Changes in Multidrug-resistant *Escherichia Coli* of Neonatal Meningitis During 2001–2019: a Study in Eastern China

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Research

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Abstract

Background and objective: Neonatal meningitis (NM) caused by Escherichia coli remains a major health problem in industrialized countries. The purpose of this study was to investigate changes in antimicrobial resistance of *E. coli* causing neonatal meningitis in a perinatal center in eastern China over the past 19 years.

Methods: This survey was investigated during three periods: 2001–2006, 2007–2012, and 2013–2019. NM is diagnosed according to the number of white blood cells in the cerebrospinal fluid and the presence of a single potential pathogenic bacterium in culture extracted from blood or cerebrospinal fluid of any newborn baby. Changes in the antimicrobial resistance of *E. coli* were analyzed.

Results: A total of 209 cases of NM were identified. *E. coli* was identified from 69 cases, of which extended-spectrum beta-lactamase (ESBL) production was found in 21 cases. *E. coli* was the main cause of neonatal meningitis in this study. The overall resistance rate of *E. coli* to third-generation cephalosporins such as cefotaxime increased from 0% in 2001–2006 to 50% in 2007–2012 and subsequently decreased to 29.0% in 2013–2019. This pattern of change is similar to that of ESBL production. Only 15.0% *E. coli* found in samples from infants with early-onset meningitis (EOM) produced ESBL, while 40.0% of *E. coli* from children with late-onset meningitis (LOM) produced ESBL.

Conclusion: We concluded that *E. coli* remains the primary pathogen of NM. Compared to LOM, the percentage of ESBL-producing multi-drug resistant *E. coli* isolated from EOM is significantly decreased. Clinicians should consider this trend when choosing appropriate and effective antibiotics as empirical treatments for NM.

1. Introduction

Bacterial meningitis is related to high mortality and morbidity[1,2]. The mortality rates vary between 10% and 15%, especially in the neonatal period[3,4]. Extraintestinal pathogenic *E. coli* remains one of the most common bacterial pathogens causing extraintestinal infections, including Neonatal meningitis (NM), septicemia, and urinary tract infections[5–7]. Early-onset meningitis (EOM) is defined as infection occurring within 7 days after birth and is usually acquired by vertical transmission from mother to infant. Late-onset meningitis (LOM) is defined as infection occurring after 7 days post-birth, generally contracted nosocomially or via community infection. Compared to LOM, infants with conditions of prolonged rupture of membranes (PROM) is more prone to develop EOM with worse outcomes because of chorioamnionitis and amniotic fluid contamination[8]. *E. coli* is the second most common pathogen and was found in 30% of all EOM in developed countries[9].

Symptoms of NM are generally non-specific, for which rapid recognition and early initiation of antimicrobial therapy before the availability of blood culture results is crucial. In the 1996 national prospective study of Menin-Gideon's disease in newborns in England and Wales, the mortality rate of

neonatal meningitis in the acute stage was 6.6%[4], while this rate was 22% in a similar study conducted in 1985[10]. Despite the overall improvement of neonatal care from 1985 to 1996, the primary difference between the two studies was an increase in the use of third-generation cephalosporins[11]. A retrospective study by Zhao et al, showed that *E. coli* is still a prominent pathogen of NM. Antibiotic treatment has always been a routine treatment for this type of infection. However, because of the emergence of drug-resistant bacteria, the curative effects of antibiotics have decreased. Presently, *E. coli* exist with different degrees of resistance to third-generation cephalosporins[12].

Although studies in developed countries have found that Group B Streptococcus (GBS), *E. coli*, and Listeria monocytogenes are major organisms in the spread of NM[13–15], the results in developing countries may differ. Data about epidemiology and antimicrobial resistance patterns of NM in developing countries are relatively rare, especially from China where the economy rapidly developed since 21st century. Almost all the reported *E. coli* isolates from Chinese neonates so far, are susceptible to amikacin, cefoperazone sulbactam and carbapenems[16,17]. As changes in multidrug-resistant *E. coli* strains occur at an increasing rate globally, the spreading of antimicrobial resistant *E. coli* is now a public health problem and big concern in China. The aim of this study was to investigate the clinical characteristics, antimicrobial resistance patterns of NM caused by *E. coli* from 2001 to 2019 in a large tertiary neonatal intensive care unit (NICU) in city Wenzhou, located in Zhejiang province of eastern China. Moreover,we compared the ratio of extended-spectrum beta-lactamase (ESBL)-producing *E. coli* of NM. We focused on comparing the ratio of ESBL-producing *E. coli* between EOM and LOM infants. Our study will be helpful in selecting more appropriate antibiotics for empirical treatment in developing countries with similar bacterial spectra and sensitivities.

2. Materials And Methods

2.1 Data Collection

Neonatal cases were defined as infants 28 days or less of age. All newborns diagnosed with purulent meningitis in the NICU of the Second Affiliated Hospital of Wenzhou Medical University and Yuying Children's Hospital during the study periods were included in this retrospective cohort study. Sensitivity and specificity test results and ESBL statuses were reported by our clinical laboratory, which conducts routine microbiological examinations according to the standards formulated by the American Clinical and Laboratory Standards Association. Because the present study covered an extended time period, bacterial species were identified by either traditional biochemical techniques or automated methods via VITEK system (Vitek 2 compact, BioMerieux, France). Initially, the manual Kirby-Bauer disk diffusion method or the recent gram-negative drug sensitivity card (BioMerieux, France) was used to determine the antibiotic sensitivity of bacterial isolates. This study covered three time periods: 2001–2006, 2007–2012, and 2013–2019. All cases were identified by registration and hospital diagnosis records and were confirmed by detailed chart reviews.

