 vs. 0.594±0.269, respectively, P=0.042). The plasma level of uPAR elevated more significantly in the MODS patients than in the non-MODS patients (4.395±0.967 vs. 3.609±1.186, respectively, P=0.017).

There was no difference in uPA level between survivors (n=33) and non-survivors (n=17) (0.526±0.246 vs. 0.552±0.277, respectively, P=0.783, Table 2, Figure 4). However, there was a significant elevation of uPAR in the non-survivors as compared with survivors (5.482±1.211 vs. 3.635±1.037, respectively, P=0.013, Table 2, Figure 4).

Correlation of plasma uPAR with APACHE II score and SOFA score is shown in Figure 5. Plasma uPAR levels were correlated positively with APACHE II score (r=0.575, P<0.001) and SOFA score (r=0.349, P=0.013). But no correlation was observed between uPA and any of these parameters (P>0.05, data not shown).

The ROC curves about the evaluation of uPA , uPAR, APACHE II score and SOFA score which are used to predict mortality are shown in Figure 6. uPAR was found to be much better to predict mortality than uPA. Their ROC curve (AUC) values were 0.67 and 0.51, respectively. AUCs for the prediction of mortality were 0.74 and 0.70, respectively, for APACHE II score and SOFA score.

Table 2. Comparison of plasma levels of uPA and uPAR among different groups (mean ±SD)

Groups	No. of patients	uPA (ng/mL)	uPAR (ng/mL)
Controls	30	0.817±0.227	1.597±0.680
Non-SIRS	35	0.761±0.269	1.806±0.578
SIRS	50	0.535±0.255$^{*\bullet}$	3.923±1.160$^{*\bullet}$
Non-sepsis	23	0.662±0.233*	3.514±0.977*
Sepsis	27	0.426±0.223$^{*\#}$	4.272±1.206$^{*\#}$
Non-MODS	30	0.594±0.269*	3.609±1.186*
MODS	20	0.446±0.208$^{*\Delta}$	4.395±0.967$^{*\Delta}$
Non-survivors	17	0.552±0.277*	5.482±1.211*
Survivors	33	0.526±0.246$^{*\blacktriangle}$	3.635±1.037$^{*\blacksquare}$

Comparison with the controls, $^{*}P$<0.05; comparison between the SIRS group and non-SIRS group,$^{\bullet}P$<0.05; comparison between the sepsis group and non-sepsis group, $^{\#}P$<0.05; comparison between the MODS group and non-MODS group, $^{\Delta}P$<0.05; comparison between the non-survivors and survivors, $^{\blacktriangle}P$>0.05; comparison between the non-survivors and survivors, $^{\blacksquare}P$<0.05.

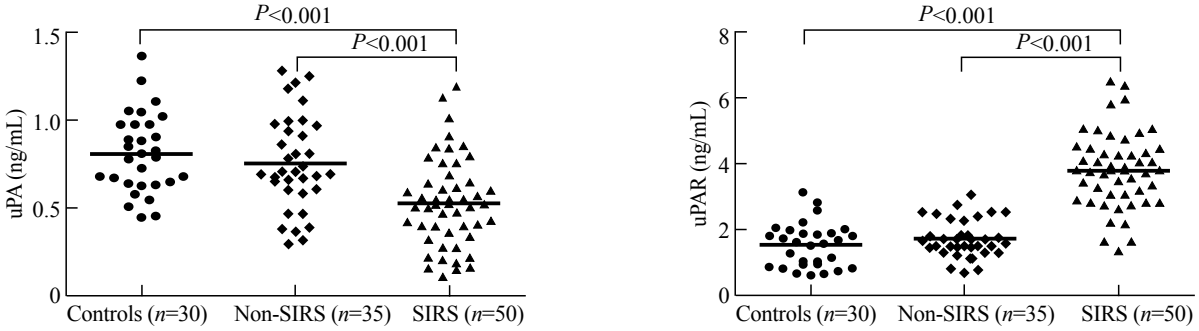

Figure 1. Plasma levels of uPA and uPAR in controls (n=30), non-SIRS group (n=35) and SIRS group (n=50) . Bars represent the means of concentrations.

