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ORIGINAL RESEARCH

Detection of common mobile genetic elements and genotyping of multidrug-resistant Gram-negative bacilli in blood specimens from septicemia patients in southern China

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Background: Integron, ISCR1 and complex class 1 integrons lead bacteria to become resistant to antibiotic regimens. The aim of this study was to detect common mobile genetic elements of multidrug-resistant Gram-negative bacilli and evaluate the genotyping of these bacilli in blood specimens from septicemia patients in southern China.

Methods: A total of 837 Gram-negative bacilli including 578 strains containing *Enterobacteriaceae* and 259 strains containing non-fermentative bacilli were investigated in blood samples collected from septicemia patients between 2011 and 2014 in southern China. Mobile genetic elements, such as class 1 integrons, the insertion sequence common region 1 (ISCR1), and complex class 1 integrons, were detected from the 837 strains.

Results: Twenty-seven types of gene cassette arrays were found among 837 strains in which 492 (58.8%) class 1 integron-positive isolates and 254 (51.6%) gene cassette-positive isolates were found, including the first description of two types, *aac*A4-bla_{IMP-1}-bla_{OXA-30}-*cat*B3 and *aac*(6′)-II-*aad*A13-*cml*A8-bla_{OXA-10}, in the corresponding species and two gene cassettes, putative helicase and aadA-like, originally detected in integrons. Twelve types of ISCR1-linked resistance gene regions in 196 ISCR1-positive bacilli and seven different types of complex class 1 integron-positive strains were obtained including four distinct complex class 1 integrons that have never been described in any species. Enterobacterial repetitive intergenic consensus (ERIC)-PCR fngerprinting showed that isolates with identical gene profles were clonally unrelated.

Conclusion: Our results indicated that we should pay more attention to enhance the quality of infection control measures and prevent hospital infection, so as to avoid the outbreak of multidrug-resistant Gram-negative bacilli.

Keywords: septicemia, multidrug-resistant, integron, IS*CR1*, complex class 1 integrons

Introduction

Septicemia is a serious medical condition where bacteria present in the blood circulatory system provoke an amplified and dysregulated immune response in the individual.[1] A wide range of Gram-negative bacilli, in which the proportion was high, has been described in septicemia patients whose diagnosis of these infections can be confirmed by blood culture.[2,3] Rapid antibiotic intervention is currently the only way to treat septicemia, which leads more and more bacteria to become resistant to antibiotic regimens, resulting in an urgent health problem worldwide.

Gram-negative bacilli confer high-level resistance to many widely used antibiotics such as β-lactams, tetracycline, aminoglycoside, and even carbapenems which can be encoded by genes that are transferable between bacteria. The rapid dissemination of

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antibiotic resistance genes by horizontal gene transfer is a major cause for concern in Gram-negative bacilli. Recent studies have shown that mobile genetic elements, including integrons, IS*CR*, which can carry one or more resistance gene cassettes in internal variable region, play an important role in the development and dissemination of antibiotic resistance.[4] Integrons, IS*CR*, carried on plasmids or chromosome are responsible for the incorporation of antibiotic resistance genes known as gene cassettes inside the mobile genetic elements, and as a result of this integration, they enhance the expression of the gene-conferring resistance to antimicrobials. Complex class 1 integrons, which have been introduced to describe the large genetic elements that associate different class 1 integrons with insertion sequence common region 1 (IS*CR1*), possess two copies 3′-CS and combine the resistance genes between class 1 integrons and IS*CR1*.[5,6] It has now been believed that the element is in association with the ability of bacteria to acquire various resistance genes.[7]

The aim of this study was to detect common mobile genetic elements of multidrug-resistant Gram-negative bacilli and evaluate the genotyping of these bacilli in blood specimens from septicemia patients in southern China.

Materials and methods

Bacterial strains and antimicrobial susceptibility testing

A total of 837 strains were prospectively and consecutively collected from 2011 to 2014 at Nanfang Hospital, a large tertiary-level teaching hospital with 2,200 beds in Guangzhou, southern China. The patients in the hospital come from Guangzhou (30%), other cities in Guangdong province (50%), and nearby province (20%). These strains were collected from blood specimens from diverse units in the hospital, and repeat isolates from the same patients were excluded. Routine biochemical identification and antimicrobial susceptibility testing were carried out using the BD Phoenix 100 Automated Microbiology System (BD, Franklin Lakes, NJ, USA). Susceptibility was determined using the minimum inhibitory concentration (MIC) break-point criteria of the Clinical and Laboratory Standards Institute M100-S27 (2017). Isolates were stored at –80°C in nutrient broth containing 30% (v/v) glycerol. The study was approved by the medical ethics committee of Nanfang Hospital, Southern Medical University, and conducted in compliance with the Declaration of Helsinki. The informed consent was exempted from committee, because of the secondary use of medical records or biological specimens obtained in clinical diagnosis and treatment. The privacy of the patient is respected. The data will be kept confidential and used only for this study.

