Chinese Medical Journal Pulmonary and Critical Care Medicine 1 (2023) 113–118

Contents lists available at ScienceDirect

Chinese Medical Journal Pulmonary and Critical Care Medicine

journal homepage: www.elsevier.com/locate/pccm

Original Article

Positive sputum culture of *Candida* spp. as a risk factor for 30-day mortality in patients with hospital-acquired pneumonia: A propensity-score matched retrospective clinical study

Yaopin Han[1,#], Yihui Zuo[2,#], Zhe Luo[3], Minjie Ju[3], Jianlan Hua[1], Binfeng He[1], Yixing Wu[1], Jing Zhang[1,*]

[1] *Department of Pulmonary and Critical Care Medicine, Zhongshan Hospital, Fudan University, Shanghai 200032, China*
[2] *Department of Clinical Medicine, Shanghai Medical College, Fudan University, Shanghai 200032, China*
[3] *Department of Critical Care Medicine, Zhongshan Hospital, Fudan University, Shanghai 200032, China*

ARTICLE INFO

Editor: Peifang Wei

Keywords:
Hospital-acquired pneumonia
Candida species
Prognosis
Mortality
Propensity score

ABSTRACT

Background: Candida species (*Candida* spp.) are commonly isolated microorganisms from lower respiratory tract (LRT) specimens of patients with hospital-acquired pneumonia (HAP); however, the clinical significance remains controversial. This study aimed to investigate the correlation between *Candida* spp. in the LRT and the clinical features and prognosis of HAP.

Methods: This retrospective analysis included eligible patients with HAP from the database of a prospective study carried out between 2018 and 2019 in nine Chinese hospitals. Data on demographics, clinical characteristics, and prognosis were collected and analyzed. Propensity score matching (PSM) was used to balance the baseline characteristics.

Results: A total of 187 HAP patients were enrolled. After PSM of severity score, 27 cases with positive sputum culture of *Candida* spp. were compared with the control group at a ratio of 1:1. The *Candida*-positive group had more bacterial isolates in blood culture than the *Candida*-negative group (39.1% [9/23] *vs.* 7.7% [2/26], $\chi^2 = 6.928$, effect size [ES] = 0.38, 95% CI: 0.12–0.61, $P = 0.008$). The proportion of patients with chronic lung diseases was significantly higher in the *Candida*-positive group (55.6% [15/27] *vs.* 22.2% [6/27], $\chi^2 = 6.312$, ES = 0.34, 95% CI: 0.07–0.59, $P = 0.012$). The 30-day prognosis of HAP was significantly different between the two groups (80.8% [21/26] *vs.* 38.5% [10/26], $\chi^2 = 9.665$, ES = 0.43, 95% CI: 0.19–0.66, $P = 0.002$). Univariable logistic regression analysis showed that LRT *Candida* spp. colonization was a risk factor for 30-day mortality of HAP (OR = 6.720, 95% CI: 1.915–23.577, $P = 0.003$).

Conclusions: *Candida* spp. in the LRT was associated with 30-day mortality of HAP. Patients with chronic underlying lung diseases tend to have *Candida* spp. colonization.

Introduction

Hospital-acquired pneumonia (HAP) is a type of pneumonia that occurs 48 or more hours after admission and is not latent at the time of admission.[1] As one of the most common hospital-acquired infections, HAP confers a great burden to the global healthcare system and greatly increases the mean length of hospital stay and the total cost of hospitalization, and even doubles the risk of death. The risk factors of HAP include older age, longer hospital stay, receiving mechanical ventilation, and intensive care unit (ICU) admission.[2–4] An epidemiological study of HAP in China revealed that *Acinetobacter baumannii* (*A. baumannii*), *Pseudomonas aeruginosa* (*P. aeruginosa*), *Staphylococcus aureus*, and *Klebsiella pneumoniae* (*K. pneumoniae*) were the top four pathogens, while other Gram-positive cocci and *Candida albicans* (*C. albicans*) also had high isolation rates.[5]

Candida species (*Candida* spp.) is the most common pathogen causing opportunistic mycosis all over the world,[6] while *C. albicans* is the most common *Candida* spp. causing bloodstream infection. Over the past two decades, the incidence rate of invasive candidiasis (IC) has been on the rise,[7] and immune dysfunction due to various causes, including central venous catheter and abdominal operation, was found to be the risk factor for IC.[8] Although the isolation of *Candida* spp. from the lower respiratory tract (LRT) is not rare, the significance of the isolation of *Candida* spp. remains controversial.[9] With the exploration of the pathogenic

* Corresponding author at: Department of Pulmonary and Critical Care Medicine, Zhongshan Hospital, Fudan University, Shanghai 200032, China.
E-mail address: jingatlas@outlook.com (J. Zhang)
# Yaopin Han and Yihui Zuo contributed equally to this work.

https://doi.org/10.1016/j.pccm.2023.04.005
Received 12 September 2021; Available online 7 June 2023

mechanism of *Candida* spp., more and more attention has been paid to the colonization of *Candida* spp. in the LRT, on which a number of studies have been conducted in recent years. These previous studies revealed the association between *Candida* spp. colonization in the LRT and worse clinical outcomes, including longer duration of mechanical ventilation, prolonged length of stay in the ICU or hospital, and higher hospital mortality.[10] Existing research has been focused on the role of LRT *Candida* spp. colonization in ventilator-associated pneumonia (VAP). However, whether HAP patients with *Candida* spp. colonization in the LRT have special clinical characteristics is a long-standing question. Thus, we believe it is necessary to conduct this study on the correlation between LRT *Candida* spp. colonization and HAP.

