Infection and Drug Resistance

Dovepress
Taylor & Francis Group

Open Access Full Text Article

ORIGINAL RESEARCH

Risk Factor Analysis and Molecular Epidemiological Investigation of Carbapenem-Resistant *Enterobacteriaceae* (CRE) Infection in Patients with Acute Pancreatitis

Fangfang Yang[1], Fang Liu[2], Xiaoji Zhao[1], Qian Chen[2]

[1]Department of Clinical Laboratory, The Third Hospital of Mianyang, Sichuan Mental Health Center, Mianyang, People's Republic of China; [2]Department of Clinical Laboratory, Chongqing Red Cross Hospital (Jiangbei District People's Hospital), Chongqing, People's Republic of China

Correspondence: Qian Chen, Chongqing Red Cross Hospital (Jiangbei District People's Hospital), Chongqing, 400000, People's Republic of China, Email qianchenfafafa@163.com

Objective: Patients with acute pancreatitis (AP) complicated by carbapenem-resistant *Enterobacteriaceae* (CRE) infection often have a higher mortality rate. However, little investigation on the risk factor analysis has been published for the AP complicated by CRE. Therefore, this study conducted a retrospective analysis of the clinical characteristics, risk factors, and molecular epidemiological features associated with CRE infection in patients with AP.

Methods: A total of 240 patients with AP were admitted to our hospital from 2011 to 2021 as the research objects, and were divided into a CRE group of 60 cases and a non-CRE group of 180 cases based on whether they were co-infected with CRE or not. Furthermore, both univariate analysis and multivariate analysis were used to analyze the risk factors of AP co-infection with CRE. In the CRE group, polymerase chain reaction (PCR) and agarose gel electrophoresis (AGE) were used to detect the expression of five common carbapenemase genes including bla_{KPC}, bla_{IMP}, bla_{VIM}, bla_{NDM}, and $bla_{OXA\text{-}48}$.

Results: The pathogenic bacteria in the CRE group are composed of *Klebsiella pneumonia* at 35.00%, *Escherichia coli* at 33.33%, *Enterobacter cloacae* at 25.00%, and *Citrobacter freundii* at 6.67%. Multivariate analysis showed that APACHE-II scores (OR=1.22), history of abdominal surgery (OR=81.82), and ERCP (OR=3.66) were independent risk factors for AP co-infection with CRE ($P<0.05$). About half (18/40) of the CRE carried carbapenemase genes. bla_{KPC} was the major carbapenemase gene.

Conclusion: There are many risk factors for AP co-infection with CRE, which can occur in patients with high APACHE-II scores, experienced ERCP, and a history of abdominal surgery.

Keywords: acute pancreatitis, carbapenem-resistant *Enterobacteriaceae* (CRE), infection, risk factors

Introduction

Acute pancreatitis (AP) is the activation of pancreatic enzymes caused by various reasons and may lead to local damage, systemic inflammatory response syndrome, and organ failure. It is one of the common gastrointestinal diseases and is potentially lethal.[1] In the United States, the AP is the main cause of hospitalization for gastrointestinal diseases, for which approximately 275,000 patients are hospitalized by AP each year and the total treatment cost reaches approximately US$2.6 billion.[2]

Biliary diseases, hypertriglyceridemia, and alcoholism are the most common causes of AP.[3] However, other causes including trauma, surgery, pregnancy, infection, drugs, inherited metabolic diseases and autoimmune diseases can also lead to AP, among which infection is closely related to the mortality of patients.[4] AP is often accompanied by two major clinical courses, the first one is systemic inflammatory response syndrome (SIRS) and the other one is compensatory anti-inflammatory response syndrome (CARS).[5] The former course usually occurs early and lasts for 1 or 2 weeks. The latter ranges from a few weeks to a few months and is characterized by the presence of local complications and related

Received: 24 October 2024
Accepted: 9 January 2025
Published: 16 January 2025

infectious complications that often occur in the later stage of the disease and usually last for a rather long time.[6] Research indicates that 80% of deaths occur in the terminal stage, as a result of infection.

The early stages of AP are sterile, but as the disease progresses, bacterial overgrowth and translocation can occur due to inflammatory reactions, intestinal barrier dysfunction, and frequent use of prophylactic antibiotics, making patients susceptible to multidrug-resistant Gram-negative bacterial (MDR-GNB) infections.[7,8] Among them, carbapenem-resistant *Enterobacteriaceae* (CRE) have attracted attention due to their high resistance rate to most antibiotics. Recently, the infection rate of CRE has increased in patients with AP concurrent infection.[9] The high mortality caused by the AP complicated by the CRE and the shortage of effective anti-infective treatment have brought great challenges to the clinic.

Carbapenemase production is the main resistance mechanism of CRE.[10] Globally, the most important carbapenemases in CRE are divided into three categories: (1) class A serine enzymes, such as KPC-type enzymes; (2) class B metalloenzymes, such as NDM, VIM, and IMP; (3) d-serine enzymes, such as OXA-48-type enzymes.[11] Some observational studies have shown that the mortality of patients with bacteremia caused by carbapenem-resistant *Enterobacteriaceae* is as high as 40% to 60.[12,13] However, little investigation on the risk factor analysis has been published for the AP complicated by the CRE. Therefore, this study conducted a retrospective analysis of the clinical characteristics and risk factors associated with CRE infection in patients with AP and also examined the molecular epidemiological features to provide references for clinical prevention and control of AP.

