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Study of antibiotic resistance in the neonatology department of the Ibn Sina University Hospital in Rabat, Morocco.

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ABSTRACT:

Nosocomial infections are a serious public health problem, leading to high rates of morbidity and mortality as well as high expenditures. **Objective:** To describe the prevalence and characteristics of nosocomial neonatal bacterial infections and their resistance to antibiotics in a group of Moroccan newborns.

Material and methods: We included hospitalized neonates managed for nosocomial bacterial infections over a period from March 1 to June 31, 2022. **Results:** The incidence rate of healthcare-associated infections during the study period was 18.7%, and the incidence density was 10 cases/1000 DH. The results show a predominance of males (65.67%), most hospitalized within the first 24 hours (82.08%), with a significant proportion of low-weight newborns (61.19%). The reasons for hospitalization were respiratory distress (71.64%), perinatal asphyxia (26.86%), neonatal bacterial infections (16.41%), prematurity (16.41%), and oesophageal atresia (14.92%). Concerning the resistance and sensitivity of germs in nosocomial infections to TBAs: *coagulase-positive Staphylococcus* showed high rates of resistance to antibiotics, particularly gentamicin (96%), teicoplanin (96%) and amoxicillin (76%). However, it remains sensitive to vancomycin (100%). *Enterobacter cloacae* showed 100% resistance to ampicillin, Piperacillin/Tazobactam, Ceftriaxone, Ceftazidime, and Ciprofloxacin, and 50% to gentamicin, erythromycin, Meropenem, Cefoxitin, Trimethoprim/sulfamethoxazole and imipenem. *Acinetobacter baumannii* showed 100% resistance to amoxicillin and gentamicin, and 88% to ciprofloxacin and imipenem. *Escherichia coli* showed total resistance to ampicillin, amoxicillin/clavulanic acid, and trimethoprim/sulfamethoxazole, 85.7% to ceftazidime and 71.4% to gentamicin and ceftriaxone. *Klebsiella pneumoniae* was 100% resistant to gentamicin and 87.5% to several TBAs, including ampicillin. The outcome was favorable in 67.16% of cases.

Keywords: Antibiotics, Neonatal, Nosocomial, Resistance.

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I. INTRODUCTION:

Newborns are susceptible to healthcare-associated infections because of their immature immune systems. The likelihood of infection is increased by some factors, including premature birth, low birth weight, mother-to-child transmission, perinatal infections, and inappropriate use of antibiotics [1, 2, 3].

Neonatal sepsis is classified according to when it occurs: early sepsis occurs before 72 hours and late sepsis occurs after 72 hours [4, 5]. Research has also shown that neonatal sepsis is a major factor in infant and young child mortality, with 7% of infant mortality and 16% of neonatal deaths linked to neonatal sepsis [6]. To reduce the occurrence of neonatal infections, it is crucial to identify the micro-organisms concerned and assess their resistance to drugs [7].

In 2013, the CDC (Centers for Disease Control and Prevention) estimated that more than 20,000 deaths in the United States alone could be attributed to resistant bacteria. In 2016, the same number of deaths linked to antibiotic resistance were reported in France [8].

Various empirical therapeutic suggestions have been put forward for late-onset sepsis, including vancomycin combined with gentamicin for late-onset nosocomial sepsis [9].

Conversely, the spread of multi-resistant organisms hinders the development of effective treatments and reduces the range of suitable antibiotics. Institutional guidelines based on the prevalence of local microbes and their antibiotic susceptibility patterns are therefore needed [10].

A global threat has emerged in the form of antibiotic resistance. In developing countries, there are increasing reports, particularly in intensive care units, of multi-resistant bacteria (MRB) causing neonatal sepsis [11].

Morocco is one of the countries that have made maternal and neonatal morbidity and mortality a priority. In 2018, infant mortality fell to 18 deaths per 1000 live births, with 75% of deaths occurring during the neonatal period (13.65 per 1000 live births) [12]. Despite this reduction, the morbidity rate remains high. It is estimated that the number of survivors is three to ten times higher than the number of neonatal deaths [13].

