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Acute kidney injury in non-severe pneumonia is associated with an increased immune response and lower survival

Raghavan Murugan[1], Vijay Karajala-Subramanyam[1], Minjae Lee[1,2], Sachin Yende[1], Lan Kong[1,2], Melinda Carter[1], Derek C. Angus[1] and John A. Kellum[1], on behalf of the Genetic and Inflammatory Markers of Sepsis (GenIMS) Investigators

[1]*The CRISMA Laboratory, Department of Critical Care Medicine, University of Pittsburgh School of Medicine, Pittsburgh, Pennsylvania, USA and* [2]*Department of Biostatistics, University of Pittsburgh Graduate School of Public Health, Pittsburgh, Pennsylvania, USA*

While sepsis is a leading cause of acute kidney injury in critically ill patients, the relationship between immune response and acute kidney injury in less severely ill patients with infection is not known. Here we studied the epidemiology, 1-year mortality, and immune response associated with acute kidney injury in 1836 hospitalized patients with community-acquired severe and non-severe pneumonia. Acute kidney injury developed in 631 patients of whom 329 had severe and 302 had non-severe sepsis. Depending on the subgroup classification, 16–25% of the patients with non-severe pneumonia also developed acute kidney injury. In general, patients with acute kidney injury were older, had more comorbidity, and had higher biomarker concentrations (interleukin-6, tumor necrosis factor, D-dimer) even among patients without severe sepsis. The risk of death associated with acute kidney injury varied when assessed by Gray's survival model and after adjusting for differences in age, gender, ethnicity, and comorbidity. This risk was significantly higher immediately after hospitalization but gradually fell over time in the overall cohort and in those with non-severe pneumonia. A significantly higher risk of death (hazard ratio 1.29) was also present in those never admitted to an intensive care unit. Hence acute kidney injury is common even among patients with non-severe pneumonia and is associated with higher immune response and an increased risk of death.

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Correspondence: *John A. Kellum, Department of Critical Care Medicine, University of Pittsburgh, 604 Scaife Hall, 3550 Terrace Street, Pittsburgh, Pennsylvania 15261, USA. E mail: kellumja@ccm.upmc.edu*

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Sepsis, an immune response to infection, is a leading cause of acute kidney injury (AKI) in severely ill patients,[1–3] and development of AKI is associated with increased risk of death.[2,4] Although several studies reported outcome of sepsis-induced AKI in critically ill patients,[1,2,4–8] less attention has been paid to infected patients with AKI who are less severely ill. For instance, community-acquired pneumonia (CAP) is a common, infectious cause of hospitalization in developed countries.[9] Most patients with CAP are not severely ill and are often treated in non-intensive care settings. However, the epidemiology and outcome of AKI in patients with non-severe CAP is unknown. Furthermore, patients with CAP who develop milder forms of AKI will not meet existing criteria for severe sepsis (i.e., sepsis with coexisting severe acute organ dysfunction),[10] and are therefore unlikely to be enrolled in therapeutic trials of sepsis.[11–13] If AKI is associated with increased risk of death in patients with non-severe CAP and in those without severe sepsis, then early recognition and treatment could improve outcome from AKI.

Inflammation is thought to have a causal role in sepsis-induced AKI in animals,[14–16] but data in humans are inconclusive. Several authors found increased concentrations of inflammatory, coagulation, and fibrinolysis markers in critically ill patients with AKI.[17–19] However, an important limitation of these studies is that it is unclear whether immune markers were associated with AKI itself, or coexisting severe organ dysfunction (severe sepsis) that may have contributed to increased immune response.[20,21] Understanding immune response associated with AKI in patients without severe sepsis is essential for development of immune modulating therapies for AKI.

Using Risk, Injury, and Failure (RIFLE) criteria to classify AKI in a large multicenter cohort of patients hospitalized with CAP, the goals of this prospective observational study

were threefold. First, to describe differences in baseline characteristics and outcomes between hospitalized CAP patients with and without AKI. Second, to examine the risk of AKI in patients with milder forms of CAP and whether development of AKI in an otherwise non-severe pneumonia is associated with adverse outcomes. We used the Pneumonia Severity Index classes I–III,[22] confusion, uremia, respiratory rate, low blood pressure, age 65 years or older (CURB-65) group 1,[23] those who never developed severe sepsis,[24] and those who were never admitted to intensive care unit, as proxies for non-severe CAP. Third, to examine immune, coagulation, and fibrinolysis pathways among patients with and without AKI, in those who do and do not develop severe sepsis.

RESULTS

Baseline characteristics of study participants

Of the 2320 patients enrolled in the study, 291 were excluded because they were discharged from the emergency department, and another 134 patients were excluded because the clinical team ruled out CAP during the first 3 days of hospitalization. Of the remaining 1895 patients with CAP, we excluded patients who were either receiving dialysis or had history of end-stage renal disease ($n=48$), and in whom serum creatinine was never obtained ($n=11$). Of the 1836 patients who formed the study cohort (Figure 1), premorbid baseline creatinine was estimated in 1745 patients using the recommended Modification of Diet in Renal Disease equation.

Of the 631 patients (34%) who met criteria for AKI, 307 (49%), 135 (21%), and 189 (30%) reached maximum RIFLE stages, respectively, during hospitalization. Of the 91 patients with known premorbid baseline creatinine, we found moderate agreement between premorbid creatinine and the estimated creatinine to classify patients with AKI (Cohen's κ coefficient = 0.70, 95% CI 0.52–0.87, $P<0.0001$). Nearly two-thirds of patients developing AKI had already done so at hospital admission ($n=399$, 63.2% of patients with AKI). Table 1 shows the demographic characteristics and severity of illness on day 1 of hospital admission stratified by AKI, and by severity of AKI. Patients with AKI were predominantly older, of white race, had more preexisting comorbidity, and had more severe CAP.