Bacterial DNA preparation

Total genomic DNA was extracted from stationary-phase broth cultures that were grown overnight in Luria–Bertani broth with the Ezup Column genomic DNA extraction kit (Sangon Biotech., Shanghai, China) according to the manufacturer's instructions.

Detection and sequencing of integrons

A multiple PCR was carried out for the detection of integrase genes located at the conserved regions of class 1 integrons,[8] followed by the amplification of the variable region of integrons.[9] To determine the identical arrays of gene cassettes, same-sized amplicons were digested with *Rsa*I and *Hinf*I (Takara Bio., Tokyo, Japan) restriction enzymes, which were dependent on the species. Amplicons showing the same restriction fragment length polymorphism pattern were deemed to be identical, and one representative product of each distinct RFLP was purified and sequenced. The resulting DNA sequences were analyzed with the Basic Local Alignment Search Tool program at the homepage of the National Center for Biotechnology Information (www.ncbi.nlm.nih.gov/blast/). All the primers used are summarized in Table S1.

Detection and sequencing of ISCR1

All strains were screened for the presence of IS*CR1* by PCR using the primers, IS*CR1*-F and IS*CR1*-R, designed to amplify orf513 segment of IS*CR1*.[10] Moreover, primers, IS*CR1*-F and sul1-R, were used to amplify the IS*CR1*-linked resistance genes.[11] RFLP and sequencing analysis of the IS*CR1*-linked resistance genes were same as the variable region of integrons.

Detection and sequencing of complex class 1 integron

The strains, whose variable region of integrons and IS*CR1*-linked resistance genes were both positive, were considered carrying complex class 1 integron. Primers were designed at variable region of integrons and IS*CR1*-linked resistance genes.[12] Then, PCR was carried out and sequencing was followed.

Enterobacterial repetitive intergenic consensus (ERIC)-PCR analysis

ERIC sequences, present in many diverse eubacterial species, using genomic sequence information obtained primarily from *Escherichia coli* and *Salmonella typhimurium,* can be utilized as efficient primer binding sites in the PCR to produce fingerprints of different bacterial genomes.[13]

ERIC-PCR of bacterial genomes was used to examine the genetic diversity of ESBL-producing *E. coli, Klebsiella pneumoniae*, carbapenem-resistant *Pseudomonas aeruginosa,* and *Acinetobacter baumannii*. PCR products were analyzed on 2% agarose gels stained with ethidium bromide. ERIC-PCR fingerprinting was analyzed by visual examination and clustering analysis based on the Dice similarity coefficient and the unweighted pair group method with arithmetic mean (UPGMA) of strains using Quantity One software (Bio-Rad Laboratories Inc., Hercules, CA, USA).

Results

Detection of integrons

The prevalence of class 1 integron gene varied among different species (Table 1). A total of 492 *intI* gene-positive strains were obtained from 837 bacterial isolates (Table 2; Figure 1), and 254 of the isolates were positive for the variable region and the inserted gene cassette sizes varied in size from 0.6 to 3.0 kb. Gene cassette arrays could be divided into 27 types according to their restriction fragment lengths of the variable region-positive strains.

Detection of IS*CR1*

A total of 349 IS*CR1*-positive strains were obtained from 837 isolates (Table 1). Among them, regions of IS*CR1*-linked resistance genes were detected in 196 IS*CR1*-positive strains (Table 3; Figure 2). The range of detected IS*CR1*-linked resistance gene regions varied in size from 2.0 kb to 5.0 kb. A total of 12 distinct IS*CR1*-linked resistance gene arrangements were observed in the 837 isolates.

Detection of complex class 1 integron

There are 31 complex class 1 integron-positive strains that were obtained, and among them a total of seven distinct resistance gene arrangements were observed from 837 isolates (Figure 3).

ERIC-PCR analysis

ERIC-PCR analysis was performed to determine the genetic relatedness of the 90 *E. coli* and 98 *K. pneumoniae.* ERIC primers generated 1–21 bands, with molecular weights ranging from 0.25 to 5.0 kb. Fingerprinting was analyzed against 81 and 91 genotypes by UPGMA using 80% similarity as a cutoff point. The results of the cluster analysis showed that these isolates were unrelated. Similarly, 44 *A. baumannii* and 40 *P. aeruginosa* also had different genotypes according to the corresponding cluster analysis. ERIC fingerprinting revealed that the isolates were unrelated (Figure 4).

More details about the drug susceptibility of dominating isolates of multidrug-resistant Gram-negative bacilli are summarized in Table S2.

