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Original Article

Analysis of pathogens, drug resistance, sensitive antibiotic treatment and risk factors of early-onset sepsis in very low birth weight infants

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Abstract: The clinical manifestations, types of infectious pathogens, and drug-resistant strains of sepsis in infants with very low birth weight (VLBWIs) vary greatly in different regions and hospitals. In order to improve the level of diagnosis and treatment, this study analyzed the distribution and drug resistance of the pathogenic bacteria of sepsis in VLBWIs in our hospital. A total of 69 cases of VLBWIs in Qilu Hospital (Qingdao), Cheeloo College of Medicine, Shandong University from January 01, 2014 to December 31, 2020 were included. Among them, 34 VLBWIs with early-onset sepsis (EOS) were assigned to the EOS group, and 14 VLBWIs with late-onset sepsis (LOS) were included in the LOS group. The distribution of pathogens and the drug resistance of antibiotics were analyzed. The results showed that fluorescent nanoparticles detected pathogenic bacteria in 48 cases, and the blood cultures were all positive. A total of 49 pathogenic bacteria were isolated, including 27 gram-negative bacteria (55.1%), 21 gram-positive bacteria (42.86%), and 1 fungus (2.04%, Candida albicans). Gram-negative bacteria comprised of E.coli and Klebsiella pneumoniae, which were highly sensitive to compound preparations containing β-lactamase inhibitors, and carbapenem antibacterial drugs, were the first choice. Gram-positive bacteria were mainly Staphylococcus epidermidis and Streptococcus agalactiae. Staphylococcus epidermidis was highly resistant to penicillins and can be treated with vancomycin. Streptococcus agalactiae was highly resistant to penicillins and can be treated with penicillin and vancomycin. Amniotic fluid pollution, intrauterine distress, premature rupture of membranes, and maternal fever were risk factors for EOS in VLBWIs, with odds ratios (ORs) of 9.369, 6.217, 5.638, and 4.267, respectively. In summary, timely and reasonable treatment should be given based on the types and drug resistance characteristics of pathogens of neonatal sepsis and the risk factors of EOS.

Keywords: Very low birth weight infants, early-onset sepsis, pathogens, drug resistance, antibiotics, risk factors

Introduction

Very low birth weight infants (VLBWIs) refer to newborns weighing less than 1500 g. With the continuous development of critical diagnosis and treatment technology, the survival rate of VLBWIs has been greatly improved [1]. However, due to the low immune function, long hospital stays, and various invasive treatment measures, the incidence of sepsis in VLBWIs is increased [2]. Sepsis is a common infectious disease in neonatology. As bacteria or fungi invade the blood circulation of the fetus, while growing and multiplying, various toxins are produced and will cause systemic infection of the fetus [3]. According to the time of onset of the newborn, neonatal sepsis can be divided into early-onset sepsis (EOS, appearing within 3 days of birth) and late-onset sepsis (LOS, appearing after 3 days of birth) [4]. EOS has the characteristics of rapid onset, rapid progress, dangerous condition, and poor prognosis, which can cause death in severe cases [5]. VLBWIs are a high-risk group of infections in the neonatal intensive care unit (NICU) due to various systemic hypoplasia, and patients with EOS have an increased risk of death than those with LOS [6]. Therefore, the identification of pathogens, drug resistance, sensitive antibiotic treatment and risk factors of EOS in VLBWIs is conducive to effective prevention and early intervention, thus improving the prognosis of patients.

