Drug resistance of healthcare-associated pathogenic bacteria and carbapenem-resistant *Acinetobacter baumannii* homology in the general intensive care unit

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Background: The objective of this study was to understand the distribution and drug resistance of healthcare-associated infection (HAI) pathogens in an intensive care unit (ICU) of a general tertiary hospital in Inner Mongolia, and to classify carbapenem-resistant *Acinetobacter baumannii* (CR-AB) in ICU patients and environmental samples. Additionally, this study aimed to provide scientific evidence for the use of clinical antibiotics and effective prevention and control measures for CR-AB outbreak.

Methods: The distribution and drug resistance of pathogens isolated from patient's samples in the ICU of 12 Hospitals from January to May 2019 were retrospectively analyzed. Meanwhile, CR-AB isolated from patients and environmental samples were collected and classified by pulsed-field gel electrophoresis (PFGE).

Results: The pathogens isolated from ICU samples, mainly Gram-negative bacteria (63.07%), were CR-AB, *Klebsiella pneumoniae*, and *Pseudomonas aeruginosa*; the main Gram-positive bacteria (22.13%) were *Enterococcus faecium* and *Staphylococcus aureus*; and fungi accounted for the remaining (14.80%). The samples mainly came from sputum (41.09%). Among non-fermenting bacteria, the resistance rates of CR-AB to piperacillin, piperacillin/tazobactam, and other treatments were higher than those of *Pseudomonas aeruginosa* (P<0.05). Meanwhile, the resistance rates to ampicillin/sulbactam and compound sulfamethoxazole were lower than those of *Pseudomonas aeruginosa* (P<0.05). The resistance rates of *Klebsiella pneumoniae* to piperacillin/tazobactam, ceftazidime, and others were higher than those of *Escherichia coli* (P<0.05). Among Gram-positive bacteria, the resistance rates of *Enterococcus faecium* to erythromycin, clindamycin, and other treatment were higher than those of *Staphylococcus aureus* (P<0.05). A total of 62 bands were obtained from 63 strains of CR-AB by electrophoresis. Also, 16 clusters (A-P) were obtained with a 74% similarity coefficient, among which K, L, and N types (more than 9 strains) were more common.

Conclusions: Gram-negative bacteria were the primary pathogens of HAI in the ICU, and their drug resistance was serious. There is homology in the PFGE typing of CR-AB. Therefore, hospitals should strengthen the surveillance of drug-resistant pathogenic bacteria. Additionally, further cleaning and disinfection measures are needed to improve environmental hygiene and prevent outbreaks of HAI.

Keywords: Carbapenem-resistant *Acinetobacter baumannii* (CR-AB); healthcare-associated infection (HAI); intensive care unit (ICU); molecular typing

Introduction

Healthcare-associated infection (HAI) is a threat to medical quality in most healthcare facilities in both developed and developing countries. The incidence rate of HAI is around 5–10%; therefore, controlling and reducing HAI has become a significant global public health question (1,2). Since patients in an intensive care unit (ICU) have serious conditions like chronic disease, they undergo invasive operations and have low immunity, and are therefore more likely to contract HAI than other patients. One study showed that the risk of HAI in the ICU is 3 times greater than that in the ordinary medical unit (3), and the ICU is a critical department in HAI surveillance (4). *Acinetobacter baumannii* (CR-AB) is a Gram-negative bacillus and has proven to be the main pathogen causing HAI. CR-AB is the most common non-fermentative bacteria which causes HAI. Its drug resistance has become increasingly severe, and is highly likely to be the main cause of HAI outbreak (5,6). CR-AB infection results in it being widely distributed in the environment of the hospital. It can further infect different sites like the urinary tract, wounds, endocardium, lungs, and blood, after longer lengths of stay (7). Carbapenem-resistant CR-AB, the most common kind of multi-drug resistant CR-AB, can easily spread and will cause intractable infection. According to an American report, 49.5% of CR-AB cases are carbapenem-resistant. CR-AB has a significant relationship with CR-AB infection death (adjusted OR: 2.49, 95% CI: 1.61–3.84). Several countries like France, Germany, Greece, and Italy have reported CR-AB outbreaks. HAI pathogen distribution and drug resistance in general ICUs of the Inner Mongolia region were detected from January to May 2019 in 12 tertiary general hospitals. In total, 63 strains of non-repetitive CR-AB were isolated from ICU patients, healthcare workers (HCW), and the environment. Pulsed-field gel electrophoresis (PFGE) and homology analysis of the 63 strains were completed to provide evidence for CR-AB infection treatment and prevention.