Materials and Methods

Study Design and Population

The study was conducted at a general hospital in southwest China, where 240 non-repetitive patients with AP co-infection were collected from April 2011 to December 2021 through the hospital's electronic case system and clinical microbiology database. Based on the presence or absence of CRE co-infection, the patients were divided into the CRE group (n=60) and the non-CRE group (n=180). It is important to note that if multiple detections of CRE occurred in the same hospitalized patient, only the first infective case was considered.

The criteria for AP include the following requirements: (1) the patients must meet the 2012 revision of the Atlanta classification;[14] (2) the hospitalized days of patients are more than 2. The exclusion criteria are as follows: (1) age < 18 years old; (2) the clinical data is incomplete. The detailed experimental process is shown in the flow chart (Figure 1).

Clinical Data Statistics and Analysis

A retrospective comparative analysis was conducted using various parameters as potential risk factors: (1) General hospitalization data including age, sex, transfer status, and etiology; (2) prognosis indicators containing death rate, shock, total hospitalization days, and ICU admission.; (3) Underlying diseases; (4) Infections during hospitalization; (5) Invasive procedures; (6) Antibacterial drug usage before detection of CRE.

Antibiotic Susceptibility Test

According to the Clinical and Laboratory Standards Institute (CLSI) guideline M45,[15] antimicrobial susceptibility testing was conducted using microdilution and disk diffusion methods.

The criteria for CRE were as follows: any isolated Enterobacteriaceae resistant to meropenem (disc diffusion diameter ≤19 mm, ertapenem ≤19 mm), ertapenem (minimum inhibitory concentration (MIC) ≥2 μg/mL) or imipenem (MIC≥4 μg/mL).

Detection of Carbapenemase Gene

Common carbapenemase genes, including Ambler's class A (KPC enzymes), class B metallo-beta-lactamase (NDM, VIM, and IMP enzymes), and class D oxacillinase (OXA-48-like enzymes), were detected in 40 strains of the CRE group. DNA templates were extracted by metal baths. The primer sequences specifically employed for detecting carbapenemase-producing isolates are presented in Table 1. Each reaction mix (20 μL) contained 10 uL PCR master

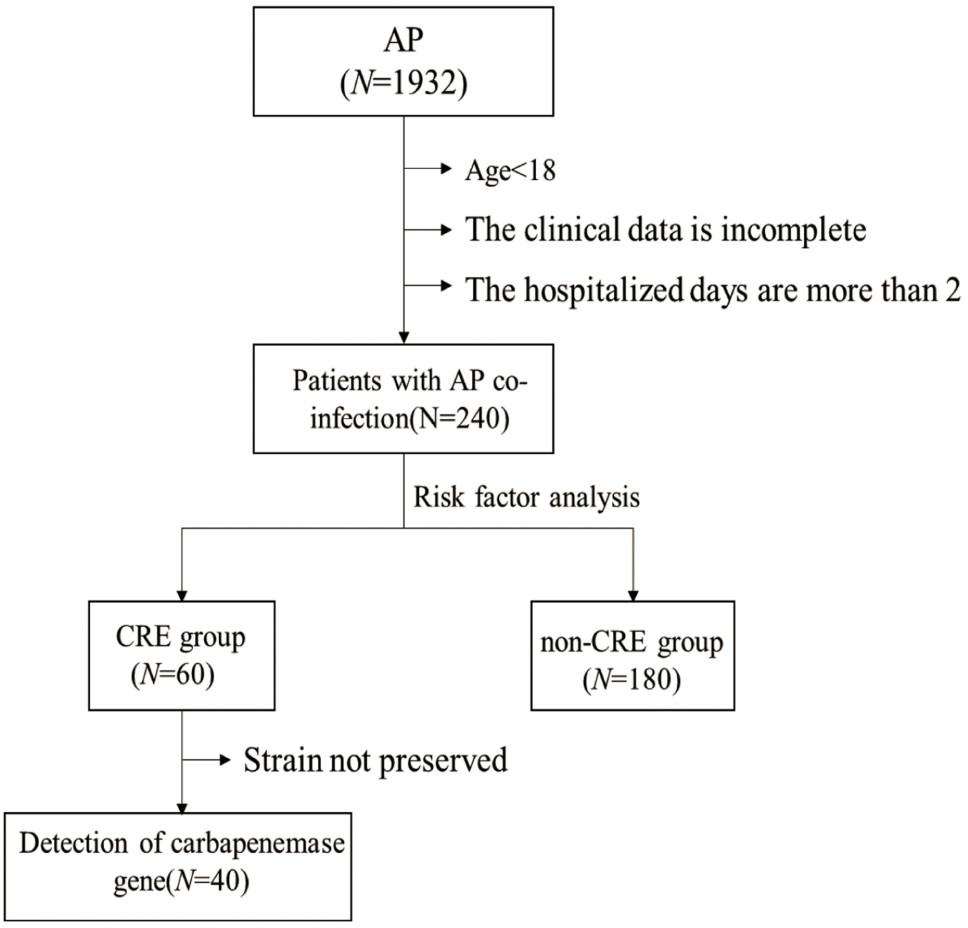

Figure 1 The flowchart of study design.
Abbreviations: AP, acute pancreatitis; CRE, carbapenem-resistant *Enterobacteriaceae*.

