Heliyon 9 (2023) e15724

Contents lists available at ScienceDirect

Heliyon

journal homepage: www.cell.com/heliyon

Research article

Antibiotic susceptibility pattern, risk factors, and prediction of carbapenem-resistant *Pseudomonas aeruginosa* in patients with nosocomial pneumonia

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ARTICLE INFO

Keywords:
Pseudomonas aeruginosa
Carbapenem-resistant
Nosocomial pneumonia
Risk factors
Predictive model

ABSTRACT

Objectives: This study was aimed at describing antibiotic susceptibility patterns and developing a predictive model by assessing risk factors for carbapenem-resistant *Pseudomonas aeruginosa* (CRPA).

Methods: A retrospective case-control study was conducted at a teaching hospital in China from May 2019 to July 2021. Patients were divided into the carbapenem-susceptible *P. aeruginosa* (CSPA) group and the CRPA group. Medical records were reviewed to find an antibiotic susceptibility pattern. Multivariate analysis results were used to identify risk factors and build a predictive model.

Results: A total of 61 among 292 patients with nosocomial pneumonia were infected with CRPA. In the CSPA and CRPA groups, amikacin was identified as the most effective antibiotic, with susceptibility of 89.7%. The CRPA group showed considerably higher rates of resistance to the tested antibiotics. Based on the results of mCIM and eCIM, 28 (45.9%) of 61 isolates might be carbapenemase producers. Independent risk factors related to CRPA nosocomial pneumonia were craniocerebral injury, pulmonary fungus infection, prior use of carbapenems, prior use of cefoperazone-sulbactam, and time at risk (≥15 d). In the predictive model, a score >1 point indicated the best predictive ability.

Conclusions: CRPA nosocomial pneumonia could be predicted by risk factor assessment particularly based on the underlying disease, antimicrobial exposure, and time at risk, which could help prevent nosocomial pneumonia.

1. Introduction

Nosocomial infection, also known as a hospital-acquired infection and intra-hospitalary infection, continues to be a serious concern in public health worldwide. The common microbes causing nosocomial infection include *Klebsiella pneumoniae*, *Acinetobacter baumannii*, and *Pseudomonas aeruginosa* [1,2]. *P. aeruginosa* is a common Gram-negative bacterium associated with nosocomial infection diseases such as pneumonia, wound infection, urinary tract infection, and invasive surgical infection [3]. *P. aeruginosa* has developed

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https://doi.org/10.1016/j.heliyon.2023.e15724
Received 28 April 2022; Received in revised form 18 April 2023; Accepted 19 April 2023
Available online 23 April 2023

resistance mechanisms after relentless exposure to carbapenems, given the increasing use of carbapenems in clinical treatments. Such occurrence has exacerbated the morbidity and mortality associated with carbapenem-resistant *P. aeruginosa* (CRPA) [4,5]. In 2016, the World Health Organization (WHO) categorized CRPA to be of critical priority in the list of pathogens that pose the highest threat to human health [6].

Nosocomial outbreaks of CRPA, which substantially burden the health system, have been reported in China [7–10]. Thus, studies of CRPA can contribute to the prevention and control of diseases associated with nosocomial infections. However, the risk factor, antimicrobial resistance pattern and the prediction of hospital-acquired pneumonia infected by CRPA have yet to be sufficiently illustrated. Here, we investigated the epidemiology, clinical presentations, and outcomes involved in CRPA nosocomial pneumonia in a teaching hospital in Sichuan, China. By analyzing collected data, we aimed to assess risk factors of CRPA nosocomial pneumonia during hospitalization through constructing a predictive model.

2. Methods

2.1. Study setting

In 2021, a retrospective case-control study was performed at the Affiliated Hospital of Southwest Medical University in Sichuan, China, a general teaching hospital and a regional medical center with 2200 beds covering patients from Yunnan, Guizhou, Sichuan, and Chongqing. A total of 21 national and provincial key disciplines and key construction disciplines were enrolled (Supplementary Table 1). This study covered nosocomial pneumonia related to *P. aeruginosa* infection in adult patients and classified the patients into the CRPA case group and the CSPA control group.

2.2. Patients and bacterial isolates

Non-repeat patients aged at least 18 y with *P. aeruginosa*-related nosocomial pneumonia were included in this study, whereas those with non-nosocomial pneumonia or nosocomial pneumonia caused by other bacteria were excluded. The inclusion and exclusion criteria for patients are shown in the flow chart (Fig. 1) [11,12].