Discussion

The current study characterized integrons and their gene cassettes. A high frequency of class 1 integrons was observed

Table 1 Number of common mobile genetic elements in multidrug-resistant Gram-negative bacilli

Bacterial species	No. of isolates	No. of *intI1* gene-positive isolates (%)[a]	No. of gene cassette-positive isolates (%)[b]	No. of isolates containing IS*CR1* (%)[c]	No. of isolates containing IS*CR1*-linked genes (%)[d]
Escherichia coli	182	168 (92.3)	82 (48.8)	53 (29.1)	31 (58.5)
Klebsiella pneumoniae	204	144 (70.6)	70 (48.6)	133 (65.2)	86 (64.7)
Klebsiella oxytoca	18	7 (38.9)	3 (42.9)	5 (27.8)	1 (20.0)
Enterobacter cloacae	63	21 (33.3)	5 (23.8)	36 (57.1)	18 (50.0)
Enterobacter aerogenes	52	17 (32.7)	6 (35.3)	23 (44.2)	11 (47.8)
Proteus mirabilis	14	6 (42.9)	3 (50.0)	2 (14.3)	1 (50.0)
Serratia marcescens	12	5 (41.7)	2 (40.0)	1 (8.3)	1 (100.0)
Salmonella typhi	15	5 (33.3)	2 (40.0)	0	–
Morganella morganii	5	1 (20.0)	1 (100)	0	–
Citrobacter freundii	13	2 (15.4)	2 (100)	3 (23.1)	1 (33.3)
Acinetobacter baumannii	89	60 (67.4)	48 (80.0)	54 (60.7)	32 (59.3)
Acinetobacter spp.	15	5 (33.3)	2 (40.0)	2 (13.3)	1 (50.0)
Pseudomonas aeruginosa	82	35 (42.7)	17 (48.6)	34 (41.5)	12 (35.3)
Pseudomonas putida	12	3 (25.0)	2 (66.7)	3 (25.0)	1 (33.3)
Burkholderia cepacia	19	4 (21.1)	3 (75.0)	0	–
Achromobacter spp.	8	4 (50.0)	2 (50.0)	0	–
Stenotrophomonas maltophilia	34	5 (14.7)	4 (80.0)	0	–

Notes: [a]Number of *intI1* gene-positive isolates/total no. of isolates. [b]Number of gene cassette-positive isolates/no. of *intI1* gene-positive isolates. [c]Number of isolates containing IS*CR1*/total no. of isolates. [d]Number of isolates containing IS*CR1*-linked gene/no. of isolates containing IS*CR1*.
Abbreviation: IS*CR1*, insertion sequence common region 1.

Table 2 Characterization of class 1 integron in multidrug-resistant Gram-negative bacilli

Bacterial species (N)	Gene cassette array	No. of isolates (%)[a]
Escherichia coli (82)	*dfr*A27; *dfr*A17-*aad*A5	4 (4.9)
	*aad*B-*aad*A2	4 (4.9)
	*dfr*A12-*orf*F-*aad*A2	1 (1.2)
	*dfr*A17-*aad*A5	62 (75.6)
	*aac*A4-*cml*A1	2 (2.4)
	*dfr*A5	4 (4.9)
	*aad*A1	2 (2.4)
	*dfr*A1-*orf*C	3 (3.7)
Klebsiella pneumoniae (70)	*dfr*A17-*aad*A5	58 (82.9)
	*dfr*A12-*orf*F -*aad*A2	1 (1.4)
	*aac*A4-*cat*B8-*aad*A1	1 (1.4)
	*dfr*A1-*aad*A5	3 (4.3)
	*dfr*A5	2 (2.9)
	*dfr*A15; *aad*B-*aad*A2	3 (4.3)
	*aac*A4-*cat*B8-*aad*A1	2 (2.9)
Klebsiella oxytoca (3)	*dfr*A17-*aad*A5	3 (100.0)
Enterobacter cloacae (5)	*dfr*A17-*aad*A5	2 (40.0)
	*aad*B-*orf*1-*cml*A1	2 (40.0)
	*dfr*A5	1 (20.0)
Enterobacter aerogenes (6)	*dfr*A17-*aad*A5	3 (50.0)
	*dfr*A1-*orf*C	1 (16.7)
	*dfr*A25	2 (33.3)
Proteus mirabilis (3)	*dfr*A17-*aad*A5	2 (66.7)
	*aad*B-*aad*A2	1 (33.3)
Serratia marcescens (2)	*dfr*A12-*orf*F-*aad*A2	2 (100.0)
Salmonella typhi (2)	*dfr*A17-*aad*A5	2 (100.0)
Morganella morganii (1)	*dfr*A1-*aad*A5	1 (100.0)
Citrobacter freundii (2)	*dfr*A1-*orf*F	2 (100.0)
Acinetobacter baumannii (48)	*dfr*A12-*orf*F-*aad*A2	7 (14.6)
	*dfr*A1-*aad*A5	21 (43.8)
	*dfr*A1-*orf*F	9 (18.8)
	*dfr*A12-*orf*F-*aad*A2-*orf*II-*orf*III	2 (4.17)
	*aac*A4-*cat*B8-*aad*A1	9 (18.8)
Acinetobacter spp. (2)	*dfr*A15	2 (100.0)
Pseudomonas aeruginosa (17)	*aad*B-bla_{PSE-1}	7 (41.2)
	*aac*A4-bla_{IMP-1}-bla_{OXA-30}-*cat*B3	1 (5.9)
	*aac*A4-*cat*B8	4 (23.5)
	bla_{VIM-2}-*aac*A4	1 (5.9)
	arr-3-*aac*A4	4 (23.5)
Pseudomonas putida (2)	*cat*B8-*aad*A1	2 (100.0)
Burkholderia cepacia (3)	*dfr*A14-*aac*A4	2 (66.7)
	*aad*A1-*aac*A6	1 (33.3)
Achromobacter spp. (2)	*aac*(6′)-II-*aad*A13-*cml*A8-bla_{OXA-10}	1 (50.0)
	bla_{IMP-1}-*aac*A6-*aad*A4	1 (50.0)
Stenotrophomonas maltophilia (4)	*aac*A7-bla_{VIM-2}-*aac*A4	1 (25.0)
	*aad*B-*aac*(6′)-II-bla_{PSE-1}	3 (75.0)