The purpose of this study was to explore the influence of LRT *Candida* spp. colonization on the prognosis of HAP and to identify the correlation between LRT *Candida* spp. isolation and clinical characteristics of HAP.

Methods

Study design and patient population

As a retrospective study, HAP patients were selected from the original database of a prospective study of our research group, which consecutively recruited patients with severe infections by attending physicians using a convenience sampling method from the respiratory department and the ICU of nine Chinese tertiary hospitals between December 2018 and December 2019. In the prospective cohort, patients diagnosed with blood infection, community-acquired pneumonia, HAP, peritonitis, acute suppurative cholangitis, acute pyelonephritis, or skin and soft tissue infections were considered to have severe infection when they met with any of the three conditions: (1) a respiratory rate ≥22 times/min, (2) a systolic blood pressure (SBP) ≤100 mmHg, or (3) an altered mental state. The most recent basic clinical data before considering HAP diagnosis and the survival outcome after a 30-day follow-up were collected (Fig. 1).[11]

Participants who met the following criteria were selected from the cohort for the current analysis: (1) Patients between 18 years and 90 years. (2) The diagnosis criteria of HAP were based on the American Thoracic Society (ATS)/Infectious Diseases Society of America (IDSA) guideline of HAP/VAP and the Chinese guideline of HAP/VAP (2018 edition),[1,12] and patients should meet three conditions: (a) Pneumonia should be acquired more than 48 hours after admission, (b) radiographic findings showed newly occurring or progressive pulmonary infiltrating lesions, and (c) patient should meet at least two of the following clinical features: a body temperature >38°C, a peripheral white blood cell count $>10\times10^9$/L or $<4\times10^9$/L, and purulent airway secretion. (3) The results of sputum culture at the time of diagnosis of HAP were available. Sputum samples were collected by deep and prolonged natural cough after gargle in the morning according to the National Guide to Clinical Laboratory Procedures (fourth edition),[13] and the qualified sputum samples were defined as a neutrophil count >25/low-power field, an epithelial cell count <10/low-power field, or the ratio of the two >2.5:1. (4) Follow-up data were available for survival at 30 days after diagnosis of HAP. Patients who had pulmonary infection, those in the latent period of infection, or those with invasive mechanical ventilation at the time of admission were excluded. Individuals unable to cooperate or with cognitive impairment were also not included in this study.

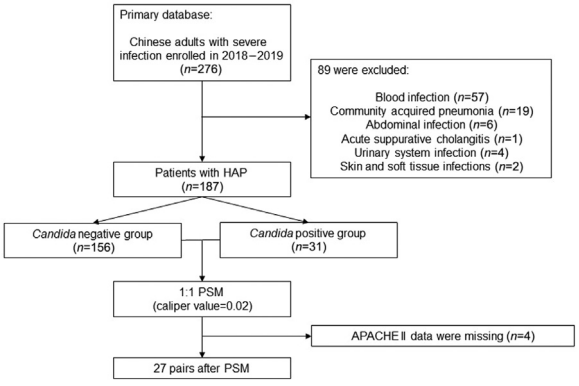

Fig. 1. Flowchart of included patients. APACHE II: Acute Physiology and Chronic Health Evaluation scoring system II; HAP: Hospital-acquired pneumonia; PSM: Propensity-score matching.

The main exposure of this study was LRT *Candida* spp. colonization. The isolation of *Candida spp.* from LRT specimen was determined as colonization if both of the following criteria were met when reviewing the medical records: (1) the pneumonia can be explained by other pathogens. (2) No antifungal therapy was administered before the enrollment and the collection of the blood and sputum specimen. The primary outcome was the 30-day mortality of HAP patients. Secondary outcomes were clinical characteristics of patients with LRT *Candida* spp. colonization.

Ethical approval

The prospective cohort study was approved by the Ethics Committee of Zhongshan Hospital Affiliated to Fudan University (Ethical Approval Number: B2018-182R). During the process of data collection and analysis, the privacy of personal information was guaranteed. Since the current study was a retrospective analysis of the aforementioned cohort, the informed consent was exempted.

Data collection

The clinical data were extracted from the original database by one researcher independently and then were checked by another researcher. If there were any differences, the original data would be reviewed for correction. The clinical data were composed of demographics (age, gender, height, and weight), symptoms and signs, past history, laboratory results on the days of inclusion and considering the diagnosis of HAP (blood routine, inflammatory markers, liver function, renal function, serum electrolytes, blood gas measurement, myocardial markers, and coagulation function), imaging results (chest X-ray and chest computed tomography [CT]), results of blood culture and blood next-generation sequencing (NGS), assessment of disease severity (shock index, Acute Physiology and Chronic Health Evaluation score II [APACHE II], and Quick Sequential Organ Failure Assessment [qSOFA]), clinical diagnosis, anti-*Candida* spp. treatment, and 30-day mortality. The final diagnosis of the causative pathogen was made by the attending physician based on all the clinical data of the patient, and the decision was confirmed by three experienced clinical experts of the study group if there was any doubt. According to the sputum culture results of *Candida* spp., all the patients were divided into the *Candida*-positive group and *Candida*-negative group.