Nosocomial pneumonia was defined as pneumonia occurring at least 48 h after admission to hospital, which is commonly classified into hospital-acquired pneumonia and ventilator-associated pneumonia [13]. The diagnosis of nosocomial pneumonia caused by *P. aeruginosa* was based on clinical manifestation, including a new or progressive and persistent infiltrate on chest radiographs and at least one systemic sign and two respiratory symptoms [14]. The clinical signs of the patients were confirmed based on sputum, alveolar lavage fluid, and other lower respiratory tract specimen with standard microbiological criteria such as: pleural fluid, flexible bronchoscopy with protected specimen brush, bronchoalveolar (BAL), transbronchial biopsy, nonbronchoscopic BAL, or tracheobronchial aspirate in intubated patients [12,15].

In the current study, 292 cases of *P. aeruginosa* infection were collected from May 2019 to July 2021. Sources of the *P. aeruginosa* isolates in our research included sputum, alveolar lavage fluid. The isolates were identified using the Microflex LT (Bruker Diagnostics Inc., United States) matrix-assisted laser-desorption/ionization time-of-flight (MALDI-TOF) mass spectrometry (MS) system.

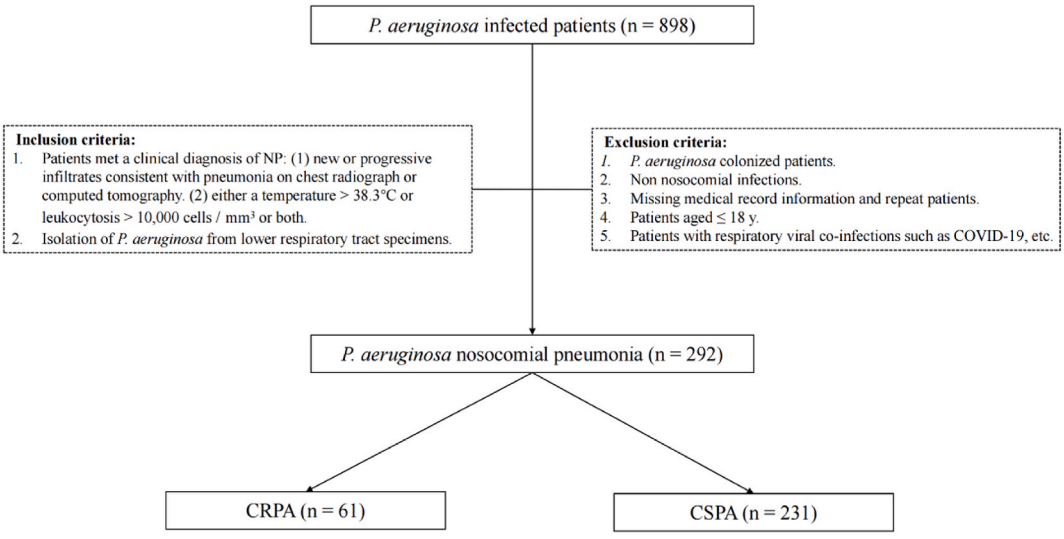

Fig. 1. Flow chart showing selection of the 292 non-duplicated *P. aeruginosa* nosocomial pneumonia. **NP:** nosocomial pneumonia; **CRPA:** carbapenem-resistant *P. aeruginosa*; **COVID-19:** coronavirus disease 2019; **CSPA:** carbapenem-susceptible *P. aeruginosa.*

2.3. Antimicrobial susceptibility testing

The minimum inhibitory concentrations (MICs) were measured using an automated system (MicroScan WalkAway 96 Plus). These agents included imipenem, meropenem, amikacin, ceftazidime, cefepime, ciprofloxacin, gentamycin, levofloxacin, ticarcillin-clavulanate, piperacillin-tazobactam, and cefoperazone-sulbactam. The results of imipenem and/or meropenem resistance (≥8 mg/mL) were confirmed by disk diffusion. The isolates with inhibitory zone diameters <19 mm were identified as CRPA. Methods and antibiotic breakpoints followed the Clinical and Laboratory Standards Institute (CLSI) guidelines, M100, 31st edition (published in 2021) [16].

2.4. Data collection

Data extracted from HIS (Hospital Information System) and LIS (Laboratory Information System) in the hospital included age, sex, underlying diseases, invasive procedure, days of hospitalization, microbiological and antibiotics sensitivity findings, history of bacterial and fungal infection, and use of antibiotics. Invasive procedures include the insertion of an indwelling gastric tube or an indwelling urethral catheter, peripheral inserted central venous catheter (PICC), tube drainage, intratracheal intubation, bronchoscopy, bronchoalveolar lavage and surgery. In the present study, time at risk was equated with the days of hospitalization before a positive culture of *P. aeruginosa*. History of bacterial and fungal pulmonary infection generally implied the detection of bacterial or fungus after a sputum culture and sputum smear before a positive culture of *P. aeruginosa*. Primary infection indicated *P. aeruginosa* was first isolated during hospitalization. The term "multidrug-resistant (MDR)" indicated that *P. aeruginosa* was non-susceptible to three or more classes of antibiotics (at least one from each class) [17].