Note: [a]Number of gene cassette-positive isolates/no. of *intI1* gene-positive isolates.

Table 3 Characterization of IS*CR1*-linked gene in multidrug-resistant Gram-negative bacilli

Bacterial species (N)	IS*CR1*-linked gene	No. of isolates (%)[a]
Escherichia coli (31)	$bla_{CTX-M-9}$+ *ins*B	31 (100.0)
Klebsiella pneumoniae (86)	*qnr*A1+ *amp*R	66 (76.7)
	*aac*A6+ arr-3	11 (12.8)
	*dfr*A12+ *aad*A2	9 (10.5)
Klebsiella oxytoca (1)	*aac*A6+ arr-3	1 (100.0)
Enterobacter cloacae (18)	$bla_{CTX-M-9}$+ *ins*B	18 (100.0)
Acinetobacter baumannii (32)	*arr*-3+ *dfr*A16+ *aad*A2	4 (12.5)
	*aad*B+bla_{PSE-1}	11 (34.4)
	DHA1+ *amp*R	4 (12.5)
	bla_{PSE-1}+ GST + ABC	12 (37.5)
	*dfr*A10	1 (3.1)
Acinetobacter spp. (1)	*sap*A-like+ *qnr*B2	1 (100.0)
Pseudomonas aeruginosa (12)	*aac*A6+ arr-3	10 (83.3)
	reductase + *qnr*B6	2 (16.7)
Pseudomonas putida (1)	DHA1+ *amp*R	1 (100.0)

Note: [a]Number of gene cassette-positive isolates/no. of *intI1* gene-positive isolates.
Abbreviation: IS*CR1*, insertion sequence common region 1.

among multidrug-resistant Gram-negative bacteria, which corroborates well with a previous report.[14,15] According to these results, most isolates carrying *intI1* genes contained gene cassettes. However, a few isolates among the *intI1*-positive strains did not contain gene cassettes. The main reasons may be the following: defects or mutations at the 3′-CS; gene cassette array in novel complex or unusual class 1 integrons; or the variable region was too long to be amplified. Of 27 different gene cassettes detected from 837 isolates, the most common type of gene cassettes included aminoglycoside resistance (*aad*A1, *aad*A2, *aad*A5, *aad*A24, *aad*A-like, *aad*B, *aac*(6′)-II, *aac*A4, and *aph*A15), followed by β-lactamase (bla_{OXA-10}, bla_{OXA-21}, bla_{PSE-1}, and bla_{VIM-1}), trimethoprim resistance (*dfr*A1, *dfr*A14, *dfr*A17, and *dfr*6), and chloramphenicol resistance (*cat*, *cat*B2, *cat*B3, and *cat*B-like). Two gene cassettes (putative helicase and *aad*A-like) were first detected in integrons, indicating that integrons can efficiently capture and integrate genes. To the best of our knowledge, the resistance gene arrangement, *aac*6-*aad*A13-*cml*A8-*aad*A1, was reported for the first time in all bacteria, indicating that the resistance gene arrangement may disseminate between different bacteria and produce new gene arrangement.

According to the results, of the 837 strains, 196 isolates contained IS*CR1*-linked resistance genes among IS*CR1*-positive strains. Eleven genes (*qnr*A1, *qnr*B2, *qnr*B6, bla_{DHA-1}, *amp*R, bla_{PER-1}, *ins*B, *sap*A-like, GST, ABC transporter, and short-chain dehydrogenase/reductase) were detected in 12 distinct IS*CR1*-linked resistance gene arrays.