At present, the detection of pathogenic bacteria mainly relies on conventional culture methods, which needs to go through various processes such as enrichment culture, selective separation, observation of morphological characteristics, physiological and biochemical reactions, and serological identification; Generally, it takes 4-7 d, and the operation is tedious as well as time and labor consuming [7]. With the continuous development of molecular biology and nanotechnology in recent years, some new detection methods have gradually been applied to microbial detection [8-11]. The use of nanoparticles for bacterial detection is a typical application of nanoparticles in the field of biology. The basic principle is to connect the nanoparticle with the bacterial identifier, and to achieve the sensitive detection of the bacteria through specific recognition of the bacterial cell by the coupling body and the fluorescence signal amplification of the nanoparticles. Magnetic nanoparticles can also be used to separate bacteria first and then to arrange detection assays [12]. Kaittanis et al. [13] has reported detection of mycobacterium avium by nuclear magnetic resonance using superparamagnetic nanoparticles bound to specific antibodies, and found a sensitivity of 1500 CFU/mL. Compared with conventional blood culture detection methods, the application of nanoparticles to bacterial detection improves detection sensitivity; Moreover, nanoparticles can detect dozens of bacteria with a short time, and the analysis and testing time is generally within a few hours [14-17].

The clinical manifestations, pathogens and drug-resistant strains of sepsis in VLBWIs vary greatly in different regions and hospitals [18]. At present, there are few studies on the application of nanoparticles in detection of sepsis bacteria in VLBWIs. In order to improve the level of diagnosis and treatment, we analyzed the distribution and drug resistance of pathogenic bacteria of sepsis in VLBWIs, and discussed the incidence and high risk factors of sepsis in hospitalized neonates, so as to provide reference for early preventive measures.

Materials and methods

Study participants

In this retrospective study, 69 cases of VLBWIs admitted to the Neonatal Care Department of Qilu Hospital (Qingdao), Cheeloo College of Medicine, Shandong University from January 01, 2014 to December 31, 2020 were selected. Among them, 48 cases of VLBWIs developed sepsis, and 49 strains of pathogenic bacteria were isolated from their blood culture-positive venous blood specimens. This study was approved by the Ethics Committee of Cheeloo College of Medicine, Shandong University (2021 No. 101701). The study was conducted with the consent of the subjects' guardians, who provided the written informed consent.

Inclusion and exclusion criteria

Inclusion criteria: All enrolled VLBWIs meet the diagnostic criteria for sepsis [19], with clinical manifestations such as fever or low body temperature, refusal to milk, non-weight gain, jaundice and shock, as well as pathogenic bacteria detected in blood culture or sterile body cavity; VLBWIs with either EOS (onset within 3 days of birth) or late-onset sepsis (onset after 3 days of birth); gestational age <32 weeks, birth weight <1,500 g. Exclusion criteria: gestational age ≥32 weeks, death within 72 hours of birth, congenital malformations and genetic metabolic diseases, or those who gave up treatment.

Identification of bacteria by fluorescent nanoparticles

Bacteria have antigens on their surfaces, which are the sites where antibodies bind to bacteria. The surface of the nanoparticle is combined with the antibody to form a nanoparticle-antibody conjugates. The surface of the nanoparticles combines with the antibody to form the antibody coupling of the nanoparticles. The nanoparticles can specifically recognize the target bacteria through the interaction between the antibodies bound to the surface of the nanoparticles and the bacterial surface antigen. Bacteria combined with a large number of nanoparticles can produce extremely strong fluorescence under the excitation of laser. High sensitivity detection of bacteria can be achieved by detecting fluorescence.

Preparation of fluorescent nanoparticles: in the presence of non-ionic surfactants (Biolab Technology Co., Ltd., Beijing, China, Cat. No. L10308-DFA), the nanoparticles were prepared by a water-in-oil microemulsion method. With RuBpy as the core and silica as the shell, the