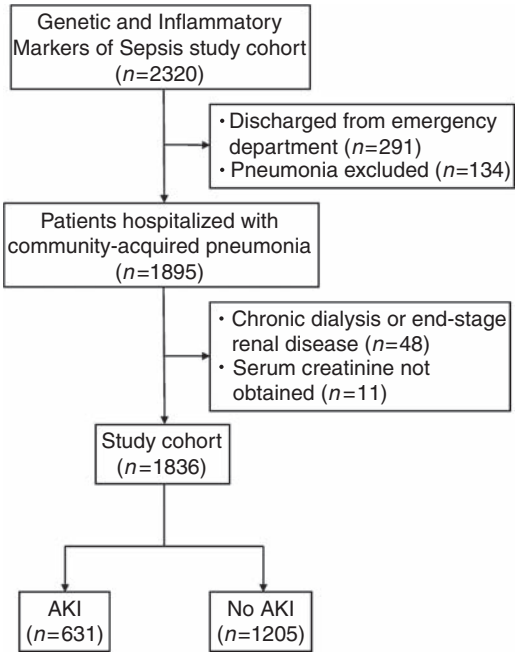

Figure 1 | Subject disposition for the Genetic and Inflammatory Markers of Sepsis study cohort. AKI, acute kidney injury

Hospital course and outcome for patients with and without AKI

Table 2 shows hospital course and outcomes for patients with and without AKI. One-third (31.1%, $n=572$) of CAP patients developed severe sepsis (i.e., had a Sequential Organ Failure Assessment (SOFA) score[10] of $\geqslant 3$ in at least one organ system during the entire hospitalization), of which more than one-half (57.5%) developed AKI ($n=329$). Although overall intensive care use was only 16%, more than a third of patients with AKI (39%) were admitted to the intensive care unit at some point during hospitalization, of which 18% received mechanical ventilation. Patients with AKI, on an average, incurred higher median hospital length of stay compared to patients without AKI (8 vs 5 days, $P<0.001$).

Patients with AKI had a higher risk of death at each time point, such as by hospital discharge (11 vs 1.3%, $P<0.001$), by 90 days (24 vs. 9.8%, $P<0.001$), and by 1 year (36.3 vs 20.1%, $P<0.001$; Table 2). Mortality increased with increased severity of AKI and was 29.6, 40.7, and 43.9% for maximum stages RIFLE, respectively, at 1 year ($P<0.001$; Figure 2a).

Using Gray's survival model, we found that the hazard ratios for association between AKI and risk of death varied significantly over 365 days after hospitalization for CAP. The unadjusted hazard ratios were highest during the first 100 days after presentation, subsequently decreased, but nevertheless persisted over the entire 1 year of follow-up (hazard ratio range 1.45–2.79, $P<0.001$; Table 4). When adjusted for differences in age, gender, race, and the Charlson Comorbidity Index the hazard ratios were attenuated (adjusted hazard ratio range 1.10–2.10, $P<0.001$; Table 4), but nevertheless remained significant up to 100 days after CAP (Figure 3a).

Similar risk of death was present when absolute (adjusted hazard ratio 1.12, 95% CI 1.04–1.20, $P=0.001$), relative (adjusted hazard ratio 1.12, 95% CI 1.04–1.19, $P=0.002$), as well as ratio (adjusted hazard ratio 1.10, 95% CI 1.04–1.17, $P=0.001$) of change in glomerular filtration rate were used in the model instead of AKI. We also found that the mortality rates were not different between patients who presented with AKI at hospital admission and those who developed AKI during hospitalization. Similarly, there was

Table 1 | Baseline characteristics of patients with AKI following CAP

Characteristic	No. (%)						
	AKI[a] (*n*=631)	No AKI (*n*=1205)	*P*-value	Risk[b] (*n*=307)	Injury[b] (*n*=135)	Failure[b] (*n*=189)	*P*-value
Age, mean (s.d.), years	73.4 (14.5)	65.2 (17.1)	<0.001	74.2 (14)	76.4 (13)	70 (15.4)	<0.001
Male gender	320 (51)	634 (53)	0.43	153 (24)	63 (10)	103 (16)	0.303
White race	542 (85)	951 (79)	0.003	273 (43)	114 (18)	155 (24)	0.085
Baseline creatinine	0.93 (0.3)	0.89 (0.2)	<0.001	0.9 (0.1)	0.9 (0.1)	0.9 (0.5)	0.042
Charlson Comorbidity Index[c]							
Mean (s.d.)	2.1 (2.2)	1.7 (2.1)	<0.001	2.1 (2.2)	2.2 (2.1)	1.8 (2.0)	0.109
Index >0	491 (77.8)	831 (68.9)	0.0001	239 (77.8)	111 (82.2)	141 (74.6)	0.266
Chronic kidney disease	24 (3.8)	9 (0.7)	<0.001	5 (1.6)	9 (6.6)	10 (5.3)	0.017
Cardiac disease	207 (38.2)	260 (24.1)	<0.001	111 (53.6)	49 (23.7)	47 (22.7)	0.116
Lung disease	225 (36)	478 (40)	0.093	117 (52)	49 (21.8)	59 (26.2)	0.293
Diabetes	151 (23.9)	210 (17.4)	0.001	69 (22.5)	42 (31.1)	40 (21.1)	0.083
Prior antibiotic use	101 (16)	222 (18.4)	0.20	52 (17)	18 (13.3)	31 (16.4)	0.541
Pneumonia severity index, mean (s.d.)[d]	105.5 (31.4)	78.5 (28.5)	<0.001	116.5 (37.6)	126.8 (34.5)	124.5 (39.4)	0.009
Pneumonia severity index class							
I and II	81 (13)	487 (40.4)	<0.001	26 (62)	4 (9.5)	12 (28.5)	0.035
III	128 (20.3)	334 (28)		52 (59.1)	17 (19.3)	19 (21.6)	
IV	283 (45)	330 (27.3)		134 (48.5)	55 (20)	87 (31.5)	
V	139 (22)	54 (4.5)		95 (42.3)	59 (26.2)	71 (31.5)	
CURB-65 score, mean (s.d.)[e]	2.4 (1.0)	1.4 (1.0)	<0.001	2.3 (1.07)	2.6 (0.9)	2.3 (1.1)	0.0042
CURB-65 group							
I	132 (21)	674 (56)	<0.001	73 (23.8)	14 (10.4)	45 (24)	0.013
II	195 (31)	331 (27.4)		92 (30)	43 (32)	60 (31.7)	
III	304 (48)	200 (16.6)		142 (46.2)	78 (58)	84 (44.5)	
APACHE III score, mean (s.d.)[f]	47 (15.2)	36.7 (11.8)	<0.001	44.8 (14)	47.1 (14)	50.3 (17)	<0.001
SOFA score, mean (s.d.)[g]	3.3 (2.2)	1.8 (1.4)	<0.001	2.8 (2)	3.6 (1.9)	4 (2.5)	<0.001
Severe sepsis on day 1[h]	165 (26.1)	102 (8.46)	<0.001	59 (19.2)	26 (19.2)	80 (42.3)	<0.001

