Surveillance of Nosocomial Infections in a Bulgarian Neonatal Intensive Care Unit

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Abstract

Introduction: Nosocomial infections (NI) are frequent complications in neonatal intensive care units (NICU) which result in high morbidity and mortality.

Aim: To determine and analyze the incidence, risk factors and etiologic agents of NI in newborns admitted in the NICU to help planning future surveillance and prevention strategies.

Materials and methods: A prospective cohort study was carried out at the NICU of St George University Hospital, Plovdiv, Bulgaria from January 1, 2017 to June 31, 2018. The number of neonates included in the study was 507. Descriptive statistics such as count, percent, mean and standard deviation was used. Chi-square test was performed to prove associations. Odds ratios, with 95% confidence intervals, were computed from the results of the binominal logistic regression analyses.

Results: Of the 507 hospitalized newborns in NICU, 48 presented with 54 NI. The incidence and the density incidence rates were 9.5% and 7.67 per 1,000 patient-days, respectively. Nosocomial infections were detected in neonates from all birth weight (BW) classes, but it was low BW and premature neonates that were at major risk to acquire them. The most common infection sites were ventilator-associated pneumonia (VAP) (67.27%), bloodstream infection (23.64%) and conjunctivitis (9.09%). Major pathogens were Gram-negative such as Klebsiella pneumoniae, E. coli, Pseudomonas aeruginosa and Acinetobacter baumannii. In the multivariate logistic regression analysis NIs were strongly associated with intubation, presence of a venous catheter, the duration of antibiotic treatment and increased CRP> 10 mg/l.

Conclusions: This report highlights the burden of NIs, identifies the major focus for future NI control and prevention programs.

Keywords

aetiology, incidence, newborn, nosocomial infection, risk factors

INTRODUCTION

Improvements in the neonatal intensive care have increased the survival of critically ill newborns.¹ At birth, newborns, especially premature and low birth weight neonates, are devoid of efficient structural barriers of a protective endogenous microbial flora and of a mature immune system.²³ Furthermore, this population of vulnerable patients is often dependent on invasive procedures that are associated with a higher risk for infections. Nosocomial infections (NI) have become a major problem in many neonatal intensive...
care units (NICUs) that complicate the hospitalization of patients and result in considerable morbidity and mortality, increased length of stay and increased health care costs.4,5 The most frequent infections in NICUs are bloodstream infections, pneumonia, and necrotizing enterocolitis (NEC); less frequent complications are infections of the eyes, mouth or skin.6,7 In developed countries, the incidence rate of nosocomial infections ranges from 6 to 9 infections per 1000 patient-days, with 3- to 20-fold higher rates in developing countries.8,9 The major pathogens of neonatal infections differ not only from country to country and from nursery to nursery, but also change within years in the same nursery.10,11 Effective surveillance is important to evaluate the epidemiology, the associated risk factors, and the causative microorganisms. There are numerous risk factors for nosocomial infection among the newborns hospitalized for intensive care. The major risk factors can be categorized into intrinsic and extrinsic factors. The intrinsic factors include characteristics such as gestational age, birth weight, severity of the disease, immunologic maturity. The extrinsic factors include hospital stay, use of invasive devices, medications, exposure to hospital environment and hospital staff, hygiene and hospital infection control practices. Understanding the risk factors associated with nosocomial infections is a prerequisite for the development of prevention strategies.

AIM

The aims of this study were to determine and analyze the incidence, risk factors and etiology of nosocomial infections in newborns admitted to neonatal intensive care unit (NICU) in order to help planning future surveillance and prevention strategies.

MATERIALS AND METHODS

A prospective cohort study was carried out at the NICU of St George University Hospital in Plovdiv, Bulgaria between January 1, 2017 and June 31, 2018. Inclusion criteria: patients admitted to the intensive care unit during the study period with medical pathology for observation, diagnosis or treatment and hospitalized for more than 48 hours. Exclusion criteria: those patients whose stay in the NICU was less than 48 hours, and patients hospitalized before the beginning of the study. The inclusion criteria were met by 507 neonates and they were included in the study. To detect any risk factor of infection either maternal, natal or postnatal, a chart for epidemiologic surveillance was developed. The information collected for the mothers was as follows: complete obstetric history of the mother, age, previ-
Prospective Surveillance of NI in NICU

RESULTS

A total of 507 hospitalized newborns and 481 mothers entered the study. 48 neonates were diagnosed with 54 nosocomial infections giving an overall nosocomial infection rate of 9.5 per 100 hospitalized patients and a nosocomial infection incidence rate of 7.67 infections per 1000 patient-days.

