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Original Paper

Serum Cystatin C Predicts AKI and the Prognosis of Patients in Coronary Care Unit: a Prospective, Observational Study

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Key Words
Cystatin C • Acute Kidney Injury • Prognosis • Coronary Care Unit

Abstract
Background/Aims: Acute Kidney Injury (AKI) is a serious clinical state associated with high morbidity and mortality, particularly in critical ill patients. We investigated the hypothesis that serum Cystatin C (sCysC) is a good predictor for AKI and may affect the short-term prognosis of coronary care unit (CCU) patients. ***Methods:*** In this prospective, observational study, we screened 412 adults admitted to the CCU from January 1, 2014 to June 1, 2015 at Zhongnan Hospital of Wuhan University. Serum samples were obtained at the time of admission, and sCysC was quantified through nephelometry. AKI was defined based on KDIGO-AKI criteria. After the patients' hospital discharge, the survivors in this study were followed up for up to 2 years. The primary endpoint was the incidence of AKI stratified by severity stage, while the second endpoints included 2-year mortality, rehospitalization and failure in renal recovery rates, as well as the progression of AKI to CKD. ***Results:*** According to the KDIGO-AKI criteria, AKI occurred in 130 (31.6%) patients. After multivariate adjustments, the highest quartile of sCysC was associated with a 9-fold increased risk of incident AKI compared with the lowest quartile. For predicting AKI, sCysC [area under the receiver operating characteristic curve (AUC=0.842)] outperformed β2-micro globulin (AUC=0.813) and the clinical model (AUC=0.777), and a cutoff of 1.255 mg/L yielded good sensitivity and specificity. After a median 19.8-month follow-up, 112 (27.2%) patients died within 2 years after admission. The sCysC independently predicted the risk of 2-year mortality [adjusted odds ratio (OR), 4.955; 95% confidence interval (95% CI), 2.853 to 8.603] and rehospitalization (OR, 3.128; 95% CI, 2.011 to 4.867), as well as renal recovery failure (OR, 3.618, 95% CI, 1.753 to 7.463). ***Conclusions:*** Serum CysC is a strong predictor of AKI and the short-term prognosis of CCU patients.

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Introduction

Acute kidney injury (AKI) is a devastating disease that affects patients, particularly those in intensive care units, and is associated with significantly increased morbidity and mortality rates [1-4]. A major barrier to improving the clinical outcomes of AKI patients is the inability to identify high-risk patients to provide immediate interventions at an early stage. Intense contemporary research has focused on validating novel biomarkers to predict AKI earlier than serum creatinine (Scr). Several biomarkers have been identified, such as neutrophil gelatinase-associated lipocalin, insulin-like growth factor-binding protein 7, tissue inhibitor of metalloproteinases-2, cystatin C (CysC) and so on [5-8]. However, the clinical utilization of these biomarkers in predicting AKI and long-term prognosis for CCU patients is limited [9, 10].

CysC is a 13-kD cysteine protease inhibitor synthesized in all types of nucleated cells at a steady state. It is freely filtered by the glomerulus, not secreted by renal tubules, and completely metabolized at the level of renal tubules [11]. Importantly, CysC is readily measured using clinical laboratory platforms and does not increase with urinary tract infection or in chronic non-renal diseases. These capabilities have made it an attractive marker for estimating glomerular filtration [12-15]. However, the receipt of glucocorticoids, thyroid function, inflammation and obesity, rather than sex or age, may affect the measurement of serum CysC[16-18]. Recent studies have further suggested that serum CysC (sCysC) may be an early predictor for AKI and the prognosis of patients in cardiac surgery [19-22], intensive care [23, 24], radiocontrast administration settings [25] and emergency departments [26].

In this study, we aimed to ① determine the accuracy of sCysC in predicting AKI in CCU patients, ② determine the relationship between sCysC and the severity of AKI and the length of hospital stay, and ③ determine the association between sCysC and the clinical outcomes of AKI patients, including all-cause mortality, rehospitalization, failure in renal recovery and progression of AKI to chronic kidney disease (CKD).

Materials and Methods

Study Design

We performed a prospective observational study involving 412 adult patients (23 to 97 years old) admitted to the CCU at Zhongnan Hospital of Wuhan University from January 1, 2014 to June 1, 2015. The following inclusion criteria were applied: (1) a CCU stay$\geqslant$48 hours, (2) at least three serum creatinine measurements during the hospital stay, and (3) at least one serum creatinine measurement over a 6-month period before admission. The exclusion criteria included pre-existing peritoneal dialysis or maintenance hemodialysis, urinary tract infection or obstruction, cancers， and a history of renal transplantation.

AKI was defined according to the KDIGO Clinical Practice Guidelines for AKI based on serum creatinine criteria. Preexisting CKD was defined as preadmission eGFR<60 ml/min per 1.73 m^2, which was calculated according to one serum creatinine measurement over a 6-month period before admission.

This study was approved by the Ethics Committee of Zhongnan Hospital of Wuhan University.

Data Collection

The following data were collected: (1) demographic characteristics,(2) preexisting medical conditions before CCU admission, such as hypertension, diabetes and CKD, (3) special medication administration before admission, for example, treatment with diuretics, angiotensin converting enzyme inhibitors/ angiotensin II type I receptor blockers (ACEI/ARB) and aspirin, (4) primary reasons for CCU admission, (5) routine biochemistry tests upon admission (blood samples were collected once immediately after CCU admission (before any in-hospital treatment) and then at the time of routine morning sampling for clinical care purposes until discharge; serum creatinine levels were measured upon admission and at least every other day for the first 3 days, and every three days thereafter), and (6) clinical outcomes, including the duration of hospitalization and the serum creatinine measurement before discharge. Acute Physiology and Chronic

Health Evaluation II (APACHEII) scores were used for all eligible participants to evaluate the severity of their diseases. This risk stratification method is a widely accepted tool used to evaluate the prognoses of adult patients in ICUs [27].

