

The characteristics of antimicrobial resistance and treatment of carbapenem-resistant Enterobacteriaceae infections in children from 2013 to 2018 in a chinese tertiary hospital

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Abstract

Background The aim of this study was to perform a retrospective analysis of prevalence and treatment of carbapenem-resistant Enterobacteriaceae (CRE) infections in children in a tertiary hospital. **Methods** The non-repeat clinical isolates of CRE in children were collected in Liaocheng People's Hospital from January, 2013 to December, 2018. The bacterial identification and antibiotic susceptibility was performed according to the standard methods. The isolated strains will be detected carbapenemases genotypes and homology analysis. All data on the culture-positive strains and associated clinical infection from different pediatric wards were reviewed. **Results** A total of 20 CRE strains isolated from pediatric patients, which derived from different infection sites and present a classification of multiple species of Enterobacteriaceae. And the production of IMP-type carbapenemase in these strains is the main reason of antimicrobial resistance. Most of the infected patients have severe comorbidities and invasive procedures, and use insensitive drugs due to the high resistance rates and medication restrictions. Nevertheless, most infected children have been treated despite the resistance of pathogens to multiple antimicrobial agents. In our follow-up survey, most children received adjuvant therapy such as human intravenous immunoglobulin, which may be an important factor in helping patients defeat pathogenic bacteria. **Conclusions** This study demonstrates a high prevalence of IMP-mediated CRE infection in pediatric patients with severe comorbidities and invasive procedures. Most children have been cured, which may be related to application of adjuvant therapy and weaker pathogenicity of the IMP-type Enterobacteriaceae.

Background

Enterobacteriaceae is a common pathogen for nosocomial infections and community-acquired infections. In the past decade, carbapenem-resistant Enterobacteriaceae (CRE) have emerged as a significant public health threat and begun to spread worldwide [1, 2]. CRE have been sporadically reported in Asia in recent years [3]. Carbapenem antibiotics have been used as the most effective drugs for the treatment of these severe infections. However, with the widespread use of imipenem and meropenem, CRE is increasing year by year. Due to the multiple drug resistance mechanisms produced by CRE, the selection of antimicrobial agents for the treatment of CRE infection poses great challenges [1, 3, 4]. A study on CRE infection showed that CRE can cause many serious infections, such as pneumonia, intra-abdominal infections, urinary tract infections, and device-related infections. Depending on the treatment, about 600 people die from CRE infection each year and can cause 18% to 48% mortality [5].

In children, CRE infection is an uncommon but very serious problem that may be associated with significant mortality and long-term hospitalization. Children with CRE infections are often very weak and severe, and have potential risk factors such as comorbidities, presence of indwelling devices, surgical history, and immunosuppressive agents [6]. In East Delhi of India, a study demonstrated that 8.7% CRE gut colonization in neonates. The risk factors for CRE gut colonization were duration of hospitalization, nasogastric (NG) tube, NG feeding, lack of breastfeeding, top feeding, ventilation, and antibiotics administration [7]. Three carbapenem-resistant *Klebsiella pneumoniae* isolates recovered from pediatric patients at a single U.S. health care facility, and no patients with these CRE isolates had a history of international travel. The isolates were resistant to carbapenems and produce an MBL carbapenemase, which not previously published as present among Enterobacteriaceae in the United States [8]. CRE infection remains a growing problem in pediatric populations. Few studies have used rigorous methods to guide children's treatment options, and the data must be inferred from adult literature. After children are infected with CRE, the choice of treatment will be more limited due to restrictions on antibiotics for children [9-12]. Here we retrieved pediatric CRE infection cases from 2013 to 2018 in a tertiary general hospital. Strain information and infection data were analyzed to help guide the control of future CRE infections in children.