Method

Bacteria strain source

(I)   Clinical sample: Phlegm, blood, urine, bile, and drainage from the central venous catheter (CVC), wound surface, enterocoelia, and thoracic cavity were collected from HAI patients from the general ICU in 12 general tertiary hospitals from January 1, 2019, to May 31, 2019. Repeated bacteria strains from the same site of the same patient were removed, while any CR-AB strains were kept.

(II) Environment sample: CR-AB strains from the ICU environment, object surface, air, and the hands of healthcare workers were collected during the same time period mentioned above.

(III) Quality control sample: *Staphylococcus aureus* ATCC 25923, *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853, *Streptococcus pneumoniae* ATCC 49619, and *Haemophilus influenzae* ATCC 49247 were provided by the laboratory department in Inner Mongolia People's Hospital. PFGE was conducted using standard a salmonella strain (H9812) as a quality control strain, and was provided by the Inner Mongolia Center for Disease Control.

(IV) Sample source: Samples were collected from 12 general tertiary hospitals distributed in 11 cities in Inner Mongolia Autonomous Region, covering almost all areas in the Inner Mongolia Autonomous Region. Inner Mongolia is found in the north part of China, and is the third-largest province in China. The east–west distance of Inner Mongolia is nearly 1,700 km.

Main reagent

The main reaguens used were Mueller-Hinton (MH), Salmonella Shigella (SS), Columbia blood, and Haemophilus selective chocolate agar culture medium; biphase blood culture; blood pathogen culture flask; Gram stain; identification and antimicrobial susceptibility test reagent; antibiotic susceptibility test discs; agarose (Seakem Gold Agarose); protease K; and restriction enzyme Xba I, Apa I, and GelRed.

Main device

The main devices used were blood culture system, Densicheck, microbial detection system, PFGE, and gel imaging system.

Strain identification and antimicrobial susceptibility test

A dominant bacteria strain from the culture medium was chosen according to the growth shape. VITEK2-compact Microbial detection system was used to conduct strain identification and antimicrobial susceptibility tests by matching reagents. Some individual strains were tested using an antibiotic disc to conduct a Kirby-Bauer (K-B)

antimicrobial susceptibility test. Judgments for the quality control of the results were according to the 2016 recommended criteria of the The Clinical and Laboratory Standards Institute (CLSI) (8).

PFGE procedure

CR-AB strains were isolated from samples of healthcare workers, HAI patients, and the environment to conduct the PFGE test. Intructions for conducting the PFGE procedure were as follows: (I) revive strain (kept at -80 ℃); (II) embed the bacteria in gel and use the corresponding Apa I endonuclease to restrict enzyme digestion on the DNA site; (III) change the small-sized gel block, including the DNA fragment, into a bigger sized gel block using 1.5% agarose gel; (IV) place a bigger sized gel block into 0.5 × TBE buffer solution to conduct PFGE; (V) dye, rinse, and image the gel block for observation and preservation.

Result determination

The National Molecular Subtyping Network for Bacterial Pathogens Surveillance (PulseNet China) software was used to recognize and dispose of the PFGE electrophoretic strip diagram. Electrophoretic strips of the test strain were located and adjusted by H9812 strain molecular weight standard, and manual adjustment was conducted when necessary. Determination of results was done according to the recommendations of Tenover (9) and Talon (10). The similarity coefficient was used to biotype from different strains. When gene homology ≥74%, 2 strains were regarded as the same cluster; when gene homology ≥85%, 2 strains were regarded as the same clone strain.

Statistical analysis

WHONET 5.6 software was used to conduct drug resistance analysis of the main pathogen bacteria, Excel 2010 was used for data statistics, and SPSS22.0 was used to conduct data analysis. Ratio (%) was used to express enumeration data, χ^2 tests or Fisher's exact test were used to comparison among groups, and P values <0.05 were regarded as statistically significant. PulseNet China software was used to analyze the PFGE image.

Ethical approval

The study was approved by the institutional ethics committee/ethics board of Inner Mongolia People's Hospital (No. 202000203L). Since this study is not involve any patient's personal information and the samples used in this study were after used samples from hospital lab, informed consent is not require in this study.

