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Original Article

Clinical significance of detecting pathogens in acute attack of bronchial asthma

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Abstract: Objective: To explore the distribution of pathogens in acute attack of bronchial asthma and their relationship with age and gender of patients, and to analyze the drug resistance. Methods: A total of 159 patients with acute attack of bronchial asthma were selected as the study subjects by retrospective study method. Sputum culture was performed and the detection of pathogens was counted. The relationship between distribution of pathogens and age, gender of patients was analyzed. The drug resistance was also performed. Results: Among 159 samples, ninety-four samples were found to be infected; the infection rate was 59.12%. One hundred and five strains of pathogens were detected, of which G^+ accounted for 22.86%, G^- accounted for 74.29%, fungi accounted for 2.86%. Of the 94 infected samples detected, the infection rate was significantly higher in patients > 60 years of age than in patients ≤ 60 years of age ($P < 0.05$), while there were no statistically significant differences in the proportion of G^+, G^- and fungal strains detected between patients ≤ 60 years of age and > 60 years of age ($P > 0.05$). There were no statistically significant differences in the infection rate and the proportion of G^+, G^- and fungal strains detected between male and female patients ($P > 0.05$). *Pseudomonas aeruginosa* had high resistance to ampicillin, amoxicillin and ceftriaxone, low resistance to gentamicin, cefoperazone/sulbactam, piperacillin, SMZ-TMP, imipenem and meropenem; *Escherichia coli* had high resistance to ampicillin, amoxicillin and gentamicin, low resistance to piperacillin, imipenem and meropenem; *Staphylococcus aureus* had high resistance to penicillin, ampicillin and erythromycin, while no resistance to gentamicin, vancomycin and rifampicin. Conclusion: Acute attack of bronchial asthma is closely related to pathogens infection, and patients > 60 years old have a higher risk of infection. For infection caused by different strains, sensitive antibacterial drugs should be selected according to *in vitro* drug sensitivity test in order to reduce the occurrence of drug resistance and improve the therapeutic effect.

Keywords: Bronchial asthma, acute attack, pathogen, distribution, drug resistance

Introduction

Bronchial asthma is a common chronic respiratory disease, mostly in children, with paroxysmal cough, dyspnea, orthopnea, chest tightness with wheezing as the main clinical manifestations, and even white foam, cyanosis, etc., in severe cases. The disease is easy to repeat and has a long course, not only affecting the growth and development of children, but also endangering the health of children [1]. The incidence of bronchial asthma in China has gradually increased because of the increasingly serious environmental pollution [2]. For patients with bronchial asthma, due to mental stimulation, environmental or climate change and respiratory tract infection and other factors, the clinical symptoms of bronchial asthma may suddenly appear as acute exacerbation, which must be clinically intervened [3]. Identifying whether acute attack of bronchial asthma is complicated by infection has important instructive significance for guiding clinical treatment and rational use of antibacterial drugs [4, 5]. This study focused on the distribution of pathogens in acute attack of bronchial asthma and their relationship with age and gender of patients. Drug resistance analysis was also performed.

Materials and methods

General data

A retrospective study method was conducted in 159 patients with acute exacerbation of bron-

chial asthma who were treated in Beijing Tongren Hospital, Capital Medical University from January 2019 to February 2020. All patients met the diagnostic criteria for acute attack of bronchial asthma in the *Guidelines for the Prevention and Treatment of Bronchial Asthma* [6]. The clinical data were derived by the electronic case system of Beijing Tongren Hospital, including 77 males and 82 females; aged 22 to 73 years old, with an average age of 47.3±5.4 years; body mass index (BMI) 22.19 to 25.04 kg/m^2, with an average of 23.54±1.02 kg/m^2; duration of bronchial asthma 2 to 11 years, with an average of 5.5±1.2 years. This study was approved by the medical Ethics Committee of Beijing Tongren Hospital, Capital Medical University and all patients signed the informed consent.

Inclusion criteria: Patients with clinical symptoms of bronchial asthma suddenly aggravated, and admitted to the hospital for treatment; the clinical data are complete.

Exclusion criteria: Patients with other respiratory diseases such as bronchial foreign bodies; patients with congenital heart disease; patients who had a history of treatment with glucocorticoids, leukotriene receptor blockers or β-agonists 2 months before enrollment.

Methods

Sputum collection and treatment: For patients with sputum, the oropharyngeal and nasopharyngeal secretions were cleared, using a disposable sterile sputum suction tube to suck the deep oropharyngeal sputum. The grayish white or colorless and transparent viscous part was removed, and the qualified sputum specimens for examination were retained. For patients without sputum, first allow them to inhale 3% hypertonic saline by atomization, then increase its concentration by 1% every 5-7 min, and maintain the concentration of 7% hypertonic saline at the highest, until about 3 mL of sputum specimens were collected, and the sputum specimens were sent for examination after the same treatment. The qualified sputum specimens were stained with gram. Sputum specimens with < 10 squamous epithelial cells and > 25 white blood cells/field were screened for sputum culture.