Methods

Setting and study design

The non-repeat clinical isolates of CRE in children were collected from Liaocheng People's Hospital, a Chinese tertiary comprehensive hospital. The children patients infected with CRE strains were from neonatal intensive care units (NICU), pediatric intensive care unit (PICU), neonatology, pediatric surgery and pediatrics ward. Patients were included if CRE pathogens identified from respiratory, blood, urine, superficial pus, bile, deep pus or sterile body fluid samples from January 1, 2013 to December 31, 2018. This study focused on bacterial strains, and retrospectively analyzed infection-related indicators and clinical data of infected hosts, which were already existing fully de-identified retrospective data. According to the Medical Ethics Committee of Liaocheng People's Hospital and local ethics regulations, this study was exempt from IRB review.

All positive CRE cultures and associated clinical infection data from different pediatric wards were reviewed. The same patient with recurrent infections were defined only once infection. In cases of concurrent infection, patients were classified according to the primary CRE infection. Patients' data were compiled until death or hospital discharge. The basic clinical and drug resistance data with CRE infection in pediatric patients were investigated, such as isolation genotype, drug susceptibility result, ward, underlying disease, invasive procedure, treatment and outcome due to CRE.

Detection of carbapenemases and molecular typing

The bacterial identification was performed using a Vitek MS automated mass spectrometry detection system, and antibiotic susceptibility was operated by Vitek 2 compact automated system. PCRs were performed for *bla*_{KPC}, *bla*_{VIM}, *bla*_{IMP}, *bla*_{NDM-1} and *bla*_{OXA-48} genes as Table 1. The amplicons were sequenced in Shanghai Sangon company, sequencing instruments and reagents were ABI-PRISM 3730 and BigDye terminator v3.1 respectively. Then the sequencing results blasted to NCBI databases (www.ncbi.nlm.nih.gov/) to identify genotype and registered to GenBank sequence database.

The homology of strains harboring carbapenemase genes were determined by using pulse-field gel electrophoresis (PFGE) analysis. The restriction enzyme was *Xba* I, and *Salmonella Braenderup* H9812 strain DNA was used as standard size markers. The genetic relationship of the test strains was generated by

the unweighted pair-group method (UPGMA). Isolates that exhibited a PFGE profile with more than 80% similarity (pulsotype, PT) were considered as closely related strains.

Definitions

Children was defined if patients were no elder than 14 years old.

CRE was defined as organisms of Enterobacteriaceae showing decreased susceptibility to any carbapenem antibiotic, including ertapenem minimum inhibitory concentration (MIC) $\geq 1 \mu\text{g/ml}$, meropenem MIC $\geq 2 \mu\text{g/ml}$, or imipenem MIC $\geq 2 \mu\text{g/ml}$, using current Clinical and Laboratory Standards Institute breakpoints [13].

Results

Infected strain and carbapenemase information

Twenty cases of CRE infection in children patients were identified: seven *Escherichia coli*, six *Klebsiella pneumoniae*, two *Klebsiella oxytoca*, two *Enterobacter aerogenes*, two *Serratia marcescens*, one *Enterobacter cloacae*. PFGE revealed that two *E. coli* and three *K. pneumoniae* had an indistinguishable PFGE pattern, while the other isolates were different strains. The production of IMP-type carbapenemase was the main antibiotic resistance mechanism. Confirmed by sequencing, twelve strains of CRE were produced IMP-4 type carbapenemase, which included four *E. coli*, two *K. pneumoniae*, two *K. oxytoca*, two *E. aerogenes*, one *S. marcescens* and one *E. cloacae*. While seven strains of CRE were confirmed to produce IMP-8 carbapenemase, including three *K. pneumoniae*, three *E. coli* and one *S. marcescens*, one strain did not detect the carbapenemase genotype.

The characteristics of antimicrobial resistance

The isolated CRE strains show highly resistant to carbapenems and cephalosporins (ertapenem 70.0%, imipenem 95.0%, meropenem 90.0%, ceftriaxone 85.0%, ceftazidime 80.0% and cefepime 75.0%). The resistance rate to trimethoprim/sulfamethoxazole, tobramycin and ciprofloxacin were relatively low (55.0%, 45.0% and 30.0%), while no resistance to tigecycline was observed.