Result

Type and distribution of pathogen bacteria

A total of 696 strains of pathogen bacteria were isolated from HAI patients in the ICU of 12 general tertiary hospitals in the Inner Mongolia region from January to May 2018. Among them were included 89 strains from Inner Mongolia People's Hospital, 79 strains from Second Hospital of Chifeng, 67 strains from the First Affiliated Hospital of Baotou Medical College, 57 strains from the Second Affiliated Hospital of Baotou Medical College, 77 strains from Tongliao City Hospital, 75 strains from the Central Hospital of Wulanchabu, 53 strains from Bayannur Hospital, 26 strains from Ordos Central Hospital, 21 strains from the People's Hospital of Wuhai, 23 strains from the Hospital of XilinGol, 52 strains from the People's Hospital of Hinggan League, and 77 strains from the Inner Mongolia Forestry General Hospital. Among them, gram-negative bacteria constituted 63.07% (439/696) of the total, and the main bacteria were CR-AB, *Klebsiella pneumoniae*, and *Pseudomonas aeruginosa*; gram-positive bacteria constituted 22.13% (154/696) of the total, and the main bacteria were *Enterococcus faecium* and *Staphylococcus aureus*; fungus constituted 14.80% (103/696) of the total, and the main strain was *Candida albicans*. The top 6 most common HAI bacteria were CR-AB, *Klebsiella pneumonia*, *Pseudomonas aeruginosa*, *Enterococcus faecium*, *Escherichia coli*, and *Staphylococcus aureus* (*Table 1*).

In total, 696 strains of pathogen bacteria were detected from sputum samples (41.09%, 286/696), midstream urine samples (23.13%, 161/696), blood samples (17.82%, 124/696), CVC samples (4.60%, 32/696), secretion samples (4.31%, 30/696), and others (*Table 2*).

Drug resistance of major pathogenic bacteria

Drug resistance of CR-AB and Pseudomonas aeruginosa
CR-AB had low drug resistance to levofloxacin (39.53%) and high drug resistance (above 80%) to other known antibiotics. CR-AB showed pan-drug resistance to piperacillin, ampicillin, cefuroxime, cefazolin, cefotetan,

1548

Liu et al. Drug resistance of HAI pathogens and CR-AB homology in ICU

Table 1 Distribution and composition ratio of pathogenic bacteria in the ICU of 12 tertiary hospitals (%)

Pathogenic bacteria	Strain	Composition ratio (%)
Gram-negative bacteria (G-)	439	63.07
Acinetobacter baumannii	172	24.71
Klebsiella pneumoniae	78	11.21
Pseudomonas aeruginosa	72	10.34
Escherichia coli	48	6.90
Enterobacter cloacae	19	2.73
Proteus mirabilis	18	2.59
Stenotrophomonas maltophilia	10	1.44
Others	22	3.16
Gram-positive bacteria (G-)	154	22.13
Enterococcus faecium	68	9.77
Staphylococcus aureus	47	6.75
Staphylococcushominis	10	1.44
Enterococcus faecalis	8	1.15
Staphylococcus epidermidis	6	0.86
Others	15	2.16
Fungus	103	14.80
Candida albicans	52	7.47
Candida tropicalis	15	2.16
Candida glabrata	13	1.87
Candida	7	1.01
Others	16	2.30
Total	696	100

Table 2 Sources and constituent ratios of pathogenic bacteria in the ICU (%)

Sample source	Strain	Constituent ratio
Sputum	286	41.09
Midstream urine	161	23.13
Blood	124	17.82
Central venous catheter	32	4.60
Secretion	30	4.31
Others	63	9.05

pseudomonas aeruginosa (χ^2 =0.320, P=0.572) (*Table 3*).

Drug resistance of *Klebsiella pneumoniae* and *Escherichia coli*

Klebsiella pneumoniae showed low drug resistance (below 50%) to piperacillin-tazobactam, cefotetan, imipenem, meropenem, amikacin, and tobramycin, but had high (above 60%) drug resistance to other antibiotics. It especially showed pan-drug resistance to ampicillin. *Escherichia coli* showed low drug resistance (below 20%) to piperacillin-tazobactam, cefotetan, imipenem, meropenem, amikacin, tobramycin, and macrodantin. *Klebsiella pneumoniae*'s drug resistance rate to piperacillin-tazobactam, ceftazidime, cefotetan, imipenem, meropenem, amikacin, tobramycin, and macrodantin was higher than that of *Escherichia coli* (P<0.05) (*Table 4*).

Drug resistance of *Enterococcus faecium* and *Staphylococcus aureus*

Among all gram-positive bacteria, *Enterococcus faecium* and *Staphylococcus aureus* constituted the largest portion of all bacteria. *Enterococcus faecium* was sensitive to tigecycline and linezolid, and showed pan-drug resistance to ampicillin, penicillin, clindamycin, levofloxacin, ciprofloxacin, and moxifloxacin. Meanwhile, *Staphylococcus aureus* was sensitive to tigecycline, vancomycin, macrodantin, and linezolid, and showed pan-drug resistance to cefoxitin. *Enterococcus faecium*'s drug resistance rate for erythrocin, clindamycin, levofloxacin, moxifloxacin, and macrodantin was higher than that of staphylococcus aureus (P<0.05) (*Table 5*).