Isolation and identification of strains: Agarose or blood agar plate was used for culture, using French Merieux automatic bacterial identification and drug sensitivity analysis system (French bioMerieux Co., Ltd., model: VITEK 2 Compact, place of origin: France). The operating instructions for strain isolation and identification were strictly followed.

Drug sensitivity test: The French Merieux automatic bacterial identification and drug sensitivity analysis system was used for *in vitro* drug sensitivity test by Kirby-Bauer method. The drug sensitivity results were determined according to the drug sensitivity result judgment criteria established by the American Society for Clinical Laboratory Standards (CLSI; 2010 version) [7]. Susceptibility test strips and culture dishes used were purchased from Nanjing Urology Biotechnology Co., Ltd., China. The drug resistance of main G^- (*Pseudomonas aeruginosa* and *Escherichia coli*) and G^+ (*Staphylococcus aureus*) was observed. The main antibacterial drugs selected for the drug resistance of G^- were ampicillin, amoxicillin, gentamicin, ceftazidime, ceftriaxone, cefoperazone/sulbactam, piperacillin, ciprofloxacin, levofloxacin, trimethoprim-sulfamethoxazole (SMZ-TMP), imipenem and meropenem. The antibacterial drugs selected for the drug resistance of G^+ were mainly penicillin, ampicillin, gentamicin, erythromycin, klytomycin, vancomycin, cefoperazone/sulbactam, rifampicin, ciprofloxacin, levofloxacin and SMZ-TMP.

Outcome measures

Detection of pathogens in sputum and infection rate; distribution characteristics of pathogens in patients of different ages and genders; drug resistance of main pathogens.

Statistical analysis

SPSS 20.0 was used for data analysis, enumeration data were expressed as percentage (%), and the distribution of pathogens in patients ≤ 60 years old and > 60 years old and male and female patients was compared using the x^2 test. $P < 0.05$ was considered statistically significant.

Results

Detection and proportion of pathogens in sputum

Among 159 samples, 94 infected samples were detected, with an infection rate of 59.12%, including 5 cases of double infection and 3

Table 1. Detection and proportion of pathogens in sputum (n, %)

Pathogens	Strains (n)	Proportion (%)
G^+	24	22.86
Staphylococcus aureus	7	6.67
Streptococcus pneumoniae	5	4.76
Staphylococcus Haemolyticus	4	3.81
Staphylococcus Epidermidis	3	2.86
Staphylococcus capitis	1	0.95
Enterococcus faecalis	2	1.90
Other	2	1.90
G^-	78	74.29
Pseudomonas aeruginosa	15	14.29
Haemophilus influenza	11	10.48
Escherichia coli	13	12.38
Acinetobacter baumannii	12	11.43
Klebsiella pneumoniae	8	7.62
Enterobacter cloacae	9	8.57
Moraxella catarrhalis	6	5.71
Other	4	3.81
Fungi	3	2.86
Candida albicans	2	1.90
Candida krusei	1	0.95
Total	105	100.00

Table 2. Pathogens infection rate at different ages (n, %)

Age	Number of subjects (n)	Number of infections (n)	Infection rate (%)
≤ 60 years	70	28	40.00
> 60 years	89	66	74.16
t			5.046
P			0.025

cases of triple infection. A total of 105 strains of pathogens were detected, of which 24 strains of G^+ were detected, accounting for 22.86%; 78 strains of G^- were detected, accounting for 74.29%; and 3 strains of fungi were detected, accounting for 2.86%, as shown in **Table 1**.

Infection rate and distribution of pathogens in patients of different ages

Statistical analysis showed that among 94 infected samples detected, the infection rate was 74.16% in patients > 60 years old, significantly higher than the infection rate of 40.00% in patients ≤ 60 years old ($P < 0.05$), while there was no statistical difference in the detection rate of G^+, G^- and fungal strains between patients ≤ 60 years old and > 60 years old ($P > 0.05$). See **Tables 2** and **3**.

Distribution and proportion of pathogens in different genders

Statistical analysis showed that among 94 infected samples detected, there was no statistical difference in the infection rate between male and female patients ($P > 0.05$), and there was also no statistical difference in the detection rate of G^+, G^- and fungal strains between male and female patients ($P > 0.05$), as shown in **Tables 4** and **5** and **Figure 1**.

