Clinical prognostic significance of serum high mobility group box-1 protein in patients with community-acquired pneumonia

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Abstract

Objective: To investigate the relationship between serum high mobility group box-1 protein (HMGB-1) levels and prognosis in patients with community-acquired pneumonia (CAP).

Methods: This prospective study included 35 patients who attended our hospital from January 2016 to December 2016. Pneumonia severity was defined by pneumonia severity index (PSI). Serum levels of C-reactive protein (CRP), cortisol, and HMGB-1 were analyzed in relation to disease severity and clinical outcome.

Results: High HMGB-1 levels were associated with high cortisol levels. High HMGB-1 and high cortisol were both significantly associated with high white blood cell count and high serum CRP, compared with low HMGB-1 and low cortisol, respectively. PSI score and 30-day mortality were also significantly higher in patients with high HMGB-1 or high cortisol levels compared with patients with low HMGB-1 or cortisol levels, respectively. CRP, cortisol, and HMGB-1 levels were all significantly higher in patients who died compared with survivors.

Conclusion: HMGB-1 was associated with clinical outcomes and was an independent risk factor for 30-day mortality in patients with CAP. Serum HMGB-1 levels were also positively correlated with serum levels of cortisol. These results demonstrate a role for HMGB-1 in CAP, and suggest possible new therapeutic targets for patients with CAP.

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Introduction

Despite widespread use of antibiotics over the last 70 years, community-acquired pneumonia (CAP) remains a serious threat to global health, with an incidence of 3.3 to 46 per 1000 per year in the elderly population.[1–3] CAP has high mortality rates of up to 48%, and is the most common cause of death in patients with severe infectious diseases.[4,5] Numerous factors have been shown to influence mortality among patients with severe CAP, including age, comorbidities, pneumonia severity, presence of bacteremia, and serum levels of inflammatory factors.[6,7]

Many inflammatory factors are thought to be associated with development and prognosis of CAP, including interleukin (IL)-1, IL-6, IL-8, IL-10, tumor necrosis factor-α, C-reactive protein (CRP), procalcitonin, and cortisol.[8,9] Some of these factors are thought to be correlated with mortality risk in CAP patients, and some factors have been implicated as independent risk factors for death in patients with severe CAP.[10–12] The nuclear high mobility group box-1 protein (HMGB-1) has demonstrated important roles in cancer and inflammatory processes such as breast cancer and sepsis.[13–15] HMGB-1 was also shown to be elevated in patients with uncomplicated pneumonia and pneumonia with severe sepsis,[16] and was associated with *Pseudomonas aeruginosa* pneumonia in patients with cystic fibrosis.[17] However, few studies have examined the relationships of HMGB-1 with prognosis and mortality in patients with CAP.

In the present study, we investigated the relationships between serum levels of HMGB-1 and prognosis in patients with CAP, and between HMGB-1 and cortisol. These results provide clinical evidence of the role of HMGB-1 in CAP, and suggest potential new therapeutic targets for patients with CAP.

Patients and methods

Patients

This prospective study included 35 inpatients who attended the Breath Internal Medicine Department at the First Affiliated Hospital of Guangxi Medical University from January 2016 to December 2016. All patients were diagnosed with CAP according to the criteria of the American Thoracic Society guidelines for pneumonia.[18] All patients were over 18 years old, and all had pulmonary infiltration diagnosed by chest X-ray and clinical symptoms including cough, purulent sputum, positive auscultation, or fever. Patients with any serious systemic or respiratory diseases at admission were excluded, including patients with pulmonary tuberculosis, bronchial asthma, chronic obstructive pulmonary disease, idiopathic pulmonary fibrosis, urinary tract infection, and cancer. The severity of pneumonia was defined according to the pneumonia severity index (PSI) as mild/moderate CAP I–III or severe CAP IV–V, as described previously.[19] Informed consent was obtained from all patients within 24 hours after admission. The present study was approved by the

Ethics Committee of the First Affiliated Hospital of Guangxi Medical University.