After hospital discharge, a 2-year follow-up examination was performed through medical records review or telephone interview, as needed, including 2-year mortality rate (cause and date of death), rehospitalization within 2 years and serum creatinine measurements taken 2 years after admission.

Biomarker Measurements

The sCysC blood samples were obtained immediately after the CCU admission and before any in-hospital treatment. All sCysC measurements were performed in the central laboratory of Zhongnan Hospital. The laboratory investigators were blinded to the sample sources and clinical outcomes. The concentration of sCysC was measured through nephelometry using a standardized clinical laboratory platform (BN ProSpec II; Siemens Healthcare Diagnostics, Marburg, Germany), according to the manufacturer's recommendations.

Outcome Definitions

The primary outcome was the incidence of AKI stratified by the severity stage, according to the KDIGO Clinical Practice Guidelines for AKI based on serum creatinine criteria [28]. Preexisting CKD was diagnosed if the patients were diagnosed with CKD or if the eGFR was less than 60 ml/min per 1.73 m^2 before hospital admission. eGFR was estimated through Cockcroft-Gault equations [29]. Secondary outcomes included: (1) all-cause mortality from admission to the endpoint of 2-year follow-up; (2) rehospitalization within 2 years (number of patients who were discharged from the hospital and readmitted to any hospital within 2 years); (3) failure in renal recovery defined as a composite end point of being alive at discharge and a discharge-SCr exceeded 25% of the baseline (preadmission value); and (4) progression of AKI to CKD defined as an eGFR<60 ml/min per 1.73 m^2 for >3 months after AKI in patients without preexisting CKD.

Statistical Analyses

SPSS 22.0 software was used for all analyses. Descriptive analyses are reported as the means ± SD or median (interquartile range) for continuous variables and proportions for categorical variables. To compare continuous variables across groups, we used a two-sample t test or a Mann–Whitney U test. Pearson's chi-squared test (χ^2) was used to compare categorical variables. To measure the sensitivity and specificity of sCysC at different cut-off values, a conventional ROC curve was generated. Spearman's correlation coefficients were calculated between the sCysC and clinical parameters. We determined the adjusted odd ratios for AKI and 2-year prognosis through multivariable logistic regression analysis. The risk factors for 2-year mortality and rehospitalization, which were assessed through univariate Cox logistic regression hazard analysis, and statistically significant variables, which were identified through univariate hazard analysis, were included in the multivariate analysis by applying multiple logistic forward Cox regression analysis to obtain the independent predictors of 2-year survival. Cumulative survival curves, as a function of time, were generated through Kaplan-Meier analysis and compared with log-rank tests. In these analyses, sCysC was modeled both as a categorical variable (categorized into quartiles) and as a continuous variable. All statistical tests were two-tailed; and a value of $P<0.05$ was considered to be statistically significant.

Results

Subject characteristics

From January 1, 2014 to June 1, 2015, a total of 412 [135 (32.8%) male and 277 (67.2%) female] adult CCU patients were screened. The mean subject age was 68 years. The primary diagnoses for these patients were acute coronary syndrome (268 patients, 65.0%), chronic coronary artery disease (81 patients, 19.7%), acute decompensated heart failure (ADHF) (16 patients, 3.9%), arrhythmias (32 patients, 7.8%) and others (15 patients, 3.6%). For the preexisting medical conditions, 208 patients (50.5%) had comorbidities associated with hypertension, and 142 patients (34.5%) had diabetes. Moreover, 59 patients (14.3%) had a history of CKD before CCU admission. Furthermore, 201 (48.8%) CCU patients

Table 1. Characteristics of CCU patients on admission. Continuous variables were expressed as mean±SD or median (25th percentile – 75th percentile, interquartile range). Categorical variables were expressed as a number (%). BMI, body mass index; ADHF, Acute decompensated heart failure; ACEI, angiotensin converting enzyme inhibitors; ARB, angiotensin II type I receptor blockers; CAG, coronary angiography; β2-MG, β2-micro globulin; hs-CRP, High Sensitive C-Reactive Protein; TSH, thyroid stimulating hormone; NT-proBNP, N-terminal pro-B-type natriuretic peptide; LVEF, left ventricular ejection fraction; APACHEII, Acute Physiology and Chronic Health Evaluation II. [a]Defined as preadmission eGFR<60 ml•min^{-1}•1.73m^{-2}; [b]Preadmission eGFR was calculated by Cockcroft-Gault equations