A total of seven distinct complex class 1 integron gene arrangements with a different gene cassette variable region composed of the 5′-CS and the 3′-CS. Enterobacteriaceae strains carrying complex class 1 integrons are becoming more

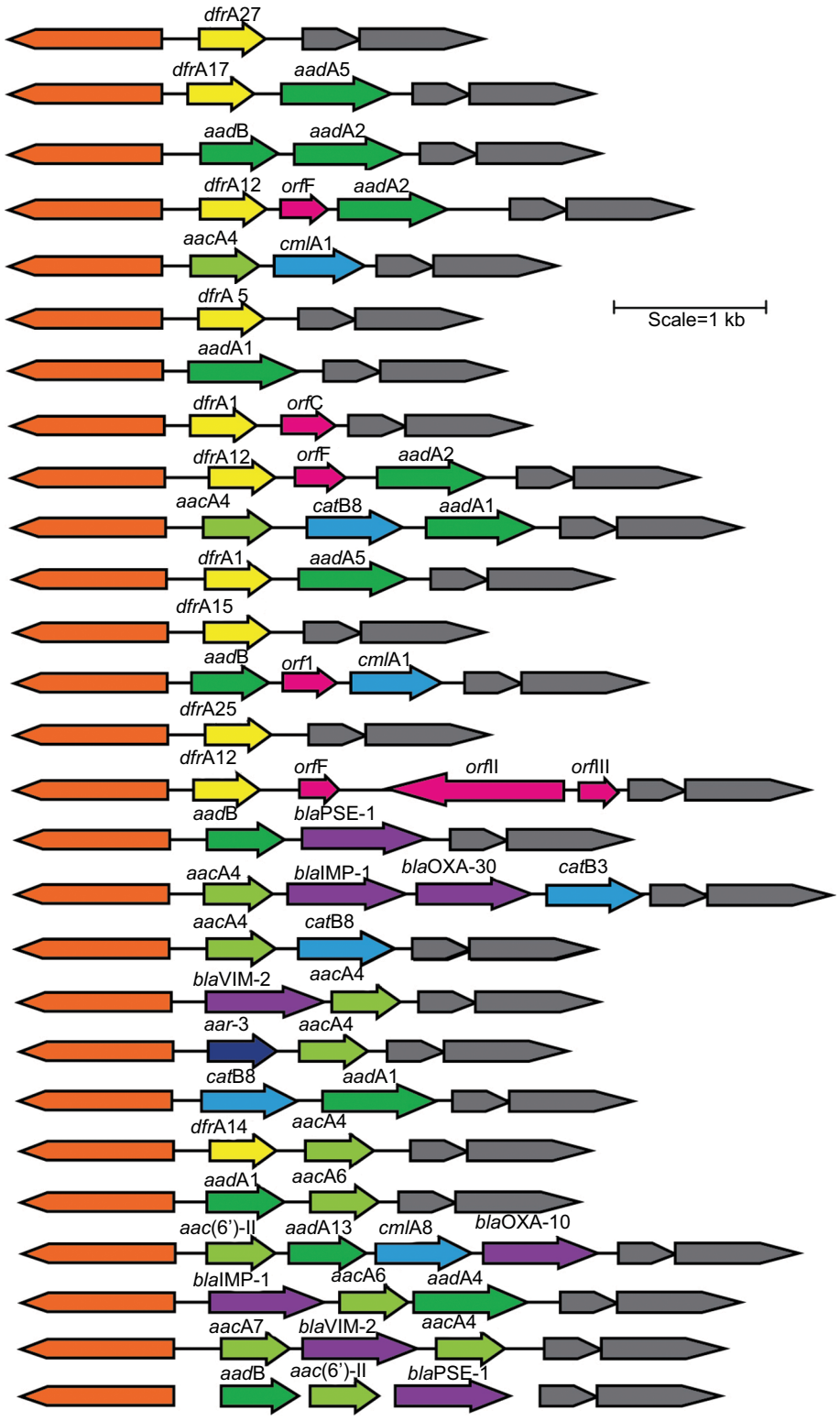

Figure 1 Twenty-seven distinct class 1 integron resistance gene arrangements.
Notes: Integrase is represented by orange boxes. qacEΔ1/sul1 is represented by gray boxes.