Statistical analysis

SPSS 26.0 software (SPSS Inc., Chicago, IL, USA) was used in the whole analysis process, and GraphPad Prism 8.0.2 for Windows (GraphPad Software Inc., San Diego, CA, USA) was applied to produce figures. An absolute standardized difference (ASD) of >0.1 was considered to indicate the imbalance of baseline. Due to the imbalance of baseline data between the *Candida*-positive and -negative group, 1:1 propensity score matching (PSM) was performed with a caliper distance of 0.02 to identify a cohort of patients with similar baseline characteristics. The propensity score was estimated using a non-parsimonious multivariable logistic regression model,[14] with *Candida* spp. colonization as the independent variable and shock index, and APACHE II score and qSOFA score as covariates. The non-replacement nearest neighbor matching method without substitution was adopted. Two groups of patients with

Table 1
Clinical scores of the *Candida*-negative *and Candida*-positive group before and after matching.

Clinical scores	After matching			Before matching		
	Candida-negative group ($n=27$)	*Candida*-positive group ($n=27$)	Absolute standardized difference	*Candida*-negative group ($n=156$)	*Candida*-positive group ($n=31$)	Absolute standardized difference
Shock index	0.90 (0.74, 1.08)	0.86 (0.72, 1.17)	0.069	0.89 (0.72, 1.12)	0.86 (0.72, 1.17)	0.066
APACHE II	18.00 (16.00, 20.00)	20.00 (12.00, 25.00)	0.045	16.00 (10.00, 20.00)	20.00 (12.00, 25.00)	0.300
qSOFA			0.053			0.124
0	2 (7.4)	1 (3.7)		17 (10.9)	1 (3.2)	
1	9 (33.3)	11 (40.7)		43 (27.6)	12 (38.7)	
2	15 (55.6)	13 (48.1)		77 (49.4)	16 (51.6)	
3	1 (3.7)	2 (7.4)		19 (12.2)	2 (6.5)	

Data are presented as median (Q_1, Q_3) or *n* (%). APACHE II: Acute Physiology and Chronic Health Evaluation scoring system II; qSOFA: Quick Sequential Organ Failure Assessment; SBP: Systolic blood pressure; Shock index: Pulse rate/SBP (mmHg).

balanced baseline were finally obtained. Further analysis of the matched data was carried out.

For continuous variables, the Shapiro–Wilk test was used to evaluate the normality of data with a sample size <50, while the Kolmogorov–Smirnov test was used for data with a sample size >50. Continuous variables were described as mean and standard deviation if they were normally distributed, and the differences were evaluated by using the Student's *t*-test. For non-normally distributed continuous variables, they were described as median (Q_1, Q_3), and the differences were evaluated by using Mann–Whitney *U* test. Categorical variables were expressed as ratio (%) or percentage (%) and were compared by using the χ^2 test or Fisher's exact test.

Moreover, patients were regrouped according to whether they survived at 30 days. Univariable logistic regression analysis was performed for the risk factors related to prognosis. The significant variables ($P < 0.05$) were included in multivariable logistic regression analysis. Risk factors were removed when missing data exceeded 20%. Bilateral $P < 0.05$ was considered statistically significant.

Results

A total of 187 HAP patients, including 138 males and 49 females, were included in the basic cohort of this study, which then were divided into the *Candida*-negative group ($n=156$) and *Candida*-positive group ($n=31$). After analyzing the clinical data of these two groups, the imbalance of the baseline was found, mainly including neutrophil percentage, lymphocyte percentage, partial pressure of carbon dioxide ($PaCO_2$), fraction of inspired oxygen (FiO_2), blood urea nitrogen (BUN), and creatinine (Cr) [Supplementary Table 1]. Therefore, 1:1 PSM was carried out with the matching variables of the clinical comprehensive scores, namely, shock index, APACHE II score, and qSOFA score, which are commonly used in clinical practice to evaluate the condition of critically ill patients and cover most of the imbalanced factors mentioned above. After matching, the baseline severity scores mentioned earlier in the two groups were balanced (*ASD* < 0.1) [Table 1]. Finally, we obtained 27 HAP patients with positive sputum culture of *Candida* spp. and 27 patients with negative sputum culture, including 37 males and 17 females (Fig. 1). Other microorganisms in sputum culture mainly consisted of *K. pneumoniae* (positive group, seven cases *vs*. negative group, two cases), *A. baumannii* (positive group, three cases *vs*. negative group, three cases), and *P. aeruginosa* (positive group, one case *vs*. negative group, two cases).

Clinical presentations of HAP patients with positive sputum culture of Candida spp. after matching