The aim of our study is to describe the incidence and characteristics of nosocomial bacterial neonatal infections and their resistance to antibiotics in a group of Moroccan newborns.

MATERIAL AND METHODS

Type and duration of the study

A prospective, descriptive, and analytical study was carried out at the National Reference Centre for Neonatology and Nutrition at the Children's Hospital, Rabat University Hospital, over a period running between March 1 to June 31, 2022.

Inclusion criteria

All admissions that developed a healthcare-associated infection were recorded. A healthcare-associated infection was defined according to the CDC definition

Data collection

We collected epidemiological data, maternal and obstetric, and clinical data on newborns, nosocomial neonatal infections, and the germs identified and their sensitivity profile.

Exclusion criteria

Newborns without symptoms of sepsis and bacteriological evidence were excluded.

Diagnostic and microbiological criteria:

A documented infection with positive bacteriological tests (including blood cultures (BC), umbilical venous catheters (UVC), cytobacteriological examination of urine (CBEU), thoracic drainage, urinary catheter, and cerebrospinal fluid (CSF). For all identified germs, an analysis of their sensitivity to antimicrobials was performed. The empirical antibiotic treatment regimens in our center include vancomycin, amikacin, or ciprofloxacin while awaiting bacteriological data.

Statistical analysis:

-Jamovi: descriptive and analytical analysis, using the Chi2 test. A p-value <0.05 was considered significant with (95% CI).

II. RESULTS:

The study included 235 neonates hospitalized at the National Reference Centre for Neonatology and Nutrition at the Children's Hospital of the University Hospital of Rabat, Morocco during the study period, including neonates suspected of having a healthcare-associated infection. **(Figure 1).**

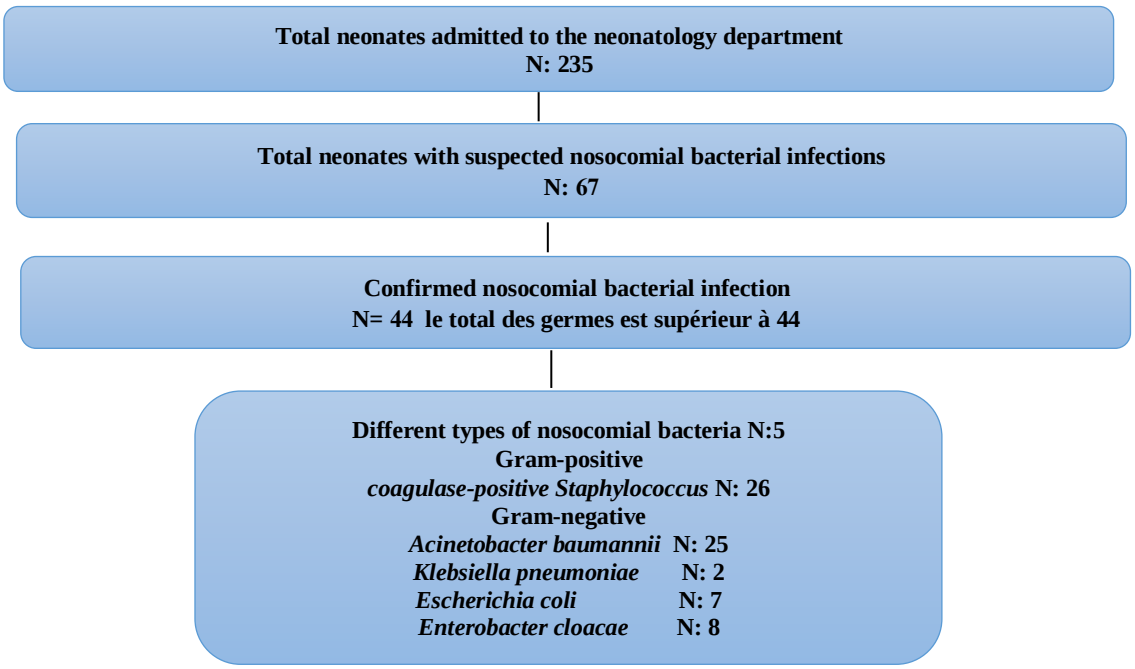

Figure 1: The study flowchart.