Detection result of CR-AB

From January to May 2019, a total of 63 strains of non-

and macrodantin. *Pseudomonas aeruginosa* had low drug resistance (below 10%) to amikacin (5.71%) and tobramycin (5.56%), and showed pan-drug resistance to ampicillin, ampicillin-sulbactam, cefuroxime, cefazolin, ceftriaxone, and cefotetan. CR-AB's drug resistance rate to piperacillin, piperacillin-tazobactam, ceftazidime, cefepime, imipenem, tobramycin, gentamicin, levofloxacin, and ciprofloxacin was higher than that of pseudomonas aeruginosa (P<0.05). Furthermore, CR-AB's drug resistance rate to ampicillin-sulbactam, which are pediatric compound sulfamethoxazole tablets, was lower than that of *Pseudomonas aeruginosa* (P<0.05). Also, CR-AB drug's resistance rate to ceftriaxone had no significant statistical difference compared to that of

Table 3 Comparison of drug resistance between *Acinetobacter baumannii* and *Pseudomonas aeruginosa*

Antibiotics	Acinetobacter baumannii	Pseudomonas aeruginosa	χ^2	P
Piperacillin	100.00	31.94	96.628	<0.001
Piperacillin-tazobactam	96.08	18.06	111.240	<0.001
Ampicillin	100.00	100.00	–	–
Ampicillin-sulbactam	90.12	100.00	4.634	0.031
Ceftazidime	97.67	26.39	144.058	<0.001
Cefepime	97.09	15.28	172.169	<0.001
Cefuroxime	100.00	100.00	–	–
Cefazolin	100.00	100.00	–	–
Ceftriaxone	97.67	100.00	0.320	0.572
Cefotetan	100.00	100.00	–	–
Aztreonam	98.52	–	–	–
Imipenem	95.93	54.17	64.589	<0.001
Meropenem	–	51.39	–	–
Amikacin	–	5.71	–	–
Tobramycin	86.63	5.56	142.647	<0.001
Gentamicin	88.37	5.56	150.965	<0.001
Levofloxacin	39.53	15.28	13.640	<0.001
Ciprofloxacin	97.67	18.06	167.982	<0.001
Macrodantin	100.00	100.00	–	–
Compound sulfamethoxazole	81.98	100.00	11.681	0.001

–, data have not been collected.

repetitive CR-AB bacteria were collected from HAI patients and the environment of 12 ICUs; 46 strains were collected from patient samples (phlegm sample), and 17 strains were collected from environment samples (*Table 6*).

PFGE typing of CR-AB

The 63 strains of CR-AB collected from HAI patient and the environment of 12 ICUs were used to conduct PFGE, and 20–30 strips were generated (*Figure 1*). BioNumerics software was used to analyze PFGE electrophoretograms. Based on the principle of 100% similarity regarded as the same PFGE type, a total of 62 types of strip were generated, and only BM17 and BM30 were 100% the same. A 74% similarity coefficient was used to conduct UPGMA cluster analysis, and resulted in 16 clusters (A–P). Finally, 16 clusters were scatter distributed: K, L, and N types constituted the

majority (more than 9 strains), I type included 5 strains, and other types only included 1–3 strains. Based on the Tenover criteria which states that >85% similarity in strips should be regarded as the same strain, we can conclude from cluster analysis (*Figure 2*) that many strains were the same. For example, BM39 and BM41 were from the same patient sample from the Central Hospital of Wulanchabu, while BM49 and BM50 were from the patient phlegm sample and surface sample of a ventilator, respectively.

Discussion

HAI is an infection caused by pathogenic microorganisms and microorganisms, which usually do not cause harm to human health but eventually result in human infection and clinical symptoms in different ways. Following the increasing amount of newly discovered antibiotics, death

Table 4 Drug resistance of *Klebsiella pneumoniae* and *Escherichia coli*

Antibiotics	*Klebsiella pneumoniae*	*Escherichia coli*	χ^2	P
Piperacillin	92.31	85.42	0.871	0.351
Piperacillin-tazobactam	46.15	4.17	24.870	<0.001
Ampicillin	100.00	91.30	1.617	0.203
Ampicillin-sulbactam	62.86	79.17	2.688	0.101
Ceftazidime	67.95	39.58	9.762	0.002
Cefepime	67.95	64.58	0.151	0.697
Cefuroxime	78.21	87.50	1.720	0.190
Cefazolin	80.77	87.50	0.969	0.325
Ceftriaxone	76.92	85.42	1.348	0.246
Cefotetan	41.03	4.17	20.489	<0.001
Aztreonam	69.23	58.33	1.823	0.177
Imipenem	42.67	2.08	24.555	<0.001
Meropenem	41.03	2.08	23.311	<0.001
Amikacin	35.90	6.52	13.317	<0.001
Tobramycin	42.31	16.67	8.900	0.003
Gentamicin	70.51	58.33	1.961	0.161
Levofloxacin	64.10	68.75	0.285	0.593
Ciprofloxacin	65.38	70.83	0.402	0.526
Macrodantin	66.67	6.25	44.096	<0.001
Compound sulfamethoxazole	70.51	56.25	2.660	0.103

