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Predictive and pathogenetic value of plasma biomarkers for acute kidney injury in patients with acute lung injury*

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Abstract

Objective—To identify biological and clinical predictors of acute kidney injury in subjects with acute lung injury.

Design—Secondary data analysis from a multicenter, randomized clinical trial.

Setting—Intensive care units in ten university medical centers.

Patients—A total of 876 patients enrolled in the first National Heart, Lung, and Blood Institute Acute Respiratory Distress Syndrome Clinical Network trial.

Interventions—Study subjects were randomized to receive a low tidal volume ventilation strategy and pharmacologic therapy with ketoconazole or lisofylline in a factorial design.

Measurements and Main Results—We tested the association of baseline levels of interleukin-6, interleukin-8, interleukin-10, von Willebrand factor, tumor necrosis factor- α , type I and II soluble tumor necrosis factor receptors (sTNFR-I and -II), protein C, plasminogen activator inhibitor-1 (PAI-1), surfactant protein-A, surfactant protein-D, and intracellular adhesion molecule-1 with subsequent acute kidney injury. Of 876 study participants who did not have end-stage renal disease, 209 (24%) developed acute kidney injury, defined as a rise in serum creatinine of >50% from baseline over the first four study days. The 180-day mortality rate for subjects with acute kidney injury was 58%, compared with 28% in those without acute kidney injury ($p < .001$). Interleukin-6, sTNFR-I, sTNFR-II, and PAI-1 levels were independently associated with acute kidney injury after adjustment for demographics, interventions, and severity of illness. A combination of clinical and biological predictors had the best area under the receiver operating characteristic curve, and the contribution of sTNFR-I and PAI-1 to this model was highly significant ($p = .0003$).

*See also p. 2866.

Conclusions—Elevations in PAI-1, interleukin-6, and the sTNFRs in subjects with acute kidney injury suggest that disordered coagulation, inflammation, and neutrophil–endothelial interactions play important roles in the pathogenesis of acute kidney injury. The combination of these biological and clinical risk factors may have important and additive value in predictive models for acute kidney injury.

Keywords

acute kidney injury; acute lung injury; acute respiratory distress syndrome; biological marker; predictive value; interleukin-6; soluble tumor necrosis factor receptor; plasminogen activator inhibitor-1

Acute kidney injury (also known as acute renal failure) in hospitalized patients is associated with high mortality, often 40–60% in the critical care setting (1–4). New therapies that reduce the morbidity and mortality of acute kidney injury are urgently needed. Dialysis only manages the symptoms, rather than treating the injury (5).

An important limitation to the treatment of acute kidney injury relates to the difficulty in identifying individuals with early disease (6). Current markers of kidney function (including serum creatinine) are metabolic waste products that accumulate after kidney injury. By the time serum concentrations of these markers are elevated, considerable tissue damage may have already occurred (7). Identification of early acute kidney injury (before changes in existing markers of kidney function) may identify a target population for the development of novel prevention and treatment strategies.

We analyzed the association between plasma biomarker levels and the subsequent development of acute kidney injury using clinical data and plasma measurements from the first Acute Respiratory Distress Syndrome (ARDS) Network clinical trial (8–10). These critically ill subjects represent a population at high risk of acute kidney injury. Biomarkers measured in this cohort include inflammatory cytokines (interleukin [IL]-6, -8, and -10), markers of endothelial injury (von Willebrand factor), markers of neutrophil–endothelial interaction (tumor necrosis factor [TNF]- α and soluble TNF receptors [sTNFR]-I and -II), markers of dysregulated coagulation (protein C and plasminogen activator inhibitor [PAI]-1), markers of epithelial cell injury (surfactant protein-A and -D), and adhesion molecules (intracellular adhesion molecule-1). Several of the biomarkers, including IL-6, IL-8, von Willebrand factor, intracellular adhesion molecule-1, sTNFR-I and -II, protein C, PAI-1, and surfactant protein-D, individually had a significant independent association with death and ventilator-free days (11–14). Specifically, we hypothesized that inflammatory cytokines, adhesion molecules, and markers of dysregulated coagulation would be associated with the development of acute kidney injury. We further hypothesized that biomarkers would have predictive value for acute kidney injury, above and beyond clinical characteristics.