1. Maternal and obstetric characteristics

The results show a predominance of women aged between 19 and 30 (53.73%), most of whom were multiparous (61.19%). Most pregnancies were between 34-36 weeks (61.19%). Around 60% of pregnancies were adequately monitored. Vaginal delivery (50.74%) and cesarean section (49.25%). 20.89% reported infections during pregnancy. Maternal antecedents included: gestational diabetes in 7.46% of cases, gravidic hypertension in 5.97%, retroplacental haematoma in 2.98%, and pre-eclampsia in 1.49% **(Table 1)**.

Table 1: Maternal and obstetrical (n = 67)

Variable	N (%)
Age (Years)	
≤ 18	**2 (2.98)**
19–30	**36 (53.73)**
31–40	**24 (35.82)**
≥ 40	**5 (7.46)**
Gestity	
Primigest	**17 (25.37)**
Multigestivity	**50 (74.62)**
Parite	
Primipare	**26 (38.80)**
Multipare	**41 (61.19)**
Gestational age	
≤ 33	**15 (22.38)**
34-36-6 D.	**41 (61.19)**
At term	**6 (8.95)**
> 41+6 D.	**5 (7.46)**
Pregnancy monitoring	
Well followed	**40 (59.70)**
Not followed	**27 (40.29)**
Type of delivery:	
vaginal delivery	**34 (50.74)**
C-section	**33 (49.25)**
Infection during pregnancy	
YES	**14 (20.89)**
NO	**53 (79.10)**

C-section *cesarean section*, , D: days;

2. Neonatal characteristics

Data from 67 neonates with suspected nosocomial infection: male: 44 (65.67%), female: 23 (34.32%). Age at hospitalization: 0-24 h: 55 (82.08%), 1-7 d: 5 (7.46%), >7 d: 7 (10.44%). Birth weight (g): Hypotrophic (<2500g): 41 (61.19%), Eutrophic (2500-4000g): 22 (32.83%), Macrosomic (> 4000g): 4 (5.97%). APGAR was defined at 1, 5, and 10 minutes **(Table 2)**.

Table 2: Neonatal characteristics (n = 67)

Variable	N (%)
Gender	
Male	**44 (65.67)**
Female	**23 (34.32)**
Age at hospitalization	
0–24 h	**55 (82.08)**
1–7 d	**5 (7.46)**
>7 d	**7 (10.44)**
Birth weight (g)	
(< 2500g)	**41 (61.19)**
(2500–4000g)	**22 (32.83)**
(> 4000g)	**4 (5.97)**
APGAR	
1 minutes	
0-3	**11 (16.41)**
4-6	**6 (8.95)**
7-10	**37 (55.22)**
NM	**13 (19.40)**
5 minutes	
0-3	**3 (4.47)**
4-6	**13 (19.40)**
7-10	**40 (59.70)**
NM	**11 (16.41)**
10 minutes	
0-3	**1 (1.49)**
4-6	**4 (5.97)**
7-10	**50 (74.62)**
NM	**12 (17.91)**
Evolution:	
Alive	**45 (67.16)**
Deceased	**22 (32.83)**

D: days; H: hours; G: gram, NM: Not measured

3. Reasons for hospitalization:

The main reasons for hospitalization were respiratory distress 48 cases (71.64%), perinatal asphyxia 18 (26.86%), early bacterial neonatal infection 14 (20.90%), prematurity 11 (16.41%), oesophageal atresia 10 (14.92%), hyaline membrane disease 6 (8.95%) and meningitis 5 (7.5%) **(Table 3)**