Data collection and measurement

Demographic data including age, sex, and antibiotic use in the 1 month prior to admission were collected from the patients' medical records. Symptoms and PSI scores were recorded. White blood cell count (WBC) was determined by routine blood tests. Blood samples were collected within 24 hours after admission and serum levels of CRP, cortisol, and HMGB-1 were determined by enzyme-linked immunosorbent assay using commercial kits (MSKBIO, Wuhan, China) according to the manufacturer's instructions. In terms of survival, all-cause death during hospitalization was considered and recorded, with a follow-up time of 30 days from the time of admission. Survival time was considered as the time from admission to the time of death or last follow-up.

Statistical analysis

Measured data were expressed by mean $\pm$ standard deviation when normally distributed, and median (range) in other instances. Rates were compared using χ^2 tests, and comparisons between two groups of continuous data were made using Student's t-test or Mann–Whitney U test. Correlations were determined by Spearman's analysis and survival analysis was performed using Kaplan–Meier curves. Relationships between serum levels of HMGB-1 and cortisol and 30-day mortality were analyzed using a logistic regression model by a stepwise method. Values of $P < 0.05$ were considered to be statistically significant. All calculations were made using IBM SPSS Statistics for Windows, Version 22.0 (IBM Corp., Armonk, NY, USA).

Results

HMGB-1 and cortisol were up-regulated in patients with severe pneumonia

Details of the 35 patients with CAP are shown in Table 1. Nineteen patients were diagnosed with mild/moderate CAP and 16 with severe CAP. There was no significant difference between the groups in terms of age, sex, antibiotic situation, or symptoms. Laboratory evaluation showed that WBC and serum levels of CRP, cortisol, and HMGB-1 were all significantly higher in patients with severe CAP compared with patients with mild/moderate CAP ($P < 0.05$). The 30-day mortality rate was also significantly higher in patients with severe compared with mild/moderate CAP ($P < 0.05$).

Relationships of HMGB-1 and cortisol with clinical outcome in patients with CAP

Serum levels of HMGB-1 were significantly correlated with serum levels of CRP and cortisol in all patients ($P < 0.05$) (Figure 1). We further investigated the relationships of HMGB-1 and cortisol with clinical outcomes of CAP by stratifying patients into high/low HMGB-1 and cortisol groups according to the median values. WBC and serum levels of CRP and cortisol were all significantly higher in the high-HMGB-1 group compared with the low-HMGB-1 group ($P < 0.05$) (Table 2). PSI stage and 30-day mortality were also significantly higher in the high-HMGB-1 group ($P < 0.05$). Similar results were found for the high- and low-cortisol groups (Table 3). These results showed that serum levels of both HMGB-1 and cortisol were associated with clinical outcomes in patients with CAP.

Table 1. Basic clinical information for all participants.

Variable	Mild/moderate CAP (n = 19)	Severe CAP (n = 16)
Age, years (mean±SD)	57.5±11.4	61.4±9.9
Sex, male:female	12:7	10:6
Antibiotics received before treatment, n (%)	10 (52.3)	8 (50)
Symptoms, n (%)		
Fever	9 (47.4)	9 (56.3)
Cough	7 (36.8)	8 (50.0)
Sputum	6 (31.6)	6 (37.5)
Shortness of breath	4 (21.1)	4 (25.0)
Chest pain	3 (15.8)	4 (25.0)
Laboratory tests		
CRP, mg/L	60 (23–178)	122.5 (84–229)*
WBC, 10^9/mL	9.6 (7.5–13.7)	12.5 (9.4–16.9)*
Cortisol, nmol/L	528 (295–764)	907 (638–1309)*
HMGB-1, ng/mL	57 (31–101)	102.5 (89–151)*
Mortality during 30 days follow-up, n (%)	4 (21.1)	7 (43.8)*