METHODS

Study Design/Patient Selection

We analyzed data from the first ARDS Network clinical trial, a factorial design, multicenter, randomized trial of a low tidal volume ventilation strategy and pharmacologic therapy with ketoconazole or lisofylline for acute lung injury. The methods and results of these trials have been reported elsewhere (8–10). Briefly, the low tidal volume ventilation strategy was factorialized with two medical interventions for acute lung injury. Specifically, the first 234 subjects were enrolled in a 2 $\times$ 2 factorial study of the low tidal volume ventilation strategy (6 vs. 12 mL/kg of ideal body weight) and 400 mg/day of ketoconazole vs. placebo. The

ketoconazole study was terminated by the Data Safety Monitoring Board for lack of efficacy (9). The last 235 subjects were enrolled in a 2×2 factorial trial of the low tidal volume ventilation strategy and $3 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$ of lisofylline vs. placebo. The lisofylline study was terminated by the Data Safety Monitoring Board for lack of efficacy. Because the low tidal volume ventilation strategy had significant benefit for patients with acute lung injury (8), the last 41 subjects in the lisofylline trial all received the low tidal volume ventilation strategy (10). The study protocols were approved by the institutional review board at each participating center. The patient or their surrogate consented to study participation, except at one hospital site where this requirement was waived. Of the participants, 876 of 902 were included in this analysis; those with end-stage renal disease (defined as the need for maintenance dialysis therapy) at study enrollment were excluded. The primary outcome was death before discharge home with unassisted breathing.

Clinical and Laboratory Variables

The clinical data have been described previously (8–10) and included chronic health conditions, laboratory data, ventilatory variables, $\text{PaO}_2/\text{FIO}_2$ ratio, and vasopressor use. Vasopressor use was defined as the use of dopamine at $\geq 6 \text{ } \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ or the use of neosynephrine, norepinephrine, or epinephrine at any dose. For each subject, the primary clinical risk factor for acute lung injury was ascertained prospectively, before randomization to ventilatory treatment group. Risk factors for acute lung injury included infectious risk factors (pneumonia and sepsis) and noninfectious risk factors (aspiration, trauma, multiple transfusions, and other), as described previously (15). Of the data elements included in the multivariable analysis, the only variable with a significant number of missing measurements was serum bilirubin (total of 62 missing measurements); these missing measurements were imputed to the mean. Baseline hypotension was defined as the use of vasopressors or a lowest systolic blood pressure of $\leq 80 \text{ mm Hg}$ recorded during the 24 hrs before study enrollment.

Definition of Acute Kidney Injury

We used the consensus definition proposed by the Acute Dialysis Quality Initiative (16) as our *a priori* definition of acute kidney injury. Because even small changes in serum creatinine are associated with increased mortality, our analysis was designed to examine the association between biomarker levels and small but significant changes in serum creatinine. Because the focus on our study was on the association of baseline biomarker levels and the subsequent development of acute kidney injury, we limited acute kidney injury to disease occurring during the first 4 days of the study. Acute kidney injury was defined as an increase in creatinine of 50% from baseline (the “risk” class of the Acute Dialysis Quality Initiative definition) at any time during the first 4 days of the study. Baseline serum creatinine was the lowest of the study measurements recorded on day 0 of the study. Peak serum creatinine was defined as the maximum value recorded on days 1–4 of the study. For some analyses, we also examined the predictive value of baseline biomarker measurements for the development of acute kidney injury by day 1 of the study.

