An-Najah National University

Faculty of Graduate Studies

BACTERIOLOGIC PROFILE AND ANTIBIOTIC SUSCEPTIBILITY PATTERN OF SEPTICEMIA IN NEONATAL INTENSIVE CARE UNITS, PALESTINE (2019-2021)

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Dedication

This study is wholeheartedly dedicated to my parents, who have been my source of inspiration and gave me strength when I thought of giving up, who continually provide their moral, spiritual, emotional, and financial support unconditionally.

To my sisters, relatives, friends, and supervisor who shared their words of advice and encouragement to finish this study.

Finally, I pray that the Almighty God, who has blessed me with wisdom, strength, mind power, protection, and abilities, would count my research among my good deeds.

Acknowledgment

In the name of Allah, the Most Merciful and Gracious

First and foremost, I thank ALLAH for inspiring and guiding me to do this work, as well as Prophet Mohammad (PBUH), who advocated for science and learning.

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Thanks are extended to IT team working at ministry of health Mrs. Alaa Abu Aisheh and Mr. Yousef Zammar, who showed a great cooperation in data extraction.

Declaration

I, the undersigned, declare that I submitted the thesis entitled:

BACTERIOLOGIC PROFILE AND ANTIBIOTIC SUSCEPTIBILITY PATTERN OF SEPTICEMIA IN NEONATAL INTENSIVE CARE UNITS, PALESTINE (2019-2021)

I declare that the work provided in this thesis, unless otherwise referenced, is the researcher's own work, and has not been submitted elsewhere for any other degree or qualification.

Student's Name: Bayan Asem Mohammad Ibrahim

Signature: 

Date: 29/12/2022

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BACTERIOLOGIC PROFILE AND ANTIBIOTIC SUSCEPTIBILITY PATTERN OF SEPTICEMIA IN NEONATAL INTENSIVE CARE UNITS, PALESTINE (2019-2021)

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Abstract

Background: Neonatal sepsis (NS) is a public health concern worldwide because of its high morbidity and mortality, in addition to the growing resistance towards World Health Organization (WHO) recommended empiric regimens. The aim of this study is to evaluate the matching status of WHO empiric antibiotic regimens with the causative pathogens of NS at three tertiary care hospitals in Palestine.

Methods: This is a retrospective study where records of neonates with suspected NS admitted to neonatal intensive care units of three tertiary care units in the period of January 2019 to December 2021 were extracted from the Ministry of Health electronic database. Univariate analysis and multivariate logistic regressions were performed for factors associated with culture-proven NS, generating odds ratios and confidence intervals.

Results: Out of 6090 suspected cases of NS, 884 (14.5%) cases had positive blood cultures. The prevalence rate of NS was 9 per 1000 live births and 4 per 1000 live births for early-onset sepsis, with an overall mortality prevalence of (17.2%). The predominantly isolated organisms were 499/884 (56.4%) Coagulase-negative *Staphylococcus* (CoNS), 119/884 (13.5%) *Klebsiella pneumonia,* and 66/884 (7.5%) *Streptococcus* spp. Moreover, most of the isolated bacteria (79.6%) didn't match any of the WHO empiric regimens and the probability of matching for both WHO regimens among EOS was found 2.4 times the matching probability among LOS by doing multiple logistic regressions. (21.6%) of the Isolated bacteria (excluding CoNS) were multi-drug resistant (MDROs). Antibiotic susceptibility for Gram-negative bacteria was

high among amikacin (63%), meropenem (70%), piperacillin-tazobactam (65.6%), and colistin (100%). Alternately, it was low for ampicillin (7.1%), cefotaxime (21.8%), and ceftazidime (29%). On the other hand, Gram-positive bacteria were sensitive to vancomycin (99.8%), and (81%) of Gram-positive other than *Staphylococcus* spp. were sensitive to ampicillin.

Conclusions: The culture-proven NS prevalence rate is 10 folds the rate of high-income countries, and the majority of isolated pathogens were resistant to empiric WHO regimens. Hence, a careful review of empiric treatment for NS is warranted. Moreover, strict infection control and antimicrobial stewardship programs should be implemented to slow down the emergence of resistant organisms among the neonatal population.

Keywords: Aantibiotic susceptibility; Empiric regimens; Neonatal sepsis; Prevalence rate of NS.