CAP: community-acquired pneumonia, SD: standard deviation, CRP: C-reactive protein, WBC: white blood cell count, HMGB-1: high mobility group box-1 protein. *$P < 0.05$, compared with mild/moderate group.
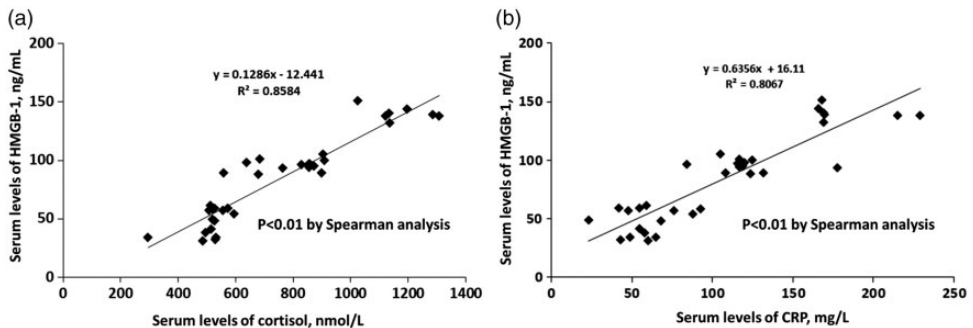

Figure 1. Correlations between serum levels of HMGB-1 and cortisol (a) and CRP (b). HMGB-1: high mobility group box-1 protein.

Relationships of HMGB-1 and cortisol with 30-day mortality in patients with CAP

We also analyzed the relationships of HMGB-1 and cortisol with 30-day mortality in patients with CAP. Patients with high serum levels of HMGB-1 or cortisol had higher 30-day mortality (lower survival) than patients with low serum levels of HMGB-1 or cortisol, respectively, according to Kaplan–Meier analysis ($P < 0.05$) (Figure 2). Further analysis showed that mean age and serum levels of CRP, cortisol, and HMGB-1 were all significantly higher in non-survivors compared with survivors

Table 2. Clinical outcomes in patients with CAP in relation to serum HMGB-1 levels.

Variable	Low HMGB-1 (n = 18)	High HMGB-1 (n = 17)
Age, years (mean±SD)	56.2±11.0	62.5±9.8
Sex, male:female	12:6	10:7
Antibiotics received before treatment, n (%)	9 (50.0)	9 (52.9)
Symptoms, n (%)		
Fever	8 (44.4)	10 (58.8)
Cough	7 (38.9)	8 (47.1)
Sputum	6 (33.3)	6 (35.3)
Shortness of breath	3 (16.7)	5 (29.4)
Chest pain	3 (16.7)	4 (23.5)
Laboratory		
CRP, mg/L	59.5 (23–132)	125 (84–229)*
WBC, 10^9/mL	9.3 (7.5–14.6)	12.0 (9.9–16.9)*
Cortisol, nmol/L	528 (295–899)	905 (684–1309)*
PSI, n (%)		
I–III	17 (94.4)	2 (11.8)*
IV–V	1 (5.6)	15 (88.2)*
Mortality during 30 days follow-up, n (%)	2 (11.1)	9 (52.9)*

CAP: community-acquired pneumonia, SD: standard deviation, WBC: white blood cell count, CRP: C-reactive protein, HMGB-1: high mobility group box-1 protein, PSI: pneumonia severity index. *$P < 0.05$, compared with the low-HMGB-1 group.

Table 3. Clinical outcomes in patients with CAP in relation to serum cortisol levels.