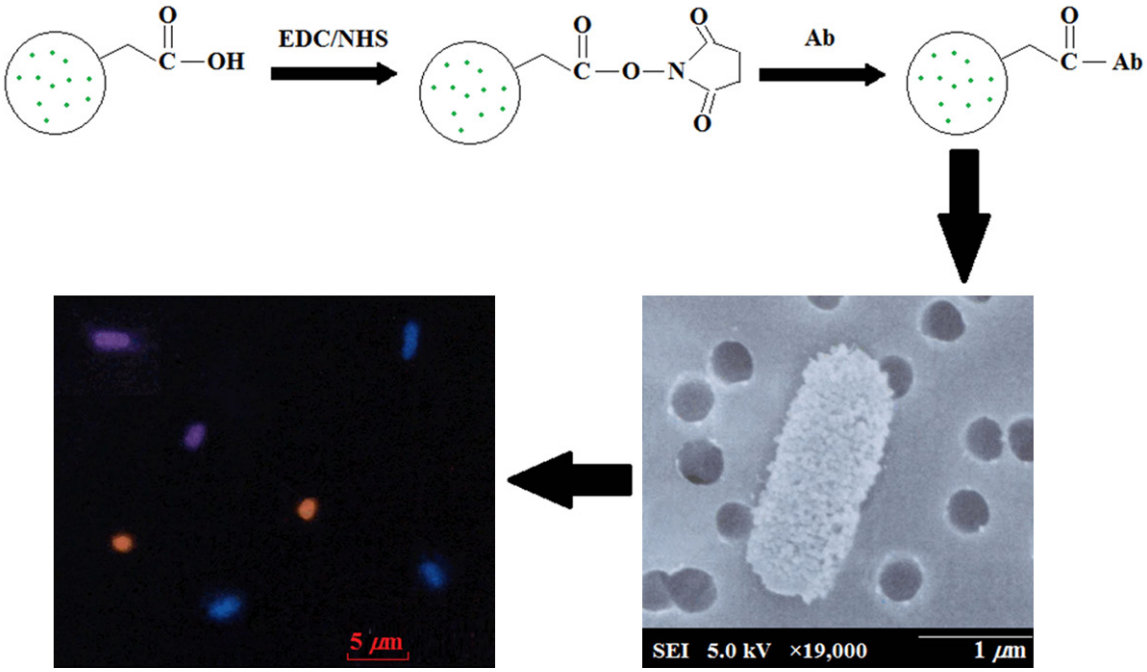

Figure 1. Carboxyl modified nanoparticles bound to antibodies and the detection of pathogenic bacteria by fluorescent nanoparticles.

core-shell structured nanoparticles were prepared. The synthesized nanoparticles were characterized by transmission electron microscopy. The carboxyl modification reagent was added to the prepared nanoparticle solution, for oscillatory reaction for 24 hours. Carboxyl modified nanoparticles (10 mg) were uniformly dispersed in the solution by ultrasonic oscillation, and centrifuged at 14,000 rpm for 10 min. After supernatant removal, 1 mL 0.1 M MES buffer (Cat. No. XG-R95485), 12 mg EDC (XG-R96742), and 33 mg Sulfo-NHS (XG-R96655) were added, all purchased from Sig Biotechnology Co., Ltd., Shanghai, China, for oscillating reaction for 20 min. Thereafter, it was centrifuged at 14,000 rpm for 10 min, and the supernatant was discarded. Subsequently, it was added with 150 μg of antibody for 3 h of oscillating reaction, and then centrifuged at 14,000 rpm for 10 min to discard the supernatant. The binding process of carboxyl modified nanoparticles and antibodies is shown in **Figure 1**. Antibody-labeled nanoparticles (20 μL) were added to 100 μL of peripheral blood or cerebrospinal fluid and phosphate buffer (Biolab Technology Co., Ltd., Beijing, China, Cat. No. GL0999-0CH). After 10 min of oscillating reaction, the mixture was centrifuged at 4,000 rpm for 10 min, and the supernatant was discarded. Scanning electron microscopy (Phenom, Shanghai, China) was used to characterize nano-particle-bacterial couplings.

Blood culture identification of strains and drug sensitivity test

Siemens MicroScan WalkAway 96 PLUS (Beckman Caulter Diagnostics, USA) and VITEK 2 compact automatic bacteria identification and susceptibility analysis system (Mérieux, France) were used for the identification and susceptibility of bacteria. Fungus identification and susceptibility used ATB-FUNGUS Fungus Identification and susceptibility test strip (BioMérieux, Shanghai, China). The results were interpreted on the NEW ATB microbial identification and susceptibility analysis system (BioMérieux, Shanghai, China). Quality control strains were provided by the Clinical Laboratory Center of the Ministry of Health.