The median age of the mothers of the studied neonates was 27.8 years; 25th percentile – 23.00 yrs., 75th percentile – 32.00 yrs (Table 1). Physician had tracked the pregnancy of 89.6% of all mothers. In almost one-fifth (18.5%) of the mothers, a pathologic condition prior to pregnancy was recorded and notably higher was the proportion of pregnant women with a pathologic pregnancy (36.2%). Among the mothers of infected neonates, the percentage of pathologic pregnancies was even higher - 51.06%. We found an association between pathologic pregnancy and NI (χ²=5.001, p=0.025).

Higher proportion of the studied neonates were delivered through cesarean section (63.5%, 322/507) and we found an association between the method of delivery and NI (χ²=17.035, p=0.000). Infection was more common in males (60.42%, 29/48) compared to females (39.58%, 19/48). Most of them were premature neonates (68.75%, 33/48) and low birth weight (66.67%, 32/48) (Table 2). There is a statistically significant difference between the mean birth weight and gestational age of infected and non-infected neonates.

Ventilator-associated pneumonia (VAP) was the most common NI (67.27%, n=36), followed by bloodstream infection (23.64%, n=13) and conjunctivitis (9.09% n=5). The median time to NI diagnose was 8.5 days. During the study period 4 neonates with NI died which resulted in lethality rate of 8.33%.

Gram-negative bacteria (74.16%) were the most prevalent pathogenic cause of nosocomial infection in our research. Four microorganisms (Klebsiella pneumoniae, E. coli, Pseudomonas aeruginosa, Acinetobacter baumannii) from the Gram-negative flora were most commonly isolated. From the Gram-positive bacteria, the leading pathogens were Coagulase-negative Staphylococcus (CoNS) (13.49%) and Enterococcus faecalis (7.87%). The distribution of pathogens is demonstrated in Table 3.

For the period of the study, 7175 patient-days were registered, the median hospital stay for the neonates being 10 days: 25th percentile - 7.00 days, 75th percentile - 17.00 days. Infected patients had a significantly longer median hospital stay (26.50 days: 25th percentile - 18.25 days, 75th percentile - 44.75 days) than non-infected patients (10.00 days: 25th percentile - 7 days, 75th percentile - 14 days) (t=7.22, p<0.0001). The use of invasive devices in patients with NI was statistically significantly longer than in patients without NI (Table 2).

Birth weight and the medical reason for admission were directly associated with the length of hospital stay. In the

Table 1. Characteristics of the mothers of neonates with or without nosocomial infection

| Variables                          | Mothers of neonates with NI (n=47) | Mothers of neonates without NI (n=434) | p-value |
|------------------------------------|------------------------------------|---------------------------------------|---------|
| Mother’s age, mean±SD              | 28.57±6.43                         | 27.75±6.67                            | 0.418   |
| Antenatal care (visits), n (%)     |                                    |                                       |         |
| Yes                                | 44 (93.7)                          | 387 (89.17)                           | 0.343   |
| No                                 | 3 (6.3)                            | 47 (10.83)                            |         |
| Maternal disease before pregnancy, n (%) |                             |                                       |         |
| Yes                                | 9 (19.15)                          | 80 (18.43)                            | 0.90    |
| No                                 | 38 (80.85)                         | 354 (81.57)                           |         |
| Maternal disease during pregnancy, n (%) |                                 |                                       |         |
| Yes                                | 24 (51.06)                         | 150 (34.56)                           | 0.025   |
| No                                 | 23 (48.94)                         | 284 (65.44)                           |         |
| PROM >18 h, n (%)                  |                                    |                                       |         |
| Yes                                | 5 (10.6)                           | 32 (7.37)                             | 0.425   |
| No                                 | 42 (89.4)                          | 402 (92.63)                           |         |
Table 2. Characteristics of the neonates with or without infection