2.5. Statistical analysis

All analyses were performed using SPSS (version 20.0; SPSS Inc., Chicago, IL, United States). To analyze the risk factors of CRPA in nosocomial pneumonia, quantitative variables were analyzed using the *t*-test; the $\chi 2$ test or Fisher's exact test was used to compare categorical variables; univariate logistic regression analysis was performed to calculate the odds ratios (OR) of all variables.

Independent risk factors could be determined by multivariate logistic regression. The most informative variables which meant factors with statistical significance in the univariate analysis were selected for inclusion in the multivariate model by using the logistic regression analysis, and the goodness-of-fit was checked using Hosmer–Lemeshow test. The β-coefficients of the significant variables in logistic regression analysis results were then used to assign values; the smallest coefficient was assigned a score of 1, and the others were given rounded scores [18]. The receiver operating characteristic (ROC) curve was used to assess the model and calculat the cutoffs. A *P*-value <0.05 was considered statistically significant. The final results were presented using the *P* values, OR, and 95% confidence intervals (95% CI).

2.6. mCIM/eCIM testing

The mCIM (modified carbapenem inactivation method) and eCIM (EDTA-modified carbapenem inactivation method) experiments were conducted with reference to the previous studies [19]. Prepare 2 tubes of TSB broth (2 mL), 1 tube containing EDTA (5 mmol/L). Take 10-μl loopful of *P. aeruginosa* colonies in 2 mL TSB broth, and then 10 μg MEM disk were immersed in the bacterial suspension, tubes were incubated for 4 h. The preparation of a 0.5 McFarland standard bacterial suspension of *Escherichia coli* ATCC25922, with

Table 1
Antibiotic susceptibility pattern of CSPA and CRPA group.

ATB	All		CSPA		CRPA		*P*[a]
	R	S	R	S	R	S	
AMK	30 (10.3)	262 (89.7)	12 (5.2)	219 (94.8)	18 (29.5)	43 (70.5)	<0.001
CAZ	68 (23.3)	224 (76.7)	41 (17.7)	190 (82.3)	27 (44.3)	34 (55.7)	<0.001
FEP	84 (28.8)	208 (71.2)	49 (21.2)	181 (78.4)	35 (57.4)	26 (42.6)	<0.001
CPF	58 (19.9)	234 (80.1)	32 (13.9)	197 (85.3)	26 (42.6)	35 (57.4)	<0.001
CN	83 (28.4)	209 (71.6)	48 (20.8)	182 (78.8)	35 (57.4)	26 (42.6)	<0.001
LVX	80 (27.4)	212 (72.6)	44 (19.0)	185 (80.1)	36 (59.0)	25 (41.0)	<0.001
PRL	53 (18.2)	239 (81.8)	37 (16.0)	194 (84.0)	19 (31.1)	42 (68.9)	0.008
TZP	50 (17.1)	242 (82.9)	29 (12.6)	202 (87.4)	21 (34.4)	40 (65.6)	<0.001
TCC	98 (37.0)	194 (66.4)	56 (24.2)	175 (75.8)	42 (68.9)	19 (31.1)	<0.001
CPS	53 (18.2)	239 (81.8)	22 (9.5)	209 (90.5)	31 (50.8)	30 (49.2)	<0.001
MEM	53 (18.2)	239 (81.8)	0	231 (100)	53 (86.9)	8 (13.1)	
IMP	60 (20.5)	232 (79.5)	0	231 (100)	60 (98.4)	1 (1.6)	

R: resistance; **S:** susceptible; **ATB:** antibiotic; **AMK:** amikacin; **CAZ:** ceftazidime; **FEP:** cefepime; **CPF:** ciprofloxacin; **CN:** gentamycin; **LVX:** levofloxacin; **PRL:** piperacillin; **TZP:** piperacillin-tazobactam; **TCC:** ticarcillin-clavulanate; **CPS:** cefoperazone-sulbactam; **MEM:** meropenem; **IMP**: imipenem.

Data are presented as n (%).

[a] Comparison between the CSPA group and the CRPA group.

sterile cotton swab evenly spread on MH medium. The disk was pasted onto the MH medium after incubation, and the diameter of the inhibition circle was measured after incubation.

The results for both the mCIM and the eCIM were interpreted as previously described [20]. PAO1 was used as a negative control strain.

Table 2
Characteristics cases caused by CSPA and CRPA in nosocomial pneumonia.