Table 3: Hospitalization diagnosis of neonates (n = 67)

Input diagnostics	N (%)
Perinatal asphyxia	18 (26.9)
Neonatal bacterial infection Early	14 (20.90)
Premature	11 (16.41)
Esophageal atresia	10 (14.92)
Transient respiratory distress	8 (11.94)
Hyaline membrane disease	6 (8.95)
Meningitis	5 (7.5)
Congenital heart disease	2 (2.5)
Postnatal infection	2 (2.5)
Anal imperforation	1 (1.5)
Convulsion	1 (1.5)
Bladder exstrophy	1 (1.5)
Epispadias	1 (1.5)
Trisomy 18	1 (1.5)

4. Nosocomial neonatal infections

Nosocomial infection was confirmed in 44 of 67 suspected cases out of a total of 235 hospitalized patients. Sex ratio M/F 1.75 (28 Male, 16 Female), with an incidence rate of healthcare-associated infections during the study period of 18.7%, and an incidence density of 10 cases/1000 DH. The most common nosocomial germs: *Coagulase-positive Staphylococcus* in the logigram, noted as negative in 38.2% of cases, *Acinetobacter baumannii* 36.8%, *Klebsiella pneumoniae* 11.8%, *E. coli* 10.3% and *Enterobacter cloacae* 3% **(Table 4).**

Table 4: Prevalence of nosocomial bacterial neonatal infections

Pathogens	N= 68
coagulase-positive Staphylococcus	**26 (38.2%)**
Acinetobacter baumannii	**25 (36.8%)**
Klebsiella pneumoniae	**8 (11.8%)**
Escherichia coli	**7 (10.3%)**
Enterobacter cloacae	**2 (3%)**

5 Types of samples:

The samples most frequently taken to detect nosocomial bacterial infections were: blood culture (BC): 79.41%, umbilical venous catheters (UVC): 10.3%, cytobacteriological examination of urine (CBEU): 1.5%, pus sample (PUS): 2.94%, chest drain: 1.5%, urinary catheter: 1.5% and cerebrospinal fluid (CSF): 2.94% **(Table 5)**.

Table 5: The different types of samples and their percentages

Type bactérien		BC	UVC	CBEU	PUS	Drainage thoracique	Sounde urinea	CSF	TOTAL
Gram-positive	***coagulase-positive Staphylococcus***	**22(84.6%)**	**3(11.53%)**					**1(3.84%)**	26
Gram-negative	***Acinetobacter baumannii***	**20 (80%)**	**2(8%)**		**2(8%)**	**1(4%)**			25
	Enterobacter cloacae			**1(50%)**			**1(50%)**		2
	E. coli	**5 (71.42)**	**2 (28.57)**						**7**
	Klebsiella pneumoniae	**7(87.5%)**						**1(12.5%)**	8
Total		**54(79.41%)**	**7 (10.3%)**	**1(1.5%)**	**2(2.94%)**	**1(1.5%)**	**1(1.5%)**	**2(2.94%)**	68

6. Resistance and sensitivity profile of *coagulase-positive Staphylococcus* to antibiotics

Antibiotic resistance: Gentamicin: 96%, teicoplanin: 96%, amoxicillin: 76%, levofloxacin: 57.6%. Antibiotic sensitivity: Vancomycin: 100%, trimethoprim/sulfamethoxazole: 38.46% **(Table 6)**.