| Characteristics                             | Yes (n=48) | No (n=459) | p-value |
|---------------------------------------------|-----------|-----------|--------|
| Mode of delivery, n (%)                     |           |           | 0.000  |
| Vaginal                                     | 20 (41.67)| 165 (35.95)|        |
| Cesarean                                    | 28 (58.33)| 294 (64.05)|        |
| Sex, n (%)                                  |           |           | 0.62   |
| Male n (%)                                  | 29 (60.42)| 260 (56.64)|        |
| Female n (%)                                | 19 (39.58)| 199 (43.36)|        |
| Gestational age, wk, n (%)                  |           |           |        |
| ≤28                                         | 14 (29.17)| 16 (3.49)  |        |
| 28-32                                       | 10 (20.83)| 23 (5.01)  | 0.000  |
| 32-36.6                                     | 9 (18.75)| 169 (36.82)|        |
| ≥37                                         | 15 (31.25)| 251 (54.68)|        |
| Birth weight, gr n (%)                      |           |           |        |
| ≤999                                        | 11 (22.92)| 15 (3.27)  |        |
| 1000-1499                                   | 12 (25)   | 24 (5.23)  | 0.000  |
| 1500-2499                                   | 9 (18.75)| 154 (33.55)|        |
| ≥2500                                       | 16 (33.33)| 266 (57.95)|        |
| 1 minute APGAR score                       |           |           |        |
| 5.3±2.7                                     | 7.2±2     |           | 0.000  |
| 5 minute APGAR score                       | 8.4±2.1   | 9.2±1     |        |
| Hospital stay, days                         | 33.63±20.42| 12.12±9.16| 0.000  |
| Duration of CVC/UVC use, days               | 7.83±6.8  | 2.5±1.9   | 0.000  |
| Duration of PVC use, days                   | 32.5±20.25| 11.04±8.75| 0.000  |
| Length of mechanical ventilation, days      | 14.65±11.75| 3.68±4    | 0.000  |
| Duration of antibiotic treatment, days      | 29.27±18.71| 10.34±7.42| 0.000  |

Table 3. Pathogens isolated in patients with NI (n=89)

| Microorganism                        | Microorganisms in patients with NI n (%) |
|--------------------------------------|------------------------------------------|
| Gram positive                        |                                          |
| Coag. (-) Staphylococcus             | 12 (13.49)                               |
| MRSA                                 | 1 (1.12)                                 |
| Enterococcus faecalis                | 7 (7.86)                                 |
| Enterococcus faecium                 | 3 (3.37)                                 |
| Overall Gram-positive                | 23 (25.84)                               |
| Gram-negative                        |                                          |
| Klebsiella pneumoniae                | 19 (21.35)                               |
| Klebsiella oxytoxa ESBL+             | 4 (4.49)                                 |
| E. coli                              | 10 (11.24)                               |
| Enterobacter spp.                    | 4 (4.49)                                 |
| Pseudomonas aeruginosa               | 9 (10.12)                                |
| Acinetobacter baumannii              | 9 (10.12)                                |
| Acinetobacter Iwoffi                 | 2 (2.25)                                 |
| Stenotrophomonas maltophilia         | 4 (4.49)                                 |
| Chryseobacterium spp.                | 3 (3.37)                                 |
| Achromobacter spp.                   | 2 (2.25)                                 |
| Overall Gram-negative                | 66 (74.16)                               |

group of neonates weighing 2499-1500 g, the proportion of patients without NI was higher, whereas in the groups of very low (<1499 g) and extremely low birth weight neonates (<999 g) the proportion of patients with NI increased significantly. Another factor that can contribute to the longer hospital stay is the reason for intensive care admission (pre-existent morbidity). Among the infected neonates, the leading pathologic conditions that resulted in hospital admission were birth asphyxia, congenital pneumonia, and bronchopulmonary dysplasia.