Infection types and patient characteristics

The sources of cultures were respiratory (50.0%), blood (30.0%), lavage fluid (10.0%) and wound secretion (10.0%) (Table 2). Neonatology and pediatrics ward were the most frequent residence of pathogen (each six cases), followed by PICU (four cases), NICU (two cases) and pediatric Surgery (two cases). The age of pediatric patients was from one day to eight years old and male accounted for the majority in these patients (15/20, 75.0%). Most patients have comorbidities such as premature, gastrointestinal dysfunction and coronary heart disease, seven patients (35.0%) experienced invasive procedures such as mechanical ventilation, bronchoalveolar lavage (Table 3).

Treatment and outcomes

The majority (15/20, 75.0%) of patients were treated with monotherapy (third-generation cephalosporin, piperacillin/tazobactam or carbapenem) for empiric treatment. Compound sulfamethoxazole was used as single-drug therapy in two cases, one improved and the other abandon treatment due to the primary disease. Although most patients received the insensitive antibacterial drugs due to the limitations of children, outcomes were still optimistic (two cured, 10.0%; thirteen improved, 65.0%). In addition to the application of antibiotics, most children (80.0%) took ancillary treatments such as human intravenous immunoglobulin.

Discussion

In recent years, along with the rapidly increasing prevalence of CRE worldwide in the adult, CRE have been reported occasionally in children [14]. Once the children infected with CRE, clinical treatment measures are very limited due to fewer antimicrobial options. Currently it is still unclear about the optimal treatment regimens for CRE because of the lack data of randomized trials evaluating in various therapeutic approaches. To date, there are no authoritative guidelines recommending medications for children infected with CRE. only case series, case reports, or cohort studies with significant heterogeneity in population and treatment regimens were evaluated [5]. Available data support combination therapy may be beneficial to patients, however, there are insufficient data to recommend a particular combination regimen [15–17]. In our study, the resistance rates of tobramycin, ciprofloxacin and tigecycline were relatively low, but no children were treated with these drugs due to serious potential side effects such as ear nephrotoxicity, malformation, tooth or joint development especially for infants. On the contrary, 75.0% of patients have been implemented insensitive monotherapy antibiotics for empiric treatment. After the drug sensitivity report was obtained, there was still no replacement of the antibacterial drug due to the limitation of selection and the improvement of clinical symptoms. Fortunately, the infection status of most children has gradually improved. In our follow-up survey, we found that most children received adjuvant therapy which may be an important factor in helping patients defeat pathogenic bacteria. Overall, antibiotic selection should be guided by local susceptibility patterns, clinical source of infection and expert consultation.

CRE have been isolated throughout Europe, parts of Asia and Africa, and in the United States, often in individuals who have traveled from endemic regions [2]. The mechanism of resistance to CRE is mainly the production of carbapenemases. Isolates that exhibited phenotypic resistance to carbapenems were tested with primers for bla genes that encode class A (KPC), class B (NDM, IMP and VIM), and class D (OXA-48) carbapenemases. In the report of pediatric infection with CRE, different bla genotype infections have also appeared. The class A carbapenemase in KPC in paediatric hospital isolated in the USA and UK [10, 11, 14]. And class B including NDM, IMP and VIM has been described in India, USA and Spain respectively [3, 8, 18]. A outbreak of OXA-48 (class D) begun in 2012 (16 cases in NICU and 1 in PICU) confirmed that all the microorganisms isolated from children belonged to the same strain (ST11) [19]. The combination of these reports, KPC is still a major genotypes carbapenemases type in CRE infections, other genotypes are relatively small. Our study confirms that the IMP type is entire type of carbapenemase, indicating that there are still large differences between regions. Therefore, this different genotype needs to be considered when formulating the corresponding antibacterial strategy. In addition, we observed that the CRE strain producing KPC appeared to be more pathogenic and reported higher mortality [8, 16]. This may suggest the weaker pathogenicity of IMP than KPC produced by Enterobacteriaceae.