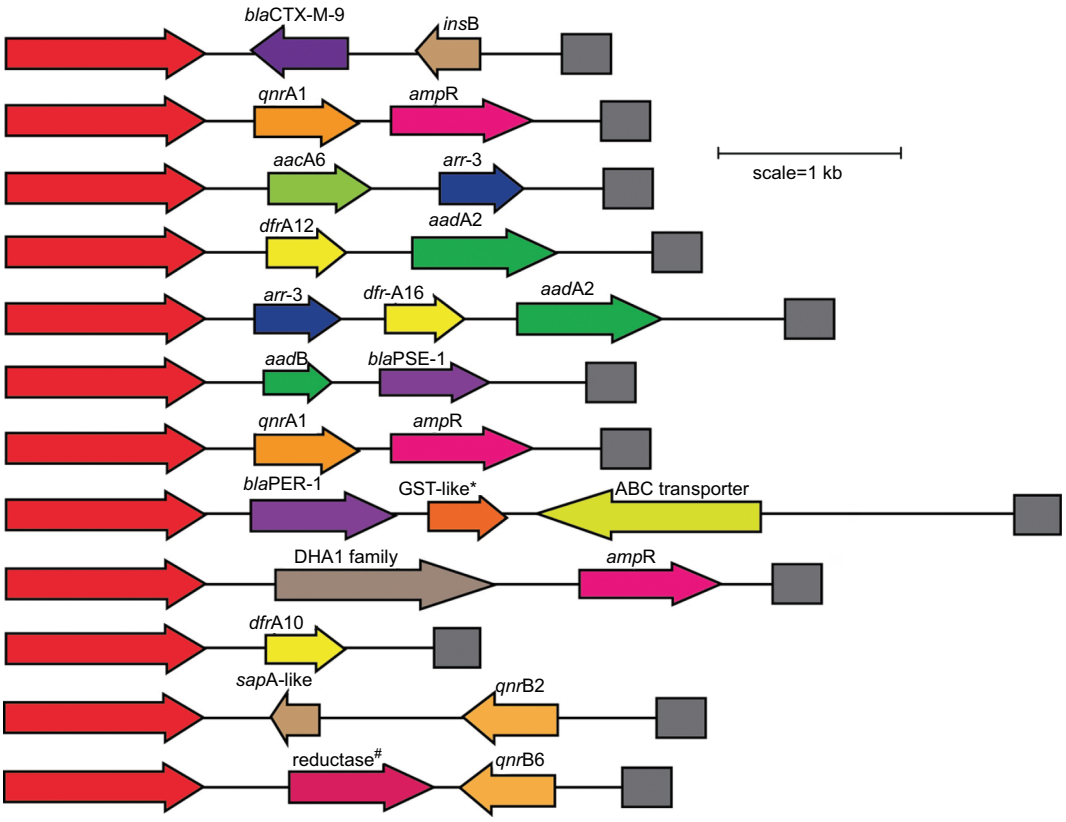

Figure 2 Twelve distinct IS*CR1*-linked resistance gene arrangements.
Notes: IS*CR1* is represented by red boxes. #Dehydrogenase/reductase; *first reported.
Abbreviations: GST, glutathione-*S*-transferase; IS*CR1*, insertion sequence common region 1.

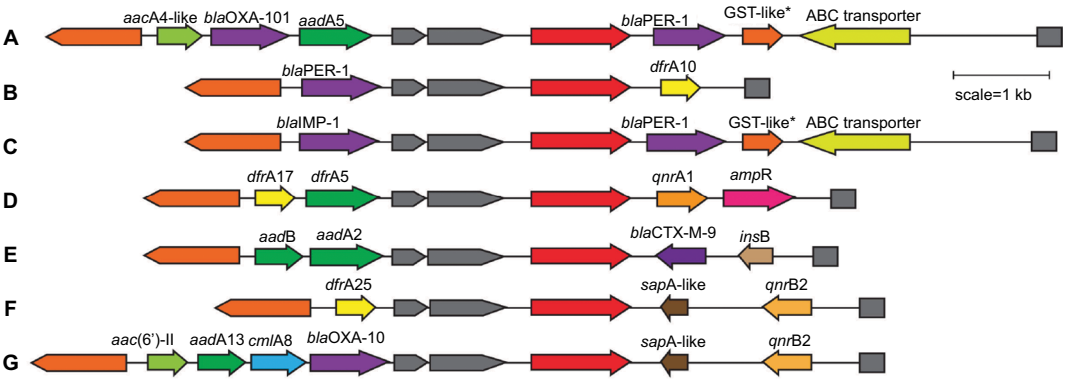

Figure 3 Seven distinct structures connecting the variable regions of the complex class 1 integron structures.
Notes: The types of structures were marked from A to G. Integrase is represented by orange boxes; the qacEΔ1/sul1 is represented by gray boxes; IS*CR1* is represented by red boxes. *First reported.
Abbreviations: GST, glutathione-*S*-transferase; IS*CR1*, insertion sequence common region 1.

common.[5,10] In this study, two distinct complex class 1 integrons (C and G type) that have never been described before.