Analysis of drug resistance of major G^-

The results of *in vitro* drug sensitivity test showed that *Pseudomonas aeruginosa* had high resistance to ampicillin, amoxicillin and ceftriaxone, with resistance > 60%, low resistance to gentamicin, cefoperazone/sulbactam, piperacillin, SMZ-TMP, imipenem and meropenem, with resistance < 10%.

The resistance of *Escherichia coli* to ampicillin, amoxicillin and gentamicin was high, and the resistance was > 50%; the resistance to piperacillin, imipenem and meropenem was low, and the resistance was < 10%. See **Table 6**.

Analysis of drug resistance of main G^+

The results of *in vitro* drug sensitivity test showed that *Staphylococcus aureus* had high resistance to penicillin, ampicillin and erythromycin, with drug resistance > 50%; it showed no resistance to gentamicin, vancomycin and rifampicin. See **Table 7**.

Discussion

Several studies have shown that bacterial endotoxin can stimulate the tracheal mucosa and increase the permeability of tracheal mucosal epithelial cells. Lipopolysaccharide of bacteria, as an allergen, can also induce asthma by stimulating the body to produce an

Table 3. Distribution of pathogens at different ages (n, %)

Age	≤ 60 years	> 60 years	t	P
Pathogens (105 strains)	30	75		
G^+ (24 strains)			0.592	0.442
Strain	5	19		
Proportion	16.67	25.33		
G^- (78 strains)			0.104	0.747
Strain	24	54		
Proportion	80.00	72.00		
Fungi (3 strains)			0.032	0.857
Strain	1	2		
Proportion	3.33	2.67		

Table 4. Pathogens infection rate by gender (n, %)

Age	Number of subjects (n)	Number of infections (n)	Infection rate (%)
Male	77	44	57.14
Female	82	50	60.98
t			0.062
P			0.803

Table 5. Distribution of pathogens by gender (n, %)

Age	Male	Female	t	P
Pathogens (105 strains)	51	54		
G^+ (24 strains)			0.745	0.388
Strain	14	10		
Proportion	27.45	18.52		
G^- (78 strains)			0.105	0.746
Strain	36	42		
Proportion	70.59	77.78		
Fungi (3 strains)			0.271	0.602
Strain	1	2		
Proportion	1.96	3.70		

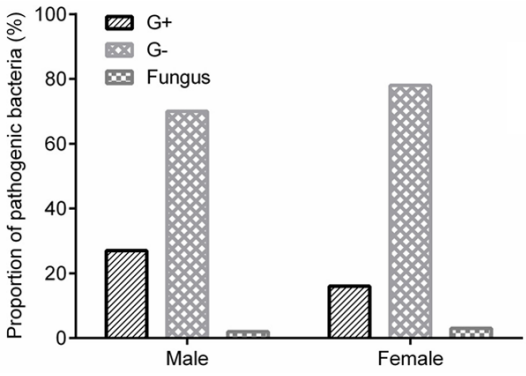

Figure 1. Proportion of pathogens in different genders.

immune response and disrupting the balance of various subtypes of lymphocytes; thus, it is believed that bacterial endotoxin is also one of the factors leading to airway hyperreactivity and inducing asthma [8-10]. Bacteria that invade the respiratory tract can act as both an infectious source and an allergen, producing specific antibodies that attach to the respiratory mucosa, resulting in allergic reactions in the body and inducing asthma [11]. Epidemiological surveys have shown that bronchial asthma caused by respiratory tract infection accounts for 70-80%, which is also the main factor inducing bronchial asthma [12]. If the infection is poorly controlled or repeatedly infected, it can also lead to acute attack of asthma. Therefore, it is believed that recurrent respiratory tract infection is one of the important factors leading to acute attack of bronchial asthma [13]. Thus, timely and effective control of infection can delay or prevent acute attack of bronchial asthma to a certain extent, while the premise is to determine whether there is pathogens infection in acute attack of bronchial asthma.

In this study, ninety-four infected samples were detected from 159 patients with acute attack of bronchial asthma, with the infection rate of 59.12%, which was slightly lower than the positive strain detection rate of 67% reported by Quan et al. [14]. It is speculated that it should be related to multiple factors such as geographical differences and local health environment. A total of 105 strains of pathogenic bacteria were detected in this study, of which G^- accounted for the highest proportion, accounting for 74.29%, followed by G^+, accounting for 22.86%, while fungi were the least detected, accounting for only 2.86%. Among