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Chapter One

Introduction

1.1 Background

Neonatal sepsis (NS) is a major public health concern on a global scale (1). It was designated a global health priority by the World Health Organization (WHO) in 2017 because of its high mortality and morbidity rates (2).

NS still kills over one million people per year (22 fatalities per 1000 live births) despite the enhanced newborn care (3). Furthermore, it is the eighth leading cause of death in children under the age of five worldwide, with a fatality rate of 11–19% (1, 3), and the third most common cause of deaths among neonates, accounting for 225 000 deaths globally every year (4).

In Palestine in 2019, the neonatal mortality rate was 10.7 deaths per 1,000 live births, but there is no precise data on neonatal deaths due to NS (5).

NS is described as a systemic infection that occurs within the first 28 days of life and is characterized by clinical signs and symptoms of infection, whether or not a bacterial pathogen is isolated in a blood culture.

However, there is no case definition for NS, and the term is used imprecisely. Additionally, neonatologists and pediatricians frequently use the term "clinical sepsis" in the context of a negative blood culture along with clinical symptoms and signs of sepsis. This term, too, lacks consensus, and there are no clear criteria for its use in research or practice (6–8).

However, antibiotics should be started immediately based on the clinical symptoms of a possible significant bacterial infection (PSBI), which is a very sensitive definition aimed at reducing the incidence of false negatives (i.e., missed cases of sepsis).

PSBI can be diagnosed by any of the following: a history of poor feeding, a history of convulsions, inactivity, a breathing rate of 60 or more breaths per minute, severe chest retractions, a fever of 37.5 °C or higher, or hypothermia of 35.5 °C or lower (9, 10).

Depending on when symptoms first appear after birth, neonatal sepsis is classified as either early-onset (EOS) or late-onset (LOS). EOS is used to describe sepsis in newborns that happens in the first 72 hours (or seven days, as some experts say), while LOS is used to describe sepsis that happens after the first 72 hours (3, 11).

EOS is typically caused by pathogen transmission from the female genitourinary tract to the baby or fetus. These pathogens are capable of ascending the vagina, cervix, and uterus, as well as infecting the amniotic fluid.

Neonatal infections can also occur in the uterus or during birth as the neonate passes through the vaginal canal. EOS is typically caused by bacteria such as Group B *Streptococcus* (GBS), *Escherichia coli*, *Staphylococcus aureus*, *Haemophilus influenzae*, and *Listeria monocytogenes*. Chorioamnionitis, GBS colonization, preterm birth, and protracted membrane rupture lasting more than 18 hours are all maternal risk factors for newborn sepsis (12).

It is a vital issue to predict the probability of EOS in neonates in order to avoid the mortality and morbidity associated with the delay in administering empiric antibiotics (13). As a result, the American Academy of Pediatrics (AAP) released a calculator for predicting EOS in neonates, where the decision on whether to start antibiotics is based on maternal, neonatal clinical factors, and the local incidence rate of EOS.

On the other hand, the AAP hoped that by implementing the calculator in addition to early detection of EOS, it would reduce unnecessary antibiotic exposure in neonates who are less likely to have EOS (14-16). However, the incidence of EOS is lacking in Palestine.

LOS is typically caused by bacterial transmission from the surrounding environment after birth, such as contact with hospital professionals or caregivers; infants who require intravascular catheter insertion or other invasive operations that damage the mucosa are more likely to develop LOS, despite the fact that a portion of LOS may also be the result of a vertically transmitted infection from the mother that is manifesting late (17).

Healthcare-associated infection prevention bundles and programs implementation are contributing significantly in lower the incidence of LOS (18).

Moreover, adjusted empiric and definitive antibiotic therapy, prompt discontinuation of antibiotics when sepsis was excluded, and antibiotic restriction policies are also vital practices for decrease incidence of both sepsis and emergence of resistant pathogens (19).

WHO has designated two empiric antibiotic regimens for treating suspected NS: Ampicillin-Gentamicin and Ampicillin-Cefotaxime, with ampicillin intended to cover potentially listeria-causing infections as well as other Gram-positive bacteria such as GBS.

However, cloxacillin is used as an alternative to ampicillin if staphylococcal infection is suspected. On the other hand, gentamicin and cefotaxime intended to cover Gram-negative bacteria (20, 21).