Abbreviations: AKI, acute kidney injury; APACHE, Acute Physiology and Chronic Health Evaluation; CURB-65, confusion, uremia, respiratory rate, low blood pressure, age 65 years or older; s.d., standard deviation; SOFA, Sequential Organ Failure Assessment.
[a]Patients were classified to have developed AKI if they met any of Risk, Injury, or Failure stages at any time during hospitalization as proposed by the Acute Dialysis Quality Initiative.[30]
[b]For severity of AKI, patients were classified according to the maximum RIFLE stage (Risk, Injury, or Failure) reached during the entire hospitalization as proposed by the Acute Dialysis Quality Initiative.[30]
[c]According to the method of Charlson *et al.*[33]
[d]Pneumonia Severity Index was measured according to criteria by Fine *et al.*[22] in the emergency department in 1546 (84.2%) subjects. There were no significant differences between subjects who did and did not have a Pneumonia Severity Index measured.
[e]According to criteria by Lim *et al.*[23]
[f]APACHE III score assessed on first hospital day regardless of whether subject was admitted to an intensive care unit or not.[34]
[g]SOFA score assessed on the first day of hospital admission according to criteria by Vincent *et al.*[10]
[h]Defined as sepsis plus acute organ dysfunction according to 2001 international consensus criteria for severe sepsis.[24]

Table 2 | Hospital course and outcomes in patients with and without AKI in the overall cohort

Characteristic[a]	No. (%)		
	AKI	No AKI	*P*-value
Developed severe sepsis	329 (52.1)	243 (20.1)	<0.001
Intensive care unit admission	245 (39)	47 (4)	<0.001
Mechanical ventilation	116 (18.4)	13 (1)	<0.001
Length of hospital stay, median (IQR)	8 (12–5)	5 (7–4)	<0.001
Hospital mortality	70 (11.1)	16 (1.3)	<0.001
90-day mortality	151 (24)	118 (9.8)	<0.001
1-year mortality	229 (36.3)	242 (20.1)	<0.001

Abbreviations: AKI, acute kidney injury; IQR, interquartile range.
[a]Hospital course and outcome in the entire cohort of 1836 patients with community-acquired pneumonia.

no difference in mortality associated with AKI among those with and without chronic kidney disease (data not shown).

Risk of AKI, hospital course, and outcome in non-severe CAP

Not surprisingly, the risk of AKI was lower in patients with non-severe CAP. Of the 1030 patients within Pneumonia Severity Index classes I–III, 20.3% of patients ($n=209$) developed AKI. Of the 806 patients within CURB-65 group 1, 16.4% of patients ($n=132$) developed AKI. Of patients who never developed severe sepsis ($n=1264$) and those never admitted to the intensive care unit ($n=1544$), the risk of AKI was 23.8% ($n=302$) and 25% ($n=386$). Patients with AKI within Pneumonia Severity Index classes I–III and CURB-65 group 1 were more likely to develop severe sepsis, be admitted to intensive care unit, and be mechanically ventilated ($P<0.001$; Table 3).

Non-severe CAP patients with AKI incurred higher length of hospital stay, in-hospital and 90-day mortality (Table 3). At 1 year, mortality varied from 17.2 to 34.2% for patients with AKI within the four non-severe CAP subgroups