Characteristic	Total（n=412）	No AKI（n=282）	AKI（n=130）	P Value
Demographic variables				
Age, yr	68.2±13.1	65.5±13.1	68.2±13.1	<0.001
Gender, male (%)	135（32.8）	93（33.0）	42（32.3）	0.893
BMI, kg/m^2	23.8±3.4	23.8±3.5	23.7±3.3.2	0.870
Preexisting clinical conditions				
CKD[a], n（%）	59（14.3）	17（6.0）	42（32.3）	0.001
Hypertension, n（%）	208（50.5）	118（41.8）	90（69.2）	<0.001
Diabetes, n（%）	142（34.5）	79（28.0）	63（48.5）	<0.001
Primary causes of admission				
Acute coronary syndrome, n (%)	268 (65.0)	179 (63.5)	89 (68.5)	0.325
Chronic coronary artery disease, n (%)	81 (19.7)	61 (21.6)	20 (15.4)	0.121
ADHF, n (%)	16 (3.9)	5 (1.8)	11 (8.5)	0.001
Arrhythmias, n (%)	32 (7.8)	23 (8.2)	9 (6.9)	0.665
Other, n (%)	15 (3.6)	14 (4.9)	1 (0.7)	0.035
Preadmission medication				
Aspirin, n（%）	366（88.8）	257（91.1）	109（83.8）	0.048
ACEI/ARB, n（%）	197（47.8）	138（48.9）	59（45.4）	0.379
Loop diuretics, n（%）	129（31.3）	73（25.9）	56（43.1）	<0.001
Preadmission renal function				
Serum creatinine, umol/L	83.1±17.1	70.9±15.4	108.4±12.9	<0.001
eGFR[b], ml/min per 1.73m^2	76.8±12.1	86.3±10.7	57.1±15.3	<0.001
CAG during hospitalization, n (%)	201 (48.8)	161(57.1)	40 (30.7)	<0.001
Urine output of 24 hours, ml	1000.0 (800.0,1200.0)	1000.0 (800.0,1200.0)	1000.0 (765.0,1300.0)	0.588
Characteristics on admission				
Systolic BP, mmHg	127.7±24.8	129.5±19.2	123.7±23.5	0.026
Diastolic BP, mmHg	74.8±15.7	77.1±12.0	70.0±10.9	<0.001
Mean arterial pressure, mmHg	92.4±17.3	94.6±13.0	87.8±13.4	<0.001
Leucocyte,×10^9/L	9.0±2.0	8.9±2.8	9.2±2.3	0.55
Hemoglobin, g/dL	123.0±22.6	130.0±18.6	109.1±23.4	<0.001
Serum Cystatin C, mg/L	1.2 (0.9, 1.7)	1.0 (0.8, 1.2)	1.7 (1.4, 2.4)	<0.001
β2-MG, ug/L	2742.2 (2010.7,4841.4)	2324.4 (1881.2,3114.7)	5090.3 (3135.0,7414.5)	<0.001
Serum bicarbonate, mmol/L	21.8±2.9	20.1±3.1	22.7±4.5	0.333
Serum albumin, g/L	37.3±4.5	38.1±4.1	35.7±5.0	<0.001
Blood sugar, mmol/L	6.6±1.7	6.5±1.9	6.9±1.3	0.477
Serum sodium, mmol/L	138±5.1	138±4.8	137±5.7	0.450
Serum potassium, mmol/L	3.9±0.6	3.9±0.5	4.1±0.7	0.001
Serum cholesterol, mg/dl	4.1±0.9	4.2±0.9	4.0±1.0	0.085
Serum triglyceride, mg/dl	1.6±1.3	1.7±1.3	1.5±1.2	0.243
hs-CRP, mg/L	19.3±7.6	16.8±7.3	24.7±7.8	0.060
Thyroxine, nmol/L	11.4±9.0	11.7±9.9	10.6±6.7	0.364
TSH, mIU/L	4.4±1.0	3.4±0.9	10.7±0.7	0.388
Troponin I, ng/mL	6.9±3.2	6.8±3.0	7.1±3.8	0.844
NT-proBNP, pg/mL	4790(1004,15845)	4710(891,16097)	5958(996,15866)	0.007
LVEF, %	58±13.3	59±12.6	54±14.3	0.006
APACHEII, points	7.9±2.7	6.8±1.8	10.4±2.2	<0.001
Length of hospital stay, day	13±7.6	11±5.6	16±9.9	<0.001

underwent coronary angiography during their hospitalization.

Serum CysC as a predictor for the primary end point

In this study, of the 412 CCU patients, AKI occurred in 130 (31.6%) patients. According to the KDIGO criteria, 72 (55.4%) patients were AKI stage I, 37 (28.5%) patients were AKI stage II, and 21 (16.1%) patients were AKI stage III. We further compared patients with and without AKI. Compared with the non-AKI patients, the AKI patients were older, had more comorbidities, had higher levels of serum CysC, β2-MG, serum potassium, and NT-proBNP, and had lower levels of MAP, serum albumin and hemoglobin. AKI occurred more frequently in the subjects with preexisting CKD and those who received aspirin and diuretics prior to admission. Moreover, the AKI patients had longer hospital stays and higher APACHEII scores compared with the non-AKI patients (Table 1). Based on Spearman's correlation coefficient, we found a positive correlation between sCysC and the length of hospital stay (r=0.320, *P*=0.015) and the APACHEII scores (r=0.440, *P*<0.001). Nevertheless, the use of renin-angiotensin-aldosterone system inhibitors (ACEI/ARB) before admission did not affect the incidence of AKI in the patients. Furthermore, except for ADHF, the