Conclusion

The study detected the molecular characteristics of Gram-negative bacilli in blood specimens from septicemia patients in southern China. Among 837 strains, two novel gene cassette arrays were found in 254 class 1 integron-positive isolates and seven different complex class 1 integron-positive strains were obtained in which two were not previously reported. The study of Gram-negative bacilli in blood specimens from hospitalized patients aids in the understanding of the propagation of Gram-negative bacilli, and effective infection control measures are urgently required to control

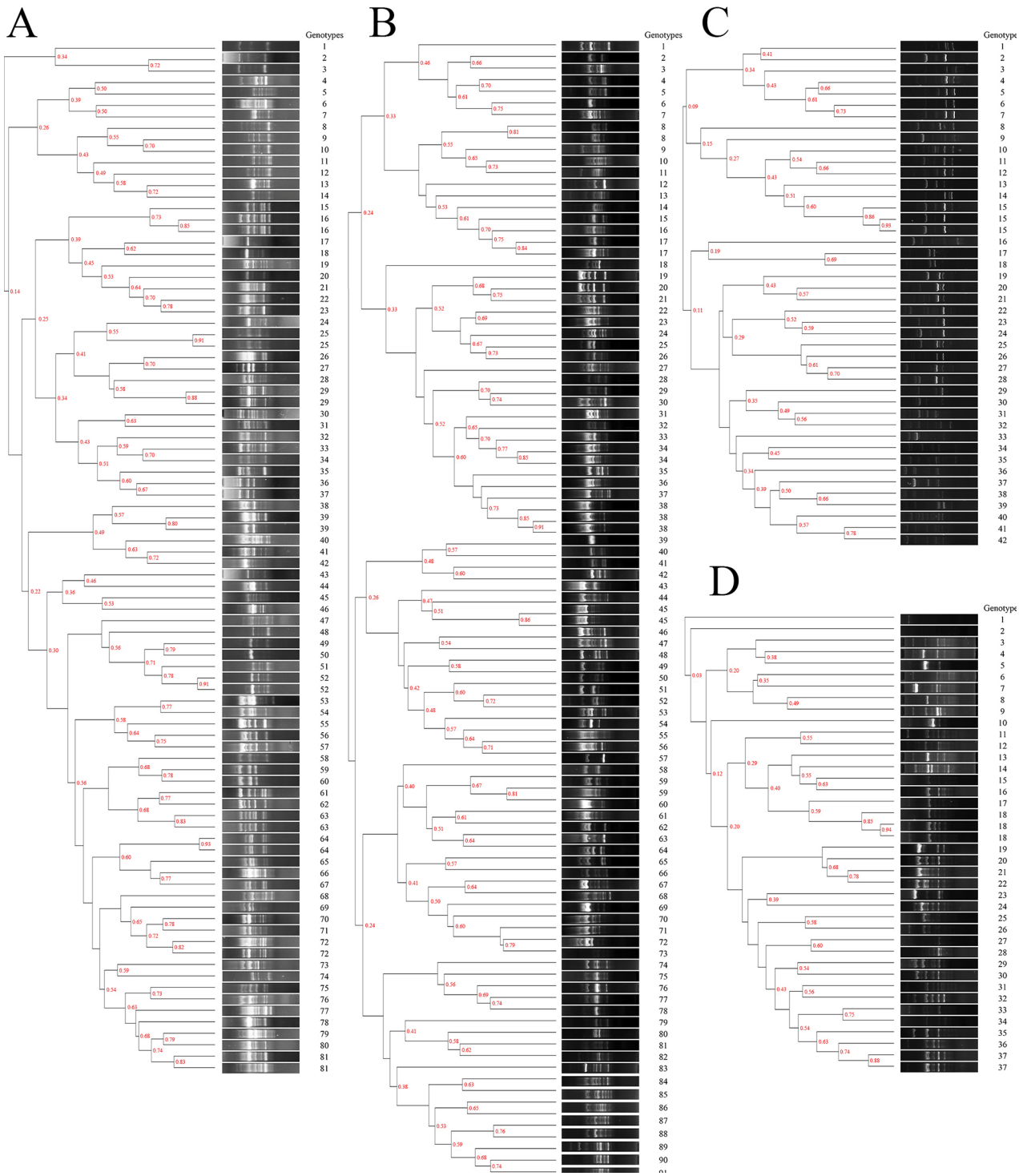

Figure 4 ERIC-PCR profile of (**A**) 90 ESBL-producing *Escherichia coli*, (**B**) 98 ESBL-producing *Klebsiella pneumoniae*, (**C**) 44 carbapenem-resistant *Acinetobacter baumannii* and (**D**) 40 carbapenem-resistant *Pseudomonas aeruginosa*.
Abbreviation: ERIC, enterobacterial repetitive intergenic consensus.

the transmission of Gram-negative bacilli in health care facilities in the country.

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Disclosure

The authors report no conflicts of interest in this work.