Biomarker Measurements

All plasma biomarker measurements have been previously described (11–14, 17). In brief, blood samples were collected on days 0, 1, and 3 according to the original study protocol. The blood was collected in EDTA-treated sterile tubes and centrifuged. Plasma was then aliquoted and frozen at -80°C . Antibodies for the IL-6, IL-8, and sTNFR-I two-antibody sandwich enzyme linked immunosorbent assays (ELISA) were obtained from R&D Systems (Minneapolis, MN). Antibodies for the IL-10 ELISA were obtained from R&D systems and BD Biosciences Pharmingen (San Diego, CA) (12). The TNF- α ELISA antibodies were obtained from BD Biosciences Pharmingen. sTNFR-II and intracellular adhesion molecule-1

measurements were made using ELISA assays from Biosource International (Camarillo, CA) (13). Surfactant protein-A was measured using a previously described sandwich ELISA. Surfactant protein-D was measured using a commercially available ELISA from Nagae Corporation (Tokyo, Japan) (11). von Willebrand factor levels were measured using a commercially available ELISA from Diagnostica Stago (Parsippany, NJ) (14). Protein C was measured using a commercially available ELISA from Helena Laboratories (Beaumont, TX), as described previously (18). PAI-1 was measured using a commercially available ELISA from American Diagnostica (Stamford, CT), as previously described (19). Measurements were made in available samples from the cohort; the number of available measurements differed by biomarker, but ranged from 361 to 755. Among subjects with specific biomarker measurements and those without measurements, there were no statistically significant differences in the acute kidney injury or death rates, ventilatory strategy randomization assignment, age, sex, race, Acute Physiology and Chronic Health Evaluation (APACHE) III score, presence of hypotension, PaO₂/FIO₂ ratio, or baseline serum creatinine, bilirubin, or platelet count (data not shown). Because there were not significant differences in acute kidney injury rates or baseline covariates between those with biomarker data and those with missing data, when data were missing, we excluded those individuals from the analysis. Regression models with multiple biomarkers are based on the subjects who had the relevant multiple measurements made. We used the baseline (day 0) measurements in this analysis.

Statistical Analysis

Baseline characteristics of subjects who did and did not develop acute kidney injury were compared. Categorical variables were expressed as proportions and compared using the chi-square test. Continuous variables were expressed as mean $\pm$ SD or median with interquartile range and were compared using Student's *t*-test or Wilcoxon's rank-sum test, where appropriate.

We first examined the association between acute kidney injury (predictor) and death (outcome) using the chi-square test. Logistic regression was used for multivariable analyses. We controlled for age, sex, race, and the trial interventions and for other covariates that reflect the severity of acute illness, including hypotension, baseline kidney function, total bilirubin, platelet count, PaO₂/FIO₂ ratio, and the presence or absence of infection. The odds ratio was expressed per 10-yr increase in age, per 50,000/mm³ decrease in platelet count, and per 50-unit decrease in the PaO₂/FIO₂ ratio. Each of these covariates was associated with death in bivariate analyses.

We next examined the association between biomarker measurements and acute kidney injury. For the logistic regression analysis, biomarker levels were log₁₀ transformed, as in previous ARDS Network analyses; hence, the odds ratios represent the increased risk of acute kidney injury per log₁₀ change in biomarker level. We initially adjusted for age, sex, race, and the trial interventions. We then controlled for other covariates that reflect the severity of acute illness, including hypotension, baseline liver function (total bilirubin), platelet count, PaO₂/FIO₂ ratio, and the presence or absence of infection. Each of these covariates was associated with acute kidney injury in bivariate analyses (see online-only table at www.ccmjournal.org).

We used logistic regression to develop predictive models for acute kidney injury, based on clinical risk factors or biomarkers. For the clinical risk factor-based model, we incorporated age, sex, and race and the severity of illness covariates used in the final multivariable model of the association of biomarkers with acute kidney injury (hypotension, baseline liver function [total bilirubin], platelet count, PaO₂/FIO₂ ratio, and the presence or absence of infection). For the biomarker-based model, markers that were associated with acute kidney

injury in the multivariable logistic regression models were incorporated (IL-6, sTNFRs, PAI-1); the individual contributions of biomarkers were tested using likelihood ratio testing. Markers that did not make a statistically significant contribution to the model were eliminated ($p > .2$). The only two biomarkers retained in the final model were PAI-1 and sTNFR-I. We developed an integrated model encompassing all of the clinical risk factors described above and the two biomarkers that were retained in the final biomarker model (PAI-1 and sTNFR-I); the contribution of biomarkers was tested using likelihood ratio testing. We compared the predictive value of our combined biomarker and clinical risk factor model for acute kidney injury during the first 4 days of the study to the predictive value of the same model during the first day of the study, as has been done in other analyses (20). Permutation testing was used to determine whether the difference in the area under the receiver operating characteristic (ROC) curves was significant (21).