Variable	Low cortisol (n = 18)	High cortisol (n = 17)
Age, years (mean±SD)	56.1±11.0	62.7±9.7
Sex, male:female	11:7	11:6
Antibiotics received before treatment, n (%)	9 (50.0)	9 (52.9)
Symptoms, n (%)		
Fever	8 (44.4)	10 (58.8)
Cough	8 (47.1)	8 (38.9)
Sputum	6 (33.3)	6 (35.3)
Shortness of breath	4 (22.2)	4 (23.5)
Chest pain	3 (16.7)	4 (23.5)
Laboratory		
CRP, mg/L	59.5 (23–132)	125 (84–229)*
WBC, 10^9/mL	9.3 (7.5–14.6)	12.0 (9.9–16.9)*
HMGB-1, ng/mL	55.5 (31–98)	101 (89–151)*
PSI, n (%)		
I–III	17 (94.4)	2 (11.8)*
IV–V	1 (5.6)	15 (88.2)*
Mortality during 30 days follow-up, n (%)	2 (11.1)	9 (52.9)*

CAP: community-acquired pneumonia, SD: standard deviation, WBC: white blood cell count, CRP: C-reactive protein, HMGB-1: high mobility group box-1 protein, PSI: pneumonia severity index. *$P < 0.05$, compared with the low-cortisol group.
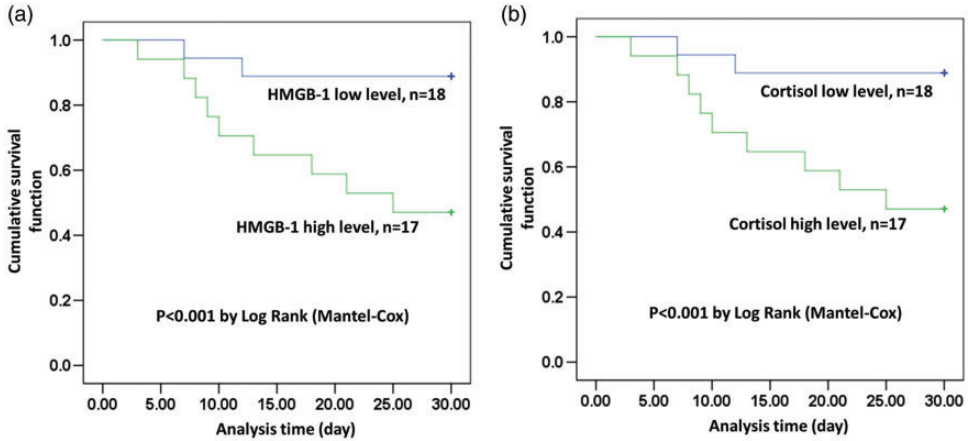
Figure 2. Kaplan–Meier survival analyses of patients with high/low HMGB-1 (a) and high/low cortisol (b). HMGB-1: high mobility group box-1 protein.

Table 4. Clinical outcomes in surviving and non-surviving patients with CAP.

Variable	Surviving (n = 24)	Non-surviving (n = 11)
Age, years (mean±SD)	54.1±8.7	70.6±3.6*
Sex, male:female	15:9	7:4
Antibiotics received before treatment, n (%)	13 (54.2)	5 (45.5)
Symptoms, n (%)		
Fever	12 (50.0)	6 (54.5)
Cough	10 (41.7)	5 (41.7)
Sputum	8 (33.3)	4 (36.4)
Shortness of breath	5 (20.8)	3 (27.3)
Chest pain	5 (20.8)	2 (18.2)
Laboratory		
CRP, mg/L	72 (23–125)	169 (117–229)*
WBC, 10^9/mL	8.4 (7.5–11.6)	14.5 (11.2–16.9)*
Cortisol, nmol/L	545 (295–909)	1123 (558–1309)*
HMGB-1, ng/mL	58.5 (31–105)	138 (88–151)*
PSI, n (%)		
I–III	15 (62.5)	4 (36.4)*
IV–V	9 (37.5)	7 (63.6)*

CAP: community-acquired pneumonia, SD: standard deviation, WBC: white blood cell count, CRP: C-reactive protein, HMGB-1: high mobility group box-1 protein, PSI: pneumonia severity index.
*$P < 0.05$, compared with surviving group.