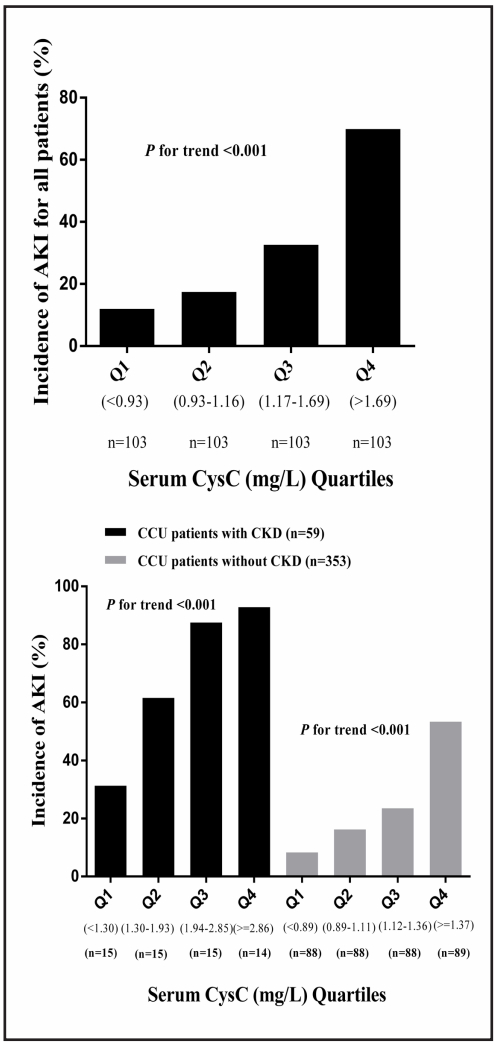


Fig. 1. Quartiles of serum CysC on the first day of admission and the incidence of AKI in all patients and patients with or without preexisting CKD. Pearson Chi-Square Values for all CCU patients, patients with preexisting CKD and patients without preexisting CKD were 91.907 (P<0.001), 20.018 (P<0.001), and 55.303 (P<0.001), respectively.

Table 2. Multivariate logistic regression analyses of sCysC as a predictor for AKI. [a]Adjusted for age, gender, BMI, hypertension, diabetes, serum CysC level, mean arterial pressure, hemoglobin, serum albumin, NT-proBNP, APACHEII, preexisting CKD, treatment with loop diuretics, treatment with ACEI/ARB, and treatment with aspirin. OR, odds ratio; 95% CI, 95% confidence interval

sCysC (mg/L) on the first day of admission	Unadjusted OR	Adjusted OR[a]	95% CI	P Value
All study participants (n=412, 103 per quartile)				
Quartile 1（<0.93）	1.0 (referent)	1.0 (referent)		
Quartile 2（0.93-1.17）	1.681	1.959	1.404 to 2.276	0.004
Quartile 3（1.18-1.69）	2.200	2.346	1.622 to 4.907	0.024
Quartile 4（>1.69）	9.866	9.592	4.413 to 20.851	<0.001
Patients with preexisting CKD (n=59, 14-15 per quartile)				
Quartile 1（<1.30）	1.0 (referent)	1.0 (referent)		
Quartile 2（1.3-1.93）	2.444	1.032	1.019-1.046	<0.001
Quartile 3（1.94-2.86）	6.910	4.414	2.631-9.902	0.031
Quartile 4（>2.86）	28.667	27.354	13.350-55.057	0.035
Patients without preexisting CKD (n=353, 88-89 per quartile)				
Quartile 1（<0.89）	1.0 (referent)	1.0 (referent)		
Quartile 2（0.89-1.11）	1.316	1.445	0.166-1.189	0.106
Quartile 3（1.12-1.37）	1.824	1.661	0.493-2.284	0.880
Quartile 4（>1.37）	5.214	4.696	2.331-9.459	<0.001

incidence of AKI was not related to any other primary diagnoses for admission to the CCU. No significant difference was found in the levels of leucocyte, hs-CRP or thyroid hormones between the AKI and non-AKI patients, and these factors have been shown to impact the level of sCysC in previous studies. sCysC levels were associated with incident AKI in all patients, regardless of preexisting CKD in this study (Fig. 1). These results were further confirmed through the multivariate analyses. After adjusting for the clinical variables, sCysC appeared to be the most powerful predictor for AKI patients with and without preexisting CKD conditions. The highest quartile of sCysC on the first day of admission was associated with a 9-fold increased risk of incident AKI compared with the lowest quartile (Table 2). When sCysC was analyzed as a continuous variable, higher sCysC concentrations were also associated with the development of AKI (OR, 6.156, 95%CI 3.638 to 10.418, *P*<0.001) in a multivariable model (Table 3).

Table 3. Multivariate logistic regression analyses: Predictors of AKI for CCU patients (n=412). [a]Adjusted for age, gender, BMI, hypertension, diabetes, preexisting CKD, serum CysC, mean arterial pressure, hemoglobin, serum albumin, NT-proBNP, APACHEII, treatment with loop diuretics, treatment with ACEI/ARB, and treatment with aspirin; OR, odds ratio; 95% CI, 95% confidence interval. Anemia was defined as the level of hemoglobin was less than 120 g/dL for men or 110g/dL for women

Variables on admission	Unadjusted OR	Adjusted OR[a]	95% CI	P Value
Age (Continuous variables)	1.060	1.025	1.002 to 1.048	0.033
sCysC (Continuous variables)	8.373	6.156	3.638 to 10.418	<0.001
Preexisting CKD (yes versus no)	6.757	3.008	1.353 to 6.689	0.007
Anemia (yes versus no)	7.608	4.268	2.283 to 7.977	<0.001
APACHEII (Continuous variables)	1.376	1.171	1.061 to 1.293	0.002