Early diagnosis of NS and quick treatment with the right empiric antibiotics are important for saving babies' lives and avoiding both short-term and long-term problems (13). So, figuring out the types of bacteria in the area and how they react to antibiotics is important for choosing a successful treatment (11).

However, the study aims to validate WHO's "empiric" antibiotic regimens for NS because of rising antimicrobial resistance locally and around the world (22).

1.2 Thesis chapters descriptions

The thesis has six chapters, summarized as follows:

Chapter One: Introduction

The study background, study justification, problem statement, study objectives, and predicted outcomes are all included in this introductory chapter.

Chapter Two: Literature review

This chapter outlines the scientific theories from previous studies on the area of this study.

Chapter Three: Methodology

This chapter gives a clear explanation of our trial and includes the study area and setting, study subjects, study design overview, sample size, methods of collecting data and measurements, management of data, as well as analysis method and ethical consideration.

Chapter Four: Results

This chapter includes actual results. The chapter includes charts, tables, and graphs as well as a narrative that describes what is considered the most relevant information.

Chapter Five: Discussion

Also included is an explanation of how our results agree or disagree with those of previous studies by quantitative analysis. In addition, the strengths and limitations of this thesis are considered, and recommendations are given for further research.

Chapter Six: Conclusion and discussion

The main findings regarding the research questions are abstracted and general conclusions are described depended on the findings of the researches presented in this thesis. This chapter deduces policy makers and researchers with recommendations.

1.3 Literature review

The incidence of suspicion of NS is high and variable among the neonatal population. A study called BARNARDS, conducted in seven low- and middle-income countries (LMICs) in the period of November 12, 2015, to February 1, 2018, suggested a rate of 166 neonates with suspected NS per 1000 live births (23); another study conducted in Iran suggested a rate of 91 neonates with suspected NS for each 1000 live births (24).

However, in a recently published systematic review and meta-analysis of the global incidence and mortality of neonatal sepsis, researchers declared the need for harmonizing the definition of NS in order to optimize diagnosis accuracy and limit measurement variations (3).

The ability to detect the causative pathogens of NS among suspected cases was found to be 25% in the BARNARDS study (23). But the detection rate varies from study to study and depends on many things, such as the method of detection (molecular or culturing), the amount of blood drawn (at least 1 cc), and the threshold for assuming a diagnosis of NS.

If more assumed diagnoses of NS with different clinical presentations are made, the ratio of culture-proven cases to suspected cases of NS will go up (25–27).

However, the incidence of culture proven NS worldwide was found to be 1–11 cases per 1000 live births, according to a systematic review published in 2018 of the global burden of pediatrics and neonatal sepsis, where the highest prevalence was found in LMICs (3).

Moreover, two large multicenter studies were conducted in Egypt and India, where they stated a rate of 8.6 and 8.9 per 1000 live births of NS, respectively (25, 28).

A systematic review on the global risk factors for EOS published in 2013 stated that the EOS rate is influenced by maternal screening for urinary and genital infections and

colonization, inadequate infection control practices prior to and during delivery, frequent vaginal examination, environmental factors (use of resuscitation tools, medical and nursing staff), and low parental education on the importance of breastfeeding and early signs of sepsis in neonates (29).

On the other hand, LOS risk factors were studied in 2010 at LMICs, where they found a major role for the neonatal center in the development of LOS, and this was due to the variable adherence to infection control practices (hand hygiene, asepsis for sterile procedures), which are crucial to prevent sepsis in hospitalized neonates.

They also found that invasive devices like central lines and mechanical ventilators were also risk factors, in addition to the use of total parenteral nutrition (30).

The EOS incidence rate was calculated separately in many studies, where a cross-sectional study of 757‧979 neonates born in 13 networks from 11 countries among high-income countries (HICs) found the rate to be near zero (0.4 per 1000 live births) in Europe, North America, and Australia (29).

Alternatively, a systematic review of EOS incidence among LMIC yielded a rate of 3.5–21 per 1000 live births, where many studies stated the need to design appropriate and context-sensitive strategies against EOS in LMIC (32, 33).

A systematic review and meta-analysis of studies published between 1979 and 2016 regarding the global incidence and mortality of NS among 12 countries (9 HICs and 3 LMICs) reported a NS mortality prevalence of 11%–19% (3).