caused by HAI has decreased sharply. Nevertheless, multi-drug resistant organisms (MDRO) have subsequently raised a considerable challenge to healthcare workers. Thus, we need to understand the local distribution of pathogens in the ICU and their drug resistance in order to choose better clinical antibiotics. According to the result of this study, gram-negative bacteria constituted the majority of the 696 strains of pathogen bacteria detected from the ICU in 12 general tertiary hospitals from January to May 2019. Non-fermentative CR-AB had the highest detection rate, which is the same as a result from the China Antimicrobial Resistance Surveillance System (11-12). Non-fermentative bacteria are conditioned pathogens that generally exist in the environment. Non-fermentative bacteria can form colonies in humans with no clinical symptoms, and can turn into pathogenic bacteria when immunity decreases.

The sputum sample constituted 41.09% of the sample total, which shows that the respiratory tract is the most common infection site in the ICU, which is similar to other findings in China (13). A possible reason for this could be that the critical condition of the patients in ICU may hve compromised consciousness, trachea intubation, tracheostomy, ventilator usage, venipuncture, or sputum aspiration damaging mucous epithelia of the respiratory tract; all contributing to declined immune function. For the resons outlined above, it is easier for bacteria to adhere to and colonize the respiratory tract (14). The 6 most common bacteria for HAI patients in the ICU were CR-AB, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Enterococcus faecium*, *Escherichia coli*, and *Staphylococcus aureus*; this composition differed from that in Chen's survey (15), while the ICU bacteria distribution differed from that of the other regions in China.

CR-AB and *Pseudomonas aeruginosa* are non-fermentative gram-negative bacteria. CR-AB can exist in the skin, respiratory tract, and urinary tract of healthy people.

Table 5 Drug resistance of *Enterococcus faecium* and *Staphylococcus aureus*

Antibiotics	*Enterococcus faecium*	*Staphylococcus aureus*	χ^2	P
Ampicillin	100.00	–	–	–
Oxacillin	–	95.56	–	–
Penicillin	100.00	97.78		0.402
Gentamycin	60.26	77.78	3.759	0.053
Cefoxitin	–	100.00	–	–
Streptomycin	78.46	–	–	–
Tetracycline	72.06	87.23	3.762	0.052
Quetiapine-Dafoe Putin	1.47	2.17		1.000
Tigecycline	0.00	0.00	–	–
Erythromycin	97.06	85.11	3.972	0.046
Vancomycin	1.47	0.00		1.000
Clindamycin	100.00	85.00	5.156	0.023
Levofloxacin	100.00	89.13	5.356	0.021
Ciprofloxacin	100.00	95.65		0.161
Moxifloxacin	100.00	74.47	16.749	0.000
Macrodantin	75.00	0.00	63.340	0.000
Compound sulfamethoxazole	–	14.89	–	–
Linezolid	0.00	0.00	–	–
Rifampicin	–	80.85	–	–

–, data have not been collected.

Table 6 Distribution and constituent ratios of CR-AB strains in samples (%)

Sample	Strain	Constitute ratio (%)
Patient sample	46	73.02
Phlegm	37	58.73
Secretion	3	4.76
Flushing fluid	2	3.17
Others	4	6.35
Environment sample	17	26.98
Around bed	7	11.11
Surface of treatment devices	3	4.76
Hand of healthcare worker	3	4.76
Treatment countertops	2	3.17
Computer keyboard	1	1.59
Nurse countertops	1	1.59

CR-AB, acinetobacter baumannii.