No significant difference was found in most of the symptoms or signs between the *Candida*-positive and -negative groups after matching, including fever, chills, cough, expectoration, and chest pain [Supplementary Table 1], except for SBP (107.00 [101.00, 130.00] mmHg *vs*. 123.00 [118.00, 138.00] mmHg, $Z = -1.991$, effect size [ES] = −0.41, 95% CI: −0.96 to 0.14, $P = 0.046$) and pulmonary rales or consolidation signs (40.7% [11/27] *vs*. 14.8% [4/27], $\chi^2 = 4.523$, ES = 0.29, 95% CI: 0.03–0.53, $P = 0.033$). Part of the laboratory examination, however, did have statistically significant difference between the two groups. Red blood cells ($[2.69 \pm 0.60] \times 10^{12}$/L *vs*. $[3.18 \pm 0.67] \times 10^{12}$/L, $t = 2.671$, ES = −0.77, 95% CI: −1.37, −0.17, $P = 0.010$), hemoglobin (7.55 [7.20, 8.85] g/dL *vs*. 9.25 [7.80, 10.48] g/dL, $Z = -2.445$, ES = −0.63, 95% CI: −1.23, −0.04, $P = 0.014$), hematocrit (HCT) ([24.51 ± 4.68]% *vs*. [29.03 ± 5.42]%, $t = 2.900$, ES = −0.90, 95% CI: −1.55, −0.24, $P = 0.006$), and albumin ([31.19 ± 6.55] g/L *vs*. [35.21 ± 7.85] g/L, $t = 2.047$, ES = −0.56, 95% CI: −1.11 to 0, $P = 0.046$) were significantly lower in the *Candida*-positive group than those in the *Candida*-negative group (Table 2), indicating a worse general condition. As described before, pulmonary rales or consolidation signs were more common in the *Candida*-positive group, which indirectly reflected more obvious lung abnormalities in the *Candida*-positive group. However, PaO_2/FiO_2 (Table 2) and radiographic results, including chest X-ray and chest CT scans [Supplementary Table 1], showed no significant difference between the two groups. *Candida* spp.-positive patients had lower platelets ($[135.77 \pm 95.27] \times 10^9$/L *vs*. $[202.57 \pm 103.10] \times 10^9$/L, $t = 2.208$, ES = −0.67, 95% CI: −1.30, −0.04, $P = 0.033$), higher D-dimer (3.63 [2.53, 2473.00] μg/L *vs*. 1.95 [0.75, 4.86] μg/L, $Z = -2.558$, ES = 0.56, 95% CI: −0.16, 1.28, $P = 0.011$), and lower fibrinogen (9.50 [4.86, 272.00] mg/dL *vs*. 193.50 [139.25, 568.75] mg/dL, $Z = -2.682$, ES = −0.97, 95% CI: −1.69, −0.25, $P = 0.007$), reflecting the imbalance of the coagulation and anticoagulation in HAP patients with positive sputum culture of *Candida* spp. (Table 2).

Patients with chronic underlying lung disease may be more prone to LRT Candida spp. colonization

To find out what kind of patients tend to have LRT *Candida* spp. colonization, we analyzed the clinical information before the onset of HAP. Details are given in Supplementary Table 1. Age and body mass index (BMI) were very close between two groups. Both groups had a high rate of antibiotic application before hospital admission and invasive operation before onset and ICU admission. In addition, there was no significant difference in the time from admission to onset in HAP patients, regardless of LRT *Candida* spp. colonization ($P > 0.05$).

The analysis of the past history suggested that patients with chronic underlying lung disease were more likely to develop *Candida* spp. colonization. In the *Candida*-positive group, there were more cases of underlying lung diseases (55.6% [15/27] *vs*. 22.2% [6/27], $\chi^2 = 6.312$, ES = 0.34, 95% CI: 0.07–0.59, $P = 0.012$), mainly including chronic obstructive pulmonary disease (COPD). Cases of COPD in those patients with *Candida* spp. colonization were markedly more that those in patients without *Candida* spp. colonization (48.1% [13/27] *vs*. 11.1% [3/27], $\chi^2 = 8.882$, ES = 0.41, 95% CI: 0.17–0.64, $P = 0.003$) (Table 2).

Table 2
Clinical characteristics of HAP patients with or without positive sputum culture of *Candida* spp. after PSM.

Items	*Candida*-negative group ($n = 27$)	*Candida*-positive group ($n = 27$)	Effect size* (95% CI)	*P*-value
Gender (male/female)	18/9	19/8	0.04 (−0.23, 0.33)	0.770
Age (years)	56.04 ± 20.36	59.70 ± 14.48	0.21 (−0.34, 0.75)	0.449
SBP (mmHg)	123.00 (118.00, 138.00)	107.00 (101.00, 130.00)	−0.41 (−0.96, 0.14)	0.046
DBP (mmHg)	71.33 ± 12.26	68.22 ± 13.30	−0.24 (−0.79, 0.30)	0.376
Rales or consolidation	4 (14.8)	11 (40.7)	0.29 (0.03, 0.53)	0.033
Underlying lung disease	6 (22.2)	15 (55.6)	0.34 (0.07, 0.59)	0.012
COPD	3 (11.1)	13 (48.1)	0.41 (0.17, 0.64)	0.003
Asthma	3 (11.1)	1 (3.7)	−0.14 (−0.35, 0.15)	0.603
Chronic interstitial lung disease	0 (0)	0 (0)	-	-
Pulmonary embolism	0 (0)	1 (3.7)	0.14 (0.11, 0.28)	1.000
Structural lung disease	1 (3.7)	0 (0)	−0.14 (−0.27, −0.11)	1.000
Laboratory examination				
RBC ($\times 10^{12}$/L)	3.18 ± 0.67	2.69 ± 0.60	−0.77 (−1.37, −0.17)	0.010
Hb (g/dL)	9.25 (7.80, 10.48)	7.55 (7.20, 8.85)	−0.63 (−1.23, −0.04)	0.014
PLT ($\times 10^9$/L)	202.57 ± 103.10	135.77 ± 95.27	−0.67 (−1.30, −0.04)	0.033
HCT (%)	29.03 ± 5.42	24.51 ± 4.68	−0.90 (−1.55, −0.24)	0.006
CRP (mg/L)	18.83 (8.75, 117.18)	79.15 (26.61, 127.43)	0.48 (−0.11, 1.07)	0.116
PCT (ng/mL)	0.33 (0.16, 13.59)	3.05 (0.37, 5.87)	−0.29 (−0.84, 0.26)	0.315
pH	7.46 (7.40, 7.47)	7.43 (7.40, 7.47)	−0.31 (−0.96, 0.34)	0.396
PaO_2 (mmHg)	129.60 (89.70, 180.45)	87.30 (60.15, 124.00)	−0.60 (−1.26, 0.06)	0.032
P/F	254.27 (147.15, 380.62)	171.50 (107.25, 347.71)	−0.52 (−1.17, 0.14)	0.152
Albumin (g/L)	35.21 ± 7.85	31.19 ± 6.55	−0.56 (−1.11, 0)	0.046
D-dimer (mg/L)	1.95 (0.75, 4.86)	3.63 (2.53, 2473.00)	0.56 (−0.16, 1.28)	0.011
Fibrinogen (mg/dL)	193.50 (139.25, 568.75)	9.50 (4.86, 272.00)	−0.97 (−1.69, −0.25)	0.007
Positive blood culture result	2 (7.7)	9 (39.1)	0.38 (0.12, 0.61)	0.008