Model discrimination was assessed using ROC curves (22). Model fit (calibration) was assessed using the Hosmer-Lemeshow goodness-of-fit test, which compares model performance (observed vs. expected) across deciles of risk. A nonsignificant value for the Hosmer-Lemeshow chi-square test suggests an absence of biased fit. In an alternate approach, we used a learning set/test set approach to calculate the area under the ROC curve. Data analysis was conducted using Stata 9.1 (StataCorp, College Station, TX). Two-tailed p values of $<.05$ were considered significant.

RESULTS

Description of the Clinical Trial Population

Of the 902 subjects in the ARDS Network factorial trials of low tidal volume ventilation, ketoconazole, and lisofylline, 26 subjects with end-stage renal disease ($n = 20$) or missing data regarding end-stage renal disease status ($n = 6$) were eliminated from our analysis. Of the remaining 876 subjects, 209 patients (24%) developed acute kidney injury during the first four days of study enrollment. Characteristics of subjects who developed acute kidney injury are compared with those who did not (Table 1). Subjects with acute kidney injury did not differ significantly from those without injury in age, sex, or race. Neither lisofylline use nor the lower tidal volume ventilation strategy was significantly associated with the development of acute kidney injury; however, a higher proportion of subjects treated with ketoconazole developed acute kidney injury during the first 4 days of the study. Baseline serum creatinine was not significantly different ($p = .45$) in those who developed acute kidney injury (mean of 1.42 mg/dL, SD 1.09 mg/dL) compared with those who did not develop acute kidney injury (mean of 1.35 mg/dL, SD 1.23 mg/dL). Subjects with acute kidney injury had more hypotension, higher bilirubin levels, lower platelet counts, and lower $\text{PaO}_2/\text{FIO}_2$ ratios at baseline.

Mortality and Acute Kidney Injury

The mortality rate among subjects with acute kidney injury was 58%, compared with 28% among those without acute kidney injury ($p < .001$). In a multivariable model adjusting for age, sex, race, interventions, vasopressor use, $\text{PaO}_2/\text{FIO}_2$ ratio, total bilirubin, platelet count, and infection, acute kidney injury was associated with a more than three-fold increase in the odds of death (Table 2).

Associations Among Biomarker Measurements and Acute Kidney Injury

Given the important association between acute kidney injury and death in this cohort, we analyzed the association among all measured biomarkers and the development of acute kidney injury. In bivariate analysis, IL-6, IL-8, IL-10, sTNFR-I and -II, intracellular adhesion molecule-1, protein C, and PAI-1 were independently associated with the

development of acute kidney injury (Table 3). All of these remained associated after adjustment for age, sex, race, and the clinical trial interventions (case-mix adjusted). IL-6, sTNFR-I and -II, and PAI-1 remained associated with the development of acute kidney injury after additional adjustment for markers of severity of illness (hypotension, baseline liver function [total bilirubin], platelet count, PaO₂/FIO₂ ratio, and the presence or absence of infection) (Table 3). Figure 1 shows box-plot summaries of median levels of IL-6, sTNFR-I and -II, and PAI-1 among those with and without acute kidney injury.