Relevant clinical data were extracted from the patients' medical records. We collected each infant's gestational age, sex, birth weight, mode of delivery, and instance of fever (> 38°C). To calculate the incidence of NM, data on the number of total live births in the hospital during these three periods were also collected. This research study was approved by the institutional ethics committee of the Second Affiliated Hospital of Wenzhou Medical College and Yuying Children's Hospital. The patients' parents consent to review their medical records which was allowed by the Institutional Ethics Committee. The treatment of confidentiality of patient data strictly follows the rules formulated by the institution and conforms to the Helsinki Declaration.

2.2 Statistical Analyses

SPSS software (version 23.0) was used for statistical analyses. The patients' basic clinical features and blood culture results and the antimicrobial susceptibilities of their relative *E. coli* strains were analyzed. The Kolmogorov–Smirnov test was used to test the normality of continuous variables. The data of normal distribution are described as means ± standard deviations and were analyzed by Student's *t*-test of variance. The data with non-normal distribution are described as medians and ranges and were analyzed using the Wilcoxon signed rank test or the Mann-Whitney *U*-test. Classification data were analyzed using the chi-square test or Fisher's exact test. A p-value of < 0.05 of the predicted variable was considered significant.

3. Results

3.1 Cases of the Neonatal Meningitis

In the studied hospital, the total number of live births was17,263 in 2001–2006, 39,202 in 2007–2012, and 66,549 in 2013–2019. 209 cases of culture-confirmed neonatal purulent meningitis were identified from 2001 to 2019, among which 69 cases of *E. coli* were isolated (including 10 cases from 2001 to 2006, 24 cases from 2007 to 2012,and 35 cases from 2013 to 2019). Of the 69 cases, 4 cases were isolated from blood cultures in other hospitals before transferred to the NICU of the studied hospital, therefore, no detailed information was obtained. Of the 209 cases of NM, 27 infants (2 from 2001–2006, 10 from 2007–2012 and 15 from 2013–2019) were borned in the studied hospital, and the other 182 cases were either transferred from other hospitals that did not have NICU services or directly admitted from the community after home birth to the studied hospital. As a result, the calculated incidence of culture-confirmed NM in the studied hospital was 0.12 per 1000 live-births in 2001–2006, and 0.26 per 1000 live-births in 2007–2012, 0.23 per 1000 live-births in 2013–2019.

3.2 Bacterial pathogens in Neonatal Meningitis

The proportions of different bacterial pathogens causing NM were compared in the three periods as shown in Table 1.The proportion of coagulate-negative staphylococci (CoNS) infections decreased gradually from 27.3% in 2001–2006 to 19% in 2007–2012 and 3.1% in 2013–2019 ($p < 0.001$). GBS rates were increased, from 3% in 2001–2006, to 13.9% in 2007–2012 and 39.2% in 2013–2019 ($p < 0.001$),

thus GBS became the most frequently isolated gram-positive bacteria from 2013 to 2019. Over the past two decades, the proportion of *E. coli* as the pathogen causing NM remained relatively stable, above 30% in the three periods and was still the primary bacterial pathogen conferring neonatal meningitis. The proportion of enterococcus causing NM decreased, although the differences were not statistically significant, similar to Staphylococcus aureus. Besides, the proportion of Klebsiella also remained relatively stable, about 3% in every period.

Table 1
Distribution of Pathogens of neonate meningitis in 2001–2006, 2007–2012 and 2013–2019 were analyzed by Pearson's chi-squared test.

Pathogens	**2001–2006 (n = 33)**	**2007–2012 (n = 79)**	**2013–2019 (n = 97)**
Gram-positive organisms			
GBS	1(3.0%)	11(13.9%)	38(39.2%)*
CoNS	9(27.3%)	15(19.0%)	3(3.1%)**
Enterococcus	4(12.1%)	8(10.1%)	6(6.2%)
Staphylococcus aureus	3(9.1%)	6(7.6%)	3(3.1%)
Other	0(0%)	4(5.1%)	4(4.1%)
Gram-negative organisms			
E. coli	10(30.3%)	24(30.4%)	35(36.1%)
Klebsiella	1(3.0%)	2(2.5%)	3(3.1%)
Other	5(15.2%)	9(11.4%)	5(5.2%)

Notes: *$\chi^2 = 24.653$, $P < 0.001$. **$\chi^2 = 16.955$, $P < 0.001$.

3.3 Clinical characteristic of *E. coli* causing Neonatal Meningitis

Table 2 shows the general characteristics of neonates with *E. coli* meningitis in three time periods. The proportion of infants with *E. coli* meningitis born at home decreased from 30% in 2001–2006 to 8.3% in 2007–2012, and no children with *E. coli* meningitis born at home during the 2013–2019 period ($p < 0.05$).

Table 2
General Characteristics of Patients with neonate *E. coli* meningitis.