Data are presented as *n/n*, mean ± standard deviation, median (Q_1, Q_3), or *n* (%). *The effect size of continuous variables is represented by *Cohen's d*, while that of categorical variables is represented by phi (φ). COPD: Chronic obstructive pulmonary disease; CRP: C-creative protein; DBP: Diastolic blood pressure; FiO_2: Fraction of inspiration oxygen; HAP: Hospital-acquired pneumonia; Hb: Hemoglobin; HCT: Hematocrit; $P/F = PaO_2/FiO_2$; P/F: Oxygenation index; PaO_2: Arterial partial pressure of oxygen; PCT: Procalcitonin; PLT: Platelet count; PSM: Propensity score matching; RBC: Red blood cell count; SBP: Systolic blood pressure.

The positive rate of blood culture was higher in the Candida spp.-positive group

The positive blood culture rate in the *Candida*-positive group was significantly higher than that in the *Candida*-negative group (39.1% [9/23] *vs*. 7.7% [2/26], $\chi^2 = 6.928$, ES = 0.38, 95% CI: 0.12–0.61, $P = 0.008$). The results of blood culture in the positive group were mainly composed of *A. baumannii* (two cases) and *Enterococcus faecium* (E. faecium)(one case), while the results in the negative group were mainly *K. pneumoniae* (two cases). Additionally, a higher positive rate of blood NGS was also found in the *Candida*-positive group (80.8% [21/26] *vs*. 55.6% [15/27], $t = 3.865$, ES = 0.27, 95% CI: 0–0.52, $P = 0.049$) (Table 2).

HAP patients with positive Candida spp. in sputum culture had higher 30-day mortality risk

The 30-day mortality was higher in the *Candida*-positive group (80.8% [21/26] *vs*. 38.5% [10/26], $\chi^2 = 9.665$, ES = 0.43, 95% CI: 0.19–0.66, $P = 0.002$). To explore the factors influencing the 30-day prognosis of HAP patients, univariable logistic regression was performed for each clinical variable [Supplementary Table 2]. Positive *Candida* spp. in sputum culture (OR = 6.720, 95% CI: 1.915–23.577, $P = 0.003$), more affected lung lobes (OR = 2.464, 95% CI: 1.405–4.323, $P = 0.002$), higher C-reactive protein (OR = 1.015, 95% CI: 1.002–1.029, $P = 0.002$), higher APACHE II score (OR = 1.122, 95% CI: 1.029–1.224, $P = 0.009$), and positive blood NGS results (OR = 3.636, 95% CI: 1.054–12.546, $P = 0.041$) were associated with the 30-day mortality in HAP patients (Fig. 2). The factors with $P < 0.05$ were included in the multivariable logistic regression analysis; however, none of these factors was found to be the independent risk factor for HAP mortality [Supplementary Fig. 1].

Discussion

Our study retrospectively analyzed the data from a multicenter, randomized, controlled prospective cohort. As a result, the causal inference in this study had a certain degree of credibility. Additionally, the prospective cohort had uniform diagnostic criteria, test criteria, and evaluation criteria, making our data more accurate than the retrospectively collected data. It was found that positive *Candida* spp. in sputum culture was a risk factor for 30-day mortality in HAP patients in the univariable analysis, which increased the risk of death by 6.720 times. In addition, more affected lung lobes, elevated C-creative protein (CRP), and a high APACHE II score were also important risk factors for HAP mortality.

Available studies on the clinical significance of LRT *Candida* were mainly performed in patients with VAP. For example, a study by Delisle *et al.*[15] in patients with VAP also revealed an independent association between *Candida* spp. colonization in LRT and hospital mortality. Studies have also demonstrated that age, APACHE II score, length of hospital stay, and *Candida* spp. isolated from the LRT were independently associated with increased hospital mortality in VAP.[10,16]

It should be noted that the concept of VAP has been separated from HAP internationally, emphasizing that HAP, different from VAP, is not associated with mechanical ventilation.[1,17] *Candida* spp. infection has been confirmed to be associated with invasive procedures, involving biofilm formation on implanted devices.[18–20] Existing studies on pulmonary *Candida* spp. colonization focused on patients with VAP, while those without mechanical ventilation have been neglected, which made us more curious about HAP patients without VAP. Therefore, the main focus of this study was on HAP patients without mechanical ventilation.