From different reports, the site of infection with CRE and the source of the specimen are not the same. At the same time, the genus of Enterobacteriaceae is different. One study by Zerr DM *et al.* on clinical epidemiology of CRE at four US pediatric hospitals showed the most common CRE pathogens were *E. coli*, followed by *K. pneumoniae* [20], which is the same to our study. But the specimen distribution in this study is mainly urine, which is different from our findings mainly in respiratory specimens. And blood infections accounted for the second place in our study. Although specimens and sources are very different in the report of CRE infection, *K. pneumoniae* and *E. coli* occupies most isolates in Enterobacteriaceae. Despite this, we have found CRE infections in several species, including two *K. oxytoca*, two *E. aerogenes*, two *S. marcescens* and one *E. cloacae*. Currently CRE in these strains reports are rare.

Much anti-infective research have evaluated the efficacy of treating CRE aspects. Available data suggest that carbapenem-containing combination therapy may be effective over monotherapy or non-carbapenem containing combination therapy [15–16]. Carbapenems remain a mainstay of pediatric therapy of CRE infections based on the available adult experience and literature despite the high resistance rates [14]. However, few data exist on the potential benefit of carbapenems in the pediatric population. One study about the risk factors for mortality with CRE bloodstream infection in children showed they were unable to assess whether combination therapy including meropenem was more effective than combination therapy without meropenem, because the mortality rate in children who received meropenem for an isolate with an MIC > 8 (100.0%, n = 8) was significantly higher than that with MIC ≤ 8 mg/L (45.5%, n =22) [17]. Although carbapenem resistance means that the strain is resistant to any carbapenems, for our study the resistance rate of ertapenem (70.0%), imipenem (95.0%) and meropenem (90.0%) is different. Accordingly, the resistance mechanism of the same isolate to different carbapenems is not the same. How to apply carbapenem antibiotics to treat CRE infection in children still needs further study.

Tigecycline-antibacterial drugs that need to be focused on because of the possibility of treating CRE infection. One study about susceptibility rates among Enterobacteriaceae isolates collected from pediatric patients (< 18 years of age) from 2004 to 2012, showed the highest susceptibility rates of tigecycline among Enterobacteriaceae isolates were *E. Coli* and *K. oxytoca* (100.0% each), followed by *Enterobacter* spp. (97.6%), *K. pneumoniae* (96.3%) and *S. marcescens* (91.5% each) in Asian region [21]. Similar results were obtained in our study, the susceptibility rate of tigecycline among CRE isolates is 100.0%. However, tigecycline is not currently indicated for the treatment of infections in patients < 18 years of age, and it should be used for pediatric patients only when no alternative antimicrobial agents are available. In pediatric settings, experience with tigecycline is limited to case reports, and along with the potential for increased mortality, association with enamel hypoplasia, tooth discoloration [22]. Purdy *et al.* recently reported the pharmacokinetic properties, safety profile, and efficacy of tigecycline in pediatric patients eight to eleven years of age. Their results show use of tigecycline among patients under eight years of age is not recommended because of possible effects on tooth development [23].

Study the risk factors of CRE infection in children, which will make infection prevention more effective. Several studies have evaluated risk factors include underlying chronic medical conditions, invasive medical devices, frequent or prolonged hospitalizations, prior antibiotic exposure, age and travel from endemic regions [24]. Nour I *et al.* [25] also have evaluated the duration of total parenteral nutrition and carbapenem use were the independent predictors for CRE acquisition. In our study, underlying disease, gender (male, 75.0%), young age and invasive procedures were considered important risk factors for CRE infection. Therefore, effective active surveillance, infection prevention and control for risk factors of CRE is an important public health issue. Adequate attention must be given to prevent the spread of CRE for children patients in each region hospitals. One study by Millar *et al.* on predictors for gut colonization of CRE in a NICU showed active surveillance of cultures from hospitalized patients and implementation of preventive efforts can reduce the risk of CRE [7]. Therefore, the future controlled studies are necessary to assess risk factors and antibiotic policies in hospital to ensure the appropriate therapeutic selection in children.