		2001–2006 (n = 10)	2007–2012 (n = 24)	2013–2019 (n = 35)	P-value
Male gender	42	6(60.0%)	15(62.5%)	21(60.0%)	NS
Gestational age (weeks)		38.2 ± 2.8	38.1 ± 3.5	36.7 ± 3.9	NS
< 37 weeks	17	2(20%)	5(20.8%)	10(28.2%)	NS
Birth weight (gm)		2850 ± 617g	3010 ± 782g	2986 ± 899g	NS
< 2500 gm	18	3(30.0%)	5(20.8%)	10(28.6%)	NS
< 1500 gm	5	0	1(4.2%)	4(11.4%)	NS
Vaginal delivery	57	10(100%)	19(79.2%)	28(80.0%)	NS
Home delivery	5	3(30.0%)	2(8.3%)	0(0)	0.005 *
fever	62	10(100%)	22(91.7%)	30(85.7%)	NS
Death	10	3(30%)	3(12.5%)	4(11.4%)	NS
Note: *P < 0.05. NS, not significant.					

3.4 Distribution of *E. coli* Neonatal Meningitis From 2001 to 2019

The varies in antibiotic susceptibility of all *E. coli* strains isolated from infants with NM in the three periods are presented in Fig. 1. As shown in Fig. 1, the overall resistance rate of *E. coli* to third-generation cephalosporins(such as ceftazidime) increased from 0% in 2001–2006 to 50% in 2007–2012 and subsequently decreased to approximately 29% in 2013–2019 ($p < 0.05$). This resistance pattern is closely related to the yield of ESBL, which also increased from 0% in 2001–2006 to 50% in 2007–2012, while in 2013–2019 the rate decreased to 29% ($p < 0.05$). The resistance rate to ceftazidime dramatically increased from 0% in 2001–2006 to 50% in 2007–2012 and decreased to 24.1% in 2013–2019 ($p < 0.05$). The resistance rate of *E. coli* to ampicillin increased from 40.0% in 2001–2006 to 83.3% in 2007–2012 and subsequently decreased to 66.7% in 2013–2019 ($p < 0.05$). There was no statistical change in the resistance rate of *E. coli* strains to gentamicin (50% in 2007–2012 compared to 40.7% in 2013–2019). All the *E. coli* in the study were susceptible to amikacin, cefoperazone sulbactam, imipenem and meropenem. The resistance rate to piperacillin tazobactam and amoxicillin clavulanic acid was very low (4.2% and 8.3%, respectively) in 2007–2012 and (3.3% and 0%, respectively) in 2013–2019. The result is [illegible] the resistance rate to piperacillin tazobactam is 7%[18].

3.5 ESBL production in years

Figure 2 shows the changes of ESBL production from 2001–2019. No ESBL-producing *E. coli was* isolated from infants in 2001–2006. ESBL-producing *E. coli was identified* from 2007–2012. Then ESBL-producing *E. coli* was significantly reduced and the rate was relatively stable since 2015.This may benefit from a stronger usage restriction of antibiotics carried out from 2013 in China.

3.6 Term and premature infants of *E. coli* Neonatal Meningitis

Figure 3 shows the results of the antibiotic susceptibility testing on meningitis-causing *E. coli* comparing data from term and premature infants. Compared to data from term infants, the composition ratio of *E. coli* encephalitis in premature infants was significantly lower (75.4–24.6%, respectively; $p < 0.01$). The resistance rate of premature infants with *E. coli* meningitis to ampicillin was 85.7%, while that of term infants was 63.8%, with no statistical difference. In isolates from premature infants, the resistance rate to levofloxacin was slightly higher than that in term infants (not significant, 33.3% vs 20.4%, $p = 0.305$). Of the *E. coli* isolates from premature infants, 40.0% were resistant to third-generation cephalosporins (such as cefotaxime), also slightly higher rate than that from term infants (31.3%, $p = 0.530$). This is similar to ESBL-producing *E. coli* which represented 40.0% of all *E. coli* isolated from preterm infants as compared to 30.0% of *E. coli* in term infants ($p = 0.639$). In general, the majority of *E. coli* strains isolated from term and premature infants with meningitis in the studied hospital were sensitive to amoxicillin-clavulanic acid, amikacin, cefoxitin, sulposhen, imipenem and meropenem.

3.7 Distribution of *E. coli* causing EOM and LOM

Figure 4 shows the results of the antibiotic susceptibility testing on *E. coli* causing meningitis grouped by EOM vs LOM among neonates. The resistance rate of the late-onset group to second-generation cephalosporins such as cefuroxime was higher than those isolated from early-onset group (43.9% vs 15%, $p < 0.05$). Compared with the rate in infants with EOM, the resistance rate of *E. coli* to third-generation cephalosporins such as cefotaxime in infants with LOM was significantly decreased (41.9% vs 15%, $p < 0.05$). Approximately one-third (37.2%) of *E. coli* isolated from infants with LOM were resistant to ceftazidime, compared with 15% of isolates from infants with EOM; However, this difference was not significant ($p = 0.086$). This difference in drug resistance is primarily because of ESBL-producing *E. coli*, which accounts for 15.0% of all *E. coli* isolates from children with EOM and 40.0% of all *E. coli* isolates from children with LOM ($p < 0.05$).