Analysis of risk factors of EOS in VLBWIs

The medical records of the newborns and the parturients were reviewed to record the gender of the child, cesarean section, premature delivery, amniotic fluid pollution, neonatal asphyxia, intrauterine distress, premature rupture of

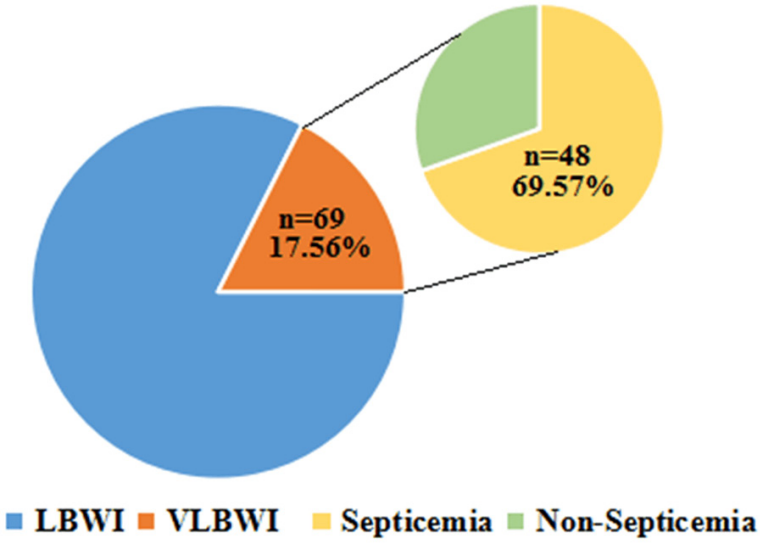

Figure 2. Incidence of sepsis in VLBWIs.

membranes, maternal fever, umbilical infection, lung infection, invasive operation, improper feeding, pregnancy-induced hypertension, gestational diabetes, and senile parturient.

Statistical analysis

SPSS23 statistical software was used to analyze the data. The measurement data were described by the $\bar{x}\pm s$, and the t test was used for comparison between groups. The categorical data were described by frequency (percentage), and the chi-square test was used for comparison between groups. $P<0.05$ was considered to be statistically significant. Binary logistic regression analysis was used to analyze the risk factors.

Results

Incidence of EOS in VLBWIs

During 2014.01.01-2020.12.31, the total number of newborns discharged from hospital was 2485, of which 393 (15.81%) were low birth weight infants and 69 (2.77%) were VLBWIs. Venous blood was collected from 69 newborns for blood culture, of which 48 were positive for blood culture, with a positive rate of 69.57% (**Figure 2**). Thirty-four cases had EOS, with an incidence rate of 49.27%.

Pathogen composition of EOS in VLBWIs

A total of 49 pathogenic bacteria were isolated from the venous blood of 48 VLBWIs with sepsis. Among the 49 pathogenic bacteria, there were 27 gram-negative bacteria (55.1%), mainly *Escherichia coli* (*E. coli*) and *Klebsiella pneumoniae* (*KPN*). There were 21 strains (42.86%) of Gram-positive bacteria, mainly *Staphylococcus epidermidis (SE)* and *Streptococcus agalactiae (SA)*. One strain (2.04%) of the fungus was *Candida albicans*. Statistical analysis showed that there was no significant difference in the constituent ratio of 49 strains of pathogenic bacteria between Gram-negative bacteria and Gram-positive bacteria ($P>0.05$), as shown in **Table 1**. There were 34 strains (69.39%) of pathogenic bacteria causing EOS, and 15 strains (30.61%) of pathogenic bacteria causing LOS. Statistical analysis revealed that the isolates of EOS were significantly higher than those of late-onset sepsis ($P<0.05$), as shown in **Table 2**.