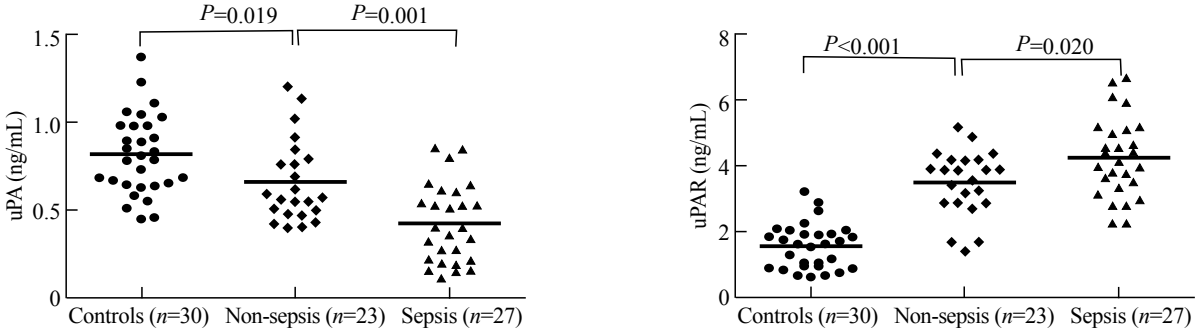

Figure 2. Plasma levels of uPA and uPAR in controls (n=30), non-sepsis group (n=23) and sepsis group (n=27). Bars represent the means of concentrations.

DISCUSSION

SIRS is an organic reaction generally associated with an infection (sepsis) or any situation that implies intense tissue damage with well-defined diagnostic criteria.[18,19] García-Fernández et al[20] have demonstrated that patients with SIRS, irrespective of the origin (infectious or noninfectious), show signs of intense endothelial damage and hypercoagulability throughout the process. The endothelial inflammation, together with a disturbance of hemostatic and tissue perfusion, leads to a progressive establishment of multiple organ dysfunction syndrome (MODS).[21]

uPA is considered to be one of the earliest mediators of fibrinolysis. It activates plasminogen into plasma, which in turn degrades fibrin and prevents its extracellular deposition. This process might be dysregulated in several diseases involving inflammation and tissue repair.[15] In the present study, the plasma level of uPA in the SIRS group was decreased more significantly than in the non-SIRS group and controls (P<0.001 and P<0.001; Table 2, Figure 1). Qiu et al[22] identified that in a clinical situation, frequently characterized by fibrin occlusion of the microcirculation, the ability of polymorphonuclear leukocytes (PMN) to express uPA activity is diminished or absent. Yu et al[23] demonstrated that dramatically increased plasminogen activator inhibitor type 1 (PAI-1) levels during staphylococcal infection led to a reduction of metabolically active uPA levels. It has been well documented that uPA is required for generation of an adequate inflammatory response. Lack of uPA impedes leukocyte recruitment, resulting in uncontrolled infection and death. However, increased total uPA levels in the infected organs have been reported in animal models of other infections.[24] This may reflect, in part, at least, the time-point in the evolution of the SIRS episode at which

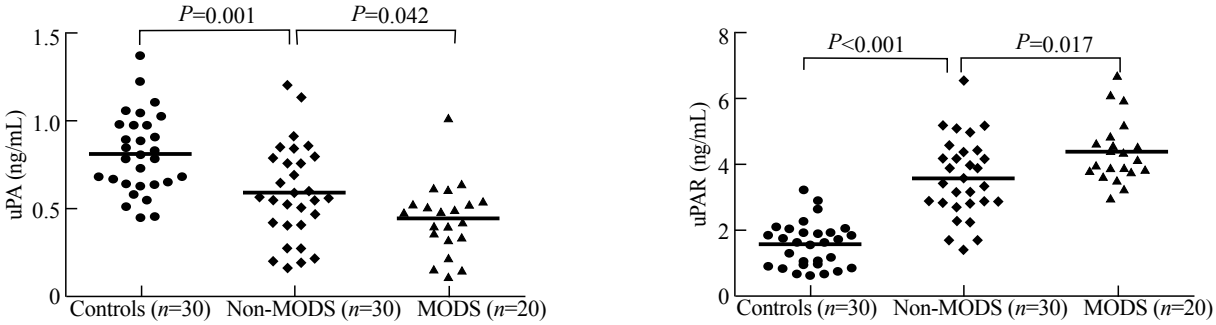

Figure 3. Plasma levels of uPA and uPAR in controls (n=30), non-MODS group (n=30) and MODS group (n=20). Bars represent the means of concentrations.