4. Discussion

Neonates are at high risk of meningitis which might lead to neurologic complications. Severe neurodisability and milder motor and psychometric impairment result from NM[19]. Despite global awareness of the risk factors of maternal and infantile infection and increased early treatments over the usative organism of NM in developed countries[20–22].From a

Large cohort studies, NM remains a substantial cause of sepsis-related morbidity and mortality in the term and near-term infant[23]. We described how meningitis-related pathogens have changed from 2001 to 2019 in a city located in China. Bacterial resistance to commonly used antibiotics has become a global problem[24], and regional differences exist[13,25]. When selecting empirical antibiotics, clinicians should consider local epidemiology (if known), early vs. late disease onset, antimicrobial resistance patterns, and availability within resource constraints[26].There are relatively rare data about NM from developing countries, and the bacterial profile may be quite different. We investigated the current research in a large third-class hospital in eastern China over the last two decades. The findings shows the ratio of meningitis caused by CoNS has significantly decreased in the NICU of studied hospital, and the ratio of GBS meningitis has significantly increased, although *E. coli* persists as the mian etiologic agent of NM in the hospital. The most common pathogens(*E. coli* and GBS) we reported here are similar to those reported by Wiswell et al[27]. A multi-center survey of neonatal purulent meningitis from 13 hospitals in northern China showed that *E. coli* was the most common pathogen of NM with a rate of 21.1% among the infants of NM[28], which is lower than that in our study (33%,69/209). We found out that GBS become the first common pathogen in our region, possibly resulted from the dramatic socioeconomic changes happened in China due to industrialization. Due to industrialization, people are more accustomed to drink milk and thus more maternal colonization in vagina and breast of pregnant women. This usually result in neonatal infection.

In the past 10 years, the annual deliveries number in our hospital has increased obviously,with approximately 10,000 births per year. This makes our center one of the largest perinatal centers in eastern China, where substantial socioeconomic changes have taken place due to industrialization. In the last decade, more babies have been born in hospitals, although occasionally some pregnant women have given birth at home. Compared with the proportion in 2001–2006, the proportion of children with *E. coli* meningitis birthed at home significantly decreased from 2007 to 2012, and there were no cases of children with *E. coli* meningitis birthed at home from 2013 to 2019 (3/10, 2/24, and 0/35, respectively). *E. coli* usually colonizes the maternal reproductive tract and can cause early neonatal infection[29,30]. However, the multidrug-resistant ESBL-producing bacteria observed in a large-scale sub-Saharan Africa study are obtained from polluted hospital environments, which increase the risk of death[18]. Our results are consistent with these findings. All five children born at home during our study periods were term infants and were infected with ESBL-negative *E. coli*. The proportion of ESBL-producing *E. coli* isolated from infants with EOM was significantly lower than that of infants with LOM (15.0% and 40.0%, respectively). In a similar way as presented in the French national survey, EOM from *E. coli* was statistically less frequent than LOM from *E. coli* (18% and 33%, respectively, $P < 0.01$) [31].Notably, the antimicrobial resistance rate of *E. coli* isolated from infants with EOM and from infants with LOM increased with regard to third-generation cephalosporins for example cefotaxime (15% and 41.9%, respectively), and to second-generation cephalosporins such as cefuroxime (15% and 43.9%, respectively), both the difference were considered significant. This may due to ESBL-producing *E. coli* infection spreads in the community and contaminated hospital environment with the mobile genetic

s. A lasted research revealed that ESBL-producing E. coli

spreaded frequently in households with babies and improving community health was helpful to prevent the spread of ESBL-producing *E. coli*[32].

The incidence of NM was between 0.12‰-1‰ in infants carried to term and 3‰ in premature infants[11,33,34]. A regional retrospective study conducted in Sweden from 1987–1996 estimated at 0.3 per 1000 live births[35]. This is in accordance with the UK and Ireland where reported an incidence of 0.38 per 1000 live births[36]. The incidence of NM much higher at 0.8–6.1 per 1000 live births, with a mortality of 40–58% in developing countries[13].The incidence of culture-confirmed NM in our perinatal center increased from 0.12 per 1000 live-births in 2001–2006 to 0.26 per 1000 live-births in 2007–2012, and a little decreased to 0.23 per 1000 live-births in 2013–2019, which is a little lower than the datas from industrialized countries. This may reason for cases of negative blood culture associated with a pleocytosis ($\geq 10\times10^6$ cells/L) in the CSF associated with typical clinical manifestations and the anti-infection treatment effect is remarkable were excluded.

Neonatal *E. coli* causing meningitis is related to high mortality and morbidity[37,38]. In a French national survey about neonatal bacterial meningitis 444 Cases from 2001 to 2007, reported the neonatal mortality rate of bacterial meningitis was 13%,while the mortality rate was twice as high in preterm (26%) than in term infants (10%),and neonatal mortality rate with *E. coli* meningitis was 12% (15/123) [31]. In 2015, it was reported that in Britain, the most common cause of neonatal meningitis caused by gram-negative bacteria was *E. coli* K1, with a mortality rate of 10–15%[39]. As reported from1997-2017 in Sweden, the pathogens with the highest NM mortality rate are gram-negative bacteria, *Klebsiella pneumoniae* (33%; 2/6), and *E. coli* (11%; 2/18) [40]. The case-mortality rate of *E. coli* meningitis in our center remained relatively unchanged with 12.5% (3/24) in 2007–2012 and 11.4% (4/35) in 2013–2019, at about 12%. Which is similar to the estimated 10% mortality rate of neonatal meningitis in developed countries[13].