Resistance of gram-negative bacteria to antibacterial drugs

The Gram-negative bacteria infected by EOS in newborns were mainly *E. coli* and *KPN*. The detection rates of *E. coli* and *KPN* were 42.85% and 35.71%, respectively. There were differences in their resistance to antibacterial drugs. *E. coli* had a high resistance rate to ampicillin, cefazolin and tetracycline antibacterial drugs. *KPN* had a higher resistance rate to ampicillin, piperacillin and tetracycline. The two pathogens were highly sensitive to imipenem, meropenem, piperacillin/tazobactam, as shown in **Table 3**.

Resistance of gram-positive bacteria to antibacterial drugs

The main Gram-positive bacteria inducing neonatal EOS were *SE* and *SA*. The detection rates of *SA* and methicillin-resistant coagulase-negative *Staphylococcus* were 38.46% and 53.84%, respectively. *Staphylococcus epidermidis* was highly resistant to cephalosporins, erythromycins and penicillins, and was sensitive to gentamicin, linezolid, minocycline, rifampicin, teicoplanin, and vancomycin. *Streptococcus agalactiae* had a high resistance rate to azithromycin,

Table 1. Composition of pathogenic bacteria of sepsis in VLBWIs

Pathogenic bacteria		Bacterial strain	Constituent ratio (%)
Gram-negative bacteria	Escherichia coli	12	24.49
	Klebsiella pneumoniae	9	18.37
	Enterobacter cloacae	3	6.12
	Pseudomonas aeruginosa	1	2.04
	Citrobacter freundii	1	2.04
	Shigella sonnei	1	2.04
Gram-positive bacteria	Staphylococcus epidermidis	8	16.33
	Streptococcus agalactiae	7	14.29
	Staphylococcus aureus	4	8.16
	Listeria monocytogenes	1	2.04
	Coagulase negative staphylococcus	1	2.04
Fungus	Candida albicans	1	2.04
Total		49	100.00
χ^2	1.020		
P	0.312		

NOTE: VLBWIs: very low birth weight infants; EOS: early-onset sepsis; LOS: late-onset sepsis.

Table 2. Composition of EOS and LOS pathogens in VLBWIs

Pathogenic bacteria		EOS		LOS		χ^2	P
		Bacterial strain	constituent ratio (%)	Bacterial strain	constituent ratio (%)		
Gram-negative bacteria	Escherichia coli	8	23.53	4	26.66		
	Klebsiella pneumoniae	6	17.65	3	20.00		
	Enterobacter cloacae	2	5.88	1	6.67		
	Pseudomonas aeruginosa	0	0.00	1	6.67		
	Citrobacter freundii	1	2.94	0	0.00		
	Shigella sonnei	1	2.94	0	0.00		
Gram-positive bacteria	Staphylococcus epidermidis	6	17.65	2	13.33		
	Streptococcus agalactiae	7	20.59	0	0.00		
	Staphylococcus aureus	1	2.94	3	20.00		
	Listeria monocytogenes	1	2.94	0	0.00		
	Coagulase negative staphylococcus	1	2.94	0	0.00		
Fungus	Candida albicans	0	0.00	1	6.67		
Total		34	100.00	15	100.00	14.731	<0.001

NOTE: VLBWIs: very low birth weight infants; EOS: early-onset sepsis; LOS: late-onset sepsis.

erythromycin, tetracycline, etc., and was sensitive to ampicillin, penicillin, rifampicin, and vancomycin. See **Table 4**.

Analysis of risk factors of EOS in VLBWIs

Thirty-four VLBWIs with EOS were assigned to the EOS group, 14 VLBWIs with LOS were included to the LOS group, and 21 VLBWIs without sepsis were used as the control group. The general information of the three groups of VLBWIs is shown in **Table 5**. Among the various variables, premature birth, amniotic fluid pollution, neonatal asphyxia, intrauterine distress, premature rupture of membranes, maternal fever, umbilical infection, lung infection, and improper feeding had statistically significant differences among the three groups. Binary logistic regression analysis was then performed with EOS as the dependent variable (1=EOS, 0=LOS and no sepsis), and the above statistically different factors as independent variables. It was found that the OR values of amniotic fluid pollution, intrauterine distress, premature rupture of membranes, maternal fever, lung infection, umbilical infection, and neonatal asphyxia