The analysis of clinical features suggested the possible causes for the increased risk of death in HAP patients with LRT *Candida* spp. colonization. To our knowledge, this is one of the first studies to analyze the clinical features of HAP patients with positive sputum culture of *Candida* spp. in detail. Previous studies have demonstrated that patients with VAP who had LRT *Candida* spp. colonization were more likely to have higher inflammatory response. Williamson *et al.*[21] reported that respiratory *Candida* spp. colonization could significantly increase PCT, CRP, and interleukin-6 (IL-6) levels in patients with severe VAP. Additionally, De Pascale and Antonelli[9] proved an increase in serum tumor necrosis factor-α caused by pulmonary *Candida* spp. colonization. These results indicated that the activation of the immune system to induce in-

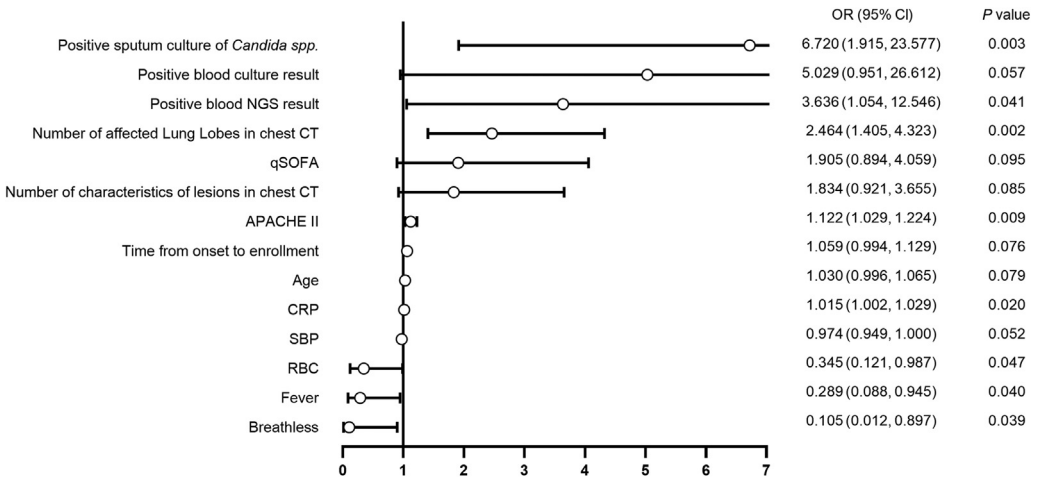

Fig. 2. Univariable logistic regression was performed for the risk factors related to prognosis. APACHE II: Acute Physiology and Chronic Health Evaluation scoring system II; CI: Confidence interval; CRP: C-creative protein (mg/L); CT: Computed tomography; NGS: Next-generation sequencing; qSOFA: Quick Sequential Organ Failure Assessment; OR: Odds ratio; RBC: Red blood cell count ($\times 10^{12}$/L); SBP: Systolic blood pressure (mmHg).

flammatory response might be the pathogenic mechanism of pulmonary *Candida* spp. Similarly, the values of CRP and PCT were numerically higher in the *Candida*-positive group in the present study, but no significant difference was found. This might be caused by the application of PSM on the clinical severity score; hence, the differences in these variates may be underestimated. It was known that *Candida* spp. in the mycelial state can damage epithelial cells and induce inflammation. The candidalysin secreted by *C. albicans* can cause invasive mucosal infection and tissue damage.[22] In our study, HAP patients with LRT *Candida* spp. colonization had more severe lung signs, reflecting lung damage caused by *Candida* spp. Moreover, other indicators of *Candida* spp. colonization in HAP patients were found in this study, including D-dimer and fibrinogen, which suggested that *Candida* spp. colonization in the LRT may increase the risk of death in HAP patients by impairing the coagulation and anticoagulation mechanism. Patients with LRT *Candida* spp. colonization had significantly lower serum albumin, possibly indicating the ability of albumin to reduce *Candida* spp. pathogenicity.[23]

We found that HAP patients with underlying lung diseases, mainly COPD, were more likely to have LRT *Candida* spp. colonization. A possible explanation is that *Candida* spp. is an opportunistic pathogen, which is most likely to attack the immunodeficient host.[8] In addition, *Candida* spp. is a common colonization bacterium in oral cavity and has been proved to be related to the occurrence of pneumonia.[18] In patients with COPD, airway epithelial cells are damaged; thus, the barrier against pathogens is weakened, creating opportunities for *Candida* spp. to colonize and cause disease.[24] COPD patients have mucosa destruction and ciliary dysfunction in the LRT, so it is more conducive for *Candida* spp. to colonize.

The positive rate of blood culture was higher in HAP patients with *Candida* spp. colonization in LRT, while the main pathogen of bloodstream infection was not *Candida* spp. The common causative pathogens in the *Candida*-positive group were *P. aeruginosa* (5/27), *A. baumannii* (3/27) and *E. faecium* (3/27), while those in the *Candida*-negative group were *Pneumocystis carinii* (3/27), *S. aureus* (2/27), and *K. pneumoniae* (2/27). These findings indicated that although the sputum culture of *Candida* spp. was positive, most HAP patients were not diagnosed with *Candida* spp. infection in clinical practice. It is generally accepted that quantitative bacterial culture of LRT can help confirm the diagnosis of bacterial pneumonia. However, such threshold of *Candida* spp. burden for the diagnosis of pneumonia has not been established; thus, *Candida* spp. pneumonia must be diagnosed by histopathology.[25] Despite the high rate of clinical isolation of *Candida* spp. from the respiratory tract, histopathologically confirmed *Candida* spp. pneumonia is rarely reported.[9] As a result, the clinical diagnosis of *Candida* spp. pneumonia is rare. Hence, it was suggested that despite the colonization of *Candida* spp. in the LRT, they are not highly invasive and cannot cause candidiasis. However, existing studies had demonstrated that there might be interaction between LRT *Candida* spp. and bacteria,[26] which was a potential risk factor for bacterial pneumonia,[27] and LRT *Candida* spp. was related to the increased risk of multi-drug resistant (MDR) bacterial isolation.[28] Moreover, antifungal treatment could reduce the risk of *P. aeruginosa* VAP.[29] This revealed the local pathogenic significance of *Candida* spp. in the LRT. Although some scholars believed that there is no need to eliminate LRT *Candida* spp.,[30] *Candida* spp. colonization, in this study, did cause adverse clinical outcomes, and worse clinical manifestations. In our view, therefore, for patients with a positive blood culture result, even though the culture result is not *Candida* spp., attention should be paid to the presence of LRT *Candida* spp. colonization.