mix (SYBR Premix Ex Taq II; Takara Bio Inc), 1 μL of each forward and reverse primer (100μM), 6 μL of molecular grade nuclease-free water, and 2 μL of DNA template. Amplification was done at 95 °C for 3 minutes as the initial step for predenaturation, followed by denaturation at 92 °C for 20 seconds, annealing at 68 °C for 20 seconds (1.5 °C decrease and 6 seconds increase per cycle) and extension at 72°C for 30 seconds were repeated for 8 cycles. Then 25 cycles of

Table 1 Primer Sequences Used in This Article

Carbapenemase Genes	Primer Sequence (5′—3′)	Amplicon Size (bp)
$bla_{KPC\text{-}f}$	CATTCAAGGGCTTTCTTGCTGC	488
bla_{KPC-r}	ACGACGGCATAGTCATTTGC	
$bla_{IMP\text{-}f}$	CATGGTTTGGTTGTTCTTGT	488
$bla_{IMP\text{-}r}$	ATAATTTAGCGGACTTTGGC	
$bla_{VIM\text{-}f}$	TTATGGAGCAGCAACGATGT	920
$bla_{VIM\text{-}r}$	CAAAAGTCCCGCTCCAACGA	
$bla_{NDM\text{-}r}$	CGGAATGGCTCATCACGATC	621
$bla_{NDM\text{-}f}$	GGTTTGGCGATCTGGTTTTC	
$bla_{OXA\text{-}48\text{-}f}$	TTGGTGGCATCGATTATCGG	438
$bla_{OXA\text{-}48\text{-}r}$	GAGCACTTCTTTTGTGATGGC	

Abbreviations: f, forward primer; r, reverse primer; bla_{KPC}, *Klebsiella pneumoniae* carbapenemase; bla_{IMP}, Imipenem-resistant *Pseudomonas*; bla_{VIM}, Verona Intergron-encoded Metallo-β-Lactamases; bla_{NDM}, New Delhi Metallo-β-Lactamases; $bla_{OXA\text{-}48}$, Oxacillinase-48.

https://doi.org/10.2147/IDR.S498829

denaturation at 92 ° C for 20 seconds, annealing at 55 °C for 20 seconds, and extension at 72 °C for 30 seconds were repeated. The final elongation was at 72 °C for 5 minutes. All PCR amplicons were analyzed by horizontal gel-electrophoresis in a 1% (weight/volume) (Bio-BAD subcell G1, USA) Tris/Acetate/EDTA 50 x concentrate buffer. The agarose was stained with 4 uL of Gel-red (Bio-Rad, USA). About 5 μL of amplicons were put into the wells and run in a 1 x Tris-acetate EDTA (TAE) at 150 volts for 30 minutes. DL2000 (Takara, AJG1332A) was used as a marker. The amplicons were visualized with the BIO-RAD GelDoo XR (Bio-Rad, USA).

Statistical Analysis

SPSS 24.0 (IBM SPSS software, Armonk, NY, USA) was used to analyze the results. Categorical variables were shown as frequencies and proportions. For continuous variables, normally distributed data were reported by mean and standard deviation. The difference between the two groups was tested by Student's *t*-test. Skewed variables were represented by median and interquartile ranges. Mann–Whitney *U*-test was used to compare the differences between the two groups. In univariate analyses, categorical variables were analyzed using either the Chi-square or Fisher's exact test. The Logistic regression method was used to determine the relationship between the potential risk factors and the pancreatitis co-infection with CRE. *P*-value <0.05 was considered significant.

Results

Sample Source and Species Distribution of CRE Infection in AP

In the CRE group, the isolates were obtained from sputum (10%), gastric juice (1.67%), urine (5%), abdominal drainage fluid (36.67%), wound secretion (10%), bile (21.67%), and blood (15%) (Table 2). The distribution of pathogens in the CRE group is listed in Table 3. It can be seen that the bacteria of AP co-infected with CRE included *Klebsiella pneumoniae* at 35.00%, *Escherichia coli* at 33.33%, *Enterobacter cloacae* at 25.00%, and *Citrobacter freundii* at 6.67% (Table 3).

Univariate Analysis of AP Complicated with CRE Infection

The results of the related risk factors analysis are listed in Table 4. It is shown that AP co-infection with CRE is related to the age, total number of hospitalized days, hospitalized days before the detection of CRE, ICU duration, days of fasting, score of APACHE II, history of abdominal surgery within 6 months, basic hyperlipidemia, diabetes, kidney disease,

Table 2 The Specimen Types of Pathogens in the CRE Group

Specimen types	Number of cases (n)	Proportion (%)
Sputum	6	10.00
Gastric juice	1	1.67
Urine	3	5.00
Ascites	6	10.00
Drainage	16	26.67
Wound secretions	6	10.00
Bile	13	21.67
Blood	9	15.00

Table 3 The Distribution of Pathogens in the CRE Group

Name of Bacteria	Number of Cases (n)	Proportion (%)
Klebsiella pneumoniae	21	35.00
Escherichia coli	20	33.33
Enterobacter cloacae	15	25.00
Citrobacter freundii	4	6.67

combined infections of abdominal, lung and urinary tract, catheterization and days of enteral nutrition, puncture, ERCP, drainage tube, gastric tube, urinary catheter, intravenous, hemofiltration, other invasive operations and the usage of antibacterial drugs before testing. The differences are statistically significant ($P<0.05$).