Contribution of Biomarkers to Predictive Models for Acute Kidney Injury

To determine the contribution of biomarkers to predictive models for acute kidney injury, we measured the areas under the ROC curve for the logistic regression models based on clinical risk factors, biomarkers, and the combination. The areas under the ROC curve for clinical risk factors and biomarkers alone were virtually identical at 0.66 (95% confidence level, 0.60–0.71) and 0.67 (95% confidence level, 0.61–0.72), respectively. The final biomarker model included only PAI-1 and sTNFR-I because these were the two biomarkers that made a significant contribution to the predictive model, based on likelihood ratio testing. Furthermore, the addition of biomarkers to the clinical risk factor–based model increased the predictive value of the model; the area under the ROC curve increased to 0.70 (95% confidence level, 0.65–0.75, $p = .0003$), compared with clinical risk factors alone (Fig. 2, *left*). In a companion analysis, using a learning set/test set procedure, the area under the ROC curve for the combined biomarker and clinical risk factor model was 0.67, compared with an area under the ROC curve of 0.62 for the clinical risk factor model alone. When the analysis was restricted to the prediction of acute kidney injury developing on day 1 (a strategy previously used in studies of biomarkers in acute kidney injury, including a study by Parikh et al. [23]), the area under the ROC curve increased further. The area under the ROC curve for the combined clinical and biological risk factor model for acute kidney injury at day 1 was 0.77 (95% confidence level, 0.71–0.83); this was significantly higher than the area under the ROC curve for days 1–4 of 0.70 ($p = .004$). The area under the ROC curve for the clinical risk factor model for acute kidney injury at day 1 was 0.72 (95% confidence level, 0.65–0.79; $p = .0006$ for the comparison with the combined biological and clinical risk factor model).

DISCUSSION

Using data from the first ARDS Network clinical trial, we tested the association between plasma biomarker levels and the development of acute kidney injury. The biomarkers measured in this cohort are representative of a number of different pathophysiological processes, and therefore, this cohort represents an efficient resource to examine and compare the associations of plasma biomarkers with the development of acute kidney injury. We found that plasma levels of IL-6, sTNFR-I and -II, and PAI-1 were significantly associated with acute kidney injury. Importantly, not all biomarkers that were associated with mortality in the original analysis were associated with acute kidney injury. For example, surfactant protein-D, which is released by injured pulmonary epithelial type II cells and is therefore a marker for lung injury (24), was associated with an increased risk of death in the cohort (11) but was not associated with acute kidney injury. This finding suggests that the associations described among biomarker levels and acute kidney injury are not simply reflections of the association between acute lung injury and death.

In our analysis, we noted that a higher proportion of subjects treated with ketoconazole developed acute kidney injury during the first 4 days of the study. In the original clinical trial (9), there was no association between ketoconazole and renal failure. This apparent difference is attributable to the time frame in which acute renal failure was defined in the two analyses (i.e., 4 vs. 28 days). Because ketoconazole was administered for up to 21 days

in the treatment protocol, in the original report, acute renal failure was examined over the first 28 study days. Using our definition of acute kidney injury (creatinine increase of >50% from baseline), there was no association with treatment with ketoconazole over 28 days. Neither the lower tidal volume ventilation strategy nor lisofylline administration was significantly associated with the development of acute kidney injury.

One of the challenges in critical care nephrology is that there is no clear consensus definition for acute kidney injury, in contrast to other diseases such as acute lung injury. We based our *a priori* definition on the Acute Dialysis Quality Initiative consensus definition (16). Specifically, we selected a graded definition because the physiologic significance of an incremental change in creatinine varies depending on baseline renal function. Also, we chose a relatively small increment in creatinine (50% increase in baseline) because even small changes in creatinine reflect kidney injury and, in turn, should be manifest at the biomarker level. A 50% increase in creatinine generally reflects a 25% reduction in the glomerular filtration rate and is associated with mortality, length of stay, and costs in hospitalized patients (25). We chose a relatively short time frame for the development of acute kidney injury because we were interested in the association between biomarkers; early elevations of biomarkers are less likely to predict late disease if a biological mechanism is postulated for the association. Lastly, because chronic kidney disease is a well-established risk factor for acute kidney injury, we chose to only exclude subjects with known end-stage renal disease from our analysis.