Due to the imbalance of baseline in the basic cohort of this study, PSM was adopted. As the sample size of this study was small, the analysis results of individual variables may not be reliable. This may also be the reason why we failed to find the independent risk factors for HAP mortality. Therefore, further studies with a larger sample size are needed. Nevertheless, this study collected the results of as many common clinical examination items as possible to make the conclusion more meaningful in clinical application. In addition, there may be correlation between the risk factors obtained by univariable regression analysis. This may be the reason why we could not find independent risk factors through the multivariate regression analysis. For example, the possible interaction mentioned earlier between *Candida* spp. and bacteria suggested that LRT *Candida spp.* colonization may be associated with positive blood NGS results. Strictly designed prospective clinical trials are needed to further explore the relationship between these risk factors.

In conclusion, positive sputum culture of *Candida* spp. might be a risk factor for the 30-day mortality of HAP and was also related to worse clinical manifestations. The coagulation and anticoagulation disorders and elevated inflammatory markers in HAP patients might be associated with LRT *Candida* spp. colonization. There was a significant correlation between *Candida* spp. colonization and chronic underlying lung diseases, especially COPD. For these patients, more attention should be paid to prevent LRT *Candida* spp. colonization.

Conflicts of interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgments

We thank the nine Chinese hospitals for their contributions to this study, namely, the Zhongshan Hospital Affiliated to Fudan University, Second Xiangya Hospital of Central South University, First Affiliated Hospital of Zhejiang University, Tongji Hospital Affiliated to Huazhong University of Science and Technology in Wuhan, First Affiliated Hospital of Wenzhou Medical University, Second Affiliated Hospital of Soochow University, Central Hospital of Minhang District in Shanghai, Fifth People's Hospital of Shanghai, and Qingdao Municipal Hospital.

Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.pccm.2023.04.005.