Performance of serum CysC for predicting AKI in subgroup analyses

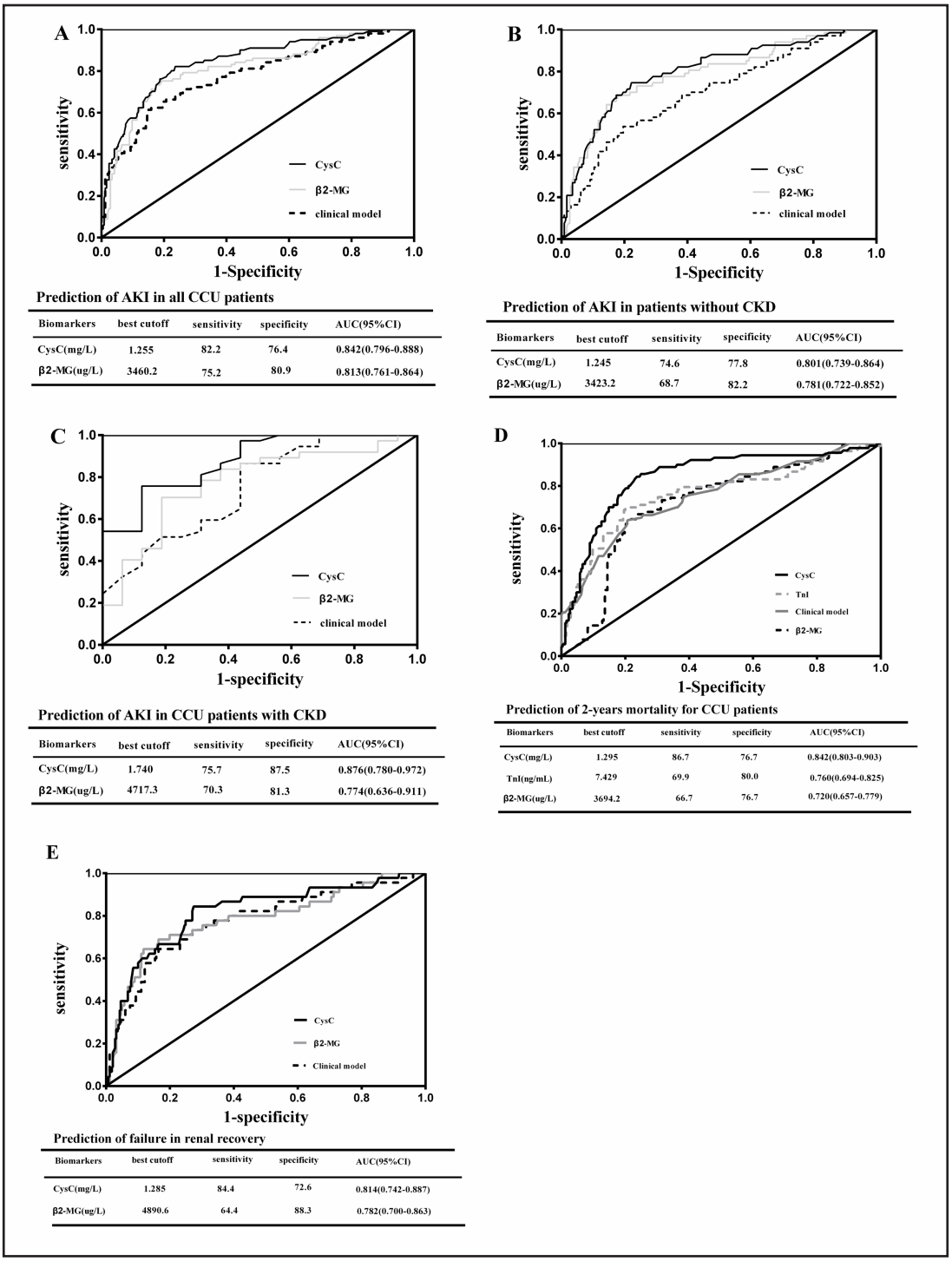
For predicting AKI, the area under the receiver operating characteristic curve (AUC) of sCysC on the first day of admission for all participants in this study was 0.842. A cutoff of 1.255 mg/L yielded good sensitivity (82.2%) and specificity (76.4%) (Fig. 2A). A positive correlation was detected between the elevated level of sCysC (⩾1.255 mg/L) and the severity of AKI (r=0.502, *P*<0.001) based on Spearman's correlation coefficient. The AUCs for sCysC in the subgroups of AKI with and without preexisting CKD were 0.876 and 0.801, respectively, which were greater than those for β2-micro globulin (0.774 and 0.787, respectively) and the clinical model (0.743 and 0.719, respectively) (Fig. 2B and C). The best sCysC cutoff value for predicting AKI was 1.740 mg/L in patients with preexisting CKD, which was significantly higher than the value in patients without preexisting CKD (1.245 mg/L).

Serum CysC as a predictor for secondary endpoints

During a median of 19.8 months [interquartile range (IQR)=17.0 to 21.5] follow-up, 112 (27.2%) patients died within 2 years after their CCU admission, with 80 (19.4%) in the sCysC⩾1.255 mg/L group and 32 (7.8%) in the group <1.255 mg/L sCysC group. A positive correlation was detected between the elevated level of sCysC (⩾1.255 mg/L) and the 2-year mortality rate of CCU patients, based on Spearman's correlation coefficient (r=0.521, *P*<0.001). An sCysC level⩾1.255 mg/L on the first day of admission was associated with a significantly increased probability of all-cause mortality (HR, 3.093, 95% CI, 1.987 to 4.184, *P*<0.001) and rehospitalization (HR, 1.783, 95% CI, 1.131 to 2.812, *P*=0.013) over the 2-year follow-up period (Fig. 4A and B).

Fig. 3A further showed an additive effect of the sCysC levels and the presence of AKI on the 2-year mortality rate for all participants. Patients who developed AKI concurrently with an sCysC concentration⩾1.255 mg/L had the highest overall mortality. As a comparison, patients with AKI alone or only with an increased sCysC level showed a relatively better cumulative survival rate. In addition, the severity of AKI significantly reduced the patient survival rate over the 2-year follow-up period for the sCysC⩾1.255 mg/L and sCysC<1.255 mg/L groups (Fig. 3B and C).