For infected infants < 60 days of age, the World Health Organization recommends using penicillins (such as ampicillin or penicillin) and aminoglycosides such as gentamicin or third-generation cephalosporins (such as ceftriaxone or cefotaxime)[13]. In the current study, 83.3% of all *E. coli* isolates from infants with meningitis from 2007 to 2012 were resistant to ampicillin, and 50% were resistant to third-generation cephalosporins (such as cefotaxime or ceftazidime). These rates were significantly higher than the percentages of resistance from 2001 to 2006, although the resistance rate of *E. coli* to ampicillin and cefotaxime or ceftazidime significantly decreased from 2013 to 2019. Our data showed that approximately one-third (21/65) of the cerebrospinal fluid isolates from infants with *E. coli* in our NICU were multidrug-resistant because of the production of ESBL. No ESBL-producing multidrug-resistant strains of *E. coli* were isolated from 2001 to 2006. An increasing number of ESBL-producing multidrug-resistant *E. coli* strains were isolated from 2007 to 2012, comprising 50% of strains (12/24), while the number of ESBL-producing multidrug-resistant *E. coli* strains significantly decreased from 2013 to 2019, comprising 29% of strains (9/31). This may be a result of China's strict control of antibiotic using over the past 10 years, particularly for children, which has influenced the formulation of a series of regulations to consensus on the diagnosis and management of neonatal

sepsis. In our NICU, the management guidelines for antibiotic using are strictly implemented. According to the guidelines of neonatal septicemia in America[41], neonatologists grasp the indications for the use and discontinuation of antibacterial drugs. When antibiotics are used in children with high-risk factors, if no infection symptoms (e.g., fever, poor reaction and feeding), exist for 48 h, and a blood culture produces no alarming results, then the use of antibiotics is to be stopped immediately. For children with meningitis, penicillin combined with antibiotics, such as Rorschach's third-generation cephalosporins such as cefotaxime, is a combination that is also used empirically, and antibiotics are adjusted according to the drug sensitivity of the *E. coli* strain.

A study on neonatal septicemia and meningitis from 26 countries in Africa between 2008 and 2018 reported a non-susceptibility of *E. coli* isolates from NM to ampicillin was 89%, and gentamicin of 47%[18]. This data is higher than that from our center, in which 83.3% from 2007 to 2012 and 66.7% from 2013 to 2019 of *E. coli* isolates were resistant to ampicillin, and a non-susceptibility to gentamicin of 40.7% between 2013 and 2019. The widespread use of carbapenems has caused a notable spread of carbapenem-resistant[42]. However, to date, all *E. coli* strains isolated from infants with meningitis in our NICU have still been sensitive to cefoperazone-sulbactam and carbapenem antibiotics.

Compared to term infants, the percentage of premature infants among *E. coli* NM cases had increased and they are more susceptible to infections. A study form France has reported *E coli* is the most common cause of late premature infants and very early premature infants[31]. A prospective French survey collected data of 325 children hospitalized globally with *E. coli* meningitis from 2001 to 2013. The results of this study showed that 65.2% of these children were born at term, 22.4% were late premature infant, and 12.5% were very early premature infant[43]. Our results were in line with those from the French study. The percentage of term infants with *E. coli* meningitis 75.4% (52/69), late premature infants11.6% (8/69), and very premature or very early premature infants accounted for 13% (9/69) of our study population. Additionally, the proportion of premature infants with *E. coli* meningitis who were birth weight < 1500 gm in our NICU increased from 4.2–11.4%, which may be the result of the comprehensive actions of many factors. With the general progress of neonatal nursing and the improvement of NICU doctors' clinical skills, an increasing number of extremely premature infants have been successfully treated. In addition, this should related to most maternal immunoglobulins do not cross the placenta before 32 weeks gestation, the extremely preterm infants are at significantly higher risk for infections.[44] Furthermore,early initiation of breastfeeding may be protective against infections which due to transfer of immunoglobulin A, however, term infants usually with breastfeeding at home[45].

Interestingly, *E. coli* is one of the most common microorganisms producing L-asparaginase, and L-glutamine releases ammonia through enzymatic reaction, which makes *E. coli* obtain acid resistance. In a recent study, which revealed the divalent cations except magnesium almost completely inhibited L-asparaginase, such as Ca^{2+}, Zn^{2+} and Cu^{2+} have been demonstrated as inhibitors of *E. cloacae* enzyme and reduce its activity by about 95 %[46]. We assume that Zn^{2+} may be a choice to supplementary

treatment of *E. coli* causing neonatal meningitis in the near future, and furthermore studies are needed to validate this suppose.