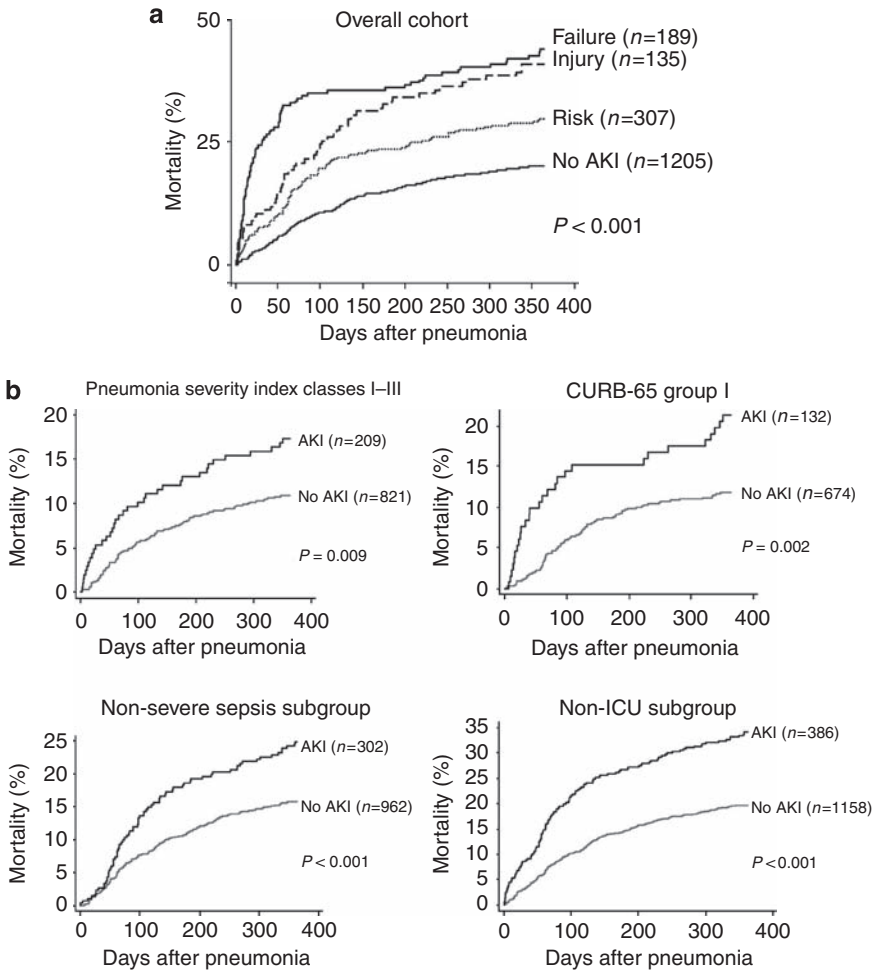

Figure 2 | One-year mortality in patients with and without AKI in the overall cohort and within non-severe CAP subgroups. (**a**) The Kaplan–Meier failure plots by maximum RIFLE stage for probability of death in the entire CAP cohort, which at 1 year was higher in patients with AKI than in patients without AKI (log rank $P<0.001$). (**b**) Failure plots for probability of death at 1 year within the four non-severe CAP subgroups. Non-severe CAP patients with AKI in each of the four subgroups had higher probability of death associated with AKI at 1 year compared to those without AKI (log rank $P<0.01$ for all subgroups). AKI, acute kidney injury; CAP, community-acquired pneumonia; RIFLE, Risk, Injury, and Failure criteria.

(Figure 2b). Patients admitted to medical wards with AKI had higher mortality compared to those without AKI (34.2 vs 19.7%, $P<0.001$). This increased risk of death was not only seen with more severe AKI. Of patients with stage Risk (61%), the mortality was higher with AKI (30.1 vs 19.6%, $P<0.001$) (hazard ratio 1.67, 95% CI 1.28–2.18, $P<0.001$).

The unadjusted hazard ratios for 1-year mortality ranged from 1.03 to 3.65 within the four non-severe CAP subgroups (Table 4). The hazard ratios remained unchanged when adjusted for age, gender, race, and the Charlson Comorbidity Index for those admitted to medical wards with AKI (adjusted hazard ratio 1.29, 95% CI 1.03–1.60, $P<0.023$). Using Gray's model, we found that the adjusted hazard ratio for death in patients within CURB-65 group 1 varied over the 1-year follow-up (adjusted hazard ratio range 0.85–2.97, $P=0.001$). The risk of death associated with AKI was high immediately after CAP in patients admitted to hospital (hazard ratio 2.73, 95% CI 1.52–4.92). Subsequently, although the hazard ratios declined, the risk of death associated with AKI nevertheless remained significant up to 50 days (Figure 3b).

Biomarker concentration in patients with and without AKI

Figure 4 shows the plasma biomarker concentrations during the first week of hospitalization among patients with and without AKI stratified by severe sepsis. Patients with AKI who developed severe sepsis had higher concentrations of interleukin-6, 10, and tumor necrosis factor at presentation to the emergency department and during the first week of CAP (Figure 4a). The geometric mean concentration on day 1 for AKI vs no AKI was—interleukin-6: 111.8 vs 42 pg/ml, $P<0.001$; interleukin-10: 8.9 vs 6.0 pg/ml, $P=0.026$; and tumor necrosis factor: 8.4 vs 4.5 pg/ml, $P<0.001$. Importantly, of patients without severe sepsis, similar patterns in interleukin-6 and tumor necrosis factor concentrations were noted in patients with and without AKI during the first week of CAP ($P<0.001$), although the magnitude of difference was