Several limitations should be considered in our research. Firstly, although this study included all children with CRE infection in our hospital for six consecutive years, the number of cases is still limited and only preliminary conclusions can be drawn. Subsequent specific treatment options need to be based on the expansion of big data in the relevant infection research area. Secondly, different physicians choose different empirical treatment options during antibiotic treatment, and this is our first concern of these CRE-infected children, so far we still have not found a uniform treatment strategy. Thirdly, our study only found that the IMP genotype in our region, which is not the same as the genotype reported in other regions, and may lead to differences in treatment options.

Conclusions

CRE infection has a prevalent trend in children patient in our hospital, and the drug-resistant genotypes detected within six years are unusually IMP-mediated. Despite the high rate of resistance and the limitations of medications for children, most patients have improved after using insensitive antibiotics. The adjuvant therapy and weaker pathogenicity of IMP-mediated CRE isolates may be responsible for desired outcomes in children. In addition, the urgent problem to be solved is to establish an optimal treatment and control program for the spread of CRE in children.

Abbreviations

CRE: carbapenem-resistant Enterobacteriaceae; ICU: intensive care units; NICU: neonatal intensive care units; PICU: pediatric intensive care unit; PFGE: pulse-field gel electrophoresis; UPGMA: unweighted pair-group method; SMA: *Serratia marcescens*; KOX: *Klebsiella oxytoca*; ECO: *Escherichia coli*; KPN: *Klebsiella pneumoniae*; EAE: *Enterobacter aerogenes*; ECL: *Enterobacter cloacae*; MIC: minimum inhibitory concentration; M: male; F: female; NRF: neonatal respiratory failure; HIE: hypoxic-ischemic encephalopathy; CPR: cardiopulmonary resuscitation; MODS: multiple organ dysfunction syndrome; CHD: congenital heart disease; ALL: acute lymphoblastic leukemia; ICH: intracerebral hemorrhage; MDS: myelodysplastic syndrome; HBV: hepatitis B virus; GERD: gastroesophageal reflux disease; AGA: acute gangrenous appendicitis; BAL: bronchoalveolar Lavage; PE: pericardial effusion; BMA: bone marrow aspiration.

Declarations

Ethics approval and consent to participate

The study was a retrospective analysis of a de-identified database. As such, it is not considered human subject research.

Consent for publication

Not applicable.

Availability of data and material

All the dataset of this study is from the Liaocheng People's Hospital's internal database. If reasonably requested, it can available from the corresponding author.

Competing interests

The authors declare that they have no competing interests.

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Authors' contributions

ZJ designed this study, participated in statistic calculations and interpretation of data. XQJ drafted this manuscript and participated in statistic calculations. FP provided funds for the isolation and identification of bacteria, as well as analysis and interpretation of drug resistance genotypes. XL participated in statistic calculations. All authors read and approved the final manuscript.

