

Original Article

Study on the flora distribution and early diagnostic value of infection biomarkers in patients with multidrug-resistant infection in intensive care unit

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Abstract: Objective: To explore the flora distribution and early diagnostic value of related infection biomarkers in patients with multidrug-resistant organism (MDRO) infection in the intensive care unit (ICU) from 2017 to 2019. Methods: From 2017 to 2019, 312 patients with infection in the ICU of The People's Hospital of Longhua in Shenzhen were studied retrospectively, including 148 patients with MDRO infection (MDRO infection group) and 164 patients with non-MDRO infection (non-MDRO infection group). Related markers of bacterial culture were submitted and tested, and serum levels of procalcitonin (PCT), C-reactive protein (CRP) and serum amyloid A (SAA) were measured in patients with MDRO infection. Results: The levels of PCT, CRP and SAA in the MDRO infection group were significantly higher than those in the non-MDRO infection group (all $P < 0.05$). In the diagnosis of MDRO infection by PCT, the area under receiver operating characteristic (ROC) curve was 0.792. When the cutoff value of PCT was 0.765 $\mu\text{g/L}$, the Youden index, specificity and sensitivity were 0.606, 0.957 and 0.649, respectively. In the diagnosis of MDRO infection by CRP, the area under ROC curve was 0.811. When the cutoff value of CRP was 32 mg/L, the Youden index, specificity and sensitivity were 0.574, 0.756 and 0.818, respectively. In the diagnosis of MDRO infection by SAA, the area under ROC curve was 0.755. When the cutoff value of SAA was 119.6 mg/L, the Youden index, specificity and sensitivity were 0.436, 0.970 and 0.466, respectively. In the combined diagnosis of MDRO infection by PCT, CRP and SAA, the area under the ROC curve was 0.842. There were differences in flora distribution of MDRO infection among different years ($P < 0.05$). The infection sites of patients were mainly in the lung, blood and surgical incision, and the distribution of MDRO infection in various sites was different ($P < 0.05$). Conclusion: The combination of CRP, PCT and SAA is valuable in the early diagnosis of MDRO infection. The majority of MDRO infections in ICU are caused by carbapenem-resistant *Acinetobacter baumannii* (CRAB) and carbapenem-resistant *Escherichia coli* (CRECO), and MDRO stains are different in different sites of infection.

Keywords: MDRO, PCT, CRP, SAA, ICU, flora distribution, early diagnosis

Introduction

The Intensive care unit (ICU) is an important place for patients who require critical care. Patients in the ICU tend to be complicated with underlying diseases and they often receive invasive procedures and related immunosuppressive treatment in their care, which has caused a higher probability of multidrug-resistant organism (MDRO) infection in the ICU than in other departments [1, 2]. Coupled with antibiotic abuse in clinic, the numbers of patients with MDRO infection in the ICU showed an

increasing trend, which seriously affects the prognosis and life quality of patients [3]. MDRO infection can cause rapid disease progression and significantly increased fatality in patients [4]. Moreover, poor efficacy of antibiotic therapy for MDRO infection increases the difficulties of clinical treatment and the economic burden for patients [5]. Research has shown that there were differences in the detection of MDRO infection in the ICU among different body areas [6, 7]. Therefore, it is instructive to understand the distribution, evolution and drug resistance of MDRO infection in the ICU for the develop-

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ment of better anti-infection treatment plans for patients. Infection biomarkers are important references for reflecting the status and severity of infection and also in guiding medication. According to this, our study explored the flora distribution and early diagnostic value of related infection biomarkers for MDRO infections the in ICU.

Materials and methods

Clinical data

A retrospective study was conducted in 312 patients with infection admitted to the ICU of The People's Hospital of Longhua from January 2017 to December 2019, including 148 patients with MDRO infection and 164 patients with non-MDRO infection. All patients were aged 18-75 years, and an average age of 68.2 ± 8.1 years. This study was approved by Ethics Committee of The People's Hospital of Longhua.

Inclusion and exclusion criteria

Inclusion criteria: Patients who met the standards for management of patients with infection in the ICU in the "Guidelines of intensive care unit design and management of China" in 2006 [8]; patients who met the diagnostic criteria of infection [8]; patients who age was between 18-75 years old; patients whose blood, urine or secretion cultures were positive.

Exclusion criteria: Patients with immune deficiency; patients with malignant tumor; patients with multiple bacterial infections; patients who had received antibiotics within 48 h.