Table 4 Univariate Analysis of AP Complicated with CRE Infection

Factor	CRE Group (n=60)	Non-CRE Group (n=180)	OR (95% CI)	*P*
Characteristic				
Sex (male)	21(65.00%)	101(56.11%)	0.69(0.38–1.26)	0.228
Age#	59.79±16.35	54.02±18.91	0.98(0.96–1.00)	0.025
History of pancreatitis	11(18.33%)	23(12.78%)	1.53(0.70–3.37)	0.288
Abdominal surgery history within 6 months#	4(6.67%)	1(0.56%)	12.79(1.40–116.76)	0.024
Fasting days#	12.92±14.62	5.87±7.20	1.07(1.04–1.11)	0.000
Biliary	27(45.00%)	105(58.33%)	0.71(0.40–1.28)	0.261
Hyperlipidemia#	15(25.00%)	22(12.22%)	2.39(1.15–4.99)	0.020
Prognosis				
Death	7(11.67%)	8(4.44%)	–	–
APACHE II score#	12.62±4.59	8.23±4.92	1.18(1.11–1.26)	0.000
Total hospitalization days#	65.50±60.48	28.25±17.71	1.04(1.03–1.05)	0.000
Hospitalization days before CRE detection#	28.27±44.81	8.43±9.12	1.08(1.05–1.11)	0.000
Before ICU admission	37(61.67%)	92(51.11%)	1.54(0.85–2.80)	0.157
ICU duration#	13.52±20.93	3.90±6.96	1.06(1.03–1.10)	0.000
Shock	10(16.67%)	26(14.44%)	1.19(0.53–2.63)	0.677
Multiple organ dysfunction	14(23.33%)	26(14.44%)	1.80(0.87–3.74)	0.113
Basic illness				
Hypertension	14(23.33%)	44(24.44%)	0.94(0.47–1.87)	0.862
Diabetes#	18(30.00%)	26(14.44%)	2.54(1.27–5.07)	0.008
Tumor	1(1.67%)	11(6.11%)	0.26(0.03–2.06)	0.202
Cardiovascular diseases	7(11.67%)	21(11.67%)	1.00(0.40–2.49)	1.000
Kidney disease#	10(16.67%)	4(2.22%)	8.80(2.65–29.26)	0.000
Respiratory diseases	10(16.67%)	16(8.89%)	2.05(0.88–4.80)	0.098
Hepatobiliary disease	46(76.67%)	135(75.00%)	1.10(0.55–2.18)	0.795
Combined infection				
Urinary tract infection#	11(18.33%)	10(5.56%)	3.82(1.53–9.51)	0.004
Lung infection#	36(60.00%)	68(37.78%)	2.47(1.36–4.49)	0.003
Abdominal infection#	44(73.33%)	49(27.22%)	7.35(3.80–14.22)	0.000
Bloodstream infection	19(31.67%)	59(32.78%)	0.95(0.51–1.78)	0.874
Skin tissue infection	8(13.33%)	11(6.11%)	2.36(0.90–6.19)	0.080
Intrusive operation				
Enteral nutrition#	33(55.00%)	33(18.33%)	5.44(2.89–10.26)	0.000
Surgical intervention	13(21.67%)	31(17.22%)	1.33(0.64–2.75)	0.442
Puncture#	28(46.67%)	23(12.78%)	5.97(3.06–11.67)	0.000
ERCP#	14(23.33%)	20(11.11%)	2.44(1.14–5.19)	0.021
Venous catheterization#	37(61.67%)	56(31.11%)	3.56(1.94–6.55)	0.000
Venous catheterization days#	14.72±29.64	3.20±8.00	1.05(1.02–1.08)	0.000
Blood flow filtration#	24(40.00%)	27(15.00%)	3.78(2.00–7.30)	0.000
Gastrojejunal tube#	39(65.00%)	75(41.67%)	2.60(1.42–4.77)	0.002
Drainage tube#	53(88.33%)	85(47.22%)	8.46(3.65–19.62)	0.000
Urinary catheter#	38(63.33%)	62(34.44%)	3.29(1.79–6.04)	0.000
Tracheal intubation	18(30.00%)	43(23.89%)	1.37(0.71–2.62)	0.347

(*Continued*)

https://doi.org/10.2147/IDR.S498829

Table 4 (Continued).

Factor	CRE Group (n=60)	Non-CRE Group (n=180)	OR (95% CI)	*P*
Usage of antimicrobial				
History of antibiotic usage[#]	56(93.33%)	142(78.89%)	3.75(1.28–10.99)	0.016
β-lactamase inhibitors	18(30.00%)	34(18.89%)	1.84(0.95–3.58)	0.073
Cephalosporins[#]	38(63.33%)	50(27.78%)	4.49(2.42–8.33)	0.000
Carbapenems[#]	39(65.00%)	45(25.00%)	5.57(2.97–10.45)	0.000
Aminoglycosides[#]	12(20.00%)	6(3.33%)	7.25(2.59–20.32)	0.000
Fluoroquinolones[#]	14(23.33%)	16(8.89%)	3.12(1.42–6.86)	0.005

Notes: [#], $P<0.05$.
Abbreviations: CI, confidence interval; OR, odds ratio; CRE, carbapenem-resistant *Enterobacteriaceae*; APACHE II, Acute Physiology and Chronic Health Status II; ICU, intensive care unit; ERCP, endoscopic retrograde cholangiopancreatography.