CR-AB showed pan-drug resistance to several kinds of antibiotics in this study. For example, it had resistance to piperacillin, ampicillin, cefuroxime, cefazolin, cefotetan, and macrodantin. Furthermore. It showed a 39.53% drug resistance to levofloxacin, which means levofloxacin is an excellent antibiotic for the clinical treatment of CR-AB infection. One study examined single or combined use of tigecycline in CR-AB infection treatment, which resulted in the appearance of tigecycline resistant strain A and B (16). Another study also showed that CR-AB will be increasingly drug-resistant to carbapenems and other antibiotics. All patients in the ICU need a ventilator or intubation tube, which can easily lead to *Pseudomonas aeruginosa* infection. According to drug sensitivity tests, *Pseudomonas aeruginosa* is sensitive to amikacin and tobramycin, with a drug resistance rate of 5.71% and 5.56%, respectively. Therefore, treatment of *Pseudomonas aeruginosa* infection should potentially first involve piperacillin, piperacillin, tazobactam, and
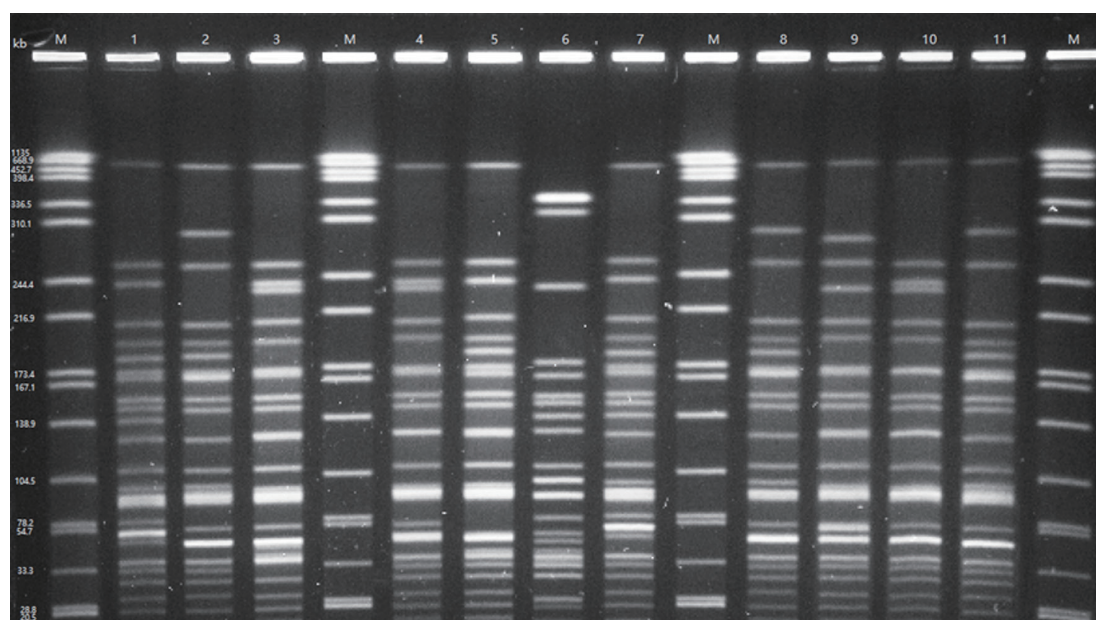
1552

Liu et al. Drug resistance of HAI pathogens and CR-AB homology in ICU

Figure 1 Electrophoresis patterns of partly carbapenem-resistant *Acinetobacter baumannii* strains. M is Marker H9812, and 1-11 is an experimental strain.

ceftazidime and avoid penicillin or cephalosporin. CR-AB, has a more severe condition of drug resistance than *Pseudomonas aeruginosa*, meaning it is more likely to have drug resistance to these kinds of antibiotics. *Klebsiella pneumoniae* showed pan-drug resistance to ampicillin, and lower than 50% drug resistance to piperacillin, tazobactam, cefotetan, imipenem, meropenem, amikacin, and tobramycin; *Escherichia coli* showed high sensitivity to piperacillin, tazobactam, cefotetan, imipenem, meropenem, amikacin, tobramycin, and macrodantin. For the 8 kinds of antibiotics mentioned above, *Klebsiella pneumoniae* had a higher drug resistance than *Escherichia coli*.

Among gram-positive bacteria, *Enterococcus faecium* and *Staphylococcus aureus* constituted the largest portion, which is consistent with a study (17) showing that *staphylococcus aureus* is a common kind of HAI pathogen bacteria, widely distributed in patient's skin and the environment of the inpatient ward. *Staphylococcus aureus* showed pan-drug resistance to cefoxitin, but no drug-resistant strain was found for tigecycline, vancomycin, macrodantin, or linezolid. *Enterococcus faecium*, on the other hand, was sensitive to tigecycline and linezolid but pan-drug resistant to ampicillin, penicillin, clindamycin, levofloxacin, ciprofloxacin, and moxifloxacin. We found that *Enterococcus faecium* had a higher drug resistance than *Staphylococcus aureus* for clindamycin, levofloxacin, moxifloxacin, and

macrodantin.

The 63 strains of CR-AB bacteria collected in this study were used to conduct homology analysis; the PFGE test figure was input into PulseNet China software to obtain a cluster analysis figure. The cluster analysis figure showed that CR-AB is sporadically across the Inner Mongolia region, with a cross-infection occurring between the patient and the environment. To better prevent cross-infection, isolating infected patients, cleaning and disinfecting the environment, and strengthening the hand hygiene of healthcare workers will have a beneficial effect. Especially when considering healthcare worker's low compliance of hand hygiene, the focus should be surveillance and prevention, since hospital cross-infection is mostly caused by contaminated hands. Therefore, it is essential to strengthen the awareness of aseptic operation and focus on training of standard operation for healthare workers. Timely and strict disinfectant medical devices (18), rational use of antibiotics, personal protection of healthcare workers, and medical waste management could also prevent outbreaks and reduce the number of HAI cases (19-21).