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Supplementary materials

Table S1 Primers used in this study

Target	Primer	Sequence (5′–3′)	Reference
intI gene	intI1-F	GCATCCTCGGTTTTCTGG	1
	intI1-R	GGTGTGGCGGGCTTCGTG	
Resistance genes associated with class 1 integrons	5′-CS	GGCATCCAAGCAGCAAG	2
	3′-CS	AAGCAGACTTGACCTGA	
IS*CR1*	ISCR1-F	ATGGTTTCATGCGGGTT	3
	ISCR1-R	CTGAGGGTGTGAGCGAG	
IS*CR1*-linked resistance genes	ISCR1-F	ATGGTTTCATGCGGGTT	4
	sul1-R	AGCCCCATACCTACAAAGCC	
3′CS-IS*CR1*	Pc-F	TATTGCTGAGGCGGACTG	5
	Pc-R	CATTGGAGGAGGTCGTTG	
*aad*A5-bla_{PER-1}	P1-F	ACTGGTCTCATTGCTCCTA	
	P1-R	ATTGGTTCGGCTTGACTC	
bla_{PER-1}-*dfr*A10	P2-F	TGTTGCCTGATGGACG	This study
	P2-R	CTTGATTACCGAATGCTCT	
bla_{IMP-1}-bla_{PER-1}	P3-F	GACGGTAAGGTTCAAGCC	
	P3-R	AGCCCAGGTATTCTGTAAAA	
*aad*A5-*qnr*A1	P4-F	CGTCGTTCTTGCTCTTGC	
	P4-R	TCTTATGGCTGACTTGATTGTAG	
*aad*A2-$bla_{CTX-M-9}$	P5-F	CGTTGCCTTGGTAGGTCC	5
	P5-R	GGTATTCAGCGTAGGTTCAGT	
*dfr*A25-sapA-like	P6-F	ACGAAGCGATGGGTAGA	
	P6-R	TGGGAGGTGCTGGATAA	
bla_{OXA-10}-sapA-like	P7-F	TTCAACAAATCGCCAGAG	This study
	P7-R	CCGCTTAACGCAACC	
ERIC-PCR	ERIC-2	AAGTAAGTGACTGGGGTGAGCG	6

Abbreviation: ERIC, enterobacterial repetitive intergenic consensus.

Table S2 Susceptibility (SIR) pattern of dominating isolates of multidrug-resistant Gram-negative bacilli

Antibiotic	Susceptibility SIR (%)											
	Escherichia coli (n=182)						*Klebsiella pneumoniae* (n=204)					
	ESBL producing (n=90)			ESBL nonproducing (n=92)			ESBL producing (n=98)			ESBL nonproducing (n=106)		
	R	I	S	R	I	S	R	I	S	R	I	S
Amikacin	11.1	0.0	88.9	3.3	0.0	96.7	16.3	0.0	83.7	6.6	0.0	93.4
Amoxicillin/clavulanic	43.3	23.3	33.3	6.5	9.8	83.7	60.2	6.1	33.7	3.8	0.0	96.2
Ampicillin	100.0	0.0	0.0	65.2	0.0	34.8	NR	NR	NR	NR	NR	NR
Ampicillin/sulbactam	60.0	25.6	14.4	15.2	30.4	54.3	92.9	7.1	0.0	7.5	0.0	92.5
Aztreonam	76.7	13.3	10.0	5.4	0.0	94.6	66.3	13.3	20.4	0.0	0.0	100.0
Cefazolin	100.0	0.0	0.0	27.2	0.0	72.8	100.0	0.0	0.0	6.6	0.0	93.4
Cefepime	63.3	13.3	23.3	5.4	0.0	94.6	57.1	9.2	33.7	2.8	0.0	97.2
Cefoperazone/sulbactam	11.1	33.3	55.6	2.2	0.0	97.8	26.5	20.4	53.1	5.7	0.0	94.3
Cefotaxime	100.0	0.0	0.0	3.3	2.2	94.6	76.5	0.0	23.5	6.6	0.0	93.4
Ceftazidime	60.0	6.7	33.3	6.5	0.0	93.5	60.2	0.0	39.8	7.5	0.0	92.5
Ciprofloxacin	73.3	6.7	20.0	25.0	0.0	75.0	33.7	6.1	60.2	11.3	0.0	88.7
Gentamicin	61.1	0.0	38.9	30.4	0.0	69.6	53.1	0.0	46.9	15.1	0.0	84.9
Imipenem	0.0	1.1	98.9	0.0	0.0	100.0	2.0	0.0	98.0	0.0	0.0	100.0
Levofloxacin	73.3	6.7	20.0	19.6	0.0	80.4	26.5	0.0	73.5	6.6	0.0	93.4
Meropenem	0.0	0.0	100.0	0.0	0.0	100.0	1.0	0.0	99.0	0.0	0.0	100.0
Piperacillin	100.0	0.0	0.0	59.8	0.0	40.2	96.9	0.0	3.1	19.8	13.2	67.0
Piperacillin/tazobactam	6.7	0.0	93.3	0.0	0.0	100.0	20.4	6.1	73.5	0.0	0.0	100.0
Tetracycline	73.3	6.7	20.0	59.8	0.0	40.2	53.1	0.0	46.9	13.2	6.6	80.2
Trimethoprim/sulfamethoxazole	70.0	NA	30.0	50.0	NA	50.0	62.2	NA	37.8	22.6	NA	77.4

Abbreviations: R, resistance; I, indeterminate; S, sensitive; NA, not available.

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