Table 6: Resistance and sensitivity of *coagulase-positive Staphylococcus* to antibiotics

Antibiotics	***coagulase-positive Staphylococcus*** **N: 26 Total: 68 germes**		
	Resistant	**Sensible**	**Intermediate**
Amoxicillin	**20 (76%)**	**1 (3.84%)**	**-**
Gentamicin	**25 (96%)**	**-**	**-**
Ampicillin	**4 (15%)**	**-**	**-**
Amoxicilline/ acide Clavulanique	**2 (7.6%)**	**-**	**-**
Piperacilline/Tazobactam	**1 (3.84%)**	**-**	**-**
Ceftriaxone	**3 (11.5%)**	**-**	**-**
Cetfazidime	**5 (19%)**	**-**	**-**
Ciprofloxacin	**2 (7.6%)**	**-**	**-**
levofloxacin	**15 (57.6 %)**	**4 (15%)**	**1 (3.84%)**
Erythromycin	**4 (15%)**	**-**	**-**
Imipinem	**4 (15%)**	**-**	**-**
Meropenem	**1 (3.84%)**	**-**	**-**
Cefepim	**2 (7.6%)**	**-**	**-**
Cefoxitin	**13 (50%)**	**-**	**-**
Vancomycin	**-**	**26 (100%)**	**-**
Téicoplanin	**25 (96%)**	**-**	**-**
Trimethoprim/sulfamethoxazole	**11 (42%)**	**10 (38.46%)**	**-**

7. Resistance and sensitivity profile of *Acinetobacter baumannii* to antibiotics:

Antibiotic resistance: Amoxicillin and gentamicin: 100%, ceftazidime, and trimethoprim/sulfamethoxazole: 96%, ciprofloxacin, and imipenem: 88%, piperacillin/tazobactam and meropenem: 76%, cefepime: 64%. Antibiotic sensitivity: Colistin: 96% **(Table 7)**.

8. Resistance and sensitivity profile of *Escherichia coli* to antibiotics

Antibiotic resistance: Ampicillin, amoxicillin/ Clavulanic acid, and trimethoprim/sulfamethoxazole: 100%, ceftazidime: 85.7%, gentamicin and ceftriaxone: 71.4%.

Antibiotic sensitivity: Imipenem: 100%, amoxicillin, and cefoxitin: 85.7%, erythromycin: 71.4% **(Table 7)**.

9. Resistance and sensitivity profile of *Enterobacter cloacae* to antibiotics

Antibiotic resistance: Ampicillin, amoxicillin/ Clavulanic acid, piperacillin/tazobactam, ceftriaxone, ceftazidime, and ciprofloxacin: 100%.

Antibiotic sensitivity: Amoxicillin: 100%; gentamicin, imipenem, trimethoprim/sulfamethoxazole **(Table 7)**.

10. Resistance and sensitivity profile of *Klebsiella pneumoniae* to antibiotics

Antibiotic resistance: Gentamicin: 100%; ampicillin, amoxicillin/clavulanic acid, ceftriaxone, ceftazidime, and trimethoprim/sulfamethoxazole: 87.5%; ciprofloxacin: 62%.

Antibiotic sensitivity: Imipenem: 75%; amoxicillin and erythromycin: 62%; piperacillin/tazobactam: 12.5% **(Table 7)**.