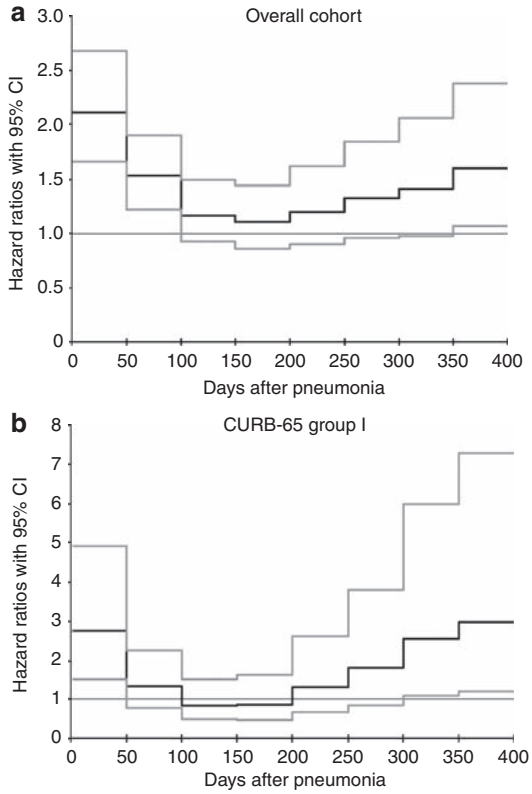

Figure 3 | Varying risk of death associated with AKI over one year after CAP hospitalization. Varying hazard ratios with 95% confidence intervals (CIs) for risk of death associated with AKI over 365 days among the overall cohort (**a**), and within the CURB-65 group 1 subgroup (**b**). Hazard ratios were estimated from Gray's model using nine time nodes and eight intervals and adjusted for age, gender, race, and Charlson Comorbidity Index. In the overall cohort, the 95% CIs are above 1 for 100 days in patients with AKI, suggesting significant increased risk of death associated with AKI. Thereafter the risk declines showing no association between AKI and risk of death (**a**). In the CURB-65 group 1 subgroup, the association between AKI and risk of death persists up to 50 days following hospitalization for pneumonia (**b**). AKI, acute kidney injury; CURB, confusion, uremia, respiratory rate, low blood pressure, age 65 years or older.

only modest. The geometric mean concentration on day 1 in those without severe sepsis for AKI vs no AKI was—interleukin-6: 48.6 vs 35 pg/ml, $P<0.03$; and tumor necrosis factor: 6.5 vs 4.9 pg/ml, $P<0.001$.

Significant derangement in coagulation occurred in patients with AKI (Figure 4b). Of patients who developed severe sepsis, higher concentration of thrombin–antithrombin complexes ($P<0.001$) and lower concentration of antithrombin persisted during the first week of CAP ($P<0.001$) in patients with AKI. Day 1 geometric mean concentration for AKI vs no AKI was 6.3 vs 4.9 μg/ml for thrombin–antithrombin complexes ($P=0.08$) and 79.8 vs 88.1 μg/ml, for antithrombin ($P<0.001$). In contrast, of patients without severe sepsis, circulating concentrations of thrombin–antithrombin complexes, factor IX, and antithrombin did not differ between those with developing and not developing AKI (Figure 4b).

Table 3 | Hospital course and outcomes in patients with and without AKI in non-severe CAP subgroups

	No. (%)		
Characteristic	AKI	No AKI	*P*-value
Pneumonia Severity Index classes I–III[a]	*(n=209)*	*(n=821)*	
Developed severe sepsis	82 (39.2)	109 (13.2)	<0.001
Intensive care unit admission	74 (35.4)	24 (3)	<0.001
Mechanical ventilation	35 (16.7)	5 (0.6)	<0.001
Length of hospital stay, median, IQR	7 (5–10)	5 (4–7)	<0.001
Hospital mortality	10 (4.8)	3 (0.4)	<0.001
90-day mortality	20 (9.6)	40 (4.9)	0.009
1-year mortality	36 (17.2)	89 (10.8)	0.011
CURB-65 group 1[b]	*(n=132)*	*(n=674)*	
Developed severe sepsis	67 (50.8)	96 (14.2)	<0.001
Intensive care unit admission	64 (48.5)	20 (3)	<0.001
Mechanical ventilation	35 (26.5)	5 (0.7)	<0.001
Length of hospital stay, median, IQR	9 (5–15)	5 (4–7)	<0.001
Hospital mortality	9 (6.8)	2 (0.3)	<0.001
90-day mortality	18 (13.6)	35 (5.2)	<0.001
1-year mortality	28 (21.2)	79 (11.7)	0.003
Non-severe sepsis cohort[c]	*(n=302)*	*(n=962)*	
Intensive care unit admission	53 (17.5)	30 (2)	<0.001
Length of hospital stay, median, IQR	6 (5–8)	5 (4–7)	<0.001
Hospital mortality	3 (0.99)	1 (0.1)	0.016
90-day mortality	36 (11.9)	67 (6.9)	0.006
1-year mortality	75 (24.8)	152 (15.8)	<0.001
Non-ICU cohort[d]	*(n=386)*	*(n=1158)*	
Developed severe sepsis	137 (35.5)	216 (18.6)	<0.001
Length of hospital stay, median, IQR	6 (4–8)	5 (4–7)	<0.001
Hospital mortality	20 (5.1)	14 (1.2)	<0.001
90-day mortality	77 (19.9)	108 (9.3)	<0.001
1-year mortality	132 (34.2)	228 (19.7)	<0.001

Abbreviations: AKI, acute kidney injury; CAP, community-acquired pneumonia; CURB-65, confusion, uremia, respiratory rate, low blood pressure, age 65 years or older; IQR, interquartile range.
[a]Hospital course and outcome in the subgroup of 1030 non-severe CAP patients as defined by Pneumonia Severity Index classes I–III at presentation to the emergency department according to criteria by Fine *et al.*[22]
[b]Hospital course and outcome in the subgroup of 806 non-severe CAP patients as defined by CURB-65 group 1 at presentation to the emergency department according to criteria by Lim *et al.*[23]
[c]Hospital course and outcome in the subgroup of 1264 non-severe CAP patients as defined by maximum Sequential Organ Failure Assessment score less than 3 during entire hospitalization according to criteria by Vincent *et al.*[10]
[d]Hospital course and outcome in the subgroup of 1544 patients who were never admitted to the ICU during the entire hospitalization.