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Tables

Table 1 Universal amplification primers of common carbapenemase genotype

| Target segment | Universal primers | Design sequence (5' - 3') | Product length | Annealing temperature |
|----------------|-------------------|---------------------------|----------------|-----------------------|
| KPC | KPC-F | GATGTCACTGTATCGCCGTCT | 739 bp | 55°C |
| | KPC-R | ATAGTCATTTGCCGTGCCATA | | |
| VIM-1 | VIM-1-F | TTATGGAGCAGCAACGATGT | 920 bp | 54°C |
| | VIM-1-R | CAAAAGTCCCCTCCAACGA | | |
| VIM-2 | VIM-2-F | AAAGTTATGCCGCACTCACC | 953 bp | 54°C |
| | VIM-2-R | TGCAACTTCATGTTATGCCG | | |
| IMP-1 | IMP-1-F | TGAGCAAGTTATCTGTATTC | 740 bp | 51°C |
| | IMP-1-R | TTAGTTGCTTGGTTTTGATG | | |
| IMP-2 | IMP-2-F | GGCAGTCGCCCTAAAACAAA | 737 bp | 54°C |
| | IMP-2-R | TAGTTACTTGGCTGTGATGG | | |
| NDM-1 | NDM-1-F | CAACGGTTTGATCGTCAGG | 370 bp | 56°C |
| | NDM-1-R | GATCCCAACGGTGATATTGTC | | |
| OXA-48 | OXA-48-F | AAGGGGACGTTATGCGTGTAT | 781 bp | 55°C |
| | OXA-48-R | ACTTCTTTTGTGATGGCTTGG | | |

Table 2 Antimicrobial susceptibility profiles of carbapenem-resistant Enterobacteriaceae isolated from pediatric patients

| Isolate ^a | Year | Specimen | Genotype | Antimicrobial agent and MIC ^b (µg/mL) | | | | | | | | |
|----------------------|------|--------------------|----------|--|----------|-----------|------------|------------|-------------------------|-------------|-------------|----------|
| | | | | Ertapenem | Imipenem | Meropenem | Gentamicin | Tobramycin | Piperacillin/tazobactam | Ceftriaxone | Ceftazidime | Cefepime |
| SMA1 | 2013 | Blood | blaIMP-4 | ≤0.5 | 4 | 2 | ≤1.0 | 2 | ≤4 | ≤1.0 | ≤1.0 | ≤ |