Multivariate Analysis of AP Complicated with CRE Infection

The results of multivariate analysis for the CRE infections in the AP are listed in Table 5. It is shown that the APACHE II score at admission, history of abdominal surgery within 6 months, and ERCP were independent risk factors of AP co-infection with CRE ($P<0.05$).

Table 5 Multivariate Analysis of AP Complicated with CRE Infection

Factor	OR	95% CI	*P*
Age	0.99	0.96–1.02	0.483
Total hospital days	1.02	1.00–1.05	0.098
Days in the hospital before detection of the bacteria	1.07	1.00–1.15	0.064
ICU duration	1.03	0.97–1.08	0.375
Fasting days	0.98	0.92–1.05	0.57
APACHE II score[#]	1.22	1.09–1.35	0.000
Abdominal surgery history (within 6 months) [#]	81.82	2.62–2555.96	0.012
Hyperlipidemia	2.23	0.58–8.64	0.246
Diabetes	1.99	0.64–6.12	0.232
Kidney disease	5.55	0.80–38.58	0.083
Hemofiltration	0.83	0.19–3.59	0.798
Puncture drainage	1.4	0.43–4.55	0.577
ERCP[#]	3.66	1.0–13.34	0.049
Drainage tube	2.14	0.44–10.46	0.346
Stomach tube	0.42	0.12–1.42	0.163
Urinary catheter	1.55	0.45–5.29	0.487
Enteral nutrition	0.44	0.12–1.63	0.217
Venous catheterization	0.73	0.18–2.94	0.654
Venous catheterization days	0.97	0.93–1.01	0.154
Urinary tract infection	0.61	0.11–3.40	0.572
Lung infection	0.38	0.11–1.26	0.112
Abdominal infection	2.83	0.95–8.47	0.063
History of antibiotic use before testing	1.24	0.22–7.17	0.808
Cephalosporins	1.28	0.38–4.29	0.694
Carbapenems	1.87	0.60–5.82	0.277
Aminoglycosides	1.56	0.26–9.24	0.626
Fluoroquinolones	1.68	0.39–7.16	0.485

Note: [#], $P<0.05$.
Abbreviations: CI, confidence interval; OR, odds ratio; APACHE II, Acute Physiology and Chronic Health Status II; ICU, intensive care unit; ERCP, endoscopic retrograde cholangiopancreatography.

Detection of Carbapenemase Gene

The results indicated that 18 (45%) of the 40 strains expressed carbapenemase genes, of which 7 expressed bla_{KPC}, and all were strains of *Klebsiella pneumoniae* (*kpn*). 7 strains expressed bla_{NDM}, including 1 strain of *kpn*, 5 strains of *Escherichia coli (eco)*, 1 in *Citrobacter freundii* (*cfr*). A total of 4 strains of *kpn* expressed both bla_{KPC} and bla_{NDM} genes simultaneously. No strains expressed the bla_{IMP}, bla_{VIM}, or bla_{OXA-48} genes. Refer to Figure 2 and Table 6 for further details.

Discussion

With a diverse range of clinical manifestations, AP is mild and transient but can rapidly progress to a severe and fatal condition. However, the majority of AP-related deaths are attributed to infection complications.[16] It has been reported that 40% to 70% of severe AP patients develop bacterial infections during the natural course of the disease, with infection being the most significant risk factor for mortality in these cases.[17] Moreover, CRE infection is a thorny problem in the treatment of AP. However, limited research has been published on this topic in recent years.

The co-infection of pathogenic bacteria in AP primarily consists of Gram-negative bacteria, particularly *Enterobacteriaceae* species which can cause respiratory tract infections, urinary tract infections, abdominal cavity infections, and surgical site infections. With the widespread use of broad-spectrum antibiotics, there has been an increase in multi-drug resistant Enterobacteriaceae strains as well.[18] In recent years, due to the emergence and spread of