In a cohort of patients who suffered a cardiac arrest and underwent successful resuscitation, PAI-1 levels were higher in those who developed acute kidney injury (defined as oliguria and a creatinine of >2.4 mg/dL or the need for renal replacement therapy) compared with those who did not (26). Indeed, of the biomarkers tested in this cohort, the association between acute kidney injury and PAI-1 was the most robust (smallest *p* value by either Wald chi-square testing or likelihood ratio testing in multivariable models with one biomarker). The finding of higher PAI-1 levels in subjects who subsequently developed acute kidney injury suggests that impaired fibrinolysis may be important in the pathogenesis of acute kidney injury. Previous studies in animal models have shown that PAI-1 messenger RNA levels in the kidney were elevated after cecal ligation and perforation in a model of sepsis-induced acute kidney injury (27). However, clinical studies of plasma biomarkers of impaired fibrinolysis have not been previously reported. Although it has become increasingly clear from animal studies that the endothelium plays a critical role in the pathogenesis of acute kidney injury (28), there was no clear association between levels of markers of endothelial injury measured in this study and the development of acute kidney injury.

Our results are in agreement with those of the Norasept II study (29), a randomized trial of a TNF- α antibody for sepsis, that also demonstrated an association between levels of sTNFR-I and -II and acute kidney injury. However, our cohort is composed of subjects with acute lung injury of diverse pathogeneses, whereas in Norasept II, study subjects were restricted to those with septic shock. Nevertheless, among the ARDS Network subjects who had a primary cause of acute lung injury other than infection, levels of sTNFR-I were also significantly associated with the development of acute kidney injury (data not shown).

Elevations in IL-6 are associated with acute kidney injury in animal models of ischemic acute tubular necrosis (30). In humans, Ahlstrom et al. (31) reported that IL-6 levels differed among patients with systemic inflammatory response syndrome with and without acute kidney injury at days 2 and 3 of systemic inflammatory response syndrome. However, in this study, the authors did not report the predictive value of IL-6 for the subsequent development of acute kidney injury. In the Norasept II study (29), there was a trend toward

higher IL-6 levels in those subjects who subsequently developed acute kidney injury, but this result did not quite achieve statistical significance. More recently, in a secondary analysis of the PROWESS clinical trial of activated protein C for the treatment of severe sepsis, Chawla et al. (32) demonstrated that baseline IL-6 levels were predictive for acute kidney injury. Our results are consistent with these findings and suggest that IL-6 is predictive in other critically ill patient populations, namely, those with acute lung injury.

Ideally, a biomarker study compares the predictive value of the biomarker for the outcome of interest to known clinical risk factors. Clinical risk factors have several important practical advantages over biomarker measurements, including ease and lower cost of measurement. Models to predict acute kidney injury in the critical care setting based on clinical risk factors are likely to have limited discriminant function on their own, as was the case with the model we developed. However, in principle, clinical risk factors can make an important contribution to integrated models that predict acute kidney injury. In our analysis, we found that clinical risk factors, and PAI-1 and sT-NFR-I, contributed significantly to the integrated model. Thus, our study highlights the potential value of an integrated approach using biomarkers and clinical risk factors in clinical studies of acute kidney injury.

This study has several strengths. The sample size was relatively large and subjects were enrolled in 20 centers across the United States. Acute kidney injury and death could be ascertained for the vast majority of subjects. Biological specimens were prospectively collected from subjects, and biomarkers that reflect a number of different pathophysiologic processes were measured. Thus, we were able to identify associations among biomarker levels and acute kidney injury that may add important insights into the pathophysiology of acute kidney injury.

There are also some limitations to our study. Because this was a study of lung injury, some of the clinical data that would have been collected in a study focused on kidney injury are missing. Although the original study was not focused on acute kidney injury, creatinine measurements were recorded during the first 28 days of the study as clinically available, which was daily for most of these patients because they were critically ill in the intensive care unit. However, we were not able to incorporate changes in blood urea nitrogen level, urine output, or need for dialysis into our definition of acute kidney injury. Moreover, had information on the use of nephrotoxic medications been available, it might have enhanced our ability to predict acute kidney injury. Despite having included a wide array of covariates, there is likely to be some residual confounding for which we cannot adjust. Because all subjects had acute lung injury, the results may not be generalizable to those with other critical illnesses. However, the pathogenesis of acute lung injury in the study cohort were diverse and included infectious and other pathogenesis. Although we identified novel predictors of acute kidney injury, the discriminatory power of the models remains modest. Additional studies in critically ill patients at risk of acute kidney injury are needed to confirm and extend these results.