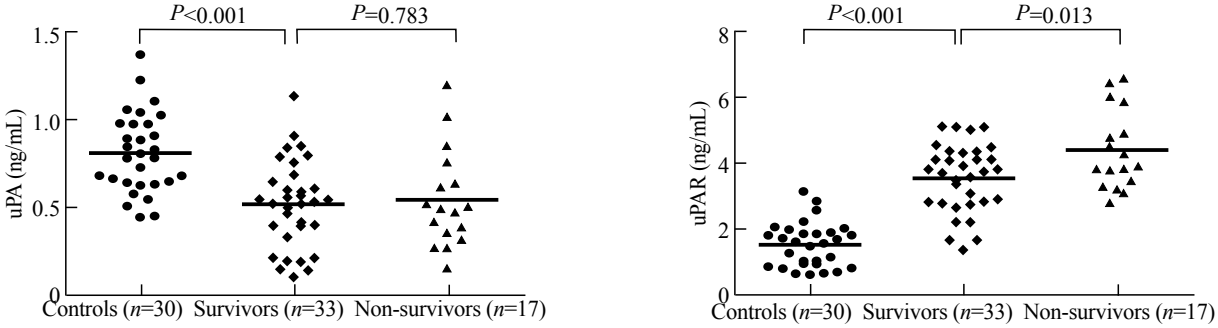

Figure 4. Plasma levels of uPA and uPAR in controls (n=30), survivors (n=33) and non-controls group (n=17). Bars represent the means of concentrations.

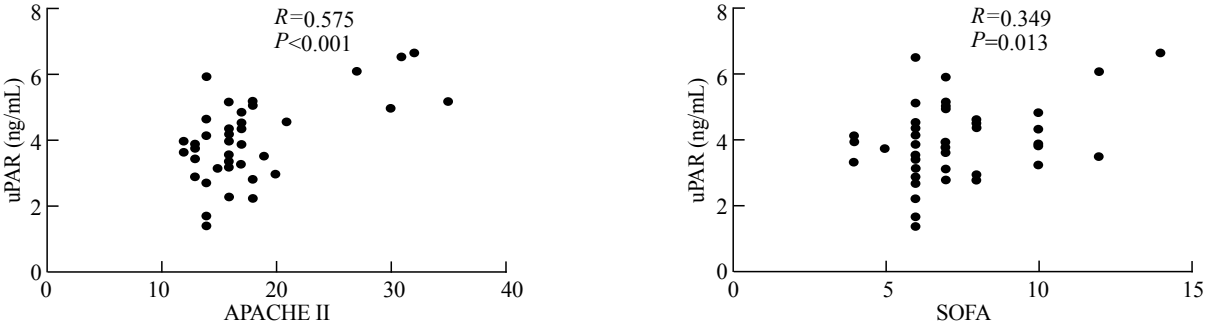

Figure 5. Correlation between plasma level of uPAR and APACHE II, SOFA score during clinical course.