Our study has also shown that sCysC played an important role in predicting failure in



Prediction of AKI in all CCU patients

Biomarkers	best cutoff	sensitivity	specificity	AUC(95%CI)
CysC(mg/L)	1.255	82.2	76.4	0.842(0.796-0.888)
β2-MG(ug/L)	3460.2	75.2	80.9	0.813(0.761-0.864)

Prediction of AKI in patients without CKD

Biomarkers	best cutoff	sensitivity	specificity	AUC(95%CI)
CysC(mg/L)	1.245	74.6	77.8	0.801(0.739-0.864)
β2-MG(ug/L)	3423.2	68.7	82.2	0.781(0.722-0.852)

Prediction of AKI in CCU patients with CKD

Biomarkers	best cutoff	sensitivity	specificity	AUC(95%CI)
CysC(mg/L)	1.740	75.7	87.5	0.876(0.780-0.972)
β2-MG(ug/L)	4717.3	70.3	81.3	0.774(0.636-0.911)

Prediction of 2-years mortality for CCU patients

Biomarkers	best cutoff	sensitivity	specificity	AUC(95%CI)
CysC(mg/L)	1.295	86.7	76.7	0.842(0.803-0.903)
TnI(ng/mL)	7.429	69.9	80.0	0.760(0.694-0.825)
β2-MG(ug/L)	3694.2	66.7	76.7	0.720(0.657-0.779)

Prediction of failure in renal recovery

Biomarkers	best cutoff	sensitivity	specificity	AUC(95%CI)
CysC(mg/L)	1.285	84.4	72.6	0.814(0.742-0.887)
β2-MG(ug/L)	4890.6	64.4	88.3	0.782(0.700-0.863)

Fig. 2. ROC analyses for predicting AKI or 2 years prognosis. Clinical model for predicting AKI or 2 years prognosis was composed of age, gender, hypertension, diabetes, mean arterial pressure, hemoglobin, serum albumin, APACHEII, treatment with loop diuretics, treatment with ACEI/ARB, and treatment with aspirin. (A-C) sCysC, β2-micro globulin and clinical model for predicting AKI in all participants (A), in patients without preexisting CKD (B), and in patients with preexisting CKD (C). ROC analysis for predicting 2-year mortality (D) and failure in renal recovery (E) in all participants.

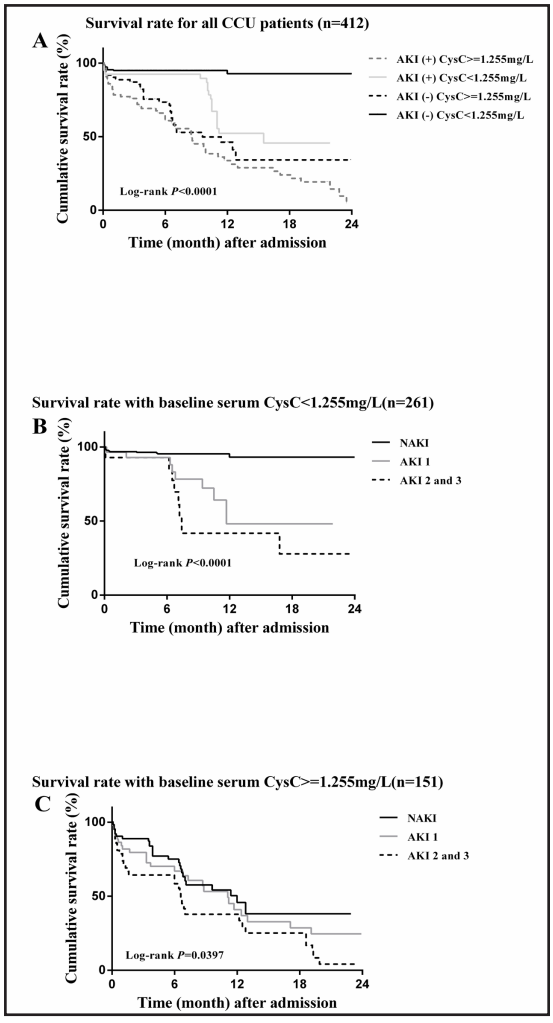


Fig. 3. (A) Kaplan-Meier survival rate of all-cause mortality from admission to 2-year follow-up according to the presence of AKI and the category of sCysC level. Survival curves according to the severity of AKI and the level of sCysC<1.255mg/L (B), and sCysC>=1.255mg/L (C).

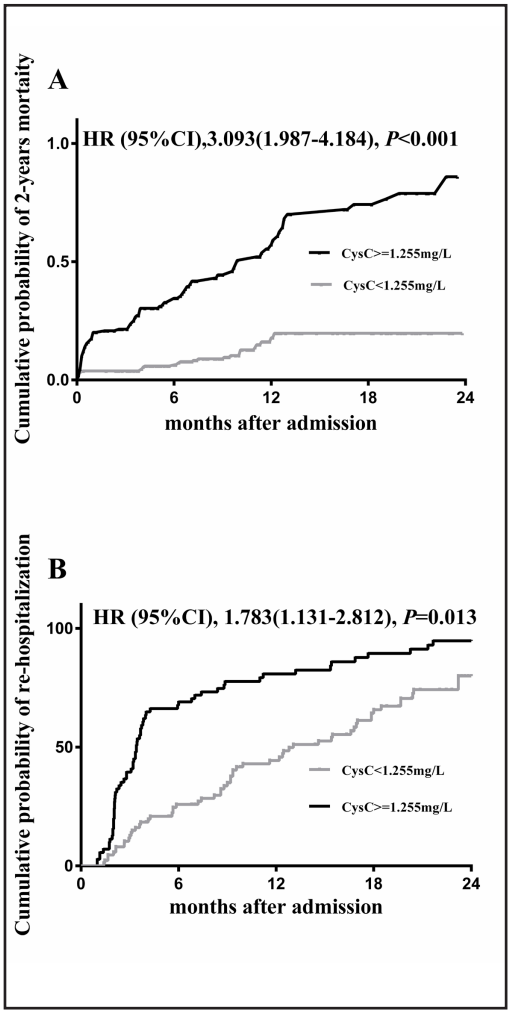


Fig. 4. (A) Cumulative probability of all-cause mortality from admission to 2-year follow-up according to the category of sCysC level. (B) Cumulative probability of re-hospitalization from discharge to one-year follow-up according to the category of sCysC.