Table 6. Analysis of drug resistance of major G^-

Antimicrobial Drugs	*Pseudomonas aeruginosa* (n=15)		*Escherichia coli* (n=13)	
	Resistant Strains (n)	Drug resistance rate (%)	Resistant Strains (n)	Drug resistance rate (%)
Ampicillin	10	66.67	9	69.23
Amoxicillin	10	66.67	10	76.92
Gentamicin	1	6.67	7	53.85
Ceftazidime	3	20.00	4	30.77
Ceftriaxone	13	86.67	5	38.46
Cefoperazone/Sulbactam	1	6.67	2	15.38
Piperacillin	1	6.67	1	7.69
Ciprofloxacin	2	13.33	4	30.77
Levofloxacin	2	13.33	3	23.08
SMZ-TMP	1	6.67	2	15.38
Imine	1	6.67	0	0.00
Melodia	0	0.00	1	7.69

Note: SMZ-TMP: trimethoprim-sulfamethoxazole.

Table 7. Analysis of drug resistance of main G^+

Antimicrobial Drugs	Staphylococcus aureus (n=7)	
	Resistant Strains (n)	Drug resistance rate (%)
Penicillin	6	85.71
Ampicillin	4	57.14
Gentamicin	0	0.00
Erythromycin	4	57.14
Kynomycin	3	42.86
Vancomycin	0	0.00
Cefoperazone/Sulbactam	1	14.29
Rifampicin	0	0.00
Ciprofloxacin	2	28.57
Levofloxacin	1	14.29
SMZ-TMP	1	14.29

Note: SMZ-TMP: trimethoprim-sulfamethoxazole.

the detected G^-, *Pseudomonas aeruginosa* was the most detected (14.29%), followed by *Escherichia coli* and *Acinetobacter baumannii* (12.38% and 11.43%, respectively). Among the detected G^+, *Staphylococcus aureus* was the most detected (6.67%), suggesting that most of the patients with acute attack of bronchial asthma in this area were accompanied by pathogens infection, and G^- induced infection was predominant, which was consistent with the results of relevant studies [15]. In this study, the infection rate and pathogen distribution of patients of different ages and genders were statistically analyzed. The results showed that among 94 infected samples detected, the infection rate of patients > 60 years old was significantly higher than that of patients ≤ 60 years old (74.16% vs 40.00%). There was no statistical difference in the infection rate between male and female patients. There was also no statistical difference in the detection rate of G^+, G^- and fungal strains in patients of different ages and genders, suggesting that patients > 60 years old with acute attack of bronchial asthma had higher risk of infection and were more likely to cause bacterial infection. There was no difference in the types of pathogens infection between patients of different ages and genders, which was consistent with the study results of Liu et al. [16]. In this study, the sample size was limited, and the pathogen strains detected were also limited, for example, there were only 3 cases of fungal infection, so there was no statistical difference. Expanding the sample size and increasing the number of detected strains could further verify whether there was a statistical difference in the detection rate of different strains in patients of different ages and genders.

Bronchial asthma is easy to have repeated attacks. Acute attack is often accompanied by bacterial infection, and airway spasm leads to airway secretions that are not easy to discharge. A large number of secretions can accu-

mulate in the airway for a long time, further aggravating the airway inflammatory response, so clinical anti-infective treatment is extremely important [17-19]. In recent years, with the irrational use of antibacterial drugs, the composition of pathogens has also gradually changed, and the number of drug-resistant strains has gradually increased, resulting in poor clinical treatment [20, 21]. Therefore, it is essential to clarify the pathogens composition and drug resistance in patients with acute attack of bronchial asthma to guide the application of clinical antibacterial drugs and improve the clinical treatment effect. The results of *in vitro* drug sensitivity test in this study showed that *Pseudomonas aeruginosa* had high resistance to ampicillin, amoxicillin and ceftriaxone, while low resistance to gentamicin, cefoperazone/sulbactam, piperacillin, SMZ-TMP, imipenem and meropenem; *Escherichia coli* had high resistance to ampicillin, amoxicillin and gentamicin, while low resistance to piperacillin, imipenem and meropenem; *Staphylococcus aureus* had high resistance to penicillin, ampicillin and erythromycin, while showed no resistance to gentamicin, vancomycin and rifampicin. Considering the cost of antibacterial drug treatment, piperacillin could be used for patients with mild symptoms, while cefoperazone/sulbactam, imipenem and meropenem could be considered for patients with severe symptoms. However, for infections caused by different strains, sensitive antibacterial drugs should be selected according to *in vitro* drug sensitivity test.

In summary, acute attack of bronchial asthma is closely related to pathogens infection, while patients > 60 years old have a higher risk of infection. For infections caused by different strains, sensitive antibacterial drugs should be selected according to *in vitro* drug sensitivity test in clinical practice to reduce the occurrence of drug resistance and improve the therapeutic effect.

Disclosure of conflict of interest

None.

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