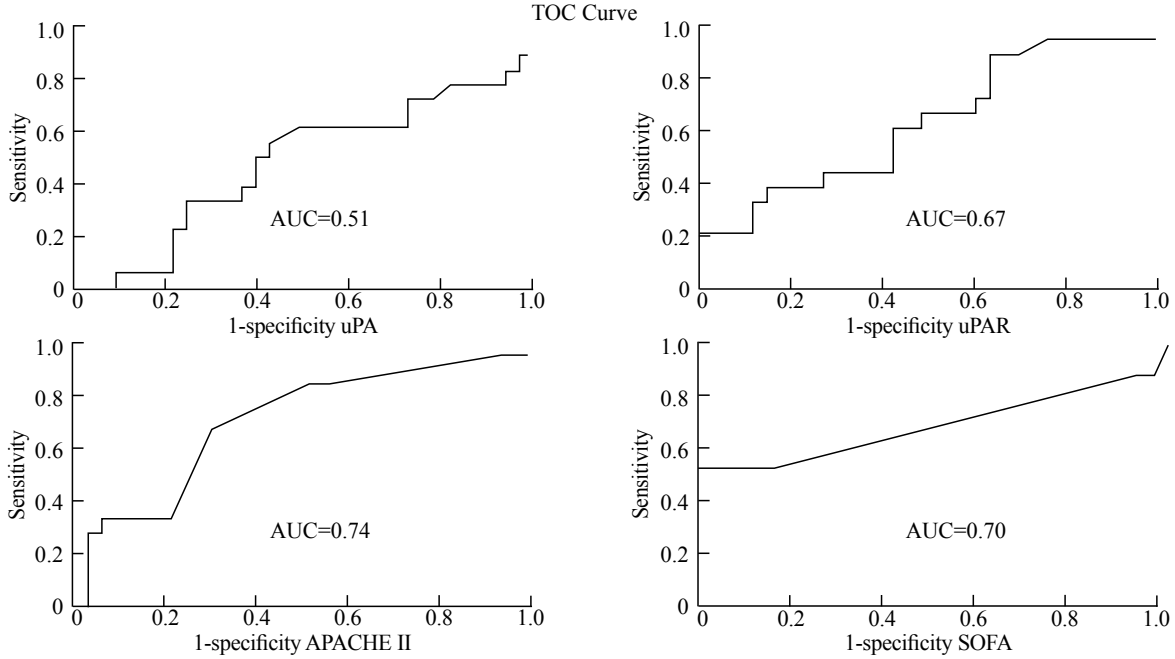

Figure 6. The ROC curves for prediction of mortality by uPA, uPAR, APACHE II score and SOFA score on the admission day in ICU.

the patients were studied. The time of their entry into the intensive care unit will be different from patient to patient. This possibility is currently under investigation.

uPAR is a multifunctional protein involved in different inflammatory responses, including cell-associated proteolysis, cell adhesion, chemotaxis, cell migration, and proliferation.[25-27] Increased uPAR concentrations were found in patients with rheumatoid arthritis and primary Sjögren's syndrome, and were found to be closely related to their poor prognosis.[28] A multicenter prospective study showed that uPAR was also elevated during pneumococcal bacteremia, and had a predictive value in the early stage of the disease.[29] In this study, we found an increased plasma level of uPAR in patients with SIRS, demonstrating an abnormality in the uPA-mediated fibrinolysis pathway. The level of uPAR was remarkably higher in sepsis patients, MODS patients and non-survivors. Plasma uPAR levels correlated positively with APACHE II score and SOFA score, and uPAR were found to be much better to predict mortality than uPA. Indeed, retrospective studies[30,31] have shown that the measurement of soluble urokinase-type plasminogen activator receptor (suPAR) levels in serum, tissue and urine of patients predicts disease severity (i.e. the death of the patients).

However, several limitations in this study are worth considering. The small number of outcomes (deaths) limits the study's ability to make definitive conclusions. ABG analyses were only performed when judged necessary by the attending physician, which could have led to underestimation of the bicarbonate score of the APACHE II model and the respiration score of the SOFA model. Nevertheless, results from the analysis omitting all ABG measurements showed only minor changes in the AUCs of the models. The fact that not all samples were collected directly at admission might weaken the results. Our results may apply only to patients suspected of required hospitalization in an ICU directly at admission, which do not include community-acquired infections, and, thus, may not be valid in these patients. Finally, patients with dementia or other mental diseases were excluded from this study (due to the demand for an informed written consent), thus, these results cannot be extrapolated to this important group of patients.

In conclusion, our results indicate that uPAR could be a predictor of poor outcome in patients with SIRS.

ACKNOWLEDGMENTS

We are grateful to the staff of the intensive care unit (ICU), Wuhan Central Hospital, Tongji Medical College, Huazhong University of Science and Technology for providing samples and clinical information. We wish to thank all the patients and volunteers who participated in this study.

Funding: None.
Ethical approval: The present study was approved by the Ethical Committee of Wuhan Central Hospital, Tongji Medical College, Huazhong University of Science and Technology, Wuhan, China.
Conflicts of interest: The authors declare that there is no conflict

of interest.

Contributors: Wu XL proposed the study,analyzed the data and wrote the first draft. All authors contributed to the design and interpretation of the study and to further drafts.

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Received February 10, 2013
Accepted after revision June 15, 2013