Significant risk factors in the univariate analysis associated with NIs are presented in Table 4. We performed a binary logistic regression to ascertain the effects of the significant variables on the likelihood that participants have NIs. The logistic regression model was statistically significant ($\chi^2(4)=100.27, p<0.0005$). The model explained 40.0% (Nagelkerke $R^2$) of the variance in NIs and correctly classified 90.1% of cases. The independent risk factors in the multivariate analysis were intubation, increased CRP, duration of antibiotic therapy (>7 days), and PVC indwelling time (>14 days) (Table 5).
Table 4. Univariate analysis of the risk factors associated with nosocomial infections

| Variables                                      | Cases with NI (n=48) | Cases without NI (n=459) | ORa  | 95%CIb  | p-valuec |
|------------------------------------------------|----------------------|--------------------------|------|---------|----------|
| Birthweight (<2000 g)                          | 26                   | 85                       | 5.2  | 2.8-9.6 | <0.0001  |
| Gestational age (<37 weeks)                    | 31                   | 193                      | 2.5  | 1.3-4.6 | 0.003    |
| Gender (male/female)                           | 29                   | 260                      | 1.2  | 0.6-2.1 | 0.617    |
| Mode of delivery (vaginal/cesarean)            | 28                   | 294                      | 0.8  | 0.4-1.4 | 0.434    |
| Reanimation                                     | 38                   | 176                      | 6.1  | 2.9-12.6 | <0.0001  |
| Intubation                                      | 30                   | 68                       | 9.6  | 5.1-18.1 | <0.0001  |
| Pathologic conditions during pregnancy or at delivery | 24                   | 150                      | 1.9  | 1.1-3.6 | 0.027    |
| CVC/UVC                                        | 46                   | 353                      | 6.9  | 1.6-28.9 | 0.002    |
| CVC/UVC indwelling time (>14 days)              | 2                    | 6                        | 26.4 | 5.2-135.2 | <0.0001  |
| PVC indwelling time (>14 days)                  | 38                   | 90                       | 15.6 | 7.5-32.5 | <0.0001  |
| Duration of MV (>7 days)                       | 28                   | 10                       | 22.9 | 8.9-58.4 | <0.0001  |
| Patient days (>14 days)                        | 40                   | 108                      | 16.2 | 7.4-35.8 | <0.0001  |
| Duration of antibiotic therapy (>7 days)        | 47                   | 236                      | 40.0 | 5.5-292.7 | <0.0001  |
| Duration of antibiotic therapy (>14 days)       | 37                   | 70                       | 17.6 | 8.6-36.2 | <0.0001  |
| Increased CRP>10 mg/l                          | 22                   | 55                       | 6.3  | 3.3-11.9 | <0.0001  |

a: odds ratio; b: 95% confidence interval; c: Pearson χ²; d: Fisher exact probability test

Table 5. Risk factors for nosocomial infections in the binary logistic regression analysis

| Variables                                      | ORa  | 95% CIb  | p-valuec |
|------------------------------------------------|------|---------|----------|
| Intubation                                      | 4.7  | 2.3-9.7 | 0.000    |
| Increased CRP>10 mg/l                          | 2.4  | 1.2-5.2 | 0.016    |
| Duration of antibiotic therapy (>7 days)        | 11.4 | 1.4-90.8 | 0.022   |
| PVC indwelling time (>14 days)                  | 4.0  | 1.8-9.1 | 0.001    |

a: odds ratio; b: 95% confidence interval

DISCUSSION

Nosocomial infections in the NICU are a relevant, major medical problem. As much as 9.5% (7.67/1000 patient-days) of the patients in our NICU were diagnosed with NI, with 47.92% of infections occurring in very low birth weight infants (<1499 g). Previous study in the same NICU in 2012 revealed a median incidence rate of NI of 12.2 per 100 hospitalized patients14 which shows a non-significant decline in the incidence rate. In our country there are a limited number of studies considering neonatal nosocomial infections. Gladilova and Riberova15 for the period 2010-2011 discovered that 2% of all hospitalized neonates were diagnosed with a nosocomial infection. A research paper from 2014 of the National Center of Infectious and Parasitic Diseases (NCIPD) identifies 1062 NIs for the whole country and an incidence rate of 1.93%.16 In this report, the information from physiological nurseries and NICUs is not separated. The incidence rates from our study are higher compared with the few published papers in the country. This can be explained by the prospective monitoring and the active registration of the cases of NI. On the other hand, the cited studies from Bulgaria are based on official data from the system for passive surveillance of NI. Another problem present in our country is that a lot of the cases of NI are not proven microbiologically.17

The incidence rates of NI reported by research from other countries vary widely: 6–50 per 100 hospitalized patients and 5–62 per 1000 patient-days18-20, which is consistent with our results. Apart from that, it is very difficult to compare data from different studies because of the differences in study methodology, populations included, infection detection methods, and definitions used for NI.