An international multisite prospective observational study at LMICs found that higher mortality rates will be observed in associations with increased resistance to empiric antibiotics, slower identification of NS, increased load and reduced staff for resuscitation, and higher virulence of multidrug-resistant organisms (MDROs) in 2022 (34).

The risk factors for a NS with MDROs are studied vigorously by many researchers, and many studies worldwide suggest a higher prevalence of MDROs among preterm neonates, where they propose that their immature immune system, prolonged hospitalization, and frequent use of invasive devices put them at higher risk for acquiring MDROs (35, 36).

However, the reasons for the emergence and spread of MDROs were studied in 2014 in a review article, where they stated two mechanisms of resistance development in bacteria: first, selective pressure due to antibiotic exposure, and second, plasmid-induced resistance, which is the most prevalent resistance mechanism among gram-negative bacteria and highly influenced by infection control policies like hand hygiene, transmission-based precautions, and hospitals' handling of outbreaks (37).

A systematic review and meta-analysis were conducted in 2017 in order to study the prevalence of extended-spectrum beta-lactamase (ESBL)-producing *Enterobacteriaceae* among NS pathogens, where it revealed a prevalence of 11% (38).

On the other hand, the prevalence of Carbapenem-resistant *Enterobacteriaceae* (CRE) was studied in a cross-sectional retrospective review of MDROs in neonates admitted to a tertiary neonatal unit between 1 January 2013 and 31 December 2015 in South Africa, where they found the CRE prevalence increasing from 2.6% in 2013 to 8.9% in 2015 (39).

Many studies suggested that neonates with MDRO-associated sepsis died at a higher rate; one study in India found that mortality among neonates with MDRO-associated sepsis was 50% higher than that of those with non-MDRO-associated sepsis (40). Furthermore, a prospective study conducted in India found a mortality rate of more than 50% among CRE neonates (41).

Because the profile of the causative pathogens for NS varies between geographical areas and hospitals within the same region (42), global efforts have been made to detect

continuous changes in the NS bacteriological profile and antibiotic susceptibility pattern at national, regional, and hospital levels (antibiograms), and this is in order to tailor empiric antibiotic therapy and establish specific national and institutional guidelines for treatment (15, 42).

A multinational, multicenter study was conducted in 2017 using a web-based survey (NeoAMR network) to collect data from 39 neonatal hospitals spanning 12 countries on four continents (42).

Where in (NeoAMR network) study they discovered that cephalosporin resistance rates in Gram-negative isolates ranged from 26% to 84%, carbapenem resistance rates ranged from 0% to 81%, and glycopeptide resistance rates ranged from 0% to 45% among Gram-positive isolates. (42).

Other studies were conducted in Bangladesh, Ethiopia, India, Pakistan, Nigeria, Rwanda, and South Africa under the leadership of an international collaboration. Between November 12, 2015, and February 1, 2018, they enrolled 36,285 neonates to evaluate the WHO's empiric regimens (Ampicillin and Gentamicin) for newborn sepsis in LMICs, where they discovered a substantial resistance rate for the isolating bacteria. 379 (97.2 percent) of 390 Gram-negative isolates were resistant to ampicillin, while 274 (70.3 percent) were resistant to gentamicin.

Susceptibility of Gram-negative isolates to at least one antibiotic in a treatment combination was observed in 111 (28.5%) to ampicillin–gentamicin; 286 (73.3%) to amoxicillin clavulanate–amikacin; 301 (77.2%) to ceftazidime–amikacin; and 312 (80%) to Piperacillin-Tazobactam–amikacin.

As a result, they found that the WHO's antibiotic guidelines for newborn sepsis may need to be revised in LMICs, where antibiotic resistance to currently recommended therapies is particularly high (43).

Several studies were conducted at the national and institutional levels to examine the bacterial profile and antibiotic sensitivity pattern of newborn sepsis, and all studies agreed on the need for continuous adaptation of pathogen bacterial profiles and antibiotic sensitivity to empiric antimicrobial therapy choices (10, 23, 43-57).

At the national level, between January 2004 and January 2005, a study was conducted in the NICUs of two governmental hospitals in Gaza City, Palestine, to determine the most frequent bacteria and antibiotic sensitivity of neonatal septicemia (58).