Table 7: Resistance and sensitivity of Gram-negative to antibiotics

Antibiotics	*Acinetobacter baumannii* N: 25 Total: *68*			*Escherichia coli* N: 7 Total:68			*Enterobacter cloacae* N:2 Total:68			*Klebsiella pneumoniae* N: 8 Total:68		
	R	S	I	R	S	I	R	S	I	R	S	I
Amoxicillin	25 (100%)	-	-	1 (14.3%)	6 (85.7%)	-	2 (100%)	-		2 (25%)	5 (62%)	(112.5%)
Gentamicin	25 (100%)	-	-	5 (71.4%)	2 (28.5%)	-	1 (50%)	1 (50%)	-	8 (100%)	-	-
Ampicillin	4 (16%)	-	-	7 (100%)	-	-	2 (100%)	-	-	7 (87.5%)	-	-
Amoxicillin/ Clavulanic acid	1 (4%)	-	-	7 (100%)	-	-	2 (100%)	-	-	7 (87.5%)	-	-
Piperacilline/Tazobactam	19 (76%)	-	-	2 (28.6%)	3 (42.8%)	(228.57%)	2 (100%)	-	-	3 (37.5%)	2 (25%)	(112.5%)
Ceftriaxon	3 (12%)	-	-	5 (71.4%)	1 (14.3%)	-	2 (100%)	-	-	7 (87.5%)	-	-
Cetfazidime	24 (96%)	-	-	6 (85.7%)	1 (14.3%)	-	2 (100%)	-	-	7 (87.5%)	-	-
Ciprofloxacin	22 (88%)	-	-	3 (42.9%)	3 (42.9%)	-	2 (100%)	-	-	5 (62%)	1 (12.5%)	-
levofloxacin	14 (56%)	-	-	-	-	-	-	-	-	1 (12.5%)	-	-
Erythromycin	2 (8%)	-	-	-	5 (71.4%)	1 (14.2%)	1 (50%)	-	-	1 (12.5%)	5 (62%)	-
Imipinem	22 (88%)	-	-	-	7 (100%)	-	-	1 (50%)	1 (50%)	1 (12.5%)	6 (75%)	-
Meropenem	19 (76%)	-	-	-	-	-	1 (50%)	-	-	-	-	-
Cefepim	16 (64%)	-	-	2 (28.6%)	1 (14.3%)	-	-	-	-	3 (37.5%)	-	-
Ceftolozane/tazobactam	3 (12%)	-	-	1 (14.3%)	-	-	-	-	-	-	-	-
Ceftazidime-Avibactam,	-	-	-	1 (14.3%)	-	-	-	-	-	-	-	-
cefoxitin,	5 (20%)	-	-	1 (14.3%)	6 (85.7%)	-	1 (50%)	-	-	2 (25%)	4 (50%)	-
Vancomycin	8 (32%)	1 (4%)	-	-	-		-	-	-	-	-	-
Téicoplanine	7 (28%)	-	-	-	-		-	-	-	-	-	-
Trimethoprim/sulfamethoxazole	24 (96%)	-	-	7 (100%)	-	-	1 (50%)	1 (50%)	-	7 (87.5%)	-	-
Colimycin	-	24 (96%)	-	-	1 (14.3%)	-	-	1 (50%)	-	-	3 (37.5%)	-

R: Résistant, **S:** Sensible, **I:** Intermédiaire

III. DISCUSSION:

Newborns, being immunocompromised individuals, are vulnerable to infections which can cause high levels of morbidity and mortality. All countries are seeking to reduce neonatal mortality to 12 or less per 1000 live births, despite intensive surveillance and prophylactic measures, nosocomial neonatal bacterial infections in NICUs remain a major global health problem [14, 15], and drug susceptibility testing provides a scientific basis for guiding the rational use of drugs and controlling infections [16].

Blood culture is considered to be the gold standard for documenting the blood-borne spread of germs at risk of infection [17].

The incidence rate of nosocomial infections in our series is 18.7%, and the incidence density is 10 cases/1000 DH. The cumulative incidence of nosocomial infections ranged from 2.70 to 10.66%. A link with the presence of a central venous catheter was found in 5% of cases. These figures are consistent with those found in developing countries, where the incidence of BN linked to central venous catheters ranged from 1.3 to 12.7% [18]. In Europe and North America, the incidence rates reported by BN surveillance ranged from 0.99 to 3.85/1000 DH [19, 8].

Our study revealed that the most common nosocomial germs in our series were *Coagulase-positive Staphylococcus* 38.2%, *Acinetobacter baumannii* 36.8%, *Klebsiella pneumoniae* 11.8%, *E. coli* 10.3%, and *Enterobacter cloacae* 3%. A Chinese study in 2022 showed rates of *Coagulase-positive Staphylococcus* 42.2%, *Acinetobacter baumannii* 15.1%, *Klebsiella pneumoniae* 56.3%, *E. coli* 26.8% and *Enterobacter cloacae 5.9%* [1], Resistance and sensitivity of *coagulase-positive Staphylococcus* to antibiotics: The highest percentages for resistance to antibiotics were: Gentamicin: 96%, teicoplanin: 96%, amoxicillin: 76%, and all *coagulase-positive Staphylococcus* isolated were sensitive to vancomycin: 100%. An Indian study conducted by Shah A. J. et al. was consistent with our findings [20].