It is worthy of note that 51.06% of the women in the study had some pathological condition during pregnancy (Table 1). An association was discovered between pathology during pregnancy and NI (χ²=5.001, p=0.025). We believe that it is a factor related to the mother that might increase the risk of NI in neonates, although in the binary logistic regression it showed to be non-significant (p>0.05).
The premature rupture of membranes >18 h is a factor discussed in the literature that might increase the risk of sepsis in newborns. According to WHO, in 8% of pregnant women PROM >18 h is being registered annually, although in some countries, such as China, the rates can be even 19%. In our study PROM was registered in 7.7% of all pregnant women and the proportion of the mothers of infected neonates with PROM was even higher - 10.42%, but we were unable to find an association with NI.

Most of the neonates included in the study were born by cesarean section (63.5%). In a similar study in Greece, the authors observed even higher proportion of cesarean section (71.7%). According to recent guidelines, cesarean births shouldn’t exceed 19 of 100 neonates. We found an association between the mode of delivery and NI and other researchers have also found a similar relation between NI and cesarean section.

Babanzono et al. have analyzed risk factors for NI in NICU and showed that the infection incidence was significantly higher in boys (OR 1.28; 95% CI 0.43-3.75). Our study also suggests higher susceptibility of male babies to NI (OR 1.2; 95% 0.6-2.1). In this study we also observed the highest incidence of NI in patients weighing less than 1499 g (Table 1). Birth weight is an important factor that can increase the risk of infection. A 3% increase in the rate of NI for every 500 g decrement in birth weight has been previously reported.

The risk of infection in our study was also inversely related to the gestational age. We found the highest incidence of NI in the group of neonates age 32 weeks or less (Table 1) and an association was observed between premature birth and NI (χ²=51.542, p=0.000). According to Glenn Mayhall gestational age is one of the key determinants for defining the risk of NI.

Pneumonia was the most frequent NI (67.27%), followed by bloodstream infection (23.64%) and conjunctivitis (9.09%). This distribution is in agreement with other studies. It is difficult to compare the results with studies from our country because there is no separate information for NICU. Gladilova and Ribarova in their research paper identified infections of the sensory organs as the most frequent infections in neonatology units, followed by respiratory, enteric and bloodstream infections. According to the literature, the bloodstream infections in the developed countries are the leading NI in NICU followed by pneumonia. These differences in distribution might be explained by the methodologies used for diagnosis, but the consistent variations between studies from developed and developing countries does suggest that the diversity in infection control standards and in clinical practices are probably also important.

The median time to NI diagnosis was 8.5 days in our study. Other authors have observed median time to diagnosis of 15-19 days. The lethality among patients with NI in our study was 8.33% and this correlates with results of other researchers. It is very difficult to differentiate what is the role of the NI over this unfavourable outcome, but in our opinion the infection has aggravated the severe pathology diagnosed in the infected neonates and has contributed to the outcome.

The risk for NI is directly associated with the length of the hospital stay. There was a statistically significant difference in the duration of hospital stay between infected and non-infected neonates. The patients with NI had almost 3-fold longer hospital stay (Table 2). We think this might be explained by the severe pathology diagnosed in the infected neonates at the time of admission and the lower birth weight of those neonates on the other side compared to the group of non-infected neonates. These factors suggest a need for a longer duration of the hospital stay. Although we found a statistically significant difference between the two groups, the logistic regression analysis didn’t outline the hospital stay as a risk factor (p>0.05).

The invasive devices are part of the advances in intensive care that improve the survival of premature newborns, but on the other hand they increase the risk for NI. The venous cannulation is one of the most frequent procedure done in NICUs because it supports the provision of fluids and medications during the hospital stay. Alongside with the importance of venous catheterization for the intensive care, the venous catheters have proved as significant risk factor for NI. In the binary logistic regression, we observed peripheral venous catheter dwelling time > 14 days as an independent significant risk factor for NI. Other studies have also outlined IV cannulation as a significant factor.