In summary, we have discovered novel associations among levels of plasma biomarkers and the subsequent development of acute kidney injury in critically ill subjects at risk of acute kidney injury. In particular, the association between elevated PAI-1 and acute kidney injury extends experimental data from animal models, suggesting that impaired fibrinolysis plays an important role in the pathogenesis of acute kidney injury. Further studies are needed to confirm these results and to identify additional biomarkers with pathogenetic value for acute kidney injury.

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Figure 1.

Box-plot summaries of plasminogen activator inhibitor-1 (*PAI-1*), soluble tumor necrosis factor receptors (*sTNFR-I* and *-II*), and interleukin-6 (*IL-6*) levels in those with and without acute kidney injury (*AKI*). The *horizontal line* represents the median, *box* encompasses the 25th through 75th percentiles, and *whiskers* encompass the 10th through 90th percentiles. In all four cases, the *p* value for the Wilcoxon's rank-sum test comparing biomarker levels in those with and without *AKI* was $<.0001$.

Figure 2.

Receiver operating characteristic (ROC) curves for predictive models for acute kidney injury. *Left*, comparison of ROC curves based on a predictive model using clinical risk factors only (*dotted line*) to a model containing clinical risk factors and biomarker levels (soluble tumor necrosis factor receptor-1 and plasminogen activator inhibitor-1; *solid line*) for acute kidney injury during the first 4 days of the study. The areas under the ROC curve are 0.66 (95% confidence level [CI], 0.60–0.71) and 0.70 (95% CI, 0.65–0.75), respectively, $p = .0003$. In all three cases, the Hosmer-Lemeshow goodness-of-fit p value was $>.30$. *Right*, comparison of ROC curves based on a predictive model using clinical risk factors only (*dotted line*) to a model containing clinical risk factors and biomarker levels (soluble tumor necrosis factor receptor-1 and plasminogen activator inhibitor-1; *solid line*) for acute kidney injury during the first day of the study. The areas under the ROC curve are 0.72 (95% CI, 0.65–0.79) and 0.77 (95% CI, 0.71–83), respectively, $p = .006$ for difference in areas under the ROC curve. In all three cases, the Hosmer-Lemeshow goodness of fit $p > .30$.

Table 1

Baseline characteristics of subjects with and without acute kidney injury (AKI)

Risk Factor	No AKI (n = 667)	AKI (n = 209)	p Value
Mean (SD) age, yrs	51.4 (17.2)	53 (17.6)	.26
Male, n (%)	402 (60%)	120 (57%)	.46
White race, n (%)	497 (75%)	151 (72%)	.52
Mean (SD) APACHE III score	79.9 (27.7)	90.4 (27.5)	<.0005
Low tidal volume ventilation, n (%)	362 (54%)	100 (48%)	.10
Ketoconazole treatment, n (%)	77 (12%)	37 (18%)	.02
Lisofylline treatment, n (%)	92 (14%)	22 (11%)	.22
Risk factors for ALI/ARDS			
Infection, n (%)	398 (60%)	142 (68%)	.03
Aspiration, n (%)	106 (16%)	23 (11%)	.08
Trauma, n (%)	79 (12%)	14 (7%)	.04
Multiple transfusions, n (%)	15 (2%)	11 (5%)	.03
Other, n (%)	69 (10%)	19 (9%)	.60
Hypotension, n (%)	225 (34%)	109 (52%)	<.0005
Median (IQR) total bilirubin, mg/dL	1 (0.6–1.8)	1.2 (0.6–2.5)	.04
Mean (SD) creatinine, mg/dL	1.35 (1.23)	1.42 (1.09)	.45
Mean (SD) platelets, 10 ³ /mL	160 (111)	143 (102)	.05
Mean (SD) maximum temperature, °C	38.5 (0.9)	38.6 (1)	.14
Mean (SD) albumin, g/dL	2.22 (0.54)	2.19 (0.56)	.52
Mean (SD) PaO ₂ /FIO ₂ ratio	141 (53)	129 (54)	.005

APACHE, Acute Physiology and Chronic Health Evaluation; ALI, acute lung injury; ARDS, acute respiratory distress syndrome; IQR, interquartile range.