Methods

Bacterial culture: The blood, urine and body secretions (sputum, pus, pleural or peritoneal effusion) of patients were collected and submitted for examination after admission by a WalkAway-40 automated microbial identification analyzer (Siemens, Japan), and the standards for specimen collection referred to the National Clinical Laboratory Procedures [9]. The diagnostic criteria for MDRO were in line with "Multidrug-resistant, extensively drug-resistant and pandrug-resistant bacteria: An international expert proposal for interim standard definitions for acquired resistance" [10].

Markers of blood infection: Two tubes containing 5 mL of venous blood from each patient were drawn from the cubital vein after they were admitted to hospital. The blood samples were collected in ethylenediaminetetraacetic acid (EDTA) sterile tubes. After the samples were stored in the refrigerator at 4°C for 15 min, the serum and plasma were separated using a centrifuge with centrifugation force of 1106.8 (xg). Then the samples were stored in the freezer at -80°C. The white blood cell count was measured by a Coulter LH750 automatic hematology analyzer (Beckman Coulter, USA). The serum levels of procalcitonin (PCT), C-reactive protein (CRP), and serum amyloid A (SAA) were measured with enzyme-linked immunofluorescence assay (Elisa) by automatic immune analyzer (Siemens, Germany).

Statistical analysis

The data were analyzed by SPSS 17.0. Continuous variables were expressed as mean \pm standard deviation ($\bar{x} \pm sd$), and independent sample t-tests were used to analyze the data. The enumeration data are expressed as percentage (%), and Pearson chi-square tests were used to analyze the data. The ROC curves were plotted, and the area under ROC curve (AUROC) and 95% confidence intervals (CI) were calculated. $P < 0.05$ was considered statistically significant.

Results

Comparison of general data of patients in both groups

There was no significant difference between the two groups in terms of sex, age and infection sites ($P < 0.05$). The length of hospital stay and fever duration in the MDRO infection group were significantly longer than those in the non-MDRO infection group ($P < 0.001$). See **Table 1**.

Comparison of related infection biomarkers

The serum levels of PCT, CRP and SAA in the MDRO group were all higher than those in the non-MDRO Group (all $P < 0.001$), and there was no significant difference in white blood cell (WBC) count between the two groups ($P < 0.05$). See **Table 2**.

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Table 1. Comparison of general data

Items	MDRO infection group (n=148)	Non-MDRO infection group (n=164)	χ^2/t	P
Sex (male:female)	86:62	96:68	0.006	0.939
Age (year)	67.9±9.2	68.1±8.4	0.201	0.841
Infection site			0.197	0.995
Pulmonary infection	109	118		
Blood infection	18	22		
Urinary tract infection	3	4		
Surgical incision infection	16	18		
Other infections	2	2		
Length of hospital stay (d)	16.7±6.8	10.2±9.2	7.034	<0.001
Fever duration (d)	10.6±6.4	7.2±4.6	5.426	<0.001

Table 2. Comparison of related infection biomarkers

Items	MDRO group (n=148)	Non-MDRO group (n=164)	t	P
PCT ($\mu\text{g/L}$)	0.97±0.60	0.41±0.22	11.281	<0.001
CRP (mg/L)	62.37±29.02	16.97±8.81	19.089	<0.001
WBC ($\times 10^9/\text{L}$)	9.82±3.98	9.52±4.78	0.599	0.550
SAA (mg/L)	113.36±63.69	60.70±34.88	9.175	<0.001

Note: PCT: Procalcitonin; CRP: C-reactive protein; WBC: White blood cell; SSA: Serum amyloid A; MDRO: multidrug-resistant organism.

Comparison of the distribution of MDRO strains among different infection sites

The infection sites of patients with MDRO infection in the ICU were mainly in the lung, blood and the surgical incision, and the distribution of MDRO infection at different infection sites was significantly different ($P<0.05$). See **Table 4**.

The diagnostic value of related infection biomarkers in MDRO infection

In the diagnosis of MDRO infection by PCT, the area under receiver operating characteristic (ROC) curve was 0.792. When the cutoff value of PCT was 0.765 $\mu\text{g/L}$, the Youden index, specificity and sensitivity were 0.606, 0.957 and 0.649, respectively. In the diagnosis of MDRO infection by CRP, the area under ROC curve was 0.811. When the cutoff value of CRP was 32 mg/L, the Youden index, specificity and sensitivity were 0.574, 0.756 and 0.818, respectively. In the diagnosis of MDRO infection by SAA, the area under ROC curve was 0.755. When the cutoff value of SAA was 119.6 mg/L, the Youden index, specificity and sensitivity were 0.436, 0.970 and 0.466, respectively. In the combined diagnosis of MDRO infection by PCT, CRP and SAA, the area under the ROC curve was 0.842. See **Table 3** and **Figure 1**.