Table 3. Resistance of major GNB to antibacterial drugs

GNB commonly used antibacterial drugs	E.coli (n=8)		KPN (n=6)	
	Bacterial strain	Drug resistance rate (%)	Bacterial strain	Drug resistance rate (%)
Ampicillin	4	50.00	4	66.66
Ampicillin/Sulbactam	0	0	2	33.33
Aztreonam	1	12.50	1	16.66
Ceftazidime	1	12.50	1	16.66
Cefoxitin	0	0	1	16.66
Cefazolin	4	50.00	2	33.33
Ceftriaxone	3	37.50	2	33.33
Cefotaxime	3	37.50	2	33.33
Cefuroxime	3	37.50	2	33.33
Cefepime	1	12.50	0	0
Ciprofloxacin	3	37.50	1	16.66
Gentamicin	3	37.50	1	16.66
Imipenem	0	0	0	0
levofloxacin	2	25.00	0	0
Minocycline	0	0	2	33.33
Meropenem	0	0	0	0
Piperacillin	2	25.00	3	50.0
Piperacillin/Tazobactam	0	0	0	0
Tetracycline	4	50.00	4	66.66

NOTE: GNB: gram-negative bacteria; E.coli: Escherichia coli; KPN: klebsiella pneumoniae.

Table 4. Resistance of major GPB to antibacterial drugs

GPB commonly used antibacterial drugs	SE (n=6)		SA (n=7)	
	Bacterial strain	Drug resistance rate (%)	Bacterial strain	Drug resistance rate (%)
Amoxicillin/clavulanic acid	4	66.66	-	-
Azithromycin	5	83.33	5	71.42
Ampicillin	5	83.33	0	0
Cefoxitin	5	83.33	-	-
Cefazolin	5	83.33	-	-
Ciprofloxacin	2	33.33	-	-
Clindamycin	2	33.33	3	42.85
Erythromycin	5	83.33	6	85.71
Gentamicin	0	0	-	-
Imipenem	3	50.00	-	-
Levofloxacin	2	33.33	3	42.85
Linezolid	0	0	-	-
Minocyline	0	0	-	-
Meropenem	4	66.66	-	-
Oxacillin	4	66.66	-	-
Penicillin	5	83.33	0	0
Rifampicin	0	0	0	0
Teicoplanin	0	0	-	-
Tetracycline	1	16.66	5	71.42
Vancomycin	0	0	0	0

NOTES: GPB: gram-positive bacteria; *SE*: *Staphylococcus epidermidis*; *SA: Streptococcus agalactiae.*

Table 5. General information on sepsis in VLBWIs

Factors	EOS group (n=34)	LOS group (n=14)	Control group (n=21)	χ^2	P
Cesarean section	18 (47.36)	6 (42.85)	10 (47.62)	0.437	0.804
Premature delivery	22 (64.70)	7 (50.0)	11 (52.38)	1.267	0.531
Amniotic fluid pollution	15 (44.11)	11 (78.57)	6(28.57)	8.582	**0.014**
Neonatal asphyxia	12 (35.29)	2 (14.28)	1 (4.76)	7.687	**0.021**
Intrauterine distress	8 (23.52)	1(7.14)	0 (0.0)	5.708	**0.032**
Premature rupture of membranes	9 (26.47)	2 (14.28)	0 (0.0)	6.824	**0.033**
Maternal fever	12 (35.29)	3 (21.42)	0 (0.0)	9.506	**0.009**
Umbilical infection	14 (41.17)	3 (21.42)	2 (9.52)	6.847	**0.033**
Lung infection	5 (14.70)	5 (35.71)	0 (0.0)	8.648	**0.013**
Invasive operation	15 (44.11)	3 (21.42)	1 (4.76)	10.405	**0.006**
Improper feeding	8 (23.52)	4 (28.57)	2 (9.52)	2.319	0.314
Pregnancy-induced hypertension	4 (11.76)	8 (57.14)	3 (14.28)	12.988	**0.002**
Gestational diabetes	13 (38.23)	3 (21.42)	2 (9.52)	5.748	0.056
Senile parturient	3 (8.82)	2 (14.28)	1 (4.76)	0.961	0.618
Cesarean section	12 (11.76)	2 (14.28)	4 (19.04)	0.556	0.757

NOTE: VLBWIs: very low birth weight infants.