Table 2

Multivariable model examining the association between the development of acute kidney injury and in-hospital mortality

Variable	Odds Ratio	95% CI	p Value
Acute kidney injury	3.36	2.35–4.81	<.001
Age ^a	1.54	1.40–1.70	<.001
Male	1.05	0.76–1.44	.79
White	0.64	0.45–0.93	.02
Low tidal volume ventilation	0.72	0.52–0.98	.04
Ketoconazole	0.71	0.43–1.16	.17
Lisofylline	1.34	0.85–2.13	.21
Hypotension	1.77	1.28–2.46	.001
Bilirubin	1.05	1.00–1.11	.04
Platelet count ^b	1.11	1.02–1.20	.01
PaO ₂ /FIO ₂ ratio ^c	1.17	1.00–1.36	.05
ALI secondary to infection	1.44	1.02–2.02	.04

CI, confidence interval; ALI, acute lung injury.

^aPer 10-yr increase in age;

^bper 50,000/mm³ decrease in platelets;

^cper 50-unit decrease in PaO₂/FIO₂ ratio. Hosmer-Lemeshow goodness-of-fit for multivariable model, $p = .80$.

Table 3

Association between biomarker levels and risk of acute kidney injury

Biomarker	No.	Bivariate OR (95% CI)	p Value	Case Mix-Adjusted OR (95% CI) ^{a,b}	p Value	Multivariable-adjusted OR (95% CI) ^{b,c}	p Value
IL-6	734	1.67 (1.34–2.10)	<.001	1.65 (1.32–2.08)	<.001	1.31 (1.02–1.70)	.04
IL-8	755	1.38 (1.16–1.63)	<.001	1.33 (1.11–1.58)	.001	1.11 (0.91–1.34)	.29
IL-10	572	1.37 (1.12–1.69)	.002	1.34 (1.08–1.66)	.006	1.16 (0.92–1.45)	.2
TNF	362	1.38 (0.92–2.07)	.12	1.38 (0.91–2.08)	.13	1.36 (0.88–2.09)	.16
sTNFR-I	543	3.79 (2.12–6.75)	<.001	3.85 (2.14–6.93)	<.001	3.02 (1.59–5.74)	.001
sTNFR-II	361	3.94 (2.02–7.69)	<.001	3.91 (1.98–7.70)	<.001	2.57 (1.20–5.55)	.02
ICAM-1	754	2.12 (1.32–3.38)	.002	2.07 (1.27–3.40)	.004	1.51 (0.88–2.58)	.14
SP-A	546	1.09 (0.76–1.54)	.65	1.11 (0.80–1.59)	.56	1.13 (0.77–1.65)	.53
SP-D	546	1.01 (0.67–1.54)	.95	1.08 (0.71–1.66)	.71	1.04 (0.66–1.65)	.87
vWF	540	1.65 (0.88–3.12)	.12	1.56 (0.83–2.95)	.17	1.22 (0.65–2.32)	.53
Protein C	755	0.43 (0.22–0.86)	.02	0.49 (0.24–0.98)	.043	0.84 (0.49–1.78)	.66
PAI-1	755	2.58 (1.81–3.69)	<.001	2.57 (1.79–3.69)	<.001	2.08 (1.42–3.05)	<.001

OR, odds ratio; CI, confidence interval; IL, interleukin; TNF, tumor necrosis factor; sTNFR, soluble TNF receptor; ICAM, intracellular adhesion molecule; SP, surfactant protein; vWF, von Willebrand factor; PAI, plasminogen activator inhibitor. Odds ratios are reported per log10 increase in biomarker level.

^a Adjusted for age, sex, race, and interventions;

^b Hosmer-Lemeshow goodness-of-fit, *p* value of >.20 in all cases;

^c adjusted for age, sex, race, interventions, PaO₂/FIO₂ ratio, platelet count, hypotension, bilirubin, and presence of infection.