Comparison of the distribution of MDRO infection strains among different years

The distribution of MDRO infection strains among different years was statistically different ($P<0.05$). See **Figure 2**.

Discussion

Bacterial culture is still the gold standard for the diagnosis of MDRO infection. However, previous studies have shown that the positive rate of bacterial culture was very low (about 15%) [11], which leads to a delay in the diagnosis and medication of MDRO infection. Moreover, patients with MDRO infection show greater resistance to antibiotics, so treatment of MDRO infection aimed at specific pathogens is needed to control the illness [12-15]. The delays in diagnosis and effective medication can cause adverse effects on the prognosis of patients. Therefore, it becomes a good clinical research direction to select relevant infection biomarkers for early diagnosis of MDRO infection.

White blood cell (WBC) count is the most commonly used indicator for infection in clinic. However, it is affected by many factors, fluctuates greatly, and increases in most patients with bacterial infection. Our study showed WBC count increased in both groups and there was no significant difference between the two groups, which suggests that it is not effective to identify MDRO infection by only WBC count. C-reactive protein (CRP) is a common indicator

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Table 3. Diagnostic value of related infection biomarkers in MDRO infection

Items	Youden Index	Cutoff value	Sensitivity	Specificity	Area under ROC curve (95% CI)
PCT (µg/L)	0.606	0.765	0.649	0.957	0.792 (0.737-0.847)
CRP (mg/L)	0.574	32	0.818	0.756	0.811 (0.761-0.861)
SAA (mg/L)	0.436	119.6	0.466	0.970	0.755 (0.700-0.811)

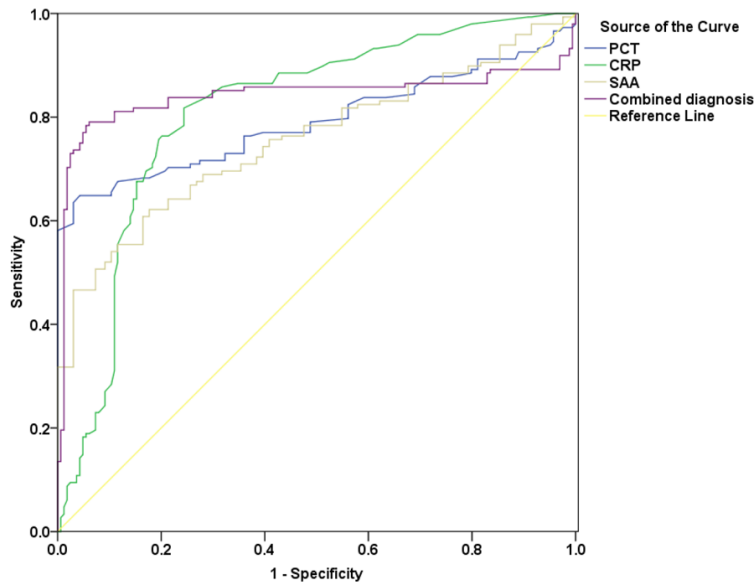


Figure 1. ROC curve of related infection biomarkers in MDRO infection. MDRO: multidrug-resistant organism.

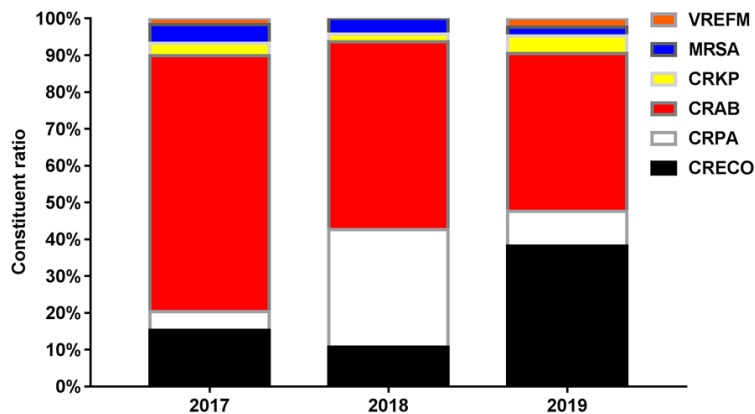


Figure 2. Comparison of distribution of MDRO infection strains among different years. CRECO: carbapenem-resistant *Escherichia coli*; CRPA: carbapenem-resistant *Pseudomonas aeruginosa*; CRAB: carbapenem-resistant *Acinetobacter baumannii*; CRKP: carbapenem-resistant *Klebsiella pneumoniae*; MRSA: methicillin-resistant *Staphylococcus aureus*; VREFM: vancomycin-resistant *Enterococcus faecium*.