Patients with severe sepsis who developed AKI had marked elevation in D-dimer and plasminogen activator inhibitor-1 concentrations during the first week of pneumonia ($P=0.001$; Figure 4c). The geometric mean concentration on day 1 in AKI vs no AKI was—D-dimer: 901 vs 643 ng/ml, $P=0.019$; and plasminogen activator inhibitor−1: 7.9 vs 4.0 ng/ml, $P=0.001$. In contrast, of patients without severe sepsis, only circulating D-dimer concentration was higher on day 1 (689.3 vs 494.9 ng/ml, $P=0.001$) and during the first week of CAP ($P<0.001$).

DISCUSSION

We found, perhaps not surprisingly, that AKI was very common in CAP. Importantly, however, even in those who appeared to have an uncomplicated course, as defined by

Table 4 | Hazard ratios for association between AKI and 1-year mortality

Characteristic	Hazard ratio for mortality in patients with AKI	95% CI	*P*-value
Unadjusted			
Overall cohort[a]	1.45–2.79	1.12–3.54	<0.001
Pneumonia Severity Index classes I–III	1.66	1.13–2.45	0.01
CURB-65 group 1[b]	1.03–3.65	0.57–8.94	0.0001
Non-severe sepsis subgroup	1.67	1.26–2.19	0.0003
Non-intensive care unit subgroup	1.94	1.57–2.41	<0.001
Adjusted[c]			
Overall cohort[a]	1.10–2.10	0.85–2.67	<0.001
Pneumonia Severity Index classes I–III	1.39	0.94–2.05	0.10
CURB-65 group 1[b]	0.85–2.97	0.47–7.29	0.001
Non-severe sepsis subgroup	1.14	0.86–1.51	0.35
Non-intensive care unit subgroup	1.29	1.03–1.60	0.023

Abbreviations: AKI, acute kidney injury; CI, confidence interval; CURB-65, confusion, uremia, respiratory rate, low blood pressure, age 65 years or older.
[a,b]Hazard ratios for association between AKI and 1-year mortality for overall cohort and for the subgroup CURB-65 group 1 was estimated from Gray's model[39] using nine time nodes and eight intervals. Range of hazard ratios and 95% CI estimates from the Gray's model are shown.
[c]Adjusted for age, gender, race, and Charlson Comorbidity Index.

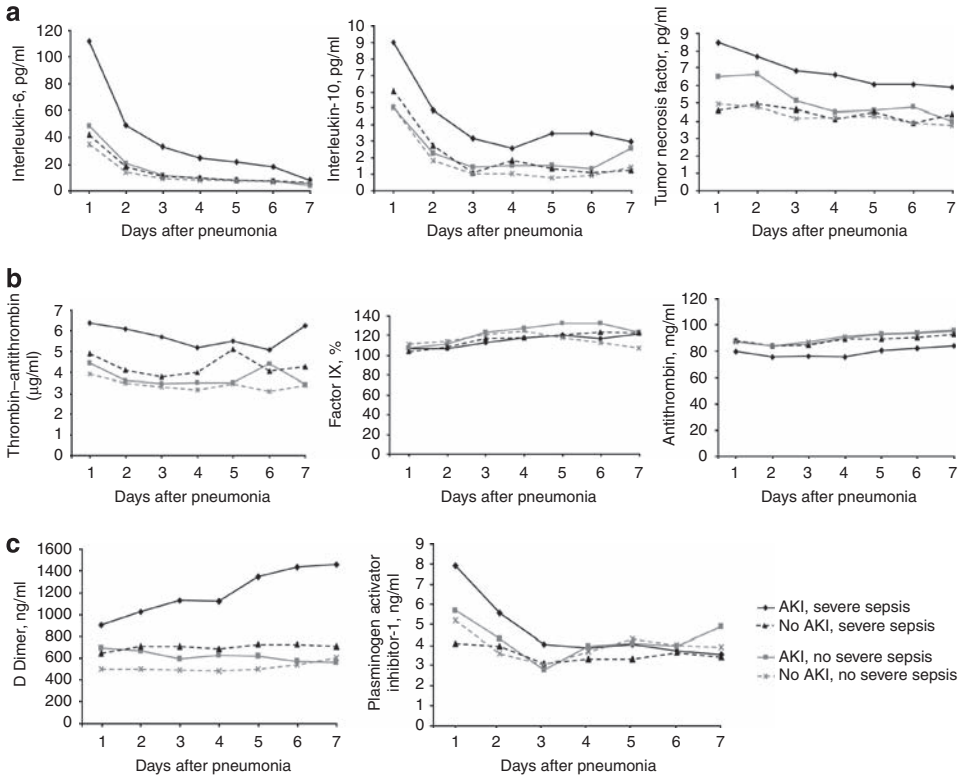

Figure 4 | Circulating inflammatory, coagulation, and fibrinolysis marker concentrations in 1380, 712, and 710 patients with and without acute kidney injury, stratified by severe sepsis during first week of CAP. (**a**) Inflammatory biomarkers (interleukin (IL)-6, -10 and tumor necrosis factor (TNF)). (**b**) Coagulation biomarkers (thrombin–antithrombin complexes, factor IX, and antithrombin (AT)). (**c**) Biomarkers of fibrinolysis (D-dimer and plasminogen activator inhibitor-1 (PAI-1)). Geometric mean plasma concentrations of IL-6, 10, TNF, TAT, D-dimer, and PAI-1were significantly increased in patients with AKI who developed severe sepsis (solid line with diamonds) when compared to patients without AKI (dashed line with triangles), during the first week of pneumonia ($P<0.001$). In contrast, plasma antithrombin concentrations were lower in patients with AKI ($P<0.001$). Of patients without severe sepsis, plasma concentrations of IL-6, TNF, and D-dimer were significantly higher in patients with AKI (solid line with squares) compared to those without AKI (dashed line with crosses) ($P<0.001$). AKI, acute kidney injury; CAP, community-acquired pneumonia.

different criteria, AKI was still very common, developing in around one-quarter of patients. In this latter group, development of AKI was associated with considerable mortality. Within those developing severe sepsis, AKI occurred in patients who also had evidence of greater activation of inflammatory, coagulation, and fibrinolysis pathways. Among the patients with less severe CAP, AKI was associated with modest but significant differences in immune and fibrinolysis activation. This study, to the best of our knowledge, is the first large-scale investigation of the

epidemiology and outcome of AKI, and immune response to CAP in patients with AKI. The result of our study suggests that AKI is common in patients with CAP even among those without severe sepsis or even severe CAP. Furthermore, the immune response to infection is different in patients with and without AKI.