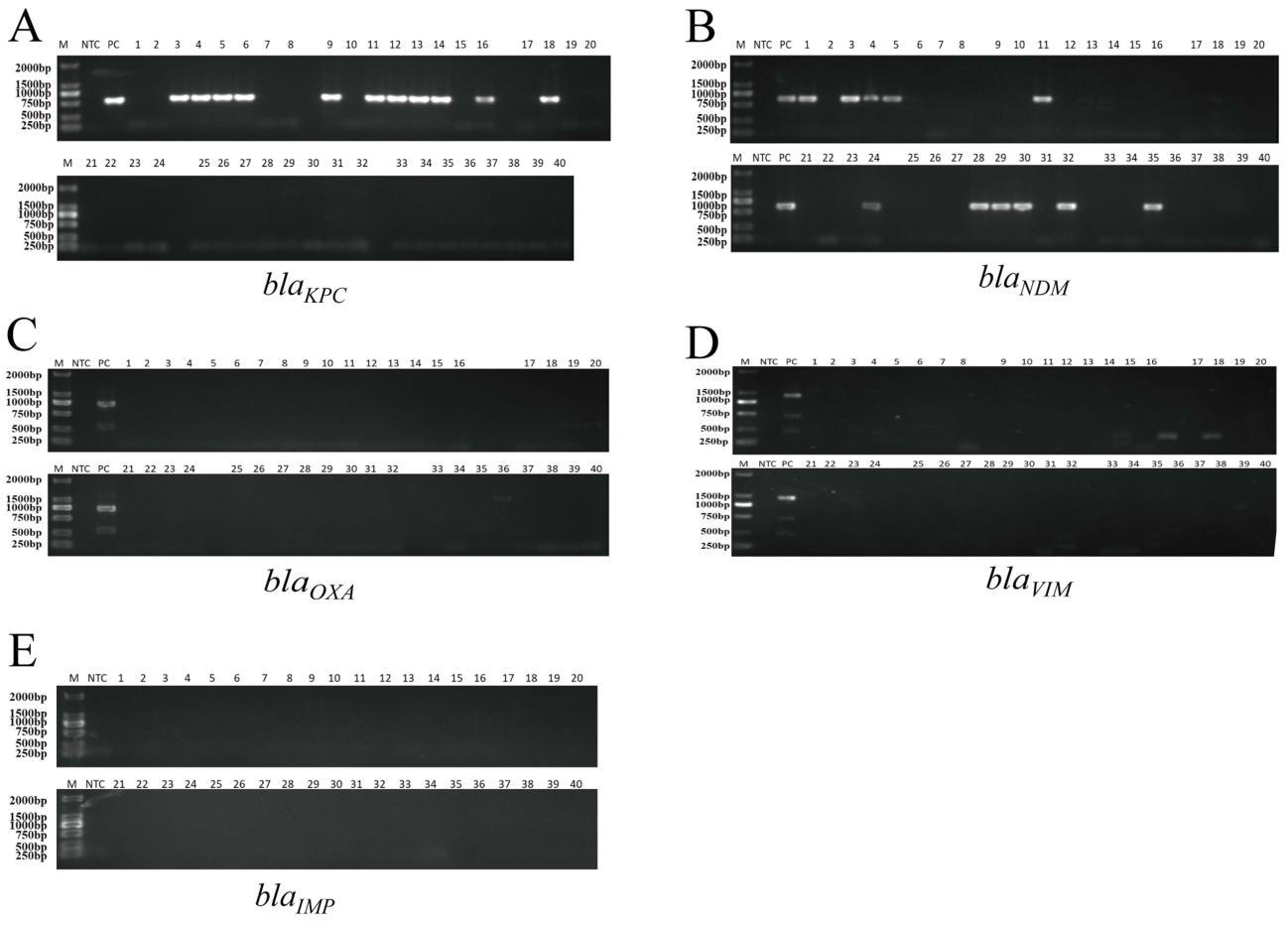

Figure 2 Detection of carbapenemase gene, including bla_{KPC} (**A**), bla_{NDM} (**B**), bla_{IMP} (**C**), bla_{VIM} (**D**), bla_{OXA-48} (**E**). Add 5μL of DNA marker into the prepared gel tank to the first well, add a no-template control to the second well, add positive sample controls (bla_{KPC}, bla_{NDM}, bla_{VIM}, bla_{OXA-48} have positive controls, bla_{IMP} has no positive control strain) to the third well, and start adding samples from the fourth well. After adding samples to each row (8 samples), leave one well empty and then add a second row (8 samples).

Abbreviations: M, maker; NTC, no template control; PC, positive sample control.

https://doi.org/10.2147/IDR.S498829

Table 6 Amplification Result of Carbapenemase Gene

Carbapenemase genes	N	Bacterial strain (N)
bla_{KPC}	7	*kpn* (7)
bla_{NDM}	7	*kpn* (1), *eco (5), cfr* (1)
bla_{KPC}+bla_{NDM}	4	*kpn (4)*
bla_{IMP}	0	–
bla_{VIM}	0	–
bla_{OXA-48}	0	-

Abbreviations: N, number; *kpn, Klebsiella pneumoniae; eco, Escherichia coli; cfr, Citrobacter freundii.*

carbapenemase genes within these strains, there has been a sharp rise in CRE rates leading to considerable difficulties in clinical anti-infection treatments.

In the study, most of the bacteria detected in patients with AP complicated with CRE was *Klebsiella pneumoniae* (35.00%), followed by *Escherichia coli* (33.33%), consistent with the prevalence of CRE in China.[19,20] As the 2021 CHINET shows, from 2005 to 2021, the resistance rates of *Klebsiella pneumoniae* to Meropenem and Imipenem increased from 2.9% and 3.0% to 24.4% and 25.3%, respectively.[21] This phenomenon may be related to the fact that *Klebsiella pneumoniae* is more prone to produce extended-spectrum β-Lactamase (ESBLS), AmpC enzyme, and carbapenemase. The primary resistance mechanisms of CRE include: (1) production of carbapenemase; (2) reduced intracellular antibiotic levels, which are attributed to decreased absorption due to changes in membrane porins and increased excretion resulting from overexpression of efflux pumps. This is often accompanied by overexpression of ESBLs and/or AmpC beta-lactamases.[22] In our study, different types of carbapenem-resistant genes were detected in 45% of the isolates. The remaining 55%, although identified as CRE in drug susceptibility testing, had a loss of carbapenem-resistance gene expression, possibly related to the production of other β-lactamases, loss of porins, and overexpression of efflux pumps.

In the univariate analysis, we found that AP combined with CRE infection was related to age, total hospitalization days, hospitalization days before CRE detection, ICU duration, fasting days, hyperlipidemia, diabetes, kidney disease, combined with abdominal cavity, lung, urinary tract infection, enteral nutrition, puncture, drainage tube, gastric tube, catheter, venous catheterization and days, hemofiltration and the use of antibiotics before detection ($P<0.05$). However, the multivariate analysis of this study showed that there was no significant difference in the above covariates, which may be related to the individual differences of patients.