They discovered that the two hospitals had a different bacteriological profile and pattern of resistance. Additionally, they emphasized the importance of escalating empiric antibiotic therapy in view of the rising prevalence of MDROs (58).

As a result, we face a challenge in determining the actual load and distribution pattern of MDROs in the neonatal population, which will affect our therapeutic options at international, national and institutional levels.

Therefore, a multicenter national study in collaboration with other international efforts was urgently required to address this concern.

1.4 Study objectives

The goal of the study is to evaluate the matching status of WHO empiric antibiotic regimens with the causative pathogens of NS during the period of January 2019 to December 2021, Palestine.

Objectives

1. Measure the bacteriologic profile of NS and its susceptibility pattern for each sepsis class.
2. Measure MDROs' trend and distribution over years and per hospitals, and their associated risk factors and mortality.

3. Measure the prevalence of NS in general and early-onset sepsis in particular in Palestine over the course of the study.
4. Measure the mortality rate of culture-proven NS and its associated risk factors.

Chapter Two

Methods

2.1 Study design and setting

Study design: a national, multicenter retrospective study.

Study Setting: This study was conducted by extracting the data from the health information system electronic database (HIS) of the Palestinian ministry of health during a three-year period from January 2019, to the end of December, 2021, for all neonates with suspected NS at the NICUs of three Palestinian tertiary governmental hospitals, selected as one hospital for each region: the northern region, the middle region, and finally the southern Palestinian regions.

2.2 Study population

Medical records which were extracted from the ministry of health central electronic database, for all neonates (N=6090) aged (0-28) days who was admitted or existing in neonatal intensive care units with suspected NS at three Palestinian tertiary governmental hospitals selected as one hospital for each region, northern region, middle region, and finally southern Palestinian regions, from January, 2019 to the end of December, 2021

Inclusion criteria

1. All medical records for neonates aged (0-28 days) of both genders, regardless of gestational age, weight, and mode of delivery, who are existed or admitted to the NICU and blood cultures were obtained due to the suspicion of NS.

2. Medical records for same patient including any new blood culture taken after 5 days of initial culture was considered as new episode of suspected sepsis case.

3. Medical records for same patient including positive blood culture with different pathogen after 48 hour of initial positive blood culture - and contamination were ruled out – were considered a new episode of NS sepsis case.

4. Medical records for same patient including positive blood cultures with different pathogen within 48 hour of initial positive blood culture - and contamination were ruled out – were considered as one episode of NS with the term of polymicrobial sepsis (8).

Exclusion criteria

1. Medical records of control blood cultures, which were taken within 5 days of initial positive blood culture to confirm treatment success and pathogen eradication from blood.