5. Conclusion

In conclusion, we studied the changes in the pattern of drug resistance in neonatal-meningitis-causing *E. coli* in a large perinatal medical center in Wenzhou located at eastern China. *E. coli* remains the major cause of neonatal bacterial meningitis despite GBS incidence has significantly increased. While the proportion of ESBL-producing multi-drug resistant *E. coli* in NM isolates has significantly decreased in recent 7 years benefit from the compliance and strict use of antibiotics. Therefore, the third-generation cephalosporins on the whole are still effective to *E. coli* NM. This study covered neonate meningitis cases across 19 years in a single hospital. The study has its limitation, such as the incidence of NM in term and premature infants were not calculated. Another limitation is that only confirmed cases before hospital discharge were calculated, further studies are needed to collect the follow-up as long-term neurologic morbidity and physical disability. The third-generation cephalosporins and penicillin would be a first option for NM when occured in developing countries each anti infection of *E. coli* and GBS. Continuous monitoring of antibiotic susceptibility in NM isolates is necessary to ensure the effectiveness of the standard empirical treatment.

Declarations

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Conflict of Interest:

There are no conflicts of interest to declare.

Authors' contributions:

YL and MZ designed the study. SC and JC conducted the data acquisition. XF and SC modify the article. YL and NJ made the tables and fugures. All authors contributed to drafting and critical revision of the work, and final approval of the manuscript.

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Figures

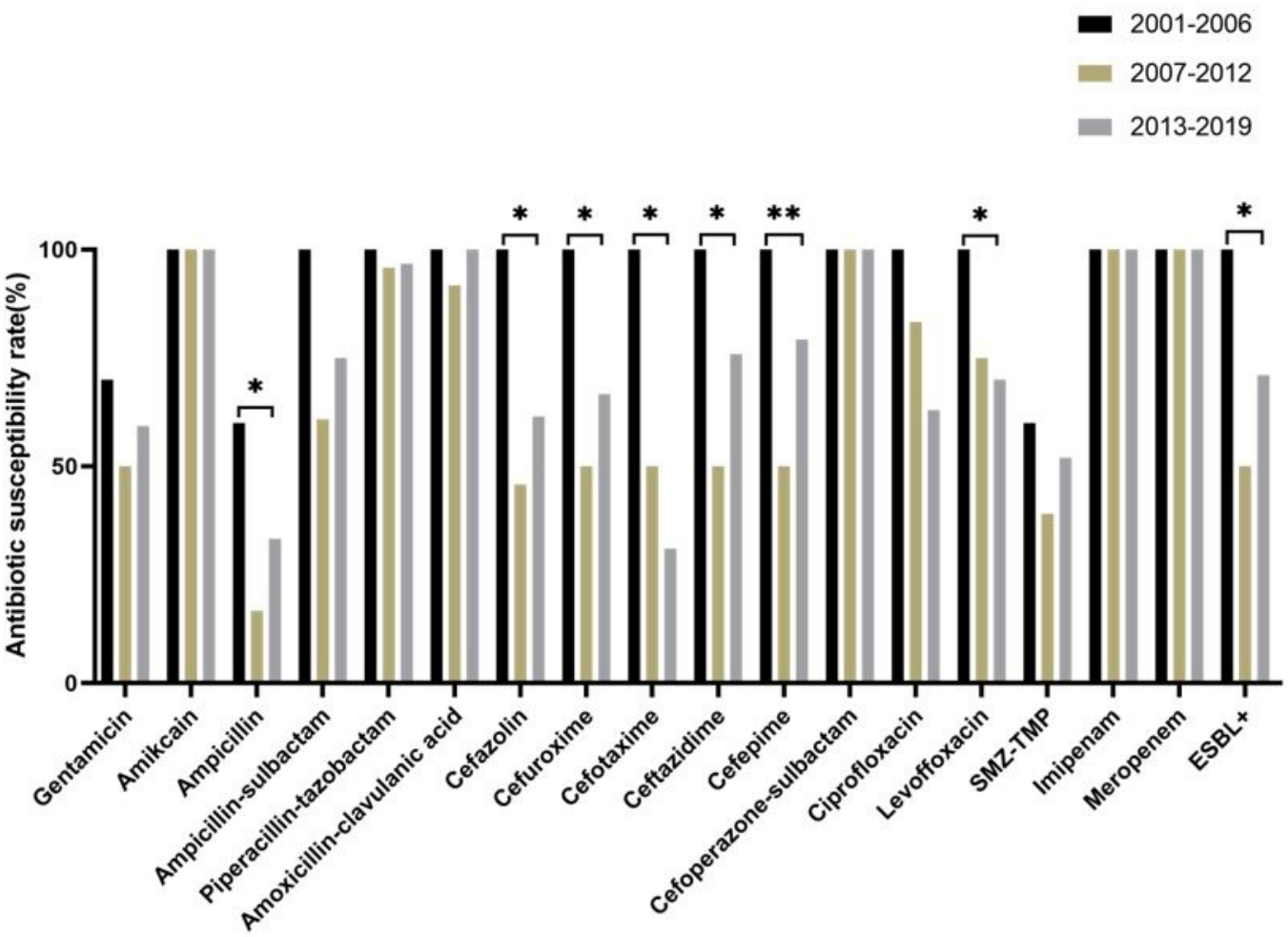

Figure 1

Antimicrobial susceptibility of all isolated E. coli in 2001–2006, 2007-2012 and 2013-2019 were analyzed by Pearson's chi-squared test. *P < 0.05. **P < 0.01.