The results of multivariate analysis showed that there were three independent risk factors for AP combined with CRE infection: (1) APACHE II score: The APACHE II scoring system is widely used in intensive care units (ICU).[23] The higher the score of the patients on admission, the more severe the clinical symptoms of the patients, and there may have been multiple organ dysfunction failure, which will further aggravate pancreatic necrosis, gastrointestinal mucosal damage, and bacterial translocation, increase the risk of infection, and be more prone to CRE infection. (2) Abdominal surgery history: Due to the large area of the abdominal cavity and the complex physiological functions, it is easy to cause some physiological functions of the patients to be destroyed during the operation. Therefore, infection is a common complication of abdominal surgery, including incision infection, and pulmonary infection. The incidence of postoperative infection is 1%−5%.[24] In addition, the decreased immunity of severely injured patients also increases the chance of CRE infection. (3) Endoscopic retrograde cholangiopancreatography (ERCP): ERCP has the advantages of reduced trauma, fewer complications, faster postoperative recovery, and shorter hospital stays. It has been widely used in the diagnosis and treatment of benign and malignant pancreaticobiliary diseases.[25] However, ERCP is an invasive surgery and may cause some related adverse events. It has been reported that the incidence of infection after ERCP was 3.58% to 13.51%, and the incidence of bacteremia was 3.56%.[26]

The emergence of carbapenemase (CHBLs) has always been the main mechanism of CRE resistance. We detected the genes of five common CHBLs with a detection rate of 45%, mainly bla_{KPC} and bla_{NDM} genes, which is consistent with

previous reports.[27] The other strains did not detect the common five drug resistance genes, which may be due to other drug resistance mechanisms, such as the lack of membrane porin, the abnormal expression of efflux pump, or the production of drug resistance genes not detected in this study, which needs further study.[28]

There are still some limitations in our research. Firstly, this is a 10-year retrospective study conducted in a single center. There may be limitations due to geographic limitations, insufficient sample size, and unobserved confounding factors. Additionally, due to the extended period, the bacterial strain was not fully preserved. Although there were 60 subjects in the CRE group, we ultimately conducted resistance gene testing on only 40 of them. Secondly, in the case of patients who had been treated in other hospitals, the data were not comprehensive enough, and the laboratory data and antibiotic use in other hospitals were not analyzed. Additionally, due to the long period of the case collation, some current laboratory projects have not been carried out at the beginning, so some potential risk factors cannot be confirmed. These will require further investigation in subsequent studies by increasing sample size and employing a prospective cohort study design to delve deeper into the findings.

Conclusions

In conclusion, to reduce the incidence of CRE infection in AP, we should evaluate critically ill patients, strengthen clinical management, isolate and protect patients with a history of abdominal surgery in the past six months, and strictly standardize the indications and aseptic techniques of ERCP. In the future, we will continue to conduct further research on the resistance mechanism of strains without carbapenem-resistant genes.

Data Sharing Statement

Data and materials associated with this work are available by request from the corresponding author.

Ethical Approval

As the study was performed with retrospective resources and will not have any impact on the diagnosis and treatment of patients, the ethics committee of the Third Hospital of Mianyang has approved the exemption of informed consent. This study was approved through the local ethics committee of the Third Hospital of Mianyang (approval number: 2024-48). This study complies with the Declaration of Helsinki.

Acknowledgments

We sincerely thank Dr. Niu Siqiang for his help on the paper.

Funding

This study was supported by the joint medical (including traditional Chinese medicine) research project by Health Commission and Science and Technology Bureau of Jiangbei District, Chongqing (202321).

Disclosure

The authors declare that they have no competing interests.

References

1. Zerem E, Kurtcehajic A, Kunosić S, et al. Current trends in acute pancreatitis: diagnostic and therapeutic challenges. *World J Gastroenterol*. 2023;29 (18):2747–2763.
2. Peery AF, Crockett SD, Murphy CC, et al. Burden and Cost of Gastrointestinal, Liver, and Pancreatic Diseases in the United States: update 2021. *Gastroenterology*. 2022;162(2):621–644. doi:10.1053/j.gastro.2021.10.017
3. Lee PJ, Papachristou GI. New insights into acute pancreatitis. *Nat Rev Gastroenterol Hepatol*. 2019;16(8):479–496.
4. Wu D, Jia Y, Cai W, et al. Impact of multiple drug-resistant Gram-negative bacterial bacteraemia on infected pancreatic necrosis patients. *Front Cell Infect Microbiol*. 2022;12(1044188). doi:10.3389/fcimb.2022.1044188.
5. Qiu M, Zhou X, Zippi M, et al. Comprehensive review on the pathogenesis of hypertriglyceridaemia-associated acute pancreatitis. *Annals Med*. 2023;55(2):2265939. doi:10.1080/07853890.2023.2265939
6. Ding L, Yang Y, Li H, et al. Circulating Lymphocyte Subsets Induce Secondary Infection in Acute Pancreatitis. *Front Cell Infect Microbiol*. 2020;10 (128). doi:10.3389/fcimb.2020.00128.