For: *Enterobacter cloacae* it was resistant to ampicillin, amoxicillin/ Clavulanic acid, piperacillin/tazobactam, ceftriaxone, cetfazidime and ciprofloxacin in 100%, sensitive to amoxicillin in 100%. A Moroccan study on the resistance of *Enterobacter cloacae* to antibiotics: Gentamicin and tobramycin 97.7%, cotrimoxazole 95.5%, ciprofloxacin 89.8% [21].

For *Acinetobacter baumannii*: resistance: amoxicillin and gentamicin: 100%, ceftazidime, and trimethoprim/sulfamethoxazole: 96%, ciprofloxacin, and imipenem: 88%, and antibiotic sensitivity: colimycin: 96%, meaning it was a multi-resistant germ. A study similar to ours revealed the same resistance profile in Egypt [22].

For *Escherichia coli*: resistance: ampicillin, amoxicillin/ Clavulanic acid and trimethoprim/sulfamethoxazole 100%, ceftazidime 85.7%, gentamicin and ceftriaxone 71.4%, sensitivity: imipenem 100%, amoxicillin and cefoxitin 85.7%, erythromycin 71.4%. A study similar to ours revealed resistance of *Escherichia coli* to the antibiotics ampicillin 50%, ciprofloxacin 50%, aztreonam 100%, cefotaxime 100%, tazocin 100%, vancomycin:100%. and sensitivity to antibiotics: imipenem 100%, amikacin 100%, penicillin 50%, azithromycin 50%, and amoxicillin/clavulanic acid 50% [23].

For *Klebsiella pneumoniae* antibiotic resistance was Gentamicin: 100%, ampicillin, amoxicillin/clavulanic acid, ceftriaxone, ceftazidime, and Trimethoprim/sulfamethoxazole: 87.5%, Ciprofloxacin: 62%, and antibiotic sensitivity was: imipineme: 75%, amoxicillin and erythromycin: 62%, cefoxi: 50%. A similar study revealed that the resistance of *Klebsiella pneumoniae* to antibiotics: ceftriaxone 34.3%, ampicillin/Sulbactam 32. 8% and cefazolin 64.1%. and sensitivity: amikacin 100%, levofloxacin 85.1%, ertapenem 100%, imipenem 100%,

ciprofloxacin 82.1%, piperacillin/tazobactam 92.5%, gentamicin 95.5% [22].

IV. CONCLUSION:

Our study aims to determine the incidence rate of nosocomial neonatal infections and to identify the germs responsible and their resistance to antibiotics. The rate of nosocomial neonatal infections and the worrying rise in bacterial resistance mean that we need to adopt effective policies to combat these infections. This highlights the importance of the appropriate use of antibiotics and the implementation of strategies to preserve their efficacy. Also, aims to improve the care of neonatal patients and to reduce the emergence and spread of resistance both in the NICU and beyond.

Ethical considerations:

The study protocols were evaluated and approved by the ethics committee at the Faculty of Medicine and Pharmacy in Rabat, Morocco (Reference number C' 64/20; date of approval, 16 February 2021) and this work is part of the project. Participation was voluntary, and each patient's guardian provided informed consent after receiving a clear explanation of the research objectives. All participants were aware that their care was not contingent on their participation in the study. Confidentiality and anonymity of their data were guaranteed.

CRediT Author Statement:

A. Al-Selwi: Conceptualization, Methodology, Formal analysis, Investigation, Writing –original draft**; El. Ilham:** Validation, Review**; El. Lamya:** Conceptualization, Investigation**; N. Amallika:** Validation**; Z. Imane:** Investigation, Validation**; A. Barkat:** Conceptualization, Validation, Supervision, Writing – review & editing, Supervision**.**

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