2. Medical records of confirmatory blood cultures taken to rule out or confirm contamination.

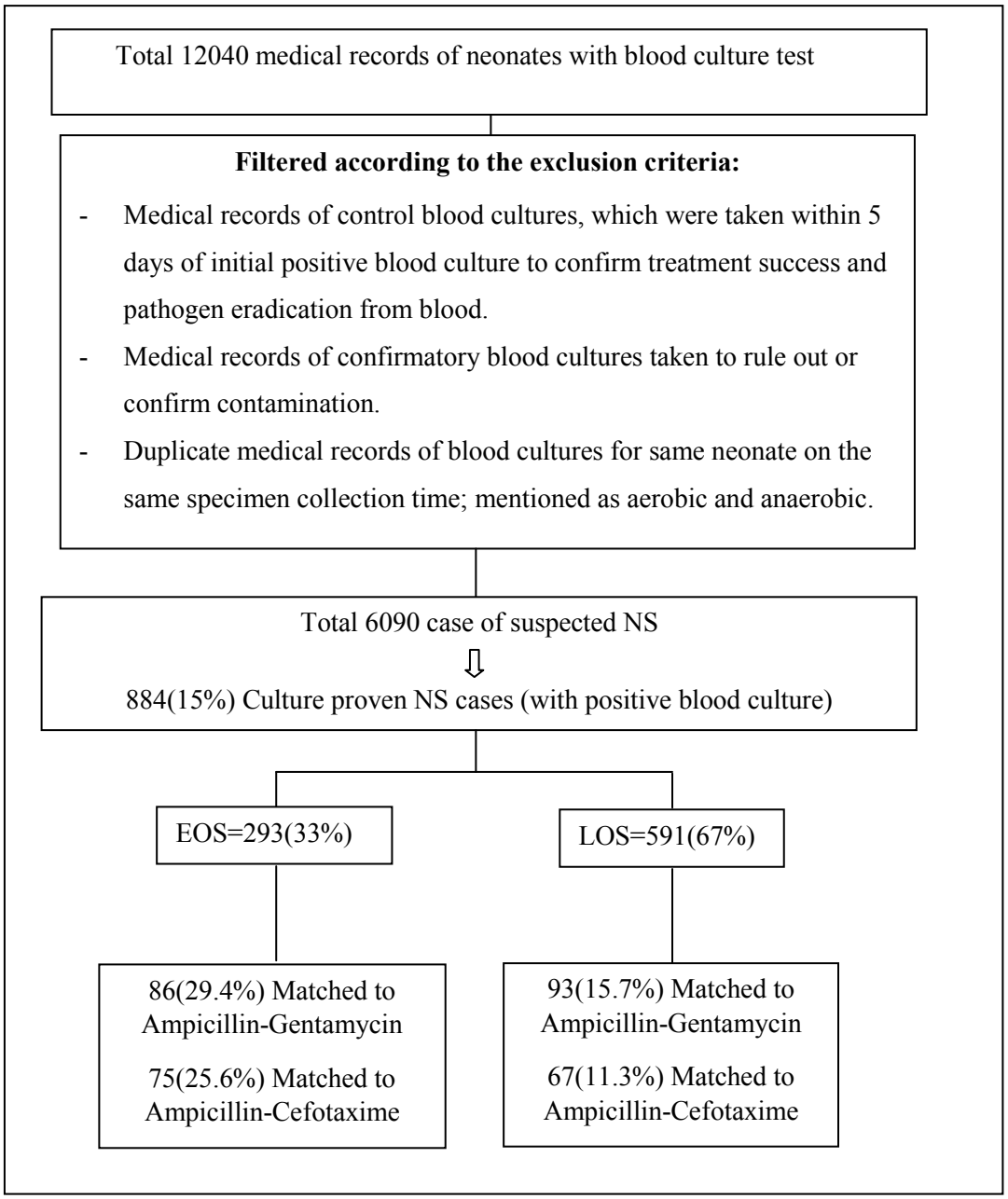
3. Duplicate medical records of blood cultures for same neonate on the same specimen collection time; mentioned as aerobic and anaerobic.

2.3 Study Procedure

The diagram shows the process of cases selection for inclusion in this study and final matching status of WHO empiric antibiotics regimens.

Figure 1

Study procedure for participants selection and final outcome



2.4 Operational variables

Background variables

Postnatal age: age after delivery

Gestational age

Term: born at ≥37 weeks of gestation

Preterm: born at <37 weeks of gestation

Gender: Female or male.

Clinical variables

Suspected neonatal sepsis: based on blood culture ordered by the pediatrician.

Culture proven sepsis: sepsis with a growth of organism in blood culture sample.

Multi-drug resistant organism(MDRO): defined as acquired non-susceptibility to at least one agent in three or more antimicrobial categories(41).

Early-onset sepsis EOS: sepsis within 72 hours of life. Except GBS within 7 days(3).

Late-onset sepsis LOS: sepsis after first 72 hours of life. Except GBS after 7 days(17).

Laboratory signs of infection:

C-reactive protein > 9 mg/dl.

White blood cells (WBC) count: < 4000 x10^9 cells/L OR >20,000 x10^9 cells/L.

Platelet count < 100,000x10^9 cells/L(42).

Suspected contaminated blood culture:

Any blood culture meets "ALL" the following criteria:

1. Coagulase-negative *Staphylococcus, Corynebacterium* species, *Bacillus* species other than *B. anthracis*, *Propionibacterium acnes, Propionibacterium* species, *Micrococcus* species, *viridians* group *streptococci*, *Aerococcus* species or *Diphtheroid* species

2. No confirmatory blood culture (second blood culture) with the same organism was taken within 5 days of initial blood culture

3. Negative laboratory signs of infection (CRP, WBC, Platelet count) (43).

2.5 Statistical analysis

Data were exported from the filtered excel sheet to SPSS version 20 for further analysis. Frequencies, proportions and summary statistics were used to describe the study population in relation to relevant variables. Binary and multivariate logistic regressions were used to assess the factors associated with neonatal sepsis class, matching status to WHO empiric antibiotics and multidrug resistant organisms. Crude odds ratio COR and adjusted odds ratio AOR were used to assess the strength of the association with 95% confidence interval. P-values of <0.05 were considered statistically significant.