References

1. Kalil AC, Metersky ML, Klompas M, et al. Management of adults with hospital-acquired and ventilator-associated pneumonia: 2016 Clinical Practice Guidelines by the Infectious Diseases Society of America and the American Thoracic Society. *Clin Infect Dis*. 2016;63:e61–e111. doi:10.1093/cid/ciw353.
2. Magill SS, Edwards JR, Bamberg W, et al. Multistate point-prevalence survey of health care-associated infections. *New Engl J Med*. 2014;370:1198–1208. doi:10.1056/NEJMoa1306801.
3. Lambert ML, Suetens C, Savey A, et al. Clinical outcomes of health-care-associated infections and antimicrobial resistance in patients admitted to European intensive-care units: a cohort study. *Lancet Infect Dis*. 2011;11:30–38. doi:10.1016/s1473-3099(10)70258-9.
4. Micek ST, Chew B, Hampton N, Kollef MH. A case-control study assessing the impact of nonventilated hospital-acquired pneumonia on patient outcomes. *Chest*. 2016;150:1008–1014. doi:10.1016/j.chest.2016.04.009.
5. Liu YN, Cao B, Wang H, et al. Adult hospital acquired pneumonia: A multicenter study on microbiology and clinical characteristics of patients from 9 Chinese cities (in Chinese). *Zhonghua Jie He He Hu Xi Za Zhi*. 2012;35:739–746. doi:10.3760/cma.j.issn.1001-0930.2012.10.009.
6. Pfaller MA, Diekema DJ. Epidemiology of invasive candidiasis: a persistent public health problem. *Clin Microbiol Rev*. 2007;20:133–163. doi:10.1128/CMR.00029-06.
7. Kullberg BJ, Arendrup MC. Invasive candidiasis. *New Engl J Med*. 2015;373:1445–1456. doi:10.1056/NEJMra1315399.
8. Leon C, Ostrosky-Zeichner L, Schuster M. What's new in the clinical and diagnostic management of invasive candidiasis in critically ill patients. *Intensive Care Med*. 2014;40:808–819. doi:10.1007/s00134-014-3281-0.
9. De Pascale G, Antonelli M. *Candida* colonization of respiratory tract: To treat or not to treat, will we ever get an answer? *Intensive Care Med*. 2014;40:1381–1384. doi:10.1007/s00134-014-3364-y.
10. Delisle MS, Williamson DR, Albert M, et al. Impact of *Candida* species on clinical outcomes in patients with suspected ventilator-associated pneumonia. *Can Respir J*. 2011;18:131–136. doi:10.1155/2011/827692.
11. Zuo YH, Wu YX, Hu WP, et al. The clinical impact of metagenomic next-generation sequencing (mNGS) test in hospitalized patients with suspected sepsis: a multicenter prospective study. *Diagnostics (Basel)*. 2023;13:323. doi:10.3390/diagnostics13020323.
12. Shi Y, Huang Y, Zhang TT, et al. Chinese guidelines for the diagnosis and treatment of hospital-acquired pneumonia and ventilator-associated pneumonia in adults (2018 Edition). *J Thorac Dis*. 2019;11:2581–2616. doi:10.21037/jtd.2019.06.09.
13. Shang H, Wang Y, Shen Z. *The national guide to clinical laboratory procedures (in Chinese)*. 4th ed. Beijing: People's Medical Publishing House; 2014:182–183.
14. Ahmed A, Husain A, Love TE, et al. Heart failure, chronic diuretic use, and increase in mortality and hospitalization: an observational study using propensity score methods. *Eur Heart J*. 2006;27:1431–1439. doi:10.1093/eurheartj/ehi890.
15. Delisle MS, Williamson DR, Perreault MM, Albert M, Jiang X, Heyland DK. The clinical significance of *Candida* colonization of respiratory tract secretions in critically ill patients. *J Crit Care*. 2008;23:11–17. doi:10.1016/j.jcrc.2008.01.005.
16. Huang Y, Jiao Y, Zhang J, et al. Microbial etiology and prognostic factors of ventilator-associated pneumonia: a multicenter retrospective study in Shanghai. *Clin Infect Dis*. 2018;67(Suppl 2):S146–S152. doi:10.1093/cid/ciy686.
17. Torres A, Niederman MS, Chastre J, et al. International ERS/ESICM/ESCMID/ALAT guidelines for the management of hospital-acquired pneumonia and ventilator-associated pneumonia: guidelines for the management of hospital-acquired pneumonia (HAP)/ventilator-associated pneumonia (VAP) of the European Respiratory Society (ERS), European Society of Intensive Care Medicine (ESICM), European Society of Clinical Microbiology and Infectious Diseases (ESCMID) and Asociacion Latinoamericana del Torax (ALAT). *Eur Respir J*. 2017;50:1700582. doi:10.1183/13993003.00582-2017.
18. Souza LCD, Mota V, Carvalho A, Correa R, Liberio SA, Lopes FF. Association between pathogens from tracheal aspirate and oral biofilm of patients on mechanical ventilation. *Braz Oral Res*. 2017;31:e38. doi:10.1590/1807-3107BOR-2017.vol31.0038.
19. Douglas LJ. *Candida* biofilms and their role in infection. *Trends Microbiol*. 2003;11:30–36. doi:10.1016/s0966-842x(02)00002-1.
20. Danin PE, Girou E, Legrand P, et al. Description and microbiology of endotracheal tube biofilm in mechanically ventilated subjects. *Respir Care*. 2015;60:21–29. doi:10.4187/respcare.02722.
21. Williamson DR, Albert M, Perreault MM, et al. The relationship between *Candida* species cultured from the respiratory tract and systemic inflammation in critically ill patients with ventilator-associated pneumonia. *Can J Anaesth*. 2011;58:275–284. doi:10.1007/s12630-010-9439-5.
22. Moyes DL, Wilson D, Richardson JP, et al. Candidalysin is a fungal peptide toxin critical for mucosal infection. *Nature*. 2016;532:64–68. doi:10.1038/nature17625.
23. Austermeier S, Pekmezović M, Porschitz P, et al. Albumin neutralizes hydrophobic toxins and modulates *Candida albicans* pathogenicity. *mBio*. 2022;12:e0053121. doi:10.1128/mBio.00531-21.
24. Guo-Parke H, Linden D, Weldon S, Kidney JC, Taggart CC. Mechanisms of virus-induced airway immunity dysfunction in the pathogenesis of COPD disease, progression, and exacerbation. *Front Immunol*. 2020;11:1205. doi:10.3389/fimmu.2020.01205.
25. Liu J, Yu YT, Xu CH, Chen DC. *Candida* colonization in the respiratory tract: what is the significance? *Front Med (Lausanne)*. 2020;7:598037. doi:10.3389/fmed.2020.598037.
26. Ricard JD, Roux D. *Candida* colonization in ventilated ICU patients: no longer a bystander!. *Intensive Care Med*. 2012;38:1243–1245. doi:10.1007/s00134-012-2587-z.
27. Tan X, Zhu S, Yan D, et al. *Candida spp*. airway colonization: a potential risk factor for *Acinetobacter baumannii* ventilator-associated pneumonia. *Med Mycol*. 2016;54:557–566. doi:10.1093/mmy/myw009.
28. Hamet M, Pavon A, Dalle F, et al. *Candida spp*. airway colonization could promote antibiotic-resistant bacteria selection in patients with suspected ventilator-associated pneumonia. *Intensive Care Med*. 2012;38:1272–1279. doi:10.1007/s00134-012-2584-2.
29. Nseir S, Jozefowicz E, Cavestri B, et al. Impact of antifungal treatment on *Candida-Pseudomonas* interaction: a preliminary retrospective case-control study. *Intensive Care Med*. 2007;33:137–142. doi:10.1007/s00134-006-0422-0.
30. Wood GC, Mueller EW, Croce MA, Boucher BA, Fabian TC. *Candida sp*. isolated from bronchoalveolar lavage: clinical significance in critically ill trauma patients. *Intensive Care Med*. 2006;32:599–603. doi:10.1007/s00134-005-0065-6.