Variables	CSPA group (N = 231)	CRPA group (N = 61)	OR (95%CI)	*P*-value
Age (year)[a]	59.61 (60.0)	58.77 (57.0)	0.995 (0.974–1.017)	0.656
Male sex	160 (69.3)	47 (77.0)	1.490 (0.771–2.879)	0.234
ICU stay	88 (38.1)	32 (52.5)	1.698 (0.962−2.995)*	0.043
Underlying diseases				
Diabetes	31 (13.4)	15 (24.6)	2.104 (1.050−4.214)*	0.033
Upper respiratory disease	47 (20.3)	12 (19.7)	0.959 (0.472–1.946)	0.907
Craniocerebral injury	161 (69.7)	56 (91.8)	4.870 (1.870−12.678)***	<0.001
Cardiovascular disease	127 (55.0)	27 (44.3)	0.650 (0.369–1.147)	0.136
Renal disease	38 (16.5)	7 (11.5)	0.658 (0.278–1.557)	0.338
Liver disease	68 (29.4)	24 (39.3)	1.555 (0.865–2.795)	0.138
Hematological diseases	87 (37.7)	31 (50.8)	1.710 (0.969–3.019)	0.063
Hypoproteinemia	92 (39.8)	33 (54.1)	1.781 (1.009–3.143)	0.045
Electrolyte metabolic disturbance	43 (18.6)	14 (23.0)	0.073 (0.010–0.540)	0.447
Epilepsy	40 (17.3)	12 (19.7)	1.169 (0.571–2.396)	0.669
COPD	25 (10.8)	5 (8.2)	1.244 (0.531–2.914)	0.548
Urinary tract infection	7 (3.0)	5 (8.2)	2.857 (0.874–9.339)	0.148
Digestive tracthemorrhage	37 (16.0)	13 (21.3)	1.420 (0.701–2.878)	0.329
Respiratory failure	24 (10.4)	11 (18.0)	1.897 (0.872–4.129)	0.102
Fracture	65 (28.1)	13 (21.3)	0.692 (0.352–1.361)	0.284
Hospitalization (days)[a]	36.85 (27.0)	44.54 (40.0)	1.011 (1.001−1.022)*	0.034
Time at risk (days)[a]	15.98 (11.0)	24.13 (17.0)	1.028 (1.012−1.045)***	<0.001
Time at risk (≥15 days)	89 (38.5)	40 (65.6)	3.039 (1.683−5.487)***	<0.001
Any invasive procedure				
Indwelling gastric tube	145 (62.8)	52 (85.2)	3.427 (1.609−7.300)**	0.001
Indwelling urethral catheter	132 (57.1)	40 (65.6)	1.429 (0.793–2.574)	0.234
PICC	62 (26.8)	16 (26.2)	0.969 (0.511–1.839)	0.924
Tube drainage	57 (24.7)	23 (37.7)	1.848 (1.016−3.360)*	0.044
Intubation intratracheal	171 (74.0)	50 (82.0)	1.595 (0.779–3.263)	0.198
Bronchoscopy	11 (4.8)	6 (9.8)	2.182 (0.773–6.159)	0.231
Bronchoalveolar lavage	7 (3.0)	6 (9.8)	3.491 (1.128–10.802)	0.052
Surgery	102 (44.2)	26 (42.6)	0.939 (0.531–1.661)	0.83
Primary infection[b]	107 (46.3)	15 (24.6)	0.378 (0.200−0.715)**	0.002
Coinfection[c]				
Klebsiella pneumoniae	50 (21.6)	27 (44.3)	2.875 (1.587−5.209)***	<0.001
Acinetobacter baumannii	43 (18.6)	19 (31.1)	1.978 (1.048−3.733)*	0.035
Escherichia coli	23 (10.0)	6 (9.8)	0.978 (0.383–2.542)	0.978
Staphylococcus aureus	22 (9.5)	10 (16.4)	0.537 (0.239–1.204)	0.127
Pulmonary fungus infection	11 (4.8)	21 (34.4)	9.756 (4.350−21.881)***	<0.001
Prior antibiotic use				
Fluoroquinolones	31 (13.4)	11 (18.0)	1.419 (0.667–3.018)	0.361
Penicillins	30 (13.0)	7 (11.5)	0.869 (0.362–2.085)	0.752
Aminoglycosides	7 (3.0)	7 (11.5)	4.148 (1.396−12.325)*	0.013
Carbapenems[d]	23 (10.0)	26 (42.6)	6.718 (3.453−13.071)***	<0.001
Antipseudomonal cephalosporins	114 (46.5)	42 (68.9)	2.269 (1.245−4.134)**	0.002
Ceftazidime	42 (18.2)	16 (28.2)	1.600 (0.826–3.100)	0.161
Cefoperazone-sulbactam	84 (36.4)	32 (52.5)	1.931 (1.093−3.413)*	0.022
MDR	67 (29.0)	53 (71.0)	14.143 (6.597−30.318)***	<0.001
Clinical outcome				
In-hospital death	4 (1.7)	2 (1.6)	1.924 (0.344–10.758)	0.961