($P < 0.05$) (Table 4). Logistic analysis identified both cortisol and HMGB-1 as independent risk factors for 30-day mortality in patients with CAP (Table 5).

Discussion

More than 5.6 million cases of CAP are reported annually in the United States,

Table 5. Correlations of serum HMGB-1 and cortisol levels with 30-day mortality in patients with severe CAP according to multivariate logistic regression analysis.

	Wald	Odds ratio	95% CI	P value
Age	4.734	2.620	(1.100–6.238)	0.030
CRP	2.025	1.280	(0.913–1.796)	0.152
Cortisol	9.029	1.007	(1.002–1.011)	0.003
HMGB-1	6.108	1.095	(1.019–1.177)	0.013
PSI	0.500	1.680	(0.399–7.075)	0.479

CAP: community-acquired pneumonia, CI: confidence interval, CRP: C-reactive protein, HMGB-1: high mobility group box-1 protein, PSI: pneumonia severity index.

accounting for more than 1 million hospitalizations, with hospital mortality rates of 5% to 48%.[20] Patients with severe CAP usually require admission to the intensive care unit; however, despite advances in supportive care, severe CAP is still associated with high mortality.[21] Serum levels of inflammatory factors have attracted much attention as risk factors for mortality among patients with CAP, and several inflammatory factors have been associated with mortality in these patients. Walters et al.[10] showed that failure to reduce CRP levels by $\geq 50\%$ at ≥ 4 days increased the probability of death in patients with CAP, and Guertler et al.[22] found that procalcitonin was also associated with long-term mortality risk among patients with CAP. In addition to these widely studied factors, HMGB-1 is a newly identified factor associated with the inflammation process, though more clinical evidence is needed to clarify the relationship between HMGB-1 and CAP prognosis. In the present study, we provide the first evidence for an association between HMGB-1 and clinical outcomes in patients with CAP, and as an independent risk factor for 30-day mortality in these patients.

The current results showed that serum levels of HMGB-1 were up-regulated in patients with severe CAP and were associated with clinical outcomes. Previous studies have demonstrated a role for HMGB-1 in pneumonia and other inflammatory processes. Wang et al.[23] showed that HMGB-1 was elevated in patients with untreated compared with treated CAP and was associated with PSI stage, while Achouiti et al.[24] demonstrated that HMGB-1 was associated with lung injury during *Staphylococcus aureus* pneumonia. Furthermore, Nosaka et al.[25] found that anti-HMGB-1 monoclonal antibody could protect against influenza A virus (H1N1)-induced pneumonia in mice.

We also showed that serum levels of HMGB-1 were positively correlated with serum levels of cortisol, and that both HMGB-1 and cortisol were associated with 30-day mortality among patients with CAP. Previous studies have revealed a role for cortisol in CAP. In a prospective study, Kolditz et al.[26] found that serum cortisol levels predicted death and critical disease independently of CRB-65 score in patients with CAP, and Omelyanenko et al.[27] demonstrated that cortisol could also be used as a potent prognostic biomarker in patients with severe CAP. In the present study, we demonstrated that serum levels of HMGB-1 were positively correlated with serum levels of cortisol; however, further insights are still needed. We also identified HMGB-1 as an independent risk factor for 30-day mortality in patients with CAP.

The present study had some limitations. First, it was a single-center study with a relatively small study population. Second,

further studies are needed to clarify the mechanisms by which HMGB-1 influences CAP and its relationship with cortisol.

In conclusion, we investigated the relationship between serum HMGB-1 levels and prognosis in patients with CAP. HMGB-1 was associated with clinical outcomes and was an independent risk factor for 30-day mortality, and serum levels of HMGB-1 were also positively correlated with serum levels of cortisol in patients with CAP. These results provide clinical evidence for the role of HMGB-1 in CAP, and may thus help to identify novel research targets for CAP treatment.

Declaration of conflicting interest

The authors declare that there is no conflict of interest.

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