Table 4. Multivariate logistic regression analyses: Predictors of rehospitalization and failure in renal. [a]Ajusted for age, gender, BMI, hypertension, diabetes, preexisting CKD, serum CysC, mean arterial pressure, hemoglobin, serum albumin, APACHEII, treatment with loop diuretics, treatment with ACEI/ARB, and treatment with aspirin; OR, odds ratio; 95% CI, 95% confidence interval. Anemia was defined as the level of hemoglobin was less than 120 g/dL for men or 110g/dL for women. [b]N=351, because 61 patients died in their hospital admission recovery

Variables on admission	Unadjusted OR	Adjusted OR[a]	95% CI	P Value
Rehospitalization (n=351)[b]				
sCysC (Continuous variables)	3.816	3.128	2.011 to 4.867	<0.001
Preexisting CKD (yes versus no)	2.756	2.260	1.121 to 6.123	0.026
Failure in renal recovery (n=130)				
Anemia (yes versus no)	4.507	3.588	2.217 to 5.806	0.001
sCysC (Continuous variables)	5.320	3.618	1.753 to 7.463	<0.001

Table 5. Multivariate logistic regression analyses: Predictors of mortality from admission to 2-year follow-up. APACHEII, Acute Physiology and Chronic Health Evaluation II. (n=412)

Variables on admission	Unadjusted OR	Adjusted OR	95% CI	P Value
Model 1				
AKI (yes versus no)	4.199	2.611	1.241 to 5.491	0.011
Treatment with aspirin (yes versus no)	5.255	3.225	1.303 to 7.979	0.011
Preexisting CKD (yes versus no)	3.976	3.182	1.223 to 8.275	0.018
sCysC (Continuous variables)	5.884	4.955	2.853 to 8.608	<0.001
Model 2				
sCysC (Continuous variables)	5.884	4.125	2.600 to 6.545	<0.001
APACHEII points (Continuous variables)	1.276	1.158	1.063 to 1.255	0.001
Troponin I (Continuous variables)	1.403	1.046	1.024 to 1.068	<0.001

Table 6. Multivariate logistic regression analyses of sCysC as a predictor for second endpoints. [a]Adjusted forage, gender, BMI, hypertension, diabetes, preexisting CKD, serum CysC, mean arterial pressure, hemoglobin, serum albumin, NT-proBNP, APACHEII, treatment with loop diuretics, treatment with ACEI/ARB, and treatment with aspirin; OR, odds ratio; 95% CI, 95% confidence interval

sCysC(mg/L) on the first day of admission	Unadjusted OR	Adjusted OR[a]	95% CI	P Value
For rehospitalization (n=412, 103 per quartile)				
Quartile 1（<0.93）	1.0 (referent)	1.0 (referent)		
Quartile 2（0.93-1.17）	1.955	1.653	0.847 to 3.127	0.122
Quartile 3（1.18-1.69）	2.477	1.932	1.932 to 3.645	0.042
Quartile 4（>1.69）	7.719	6.233	3.196 to 12.157	<0.001
For failure in renal recovery (n=412, 103 per quartile)				
Quartile 1（<0.93）	1.0 (referent)	1.0 (referent)		
Quartile 2（0.93-1.17）	0.926	1.150	0.243 to 5.431	0.860
Quartile 3（1.18-1.69）	3.902	3.246	0.957 to 11.013	0.059
Quartile 4（>1.69）	17.927	13.514	4.292 to 42.546	<0.001
For 2-years mortality (n=412, 103 per quartile)				
Quartile 1（<0.93）	1.0 (referent)	1.0 (referent)		
Quartile 2（0.93-1.17）	0.206	0.114	0.012 to 1.102	0.061
Quartile 3（1.18-1.69）	1.622	3.728	1.036 to 13.417	0.044
Quartile 4（>1.69）	8.120	14.246	3.168 to 64.060	0.001

renal recovery and rehospitalization in patients with incident AKI. In the group of patents with sCysC concentrations<1.255 mg/L upon admission, the rate of failure in renal recovery at discharge was only 3.4%, and this rate significantly increased to 31.4% in the group of patients with sCysC concentrations⩾1.255 mg/L. Similarly, the rate of rehospitalization in these patients also increased from 24.8% to 57.0%, according to the elevated level of sCysC. These results were confirmed through conditional multivariable logistic regression. After adjusting for clinical risk factors, sCysC concentration was the most powerful risk factor for failure during the renal recovery period (OR, 3.618, 95% CI 1.753 to 7.463, *P*<0.001) and rehospitalization (OR, 3.128, 95% CI 2.011 to 4.867, *P*<0.001) (Table 4). Moreover, when sCysC was analyzed as a categorical variable, the highest quartile of sCysC on the first day of admission was also associated with a 13-fold increased risk for failure in renal recovery and a 6-fold increased risk of rehospitalization compared with the lowest quartile (Table 6).