COPD: chronic obstructive pulmonary disease; **PICC:** peripherally inserted central catheter; **MDR:** multidrug resistance; **CSPA:** Carbapenem-susceptible *Pseudomonas aeruginosa*; **CRPA:** Carbapenem-resistant *Pseudomonas aeruginosa*; **OR:** odds ratio; **CI:** confidence interval.
*$P < 0.05$.
**$P < 0.01$.
***$P < 0.001$.
[a] Mean (interquartile range).
[b] *P. aeruginosa* was isolated during for the first time during hospitalization.
[c] Other lung bacteria were all isolated from respiratory tract specimens.
[d] Carbapenems include imipenem and meropenem.

3. Results

3.1. Study population

A total of 898 patients with *P. aeruginosa* from the Affiliated Hospital of Southwest Medical University were identified during the study period. However, only 292 (32.5%) adults suffered from nosocomial pneumonia, with 61 (20.9%) classified as CRPA and 231 as CSPA (79.1%).

The average age of the patients was 60.6 y, and most of them were male (70.9%); 32.2% of the patients came from the neurosurgery department, and 91.4% were exposed to an invasive procedure before they were diagnosed with *P. aeruginosa* infection. The top five underlying diseases were as follows: craniocerebral injury (74.3%), cardiovascular disease (52.7%), hypoproteinemia (42.8%), hematological diseases (40.4%), and liver diseases (31.5%). All positive specimens were collected from the respiratory tract and 97.4% from sputum samples.

3.2. Differences in antibiotic susceptibility pattern

The antibiotic susceptibility patterns of patients with CRPA and CSPA infection are presented in Table 1. Among 292 strains, amikacin was the most effective antimicrobial, exhibiting a susceptibility of 89.7% (262/292); while ticarcillin-clavulanate had a susceptibility of 66.4% (194/292). In the CRPA group, 86.9% (53/61) were resistant to meropenem, and 98.4% (1/61) were resistant to imipenem. Compared with the CSPA group, the CRPA group was more significantly resistant to amikacin ($P < 0.001$), ceftazidime ($P < 0.001$), cefepime ($P < 0.001$), ciprofloxacin ($P < 0.001$), gentamycin ($P < 0.001$), levofloxacin ($P < 0.001$), piperacillin ($P = 0.008$), piperacillin-tazobactam ($P < 0.001$), ticarcillin-clavulanate ($P < 0.001$), and cefoperazone-sulbactam ($P < 0.001$). Then we did mCIM and eCIM for detecting carbapenemases producing stain (Supplementary Fig. 1). Of the 61 CRPA strains, 9 (14.7%) *P. aeruginosa* were positive for mCIM only and 19 (31.1%) strains had positive results for both mCIM and eCIM (Supplementary Table 2).

3.3. Risk factors for CRPA infection in nosocomial pneumonia

The characteristics of the 292 patients and the results of univariate analysis for both the CSPA and CRPA groups are listed in Table 2. Several differences in clinical variables were found. Patients had a history of ICU stay ($P = 0.043$) and with underlying diseases, such as diabetes ($P = 0.033$), craniocerebral trauma ($P = 0.003$), and hypoproteinemia ($P = 0.045$) were found significantly more in the CRPA group than in the CSPA group. Similarly, the length of hospitalization in days ($P = 0.034$) and the time at risk ($P < 0.001$) were longer in the CRPA group than in the CSPA group. In addition, CRPA infection was significantly higher in patients with indwelling gastric tube ($P = 0.035$) or tube drainage ($P = 0.042$), combined with *K. pneumoniae* ($P < 0.001$) or *A. baumannii* ($P = 0.033$) or pulmonary fungus infection ($P < 0.001$) and MDR cases ($P < 0.001$). However, patients with primary infection of *P. aeruginosa* ($P = 0.002$) were sevenfold greater in the CSPA group than in the CRPA group. The multivariate analysis in Table 3 shows that craniocerebral injury (OR, 3.111; 95% CI, 1.081–8.950), pulmonary fungus infection (OR, 3.636; 95%CI 1.068–12.384), prior use of carbapenems (OR, 5.083; 95%CI 1.400–18.463), prior use of cefoperazone-sulbactam (OR, 16.276; 95%CI, 4.023–65.769), and time at risk (≥15 d) (OR, 7.200; 95%CI, 2.080–24.924) may be correlated with CRPA infection in nosocomial pneumonia patients. And our data profile showed that time at risk (≥15 d) and craniocerebral injury were the two greatest risk factors for nosocomial pneumonia infection in CRPA in all age groups stratified by age. And the association of several other risk factors with infection increased with increasing age as shown in Fig. 2.