An overall 34% incidence of AKI in patients with CAP is very similar to that reported for critically ill patients.[5] Our estimate is likely to rise further with increasing prevalence of chronic kidney disease as the populations in the developed world and even in parts of the developing world age. Development of AKI was associated with high risk of death early on, declining subsequently, yet persisting up to 100 days after hospitalization for CAP. Our findings are surprising given that most patients with AKI were discharged by 8 days, suggesting that the risk of death persists even though the clinical signs have resolved and subjects appeared to be stable for discharge after CAP.[25]

We also found that AKI was associated with a significant increase in the risk of death even in patients with non-severe CAP. We chose four different definitions for 'non-severe' CAP. Pneumonia Severity Index and CURB-65 are widely validated severity indices for CAP. We also used absence of severe sepsis and lack of intensive care use as two additional methods to identify patients who were less severely ill throughout hospitalization with pneumonia. Surprisingly, we found that many patients with non-severe CAP admitted to medical wards developed AKI and although most had mild AKI (Risk), there was still an increased risk of death. This finding is consistent with other studies of AKI showing that small changes in kidney function are associated with higher mortality.[26–28] Similarly, of the lower severity patients defined by CURB-65, development of AKI was still associated with higher mortality. Importantly, these non-severe CAP patients with AKI would not have met inclusion criteria for any of the major sepsis clinical trials and yet the presence of AKI was associated with a significant increase in mortality. We speculate that this group of patients may be better suited to the study of novel therapies for AKI because their comorbidities are fewer and the risk of death attributable to AKI may be more readily modifiable than that for AKI in severely ill patients.

As expected, we found higher concentrations of inflammatory, coagulation, and fibrinolysis markers in patients with AKI who developed severe sepsis similar to other studies.[17,29] However, a key finding of our study is that patients with AKI but without severe sepsis also had higher inflammatory and fibrinolysis markers. These observations suggest that the immune response in CAP is different in patients with AKI and may have a bidirectional relationship. Higher immune response might have caused AKI or might have occurred as a consequence of AKI (e.g., decreased elimination, increased cytokine release) or both. Because many of our patients had AKI on day 1, we were unable to delineate a temporal association to support any cause–effect relationship between AKI and immune response.

There are important limitations to our study. First, as is the case with most studies of AKI, we did not know premorbid creatinine in many patients and relied on screening for chronic kidney disease by medical history and estimated creatinine according to recommended guidelines.[30] This might have resulted in an overestimate in the incidence of AKI if there was a higher than reported underlying prevalence of chronic kidney disease among our patients. Nevertheless, we found that there was a moderate concordance between known premorbid creatinine and estimated baseline creatinine in classifying patients with AKI, suggesting that our results are less likely to be affected by misclassification bias.[31]

Second, we were unable to control for fluid, hemodynamic, and other concurrent interventions that could have influenced course of AKI, biomarker patterns, and outcomes. Third, there is no gold standard for the term 'non-severe CAP'. We therefore presented results defining non-severe CAP in four ways. We chose these definitions on the basis of clinical face validity, but recognize that the definitions are arbitrary. Fourth, we did not account for the competing risk between mortality associated with sepsis-induced AKI and subsequent decline in kidney function, ultimately reflected as either chronic kidney disease or end-stage renal disease, due to smaller cohort and shorter duration of follow-up.

Our study also has several strengths. First, being a multicenter study, our finding of high risk of AKI and its association with adverse outcomes independent of severity of CAP is highly generalizable. Second, we showed that the risk of death associated with AKI persists long after discharge following CAP hospitalization, though the mechanism responsible for late death is unclear in patients with AKI. Third, by considering subjects with CAP alone, we enrolled a more homogenous group of patients with early sepsis (CAP), unlike earlier studies that enrolled patients with different infections and at varying time points. Fourth, we were able to quantify that most sepsis-induced AKI occurs before hospitalization and therefore were able to examine the timing of AKI. Fifth, we carefully measured biomarkers from the time of admission when biomarker activation is generally highest and when concentrations are least likely to be modified by therapy. We stratified biomarker analysis by severe sepsis to account for the independent relationship between AKI and biomarkers in the absence of coexisting organ dysfunction; unlike many other studies that measured cytokines late and after coexisting organ dysfunctions were already evident.

In summary, the results of our study show that AKI occurs in approximately one-third of patients with CAP, and in a quarter of patients with non-severe CAP. Occurrence of AKI is associated with higher concentrations of immune and fibrinolysis markers even in those without severe sepsis, and an increased risk of death. Our study underscores the importance of increased awareness of this high incidence of AKI and mortality after pneumonia, and the need for development of prevention/treatment strategies especially for patients with non-severe CAP who have not typically been

included in sepsis trials. In addition, further research is needed to understand the mechanism of death after AKI, particularly as the risk appears to persist long after AKI and CAP have resolved.