7. Yang N, Ke L, Tong Z, et al. The effect of thymosin α1 for prevention of infection in patients with severe acute pancreatitis. *Expert Opin Biol Ther*. 2018;18(sup1):53–60. doi:10.1080/14712598.2018.1481207
8. Párniczky A, Lantos T, Tóth EM, et al. Antibiotic therapy in acute pancreatitis: from global overuse to evidence based recommendations. *Pancreatology*. 2019;19(4):488–499. doi:10.1016/j.pan.2019.04.003
9. Lin H, Wang P, Lu Y, et al. Clinical and Genomic Characteristics of Carbapenem-Resistant Klebsiella pneumoniae Isolates from Acute Pancreatitis with Infection in China. *Clin Lab*. 2023;69(7). doi:10.7754/Clin.Lab.2022.221017.
10. Ma J, Song X, Li M, et al. Global spread of carbapenem-resistant Enterobacteriaceae: epidemiological features, resistance mechanisms, detection and therapy. *Microbiol Res*. 2023;266(127249):127249. doi:10.1016/j.micres.2022.127249
11. Nordmann P, Dortet L, Poirel L. Carbapenem resistance in Enterobacteriaceae: here is the storm! *Trends Mol Med*. 2012;18(5):263–272. doi:10.1016/j.molmed.2012.03.003
12. Daikos GL, Tsaousi S, Tzouvelekis LS, et al. Carbapenemase-producing Klebsiella pneumoniae bloodstream infections: lowering mortality by antibiotic combination schemes and the role of carbapenems. *Antimicrob Agents Chemother*. 2014;58(4):2322–2328. doi:10.1128/AAC.02166-13
13. Wu D, Xiao J, Ding J, et al. Predictors of Mortality and Drug Resistance Among Carbapenem-Resistant Enterobacteriaceae-Infected Pancreatic Necrosis Patients. *Infect Dis Ther*. 2021;10(3):1665–1676. doi:10.1007/s40121-021-00489-5
14. Banks PA, Bollen TL, Dervenis C, et al. Classification of acute pancreatitis--2012: revision of the Atlanta classification and definitions by international consensus. *Gut*. 2013;62(1):102–111. doi:10.1136/gutjnl-2012-302779
15. Institute CALS. Methods for antimicrobial dilution and disk susceptibility testing of infrequently isolated or fastidious bacteria [M]. *CLSI Guideline M45*. 2015;2015:1.
16. Wu D, Lu W, Huang Y, et al. The impact of multi-drug resistant Pseudomonas aeruginosa infections on acute pancreatitis patients. *BMC Infect Dis*. 2023;23(1):340. doi:10.1186/s12879-023-08230-y
17. Lv C, Zhang ZX, Ke L. Early prediction and prevention of infected pancreatic necrosis. *World J Gastroenterol*. 2024;30(9):1005–1010. doi:10.3748/wjg.v30.i9.1005
18. Lu JD, Cao F, Ding YX, et al. Timing, distribution, and microbiology of infectious complications after necrotizing pancreatitis. *World J Gastroenterol*. 2019;25(34):5162–5173. doi:10.3748/wjg.v25.i34.5162
19. Yu H, Qu F, Shan B, et al. Detection of the mcr-1 Colistin Resistance Gene in Carbapenem-Resistant Enterobacteriaceae from Different Hospitals in China. *Antimicrob Agents Chemother*. 2016;60(8):5033–5035. doi:10.1128/AAC.00440-16
20. Zhang R, W CE, Zhou H, et al. Prevalence and genetic characteristics of carbapenem-resistant Enterobacteriaceae strains in China. *Lancet Infect Dis*. 2017;17(3):256–257. doi:10.1016/S1473-3099(17)30072-5
21. CHINET.Available from: https://www.chinets.com/Document/Index. Accessed Feb 11, 2022.
22. Codjoe FS, Donkor ES. Carbapenem Resistance: a Review. *Med Sci*. 2017;6(1):1. doi:10.3390/medsci6010001
23. Al-Hadeedi S, Fan ST, Leaper D. APACHE-II score for assessment and monitoring of acute pancreatitis. *Lancet*. 1989;2(8665):738. doi:10.1016/S0140-6736(89)90795-2
24. Liu Z, Dumville JC, Norman G, et al. Intraoperative interventions for preventing surgical site infection: an overview of Cochrane Reviews. *Cochrane Database Syst Rev*. 2018;2(2):Cd012653. doi:10.1002/14651858.CD012653.pub2
25. Mayumi T, Okamoto K, Takada T, et al. Tokyo Guidelines 2018: management bundles for acute cholangitis and cholecystitis. *J Hepato-Biliary-Pancreatic Sci*. 2018;25(1):96–100. doi:10.1002/jhbp.519
26. Du M, Suo J, Liu B, et al. Post-ERCP infection and its epidemiological and clinical characteristics in a large Chinese tertiary hospital: a 4-year surveillance study. *Antimicrob Resist Infect Control*. 2017;6(131). doi:10.1186/s13756-017-0290-0.
27. Muscarella LF. Risk of transmission of carbapenem-resistant Enterobacteriaceae and related "superbugs" during gastrointestinal endoscopy. *World J Gastrointestinal Endoscopy*. 2014;6(10):457–474. doi:10.4253/wjge.v6.i10.457
28. Sader HS, Carvalhaes CG, Arends SJR, et al. Aztreonam/avibactam activity against clinical isolates of Enterobacterales collected in Europe, Asia and Latin America in 2019. *J Antimicrob Chemother*. 2021;76(3):659–666. doi:10.1093/jac/dkaa504