3.4. Predictive model for CRPA nosocomial pneumonia

Results from the multivariate analysis were also used to establish the clinical prediction rule; 5 independent factors were assigned points referring to their regression coefficient (Table 4). The area under the curve (AUC) of the ROC curve was used to evaluate the diagnostic efficiency of the model (Fig. 3). The AUC was 0.777 (95% CI (0.708–0.847), indicating adequate ability to predict CRPA infection with the occurrence of *P. aeruginosa* in nosocomial pneumonia. Table 5 shows the sensitivity and specificity at different cut-off points, corresponding to different clinical situations. For example, patients with a history of cefoperazone-sulbactam use and time

Table 3
Multivariate analysis of risk factors for CRPA in patients with nosocomial pneumonia.

Risk Factor[a]	OR	(95% CI)	*P*-value
Time at risk (≥15 days)	7.200	2.080–24.924	0.002
Craniocerebral injury	3.111	1.081–8.950	0.035
Pulmonary fungus infection	3.636	1.068–12.384	0.039
Prior use of carbapenems[b]	5.083	1.400–18.463	0.013
Prior use of CPS	16.267	4.023–65.769	<0.001

CPS: cefoperazone-sulbactam; **OR:** odds ratio; **CI:** confidence interval.
[a] Selected variables with $P < 0.05$ in univariate analysis of Table 1 were include in a multivariate regression model.
[b] Carbapenems include imipenem and meropenem.

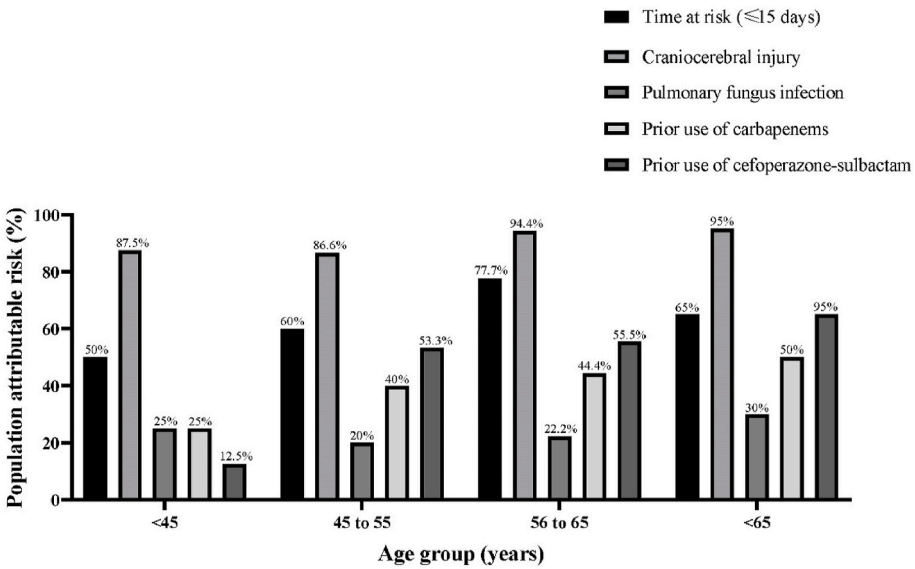

Fig. 2. Population-attributable risk percentages for incident CRPA nosocomial pneumonia. Models were associated with combinations of five independent risk factors: craniocerebral injury, pulmonary fungus infection, prior use of carbapenems, prior use of cefoperazone-sulbactam, and time at risk (≥15 d).

Table 4
Predictive factors for CRPA in patients with nosocomial pneumonia.

Factors	Points
Time at risk (≥15 days)	2
Craniocerebral injury	1
Pulmonary fungus infection	1
Prior use of carbapenems	1
Prior use of cefoperazone-sulbactam	2

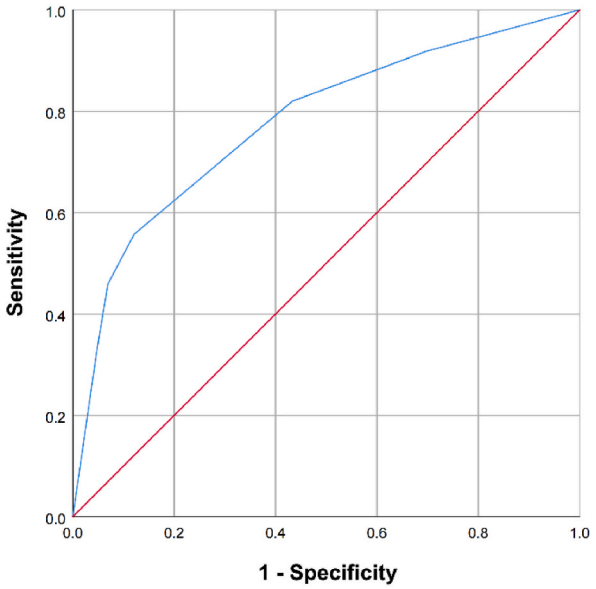

Fig. 3. ROC curve for predicting CRPA nosocomial pneumonia. ROC curve was used to assess the predictive ability of the model. Area under the curve = 0.777 (95% CI 0.708–0.847) ($P < 0.001$).