Intubation in our study showed to be a significant risk factor in the binary regression analysis (OR 4.7; 95% CI 2.3-9.7). Mohammed et al. have also found that endotracheal intubation increases the risk for NI (OR 5.43; 95% CI 3.46-8.5).

The duration of antibiotic treatment longer than 7 days was a significant risk factor in our study (OR 11.4; 95% CI 1.4 - 90.8). Kuppala et al. has also showed that antibiotic therapy > 5 days increases the risk for NI (2.66; 95% CI 1.12-6.30).

The prevalence of Gram-negative microorganisms was statistically significant compared to Gram-positive microorganisms (74.16% and 25.84%, respectively). We found that Klebsiella spp. were the most frequently isolated microorganism in patients with NI (25.84%) followed by CoNS (13.49%), E. coli (11.24%) and Enterococcus spp. (11.24%). For our country, a study in 2014 outlined Klebsiella pneumoniae and Coagulase negative Staphylococcus as the leading pathogens in the neonatal units. Authors from Brazil, Italy, and Egypt have also observed Klebsiella spp. in the highest proportions among patients with NI. Gram-negative bacteria (Klebsiella spp. E. coli, Pseudomonas aeruginosa, Acinetobacter baumannii) were the most common agents causing the leading NI-VAP. Although a shift has been noted in developed countries in the last years towards Gram-positive bacteria as agents causing pneumonia Gram-negative bacteria still remain the leading pathogens in less developed countries.
CONCLUSIONS

The study outlines the major characteristics of NI in one of the largest neonatal intensive care units in the country. The most frequent pathogens causing nosocomial infections belonged to the Gram-negative flora: *Klebsiella pneumoniae*, *E. coli*, *Pseudomonas aeruginosa*, and *Acinetobacter baumannii*. Significant risk factors for NI were intubation, PVC indwelling time more than 14 days, duration of antibiotic treatment longer than 7 days and increased CRP > 10 mg/dl. Future interventions should focus on developing training programs and applying bundles for prevention of ventilator-associated pneumonia.

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Эпидемиологический надзор за нозокомиальными инфекциями в отделении интенсивной терапии новорожденных в Болгарии

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Резюме

Введение: Нозокомиальные инфекции (НИ) – частые осложнения в отделениях интенсивной терапии новорожденных (ОИТН), которые приводят к заболеваемости и смертности.

Цель: Выявить и проанализировать частоту, факторы риска и этиологические агенты НИ у новорожденных для поддержки будущих стратегий мониторинга и профилактики.

Материалы и методы: Проспективное когортное исследование проведено в ОИТН университетской больницы «Св. Георги», Пловдив, Болгария, с 1 января 2017 г. по 31 июня 2018 г. В исследование было включено 507 новорожденных. Использовались такие описательные статистические данные, как количество, процент, среднее значение и стандартная ошибка. Для подтверждения ассоциаций был проведен тест хи-квадрат. Отношение вероятностей с интервальной конфиденциальностью 95% было рассчитано по результатам биномиального логистического регрессионного анализа.

Результаты: Из 507 новорожденных, госпитализированных в ОИТН, 48 поступило с 54 НИ. Процент и частота заболеваний составили 9.5% и 7.67 на 1000 пациенто-дней соответственно. Нозокомиальные инфекции были обнаружены у новорожденных всех весовых категорий (ВК), но именно новорожденные низких ВК и недоношенные дети были подвержены высокому риску заражения. Наиболее частыми инфекциями были вентиляторассоциированная пневмония (ВАП) (67.27%), инфекция кровотока (23.64%) и коньюнктивит (9.09%). Основными возбудителями болезни были грамотрицательные бактерии, такие как Klebsiella pneumoniae, E. coli, Pseudomonas aeruginosa и Acinetobacter baumannii. В многомерном логистическом регрессионном анализе НИ были тесно связаны с интубацией, наличием венозного катетера, продолжительностью лечения антибиотиками и повышенным уровнем С-реактивного белка CRP > 10 mg/l.

Заключение: В этом докладе подчеркивается тяжесть НИ, определяются основные направления будущих программ контроля и профилактики НИ.

Ключевые слова: этиология, заболеваемость, новорожденный, нозокомиальная инфекция, факторы риска