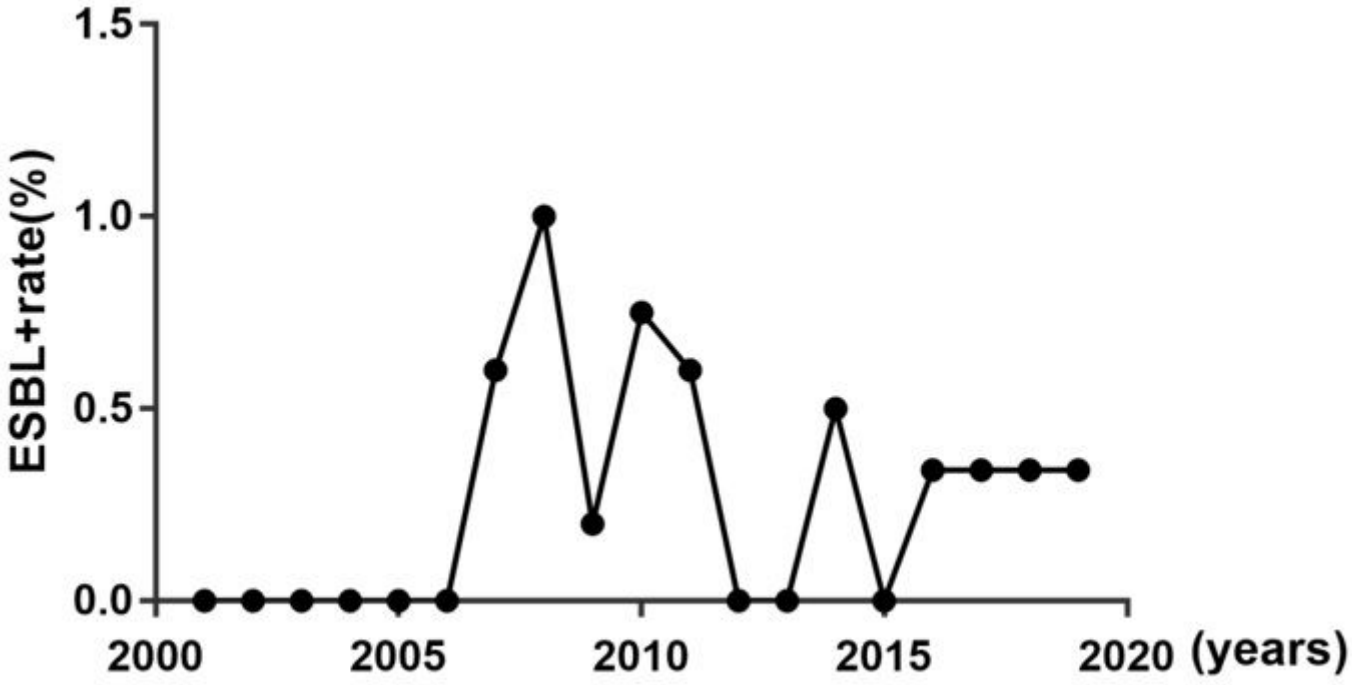

Figure 2

The change of ESBL production from 2001-2019.