Table 5
Sensitivity and specificity of different operating cut off points.

	Sensitivity	Specificity	PLR	NLR	YI
>1[a]	55.70%	87.90%	4.60	2.04	0.44
>3	32.80%	95.20%	6.83	0.71	0.28
>5	82.00%	56.70%	1.89	0.32	0.39
>6	45.90%	93.10%	6.65	0.58	0.39

PRL: positive likelihood ratio; **NLR:** negative likelihood ratio; **YI:** Youden index.
[a] Cut-off point determined by Youden index.

of risk ≥15 days (points = 3, specificity = 95.20%) had better specificity for predicting CRPA nosocomial pneumonia than patients with pulmonary fungal infection and craniocerebral injury (points = 2, specificity = 87.90%).

4. Discussion

As the second most common hospital infection, nosocomial pneumonia primarily results from *P. aeruginosa* infection [21,22]. However, few studies have thus far been conducted to distinguish risk factors for CRPA in patients with nosocomial pneumonia. This retrospective case-control study described the clinical characteristics, risk factors, and antibiotic sensitivity pattern in nosocomial pneumonia patients with CRPA infection. A predictive model was then established based on independent risk factors.

As the most widely used semisynthetic aminoglycoside, amikacin is used to treat various infections caused by aerobic Gram-negative bacteria, such as *P. aeruginosa* [23]. It was the second effective antimicrobial agent against urinary tract infection caused by Gram-negative pathogens between 2010 and 2014 in China [24]. In the current study, antimicrobial susceptibility testing suggested that amikacin exerted the best antibacterial effect, consistent with other reports [25–28]. Our results showed more MDR *P. aeruginosa* in the CRPA group. This could also explain the significantly higher resistance rate of CRPA to the measured antimicrobials in our research which has been reported in some literature [29–31]. The higher number of MDR *P. aeruginosa* in CRPA is due to the higher use of carbapenems in MDR *P. aeruginosa* which results in being more susceptible to carbapenem resistance [32].

Some underlying diseases, invasive procedures, and prior use of antibiotics have been found to be associated with CRPA acquisition in healthcare-associated infection, which was consistent with our study [33]. Our study showed that three underlying diseases could increase the risk of CRPA infection: diabetes, hypoproteinemia, and craniocerebral injury. Recent research has suggested that the impaired immunity of diabetic patients, which principally leads to increasing susceptibility to infections and failure to completely eradicate persistent infections [34]. This occurrence can potentially result in greater exposure to antimicrobial agents such as carbapenems, thus increasing the prevalence of CRPA. Patients with hypoproteinemia also tend to be infected with CRPA because of reduced protein levels in the immune system [35]. The relatively long prognosis time of open craniocerebral injury may also increase susceptibility to CRPA infection [36]. Patients with CRPA infection underwent more invasive procedures in our study, compared with the CSPA group. This difference could be attributed to the tendency of invasive catheters to increase sensitivity to secondary infection with MDR pathogens, such as *P. aeruginosa*, *A. baumannii*, *Escherichia coli*, and *Enterococcus* spp [37]. We also identified previous use of meropenem as an independent factor in CRPA infection, as carbapenem use is generally observed as a factor strongly related to CRPA infection [38,39]. Previous use of antipseudomonal cefalosporins can be associated with carbapenem resistance [33], which was also observed in our study.

A significant difference in length of intensive care unit (ICU) stay was observed between the CSPA and CRPA groups. According to a three-year descriptive study in Peru, *P. aeruginosa* presented higher than 50% resistance to carbapenems, with an even higher level in ICUs, which might be related to the irrational and widespread use of carbapenems in ICUs [40]. In addition, patients once infected with *P. aeruginosa* were likely to be infected with CRPA probably because of longer antibiotic exposure. This finding could also explain the significant difference in length of stay between the CSPA and CRPA groups. MDR was also shown to be another underlying risk factor, based on previous studies revealing the presence of MDR phenotypes in CRPA [29,41–44].