| SMA2 | 2013 | Blood | blaIMP-8 | ≤0.5 | 4 | 2 | ≤1.0 | 2 | ≤4 | 2 | ≤1.0 | ≤ |
| KOX1 | 2013 | Blood | blaIMP-4 | ≥8 | 8 | ≥16 | ≥16 | 8 | ≥128 | ≥64 | ≥64 | ≥ |
| ECO1 | 2014 | Respiratory | blaIMP-4 | 2 | ≤1.0 | ≥16 | ≤1.0 | ≤1.0 | 64 | ≥64 | ≥64 | : |
| KPN1 | 2014 | Respiratory | blaIMP-8 | ≥8 | ≥16 | ≥16 | ≥16 | 8 | ≥128 | ≥64 | ≥64 | ≥ |
| ECO2 | 2014 | Lavage fluid | blaIMP-4 | ≤0.5 | 4 | ≥16 | ≥16 | 8 | 64 | ≥64 | ≥64 | |
| ECO3 | 2015 | Blood | blaIMP-8 | ≥8 | ≥16 | ≥16 | ≥16 | 2 | ≥128 | ≥64 | ≥64 | : |
| KOX2 | 2015 | Blood | blaIMP-4 | ≥8 | ≥16 | ≥16 | ≥16 | 4 | 64 | ≥64 | ≥64 | |
| EAE1 | 2016 | Respiratory | blaIMP-4 | ≤0.5 | 4.0 | 4 | ≤1.0 | ≤1.0 | ≤4.0 | ≤1.0 | ≤1.0 | ≤ |
| KPN2 | 2016 | Respiratory | blaIMP-8 | ≥8 | ≥16 | ≥16 | ≥16 | 2.0 | ≥128 | ≥64 | ≥64 | ≥ |
| KPN3 | 2017 | Respiratory | blaIMP-4 | ≥8 | ≥16 | ≥16 | ≥16 | ≥16 | ≥128 | ≥64 | ≥64 | ≥ |
| ECO4 | 2017 | Respiratory | blaIMP-8 | ≥8 | ≥16 | ≥16 | ≥16 | ≥16 | ≥128 | ≥64 | 16 | ≥ |
| KPN4 | 2017 | Lavage fluid | None | 4.0 | 4.0 | ≥16 | ≤1.0 | ≤1.0 | 8.0 | ≥64 | ≥64 | ≥ |
| EAE2 | 2017 | Respiratory | blaIMP-4 | ≤0.5 | 4.0 | ≤1.0 | ≤1.0 | ≤1.0 | ≤4.0 | ≤1.0 | ≥64 | ≤ |
| ECO5 | 2017 | Wound secretion | blaIMP-4 | ≥8 | 4.0 | ≥16 | ≥16 | ≥16 | ≥128 | ≥64 | ≥64 | ≥ |
| KPN5 | 2017 | Respiratory | blaIMP-8 | ≥8 | ≥16 | ≥16 | ≥16 | 8 | ≥128 | ≥64 | ≥64 | |
| ECO6 | 2018 | Respiratory | blaIMP-4 | ≥8 | 8 | ≥16 | ≥16 | ≥16 | ≥128 | ≥64 | ≥64 | ≥ |
| ECL1 | 2018 | Blood | blaIMP-4 | ≤0.5 | 4.0 | ≤1.0 | ≤0.5 | 4.0 | 16.0 | 32.0 | 16.0 | ≤ |
| KPN6 | 2018 | Respiratory | blaIMP-4 | ≥8 | ≥16 | ≥16 | ≥16 | 8 | ≥128 | ≥64 | ≥64 | ≥ |
| ECO7 | 2018 | Wound secretion | blaIMP-8 | ≥8 | ≥16 | ≥16 | ≤1.0 | ≤1.0 | ≥128 | ≥64 | ≥64 | 1 |