Conclusions

In conclusion, HAI pathogenic bacteria in the ICUs of Inner Mongolia have multi-drug resistance. G-bacteria

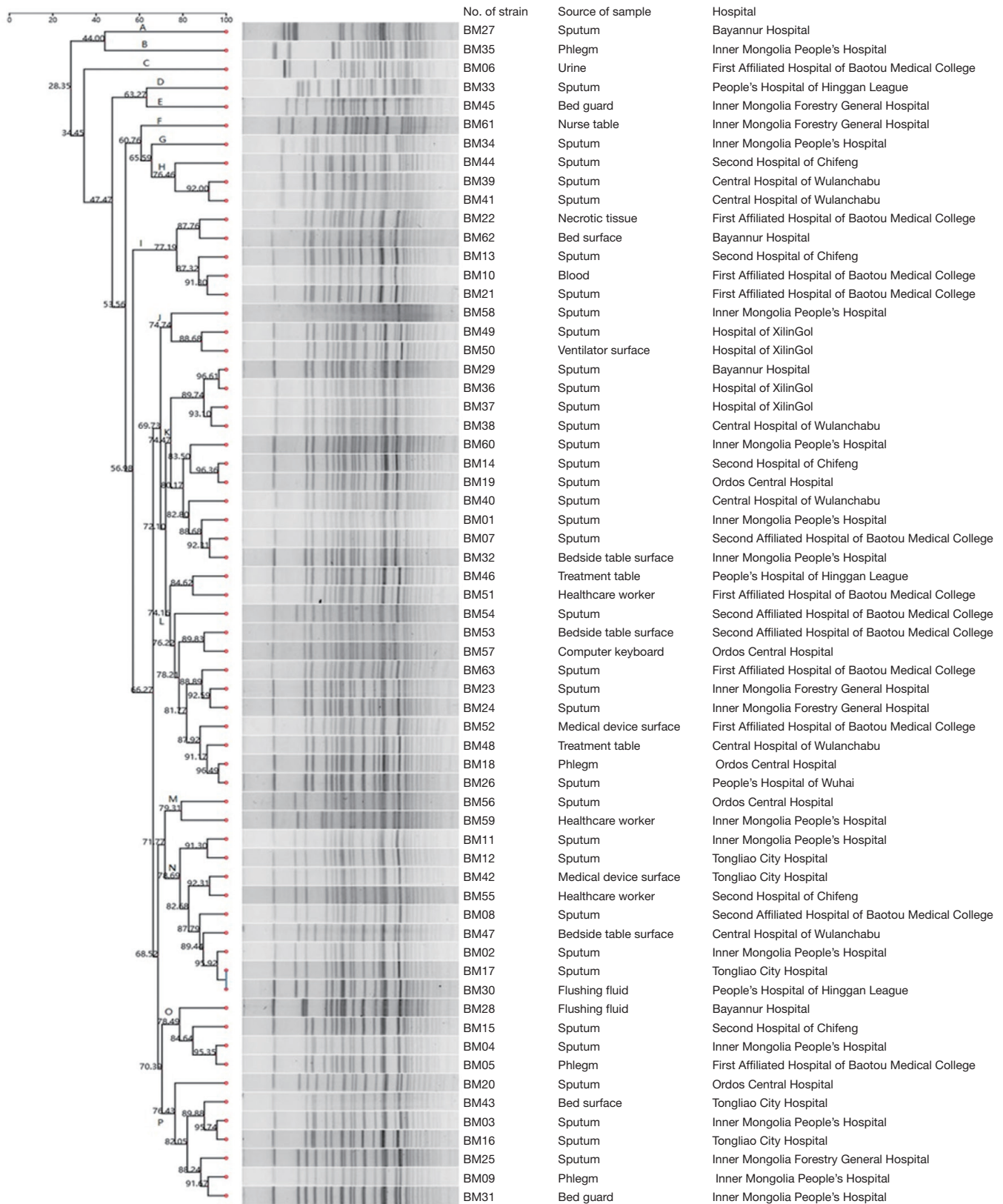| No. of strain	Source of sample	Hospital
BM27	Sputum	Bayannur Hospital
BM35	Phlegm	Inner Mongolia People's Hospital
BM06	Urine	First Affiliated Hospital of Baotou Medical College
BM33	Sputum	People's Hospital of Hinggan League
BM45	Bed guard	Inner Mongolia Forestry General Hospital
BM61	Nurse table	Inner Mongolia Forestry General Hospital
BM34	Sputum	Inner Mongolia People's Hospital
BM44	Sputum	Second Hospital of Chifeng
BM39	Sputum	Central Hospital of Wulanchabu
BM41	Sputum	Central Hospital of Wulanchabu
BM22	Necrotic tissue	First Affiliated Hospital of Baotou Medical College
BM62	Bed surface	Bayannur Hospital
BM13	Sputum	Second Hospital of Chifeng
BM10	Blood	First Affiliated Hospital of Baotou Medical College
BM21	Sputum	First Affiliated Hospital of Baotou Medical College
BM58	Sputum	Inner Mongolia People's Hospital
BM49	Sputum	Hospital of XilinGol
BM50	Ventilator surface	Hospital of XilinGol
BM29	Sputum	Bayannur Hospital
BM36	Sputum	Hospital of XilinGol
BM37	Sputum	Hospital of XilinGol
BM38	Sputum	Central Hospital of Wulanchabu
BM60	Sputum	Inner Mongolia People's Hospital
BM14	Sputum	Second Hospital of Chifeng
BM19	Sputum	Ordos Central Hospital
BM40	Sputum	Central Hospital of Wulanchabu
BM01	Sputum	Inner Mongolia People's Hospital
BM07	Sputum	Second Affiliated Hospital of Baotou Medical College
BM32	Bedside table surface	Inner Mongolia People's Hospital
BM46	Treatment table	People's Hospital of Hinggan League
BM51	Healthcare worker	First Affiliated Hospital of Baotou Medical College
BM54	Sputum	Second Affiliated Hospital of Baotou Medical College
BM53	Bedside table surface	Second Affiliated Hospital of Baotou Medical College
BM57	Computer keyboard	Ordos Central Hospital
BM63	Sputum	First Affiliated Hospital of Baotou Medical College
BM23	Sputum	Inner Mongolia Forestry General Hospital
BM24	Sputum	Inner Mongolia Forestry General Hospital
BM52	Medical device surface	First Affiliated Hospital of Baotou Medical College
BM48	Treatment table	Central Hospital of Wulanchabu
BM18	Phlegm	Ordos Central Hospital
BM26	Sputum	People's Hospital of Wuhai
BM56	Sputum	Ordos Central Hospital
BM59	Healthcare worker	Inner Mongolia People's Hospital
BM11	Sputum	Inner Mongolia People's Hospital