MATERIALS AND METHODS

Study design and selection of participants

The analyses were based on all hospitalized patients enrolled in the Genetic and Inflammatory Markers of Sepsis (GenIMS) study.[32] GenIMS is a large, multicenter, observational cohort study of subjects with CAP presenting to the emergency departments of 28 teaching and nonteaching hospitals in the United States between November 2001 and November 2003. Eligible patients were $\geqslant 18$ years old and had a clinical and radiological diagnosis of CAP using criteria by Fine *et al.*[22] Exclusion criteria included transfer from another hospital, discharge from an acute care hospital within the previous 10 days, pneumonia within the previous 30 days, chronic dependency on mechanical ventilation, cystic fibrosis, active pulmonary tuberculosis, admission for palliative care, prior enrollment in the study, incarceration, and pregnancy. The institutional review boards at all participating sites approved the study, and we obtained written informed consent from all participants or their proxies.

Methods of data collection

We gathered baseline and sequential clinical information by structured patient or proxy interviews. We prospectively ascertained comorbid conditions using the Charlson Comorbidity Index,[33] and severity of illness using the Acute Physiology and Chronic Health Evaluation III score,[34] and Pneumonia Severity Index.[22] We calculated CURB-65 score[23] retrospectively with altered mental status or a new change in Glasgow Coma Scale score as proxy measures for confusion.[35] We defined severe sepsis as infection and acute organ dysfunction, following international consensus criteria.[24] We defined acute organ dysfunction as a new SOFA score of three or greater in any of six organ systems.[10]

Definition of acute kidney injury

We classified AKI using the maximum RIFLE stages, as proposed by the Acute Dialysis Quality Initiative[30] at any time during hospitalization. The RIFLE stage was determined based on the worse of either serial serum creatinine or urine output. For patients with no known premorbid creatinine and no known medical history of chronic kidney disease, we estimated premorbid creatinine using the Modification of Diet in Renal Disease equation,[36] as recommended by the Acute Dialysis Quality Initiative. We then selected the lower creatinine value from either the hospital admission creatinine or the Modification of Diet in Renal Disease creatinine as the baseline value.[37] Patients were classified as stage Risk, if serum creatinine was 1.5 times the baseline creatinine, or urine output <0.5 ml/kg/h for 6 h; stage Injury, if serum creatinine was twice the baseline, or urine output <0.5 ml/kg/h for 12 h; stage Failure, if serum creatinine was thrice the baseline, or creatinine $\geqslant 4$ mg/dl with an acute rise >0.5 mg/dl, or urine output <0.3 ml/kg/h for 24 h, or anuria for 12 h.[30]

Criteria for non-severe CAP

We used four different sets of criteria to classify the severity of CAP.[22–24,38] We defined non-severe CAP as classes I–III for Pneumonia Severity Index calculated at admission and group 1 for CURB-65 to identify patients who had non-severe CAP at hospital admission. We also chose patients who did not develop severe sepsis according to the SOFA score of less than three in any of six organ systems, and those never admitted to the intensive care unit to identify patients who were less severely ill with CAP throughout index hospitalization.

Outcome variables

Our primary outcome was 1-year all-cause mortality. Secondary outcomes included length of stay, in-hospital and 90-day mortality. Study coordinators ascertained deaths in hospital and post-discharge mortality was ascertained by telephone and National Death Index search.

Laboratory procedures

Blood was drawn for cytokines (tumor necrosis factor, interleukin-6, -10), coagulation (thrombin–antithrombin complexes, antithrombin, factor IX), and fibrinolysis (plasminogen activator inhibitor-1, and D-dimer) markers in the emergency department and daily for the first week, while a subject remained in hospital. Detailed sample handling and laboratory procedures have been described elsewhere.[32] We rechecked all outlier values and masked laboratory personnel to clinical data. Although we analyzed inflammatory markers in 1380 patients, for logistical and cost reasons, coagulation and fibrinolysis markers were assessed only in a random subset of 712 and 710 patients.

Statistical analysis

We conducted univariate comparisons using χ^2-tests, Student's *t*-tests, or their nonparametric equivalents, as appropriate. We assessed 1-year mortality by comparing Kaplan–Meier failure plots using log-rank test. We conducted survival analyses using Cox proportional hazard model to estimate hazard ratios for association between AKI and 1-year mortality. We used Gray's model to estimate hazard ratios for nine time nodes and eight intervals, if hazards failed Cox's proportionality assumption.[39] We adjusted for potential confounders by expanding the models to include age, sex, race/ethnicity, and the Charlson Comorbidity Index.

We present sequential biomarker data as plots of daily means stratified by development of AKI and severe sepsis. We assumed a log-normal distribution of biomarkers and analyzed data in natural log scale. We constructed Tobit models to account for truncated data that fell below detection thresholds,[40] and generated daily geometric means to compare the concentration between groups for a single time point. For sequential data, we conducted regression analysis with mixed models that accounted for correlation of repeated measures over time,[41] incorporating Tobit models as necessary.[42] Models included linear and quadratic terms to allow evaluation of trends. All analyses were performed using SAS 9.0 (SAS Institute, Cary, NC, USA) assuming statistical significance at $P<0.05$.

DISCLOSURE

All the authors declared no competing interests.

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Author contributions: Murugan, Lee, Kong and Kellum had had full access to all of the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis. Study concept and design: Murugan, Kellum, and Angus. Acquisition of data: Kellum and Angus. Cytokine analysis: Carter. Analysis and interpretation of data: Murugan, Kellum, Yende, Karajala-Subramanyam, Lee, Kong, and Angus. Drafting of the paper: Murugan, Karajala-Subramanyam, Kellum, Yende, and Angus. Critical revision of the paper for important intellectual content: Kellum, Yende, Angus, Kong, Lee, and Carter. Statistical analysis: Lee and Kong. Obtainment of funding: Kellum and Angus. Administrative, technical, or material support: Kong, Kellum, and Angus. Study supervision: Kellum and Angus.

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