^a SMA, *Serratia marcescens*; KOX, *Klebsiella oxytoca*; ECO, *Escherichia coli*; KPN, *Klebsiella pneumonia*; EAE, *Enterobacter aerogenes*; ECL, *Enterobacter cloacae*.

^bMIC, minimum inhibitory concentration

Table 3 Characteristics of pediatric patients with carbapenem-resistant Enterobacteriaceae

| Patient number/ Isolate ^a | Ward ^b | Underlying disease ^c | Invasive procedure ^d | Infection due to CRE ^e | Directed antibiotic therapy | Outcome |
|---|-------------------|---|---------------------------------|--------------------------------------|--|-------------------|
| 1/ SMA1 | NICU | Premature infant, neonatal pneumonia, NRF, HIE | Mechanical ventilation | Neonatal pneumonia | Piperacillin/tazobactam, 0.10g, bid, 14 days | Improve |
| 2/ SMA2 | NICU | Premature infant, neonatal pneumonia | None | Neonatal pneumonia | Piperacillin/tazobactam, 0.06g, bid, 19 days | Unknown |
| 3/ KOX1 | PICU | Sepsis, intracranial infection, liver damage | None | Sepsis, intracranial infection | Ceftriaxone, 1g, qd, 14 days and meropenem, 0.4g, q8h, 18 days | Unknown |
| 4/ ECO1 | Neonatology | Bronchopneumonia, NRF | Mechanical ventilation | Bronchopneumonia | Imipenem, 0.03g, q6h, 3 days | Cure |
| 5/ KPN1 | PICU | Bronchopneumonia, CPR, HIE, MODS | Mechanical ventilation | Bronchopneumonia | Ceftriaxone, 0.5g, qd, 10 days | Abandon treatment |
| 6/ ECO2 | PICU | Severe pneumonia, CHD | BAL | Severe pneumonia | Ceftizoxime, 0.25g, q8h, 8 days | Improve |
| 7/ ECO3 | PICU | Septicemia, ALL, Bronchopneumonia | Mechanical ventilation | Bronchopneumonia, septicemia | Amikacin, 0.1g, q12h and caspofungin 31.25mg, qd, 3 days | Abandon treatment |
| 8/KOX2 | Pediatrics | Intracranial infection, sepsis | None | Intracranial infection, sepsis | Meropenem, 0.4g, q8h, 7 days and ceftriaxone, 1.5g, qd, 7 days | Improve |
| 9/ EAE1 | Pediatrics | Severe pneumonia, gastrointestinal, dysfunction | None | Severe pneumonia | Ceftriaxone, 0.5g, qd, 7 days | Improve |
| 10/KPN2 | Neonatology | Neonatal pneumonia, ICH | None | Neonatal pneumonia | Piperacillin/tazobactam, 0.27g, bid, 10 days | Improve |
| 11/KPN3 | Pediatrics | Bronchopneumonia, sepsis, MDS | Drainage of PEI BMA | Bronchopneumonia, sepsis | Compound sulfamethoxazole, 0.48g, q12h, 6 days | Abandon treatment |
| 12/ECO4 | Pediatrics | Bronchopneumonia, CHD | None | Bronchopneumonia | Compound sulfamethoxazole, 70mg, q12h, 7 days | Improve |
| 13/KPN4 | Neonatology | Severe pneumonia, sepsis, CHD | None | Severe pneumonia, sepsis | Piperacillin/tazobactam, 0.22g, bid, 7 days | Improve |
| 14/ EAE2 | Neonatology | Acute laryngitis, neonatal, pneumonia | None | Acute laryngitis, neonatal pneumonia | Cefoperazone/sulbactam, 0.2g, q12h, 7 days | Improve |
| 15/ ECO5 | Pediatric surgery | AGA, peritonitis, HBV | None | AGA, peritonitis | Imipenem, 0.27g, q6h, 6 days | Improve |
| 16/ KPN5 | Pediatrics | Severe pneumonia, gastrointestinal, dysfunction | None | Severe pneumonia | Piperacillin/tazobactam, 0.5g, bid, 7 days | Improve |
| 17/ ECO6 | Pediatrics | Bronchopneumonia, gastrointestinal, dysfunction | None | Bronchopneumonia | Cefoperazone/sulbactam, 0.4g, bid, 11 days | Improve |
| 18/ ECL1 | Neonatology | Neonatal pneumonia, hyperbilirubinemia | None | Neonatal pneumonia | Cefoperazone/sulbactam, 0.14g, q12h, 10 days | Improve |
| 19/KPN6 | Neonatology | Premature infant, bronchopneumonia, GERD | None | Bronchopneumonia | Ceftizoxime, 0.12g, q8h, 6 days | Improve |
| 20/ ECO7 | Pediatric surgery | AGA, peritonitis | Laparoscopic appendectomy | AGA, peritonitis | Cefoperazone/sulbactam, 0.8g, bid and metronidazole, 0.5g, q8h, 5 days | Cure |

^a SMA, *Serratia marcescens*; KOX, *Klebsiella oxytoca*; ECO, *Escherichia coli*; KPN, *Klebsiella pneumoniae*; EAE, *Enterobacter aerogenes*; ECL, *Enterobacter cloacae*.

^b NICU, neonatal intensive care units; PICU, pediatric intensive care unit.

^c NRF, neonatal respiratory failure; HIE, hypoxic-ischemic encephalopathy; CPR, cardiopulmonary resuscitation; MODS, multiple organ dysfunction syndrome; CHD, congenital heart disease; ALL, acute lymphoblastic leukemia; ICH, intracerebral hemorrhage; MDS, myelodysplastic syndrome; AGA, acute gangrenous appendicitis; HBV, hepatitis B virus; GERD, gastroesophageal reflux disease.

^d BAL, bronchoalveolar Lavage; PE, pericardial effusion; BMA, bone marrow aspiration.

^e CRE, carbapenem-resistant Enterobacteriaceae