Univariate analysis indicated that coinfection of *K. pneumonia* or *A. baumannii* were two potential risk factors associated with CRPA acquisition, similar to the finding by Di et al. [45]. However, these two risk factors have not been elaborately discussed in other studies. The multivariate results demonstrated that fungal infection was an independent risk factor in CRPA nosocomial pneumonia, although few studies have proved the correlativity between them. Our finding is supported by previous research. Both *P. aeruginosa* and *Candida albicans* are opportunistic pathogens and frequently co-isolated in polymicrobial infections [46,47]. In addition, a previous study has associated *Candida* "colonization" of the respiratory tract with *P. aeruginosa* pneumonia [48].

So far, diagnosis of *P. aeruginosa* in nosocomial pneumonia has been a challenge in the clinical setting [22]. We first developed a predictive model that exhibited good discriminatory power of CRPA nosocomial pneumonia. To facilitate the rapid identification of nosocomial pneumonia CRPA infections, we developed a quick scoring prediction rule based on a few straightforward clinical factors. Cut off point equaled 5 was the common scenario in CRPA group due to the highest sensitivity of 82.00% (n = 50), it included 5 situations: i) patients had concurrent craniocerebral injury, pulmonary fungus infection, prior use of carbapenems and time at risk (≥15 days); ii) patients had concurrent craniocerebral injury, pulmonary fungus infection, prior use of carbapenems and prior use of cefoperazone-sulbactam; iii) patients had time at risk (≥15 days), prior use of cefoperazone-sulbactam and craniocerebral injury; iv) patients had time at risk (≥15 days), prior use of cefoperazone-sulbactam and pulmonary fungus infection; v) patients had time at risk (≥15 days), prior use of cefoperazone-sulbactam and prior use of carbapenems. The model helped provide a chance to select CRPA

infection in nosocomial pneumonia patients in time or even ahead of time, improving clinical antibiotic therapy. Compared with other published models, our study included a wide and uniform age distribution and a balanced male to female ratio, so that the prediction model was somewhat accurate in area of Southern Sichuan. However, this was a single-center study, which led to model construction being dominated by the local area. Notably, some factors could not be controlled in advance: we could only control the antibiotic use of patients during hospitalization; the final diagnosis could be influenced by doctors subjectively; and the length of stay at other hospitals was not calculated. Although the scoring tool had been used in some studies [18,49], its practicability has rarely been reported.

In addition, some microbial factors might influence the occurrence of CRPA pneumonia by affecting drug resistance. Previous studies had revealed that twitching motility showed negative correlation with MDR/extensively drug-resistant (XDR) status, while hemolysin-production showed significant positive [50]; pyocyanin production was shown to be higher among MDR isolates in a Hungarian and Italian tertiary care hospital [51]; genotypes and virulence factor carriage were also correlated with carbapenem-resistant *P. aeruginosa* [52,53]. In addition, biofilm production could influence the XDR phenotype of *P. aeruginosa* by affecting the persistence and endemic spread of *P. aeruginosa* [53]. In conclusion, these microbiological parameters could affect the antibiotic resistance rate of *P. aeruginosa* and they could further influence the prediction model. All of the above microbiological parameters might be associated with nosocomial pneumonia CRPA infection. However, further experiments should be performed to demonstrate the role of these microbiological parameters in nosocomial pneumonia CRPA prediction model for different study groups and the different sources of *P. aeruginosa*.

In conclusion, this study identified five predictive factors contributing to CRPA nosocomial pneumonia: craniocerebral injury, pulmonary fungal infection, prior use of carbapenem, prior use of cefoperazone-sulbactam, and time at risk ($\geq$15 d). Our findings suggest that compared with CSPA, CRPA exhibits higher antibiotic resistance and thus should be paid more attention in the clinical setting. Further, CRPA can be prevented by reducing the unnecessary use of antibiotics.

Author contribution statement

Yao Liu: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Yifei Xu: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Wrote the paper.

Shu Wang: Performed the experiments.

Zhangrui Zeng; Yinhuan Ding; Zhaoyinqian Li; Jinbo Liu: Contributed reagents, materials, analysis tools or data.

Data availability statement

The authors do not have permission to share data.

Funding source

This work was supported by the Science and Technology Project of Science & Technology Department of Sichuan Province under Grant [No. 2021YFH001, No. 2022YFQ0093], Health Commission of Sichuan Province under Grant [No. 19PJ293] and Science and Technology Project of Southwest Medical University under Grant [No. 2022QN088].

Ethical approval statement

This study was approved by Ethics Committee of the Affiliated Hospital of Southwest Medical University (Luzhou, China) (approval number KY2022267).

Acknowledgments

We want to thank microbiology laboratories and other participating departments in the Affiliated Hospital of Southwest Medical University.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.heliyon.2023.e15724.

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