Furthermore, sCysC was an independent predictor for 2-year mortality, as a continuous variable (Table 5) and as a categorical variable (Table 6). In addition, sCysC on the first day of admission outperformed Troponin I or β2-micro globulin when predicting 2-year mortality rates (AUC, 0.842, 95% CI, 0.803 to 0.903), as well as failure during renal recovery (AUC, 0.814, 95% CI, 0.742 to 0.887) (Fig. 2D and E).

sCysC might also play a role in predicting the progression from AKI to CKD. Among the AKI patients without preexisting CKD, an sCysC concentration ⩾1.255 mg/L led to a higher incidence of AKI progression to CKD than that in patients with sCysC concentrations <1.255

mg/L (73.1% vs. 25.0%). Due to the limits of this sample size, we were unable to conduct subgroup analyses.

Discussion

Our study showed that the incidence of AKI in CCU patients was 31.6%. A previous prospective study by Chen, et al. has also shown that AKI occurred in 28.7% of patients who were admitted to the CCU due to acute myocardial infarction [10]. Nevertheless, in patients with ADHF, Yang et al. have shown that the incidence of AKI further increased to 47.6%[30]. This higher incidence of AKI in ADHF patients has been referred to as acute cardiac and renal dysfunction [31-34].

The diagnosis of AKI, particularly in CCU patients, is currently delayed and inaccurate, which largely contributes to the poor clinical outcomes of AKI and results in great challenges in preventing and treating this kidney disease worldwide [35]. As opposed to acute coronary syndrome, in which the discovery of biomarkers, such as troponin, has completely advanced clinical care by establishing early diagnosis, specific early biomarkers for AKI have been lacking, in terms of predicting the severity of AKI and guiding its treatment. In this observational study, we found that sCysC levels measured on the first day of admission were a powerful predictor for AKI and the short-term prognosis of CCU patients. These findings are consistent with previous results that have been reported in two other clinical studies [36, 37]. More importantly, despite evidence that β2-micro globulin is useful for predicting prognosis in kidney disease, cardiovascular outcomes and death [38-40], the performance of sCysC in predicting AKI and short-term prognosis was superior to β2-micro globulin in our study.

A bidirectional relationship between AKI and CKD has been suggested by recent studies. The non-recovery of AKI is associated with its progression to CKD; however, CKD patients have an increased risk of developing AKI[41-44]. In this study, we prospectively screened CCU patients whose serum creatinine measurements were available for the 6-month period prior to admission. This design allowed us to determine the predictive performance of sCysC in patients with and without preexisting CKD. Our results showed that compared with patients without preexisting CKD, the level of sCysC on the first day of admission was remarkably higher ($\geqslant$1.255 mg/L) in patients with preexisting CKD. Note that the best cutoff value of sCysC was also higher in these patients. Moreover, in patients without preexisting CKD, an elevated sCysC level significantly increased the incidence of AKI progression to CKD. These distinctive characteristics of sCysC make it a valuable and unique predictor for AKI, particularly for AKI with preexisting CKD and AKI progression to CKD.

Identifying patients who are at higher risk of poor prognosis is still an enormous challenge in clinical practice. In our study, during the 2-year follow-up period, patients with sCysC levels$\geqslant$1.255 mg/L had significantly higher 2-year mortality, rehospitalization and failure of renal recovery rates than those with sCysC levels<1.255 mg/L. Consistently, a recent study has also found a positive correlation of an elevated sCysC with poor cardiorenal outcomes in patients with acute heart failure [36]. These data suggest that measuring sCysC on the first day of admission could be used to assess the 2-year prognosis of CCU patients, who are often associated with poor in-hospital and post-discharge outcomes. More importantly, we compared the prognostic performance of sCysC with Troponin I, a validated marker for prognosis of acute coronary syndrome. sCysC performed substantially better than Troponin I in predicting 2-year mortality rates (AUC=0.842 versus AUC=0.760). These results suggest that sCysC could be an independent predictor for 2-year mortality, as well as rehospitalization and failure in renal recovery for CCU patients,

This study has several strengths. First, we employed a prospective observational design and a rigorous protocol for patient screening in CCU and performed sCysC measurements in a blinded manner. Second, serum creatinine measurements before admission were available

for the patients in the study, which allowed us to determine the predictive performance of sCysC in subgroups with and without prior CKD. Third, we provided convincing evidence that sCysC outperformed β2-MG and clinical models and served as a powerful predictor of AKI in CCU patients. Finally, the use of sCysC as a predictor for short-term prognosis was assessed during the 2-year follow-up of these CCU patients, further broadening the clinical implications of sCysC in disease diagnosis and prognosis.

This study also has some limitations. First, it was a single center study of CCU patients. Second, we measured the sCysC levels on the first day of admission but did not follow-up t possible sCysC changes in patients during their hospital stays. Unfortunately, urinary CysC was not measured either. Furthermore, the diagnosis of AKI was based on an increase in serum creatinine, which may result in the use of using a defective outcome variable to analyze the performance of novel biomarkers. Evidence of AKI on renal biopsy would be the gold standard, but it was not practicable in our study. Similar to most previous AKI studies, we were not able to use urine output for the AKI diagnosis because an indwelling urinary catheter was not present in most patients in this study.

In summary, this cohort study showed that the sCysC can serve as an early predictor for the development of AKI in CCU patients, as well as their short-term prognoses. If confirmed further, sCysC may provide a unique opportunity to impact dramatically the management of AKI by delivering diagnostic, severity, and prognostic information at an early time-point following a renal insult.

Disclosure Statement

The authors of this manuscript state that they do not have any Disclosure Statements and nothing to disclose.

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