Table 6. Analysis of risk factors for EOS in VLBWIs

	B	S.E	wald	P	OR	95% CI lower limit	95% CI upper limit
Premature delivery	0.502	0.730	0.473	0.491	1.652	0.395	6.911
Amniotic fluid pollution	2.237	0.977	5.244	0.022	**9.369**	1.380	63.584
Neonatal asphyxia	0.712	1.180	0.365	0.546	**2.039**	0.202	20.601
Intrauterine distress	1.827	1.041	3.08	0.079	**6.217**	0.808	47.841
Premature rupture of membranes	1.730	0.976	3.141	0.076	**5.638**	0.833	38.183
Maternal fever	1.451	0.928	2.445	0.118	**4.267**	0.692	26.302
Umbilical infection	0.733	0.911	0.647	0.421	**2.081**	0.349	12.416
Lung infection	1.036	0.883	1.374	0.241	**2.817**	0.499	15.916
Improper feeding	-0.945	0.905	1.090	0.296	0.389	0.066	2.291
Constant	-1.889	0.663	8.114	0.004	0.151		

NOTE: EOS: early-onset sepsis; VLBWIs: very low birth weight infants.

were all >2, which were 9.369, 6.217, 5.638, 4.267, 2.817, 2.081, and 2.039, respectively. See **Table 6**.

Discussion

Neonatal sepsis is characterized by low specificity of early symptoms, rapid onset, and rapid progress; Moreover, blood culture materials are not easy to obtain and specimens are easy to be contaminated. Given the long culturing cycle, it poses a greater challenge for the early diagnosis and treatment of the disease [20].

The clinical manifestations, types of infectious pathogens, and drug-resistant strains of sepsis in VLBWIs are quite different in different regions and hospitals [21]. In this study, we analyzed the distribution and drug resistance of the pathogenic bacteria of sepsis in VLBWIs in our hospital. 69 cases of VLBWIs had a positive rate of 69.57% for bacterial detection. The main pathogens of infection in VLBWIs were gram-negative bacteria (55.1%), gram-positive bacteria (42.86%), and fungi (2.04%). Among them, *E.coli* accounted for 24.49%, *KPN* accounted for 18.37%, followed by *SE* and *SA,* and most of the pathogens were isolated from EOS. The results suggest that the bacterial infections of VLBWIs in this hospital are mainly *E.coli, KPN, SE,* and *SA*. The research from Lamba et al. [22] has shown that Gram-negative bacteria are the main cause of neonatal

sepsis, among which *coagulase-negative staphylococci* are the most common, followed by *Klebsiella*, reflecting the differences in the distribution of neonatal infection pathogens in different regions.

According to the time of onset, neonatal sepsis can be divided into EOS and LOS. EOS usually occurs within 3 days after birth, and is mostly related to prenatal or intrapartum infections [23]. While LOS occurs 3 days after birth and is mostly related to acquired infection, with a lower fatality rate than EOS [24]. It was found in this study that in neonatal sepsis caused by pathogenic bacteria, EOS (49.27%) was more prevalent than LOS (20.29%). The proportion (in descending order) of bacteria causing EOS was *E.coli*, *SA*, *KPN*, and *SE*. While *E.coli*, *KPN*, *SA*, and *SE* were the bacteria causing LOS in descending order. In addition, *Pseudomonas aeruginosa* mainly causes late-onset infections. Fungal infections can cause late-onset infections. *Citrobacter freundii*, *Shigella sonnei*, *SA*, *Listeria*, and *coagulase-negative staphylococci* mainly cause early-onset infections.