BM12	Sputum	Tongliao City Hospital
BM42	Medical device surface	Tongliao City Hospital
BM55	Healthcare worker	Second Hospital of Chifeng
BM08	Sputum	Second Affiliated Hospital of Baotou Medical College
BM47	Bedside table surface	Central Hospital of Wulanchabu
BM02	Sputum	Inner Mongolia People's Hospital
BM17	Sputum	Tongliao City Hospital
BM30	Flushing fluid	People's Hospital of Hinggan League
BM28	Flushing fluid	Bayannur Hospital
BM15	Sputum	Second Hospital of Chifeng
BM04	Sputum	Inner Mongolia People's Hospital
BM05	Phlegm	First Affiliated Hospital of Baotou Medical College
BM20	Sputum	Ordos Central Hospital
BM43	Bed surface	Tongliao City Hospital
BM03	Sputum	Inner Mongolia People's Hospital
BM16	Sputum	Tongliao City Hospital
BM25	Sputum	Inner Mongolia Forestry General Hospital
BM09	Phlegm	Inner Mongolia People's Hospital
BM31	Bed guard	Inner Mongolia People's Hospital

Figure 2 Cluster analysis of 63 strains of carbapenem-resistant *Acinetobacter baumannii* (ApaI). Sixteen clusters were scatter distributed.

were the primary pathogens of HAI in the ICU, which is similar to findings in other regions (22,23). Therefore, the connection between the HAI management department and the clinical microbiology lab should be strengthened. Healthcare workers should promptly and periodically investigate any pathogenic bacteria of inpatients to dynamically observe drug resistance of ICU pathogen bacteria. Clinical doctors could choose antibiotics according to a drug sensitivity test which could simultaneously prevent a drug-resistant bacteria epidemic. It is necessary to ensure that the right-hand hygiene method is used, while strict adherence to aseptic operation and immediate disinfection of environment and air can effectively reduce cross-infection between patients, further preventing CR-AB outbreak in healthcare facilities.

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Footnote

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Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013). The study was approved by the institutional ethics committee/ethics board of Inner Mongolia People's Hospital (No. 202000203L). Since this study is not involve any patient's personal information and the samples used in this study were after used samples from hospital lab, informed consent is not require in this study.

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