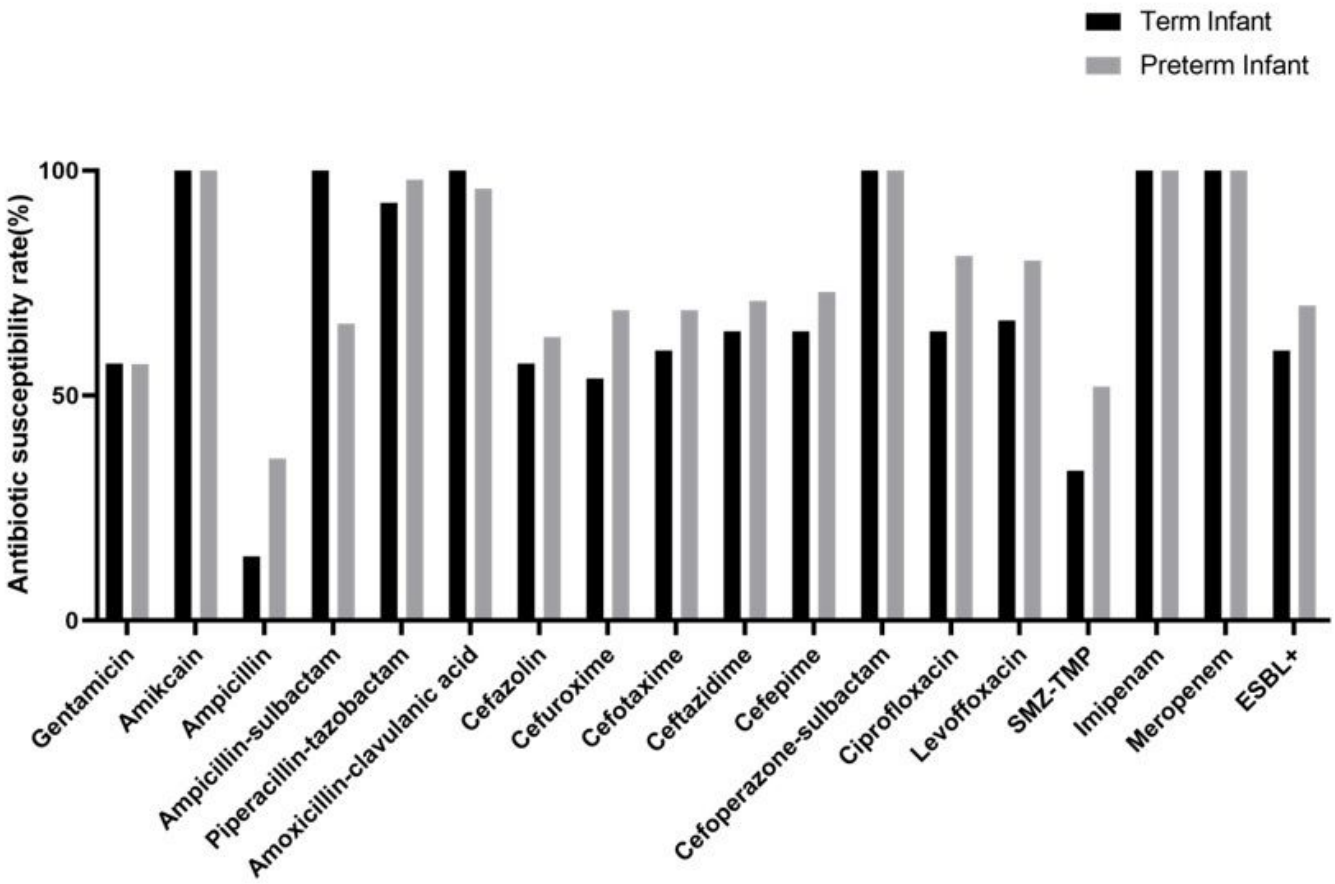

Figure 3

Antimicrobial susceptibility of all isolated E. coli from term and preterm infants. There is no significant difference between the two groups.

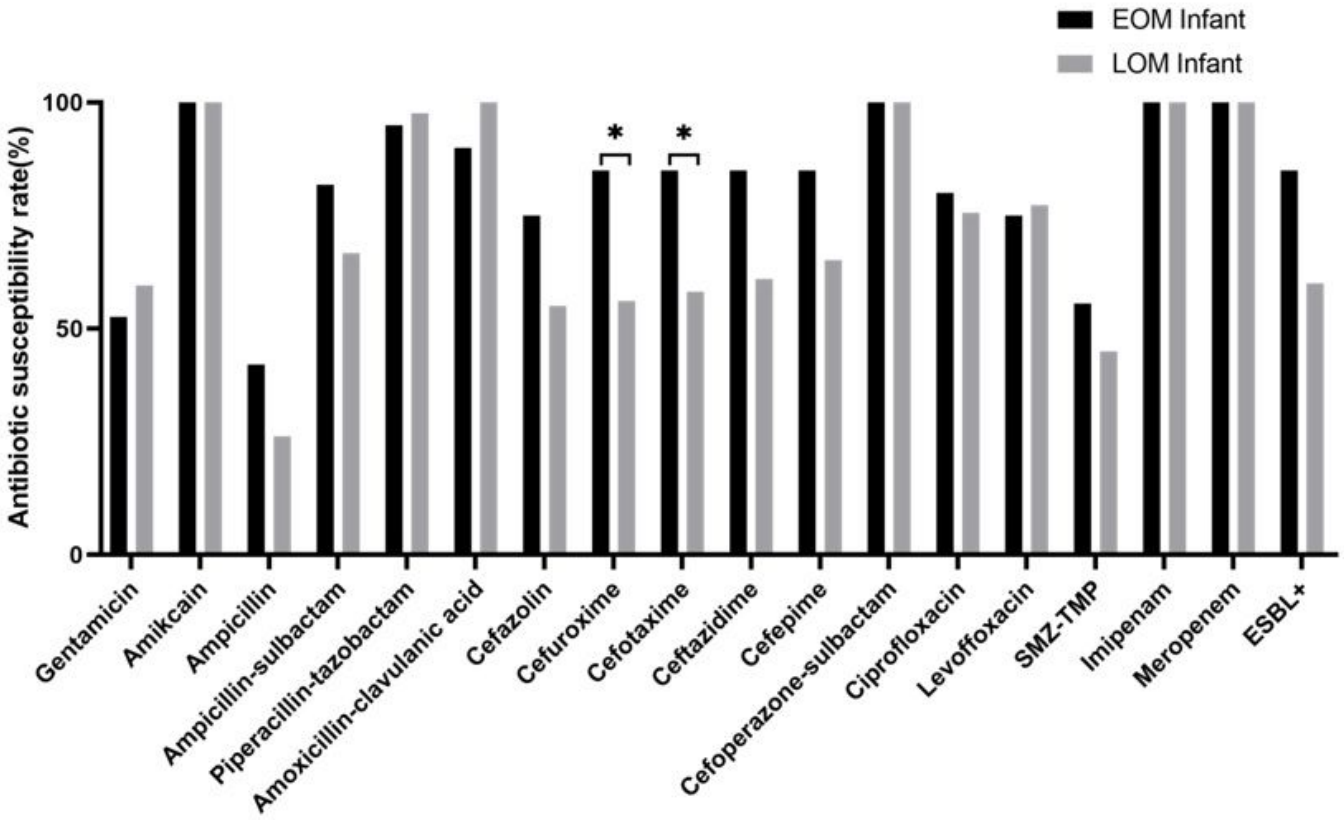

Figure 4

Antimicrobial susceptibility of all isolated E. coli from EOM and LOM. *P < 0.05.