E.coli has a high resistance rate to ampicillin, cefazolin and tetracycline antibacterial drugs. *KPN* is highly resistant to ampicillin, piperacillin and tetracycline. These two pathogens are highly sensitive to imipenem, meropenem, and piperacillin/tazobactam. Gram-negative bacteria can use carbapenems, and β-lactamase inhibitors as the first choice for the treatment of neonatal sepsis. *SE* has a high resistance rate to cephalosporins, erythromycins and penicillins; It is highly sensitive to gentamicin, linezolid, minocycline, rifampicin, teicoplanin, and vancomycin, and can be used as the drug of choice against Staphylococcus epidermidis. *SA* has a high resistance rate to azithromycin, erythromycin, tetracycline, etc; It is highly sensitive to ampicillin, penicillin, rifampicin, and vancomycin, and can be used as the drug of choice against Streptococcus agalactiae. In summary, we should select antibiotics according to the distribution characteristics of pathogens and drug susceptibility to avoid abuse of antibiotics and reduce the production of drug-resistant strains [25].

The main risk factors of EOS in VLBWIs are amniotic fluid pollution, intrauterine distress, premature rupture of membranes, and maternal fever. Amniotic fluid pollution: When the fetus has hypoxic acidosis in the uterus, the intestinal peristalsis is hyperactive, which promotes the excretion of meconium into the amniotic fluid. Prolonged exposure of the fetus to amniotic fluid containing meconium will increase the occurrence of fetal intrauterine infection [26]. Therefore, oxygen inhalation, monitoring, fluid infusion and symptomatic treatment should be carried out when amniotic fluid pollution occurs during pregnancy and childbirth. Intrauterine distress is a syndrome mainly characterized by fetal hypoxia. Intrauterine distress can cause amniotic fluid pollution, as well as abnormalities in the fetal mucosal barrier and function, which can increase the risk of infection. Premature rupture of membranes is closely related to bacterial infection. Phospholipase A produced by genital tract bacteria such as Escherichia coli can decompose collagen on fetal membranes, leading to the destruction of membrane structure [27]. Long-term use of broad-spectrum antibiotics should be avoided during pregnancy and childbirth, otherwise, it can lead to flora disequilibrium and premature rupture of membranes. It has been shown that premature rupture of membranes is the main risk factor for neonatal sepsis [28, 29]. Women in the third trimester of pregnancy are susceptible to infection with Streptococcus agalactiae. Once infection occurs, it is easily transmitted vertically between mother and child, causing neonatal pneumonia and sepsis [30, 31]. If the mother's body temperature exceeds 38°C during childbirth, infection should be considered. Studies have confirmed that mothers with EOS have fever during delivery [32]. Early maternal temperature screening can provide evidence for the preventive use of antibiotics for mothers and newborns.

Conclusion

In this retrospective analysis, 48 of the 69 VLBWIs developed sepsis, 34 of which had EOS. Gram-negative bacteria are mainly *E.coli* and *KPN*. The two pathogenic bacteria have high sensitivity to β-lactamase inhibitor-containing compound preparations, and carbapenem antibacterial drugs, and can be used as the first choice. Gram-positive bacteria are mainly *SE* and *SA*. *SE* is highly resistant to penicillins and can be treated with vancomycin. *SA* is highly resistant to erythromycin and can be

treated with penicillin and vancomycin. Amniotic fluid pollution, intrauterine distress, premature rupture of membranes, and maternal fever are risk factors for EOS in VLBWIs. In summary, timely and reasonable treatment should be based on the types and drug resistance characteristics of neonatal sepsis pathogens and the risk factors of EOS. Given the small sample size resulting from the relatively low proportion of VLBWIs in our hospital in the past 7 years, there may be some errors in the distribution of pathogens detected. Therefore, we continue to collect and observe the types and drug resistance characteristics of pathogenic bacteria in VLBWIs in our hospital to improve the diagnosis and treatment.

Disclosure of conflict of interest

None.

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