in clinic and it is synthesized in the liver under the function of interleukin-6 and other inflammatory factors [16]. However, clinical studies have found that the level of CRP can increase in

the case of infection, oxidative stress and body injury [17, 18]. Therefore, its diagnosis specificity for infection is very poor; however, another study showed a positive correlation between CRP and the severity of infection [19]. In our study, CRP increased significantly in patients with MDRO infection, which may be related to the severe illness and poor treatment effect of MDRO infection.

Procalcitonin (PCT) is a specific indicator of bacterial infection [20], so the increase of PCT level indicates that there is bacterial infection. The results in our study showed that the increase of PCT level was more obvious after MDRO infection, which indicates MDRO infection is a severe bacterial infection. Serum amyloid A (SAA) is an indicator of acute bacterial infection [21], which increases in the early stage of MDRO infection, and it is also one of the indicators reflecting the severity of MDRO infection. Our study found that the level of SAA significantly increased in patients with MDRO infection, suggesting that MDRO infection is more serious than general infections. In our study, we combined CRP, PCT and SAA to diagnose MDRO infection in the early stage of the disease. The results showed that the combined AUC value for diagnosis was higher than that by using only one indicator. The combination of CRP, PCT and SAA is clinically valuable in the early diagnosis of MDRO infection.

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Table 4. Distribution of MDRO strains among the different infection sites

Items	Pulmonary infection	Blood Infection	Urinary tract infection	Surgical incision infection	Other infections	χ^2/F	P
CRECO	22	6	0	1	1	33.925	0.027
CRPA	19	2	0	1	0		
CRAB	63	7	2	10	1		
CRKP	2	2	1	0	0		
MRSA	2	1	0	3	0		
VREFM	1	0	0	1	0		

Note: CRECO: carbapenem-resistant *Escherichia coli*; CRAB: carbapenem-resistant *Acinetobacter baumannii*; CRAB: carbapenem-resistant *Acinetobacter baumannii*; CRKP: carbapenem-resistant *Klebsiella pneumoniae*; MRSA: methicillin-resistant *Staphylococcus aureus*; VREFM: vancomycin-resistant *Enterococcus faecium*; MDRO: multidrug-resistant organism.

MDRO infections increase due to antibiotic abuse [22, 23]. Patients with severe illness or those who receive major surgeries in ICU are often treated with antibiotics. Moreover, the space and the distance between beds in the ICU is small, and the resistance of ICU patients is worse than that of common patients. Therefore, MDRO strains are easily spread among patients in the ICU [24]. The results in our study showed that the majority of MDRO infections in the ICU were caused by carbapenem-resistant *Acinetobacter baumannii* (CRAB) and carbapenem-resistant *Escherichia coli* (CRECO), which is consistent with the results in previous studies [25]. CRAB is the most common MDRO strain in ICU patients, and CRAB exhibits high drug resistance due to its own characteristics [26]. In recent years, with the implementation of MDRO infection control, the incidence of CRAB infection showed a decreasing trend, although it is still high. However, the incidence of CRECO infection is on the rise, which may be related to irrational use of antibiotics and the introduction of community infection, indicating related monitoring and management of CRECO infection is needed to be strengthened. In our study, we also found there were differences in MDRO infection among the different infection sites, which may be associated with invasive procedures in ICU patients. Therefore, invasive procedures should be carried out according to specifications [27].

Limitations and prospects: This study is a retrospective study, and further prospective study is needed. Moreover, the sample size in our study is small; further, a multi-center study to expand the sample size should be conducted. Last but not least, this study did not implement related interventions for MDRO, and fur-

ther interventions can be implemented to study the prevention of MDRO infection.

In conclusion, the combination of CRP, PCT and SAA is valuable in the early diagnosis of MDRO infection. The majority of MDRO infections in the ICU are caused by CRAB and CRECO; and MDRO stains are different in different sites of infection.

Disclosure of conflict of interest

None.

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