2.6 Ethical considerations

The study has been approved by the research committee and the Institutional Review Board (IRB) at An-Najah National University. All extracted patient records were represented as coded numbers without names. Furthermore, data confidentiality was maintained by restricting data access to the researchers only.

Chapter Three

Results

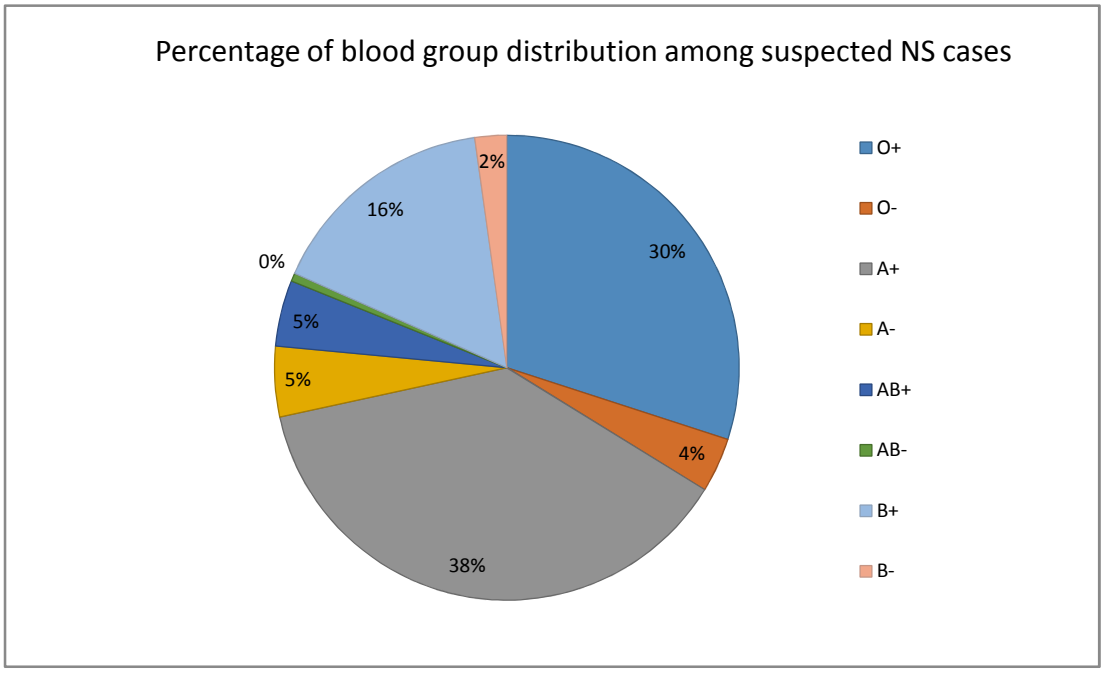
3.1 Characteristics of all study participants with suspected NS

A total of 6090 suspected NS cases were reviewed**.** There were 3464 (57%) males and 2626 (43%) females. One-third of the cases were preterm infants (33%), and two-thirds were full term (67%). The percentage of early infant age (0-7days) was 58% when they were suspected NS, however 51% of the neonates were 0-3 days old therefore they were classified to have suspected EOS. However, results showed that for each 1000 live births there were 100 neonates who had suspected NS.

Out of the total 6090 suspected NS cases, only 3132 neonates (51%) were tested for blood group, it was found that 1184 neonate had A+ blood group which makes (38%) of all tested neonates.

Figure 2

Percentage of blood groups distribution among suspected NS cases admitted to the NICU of three tertiary care hospitals in Palestine, January 2019 to December 2021, N =3132.



3.2 characteristics of culture proven NS

Among all 6090 cases, 884 cases (14.5%) were culture proven NS.

Figure 3

Percentage of suspected NS cases vs. Culture Proven NS cases for three tertiary care hospitals in Palestine, January 2019 to December 2021.

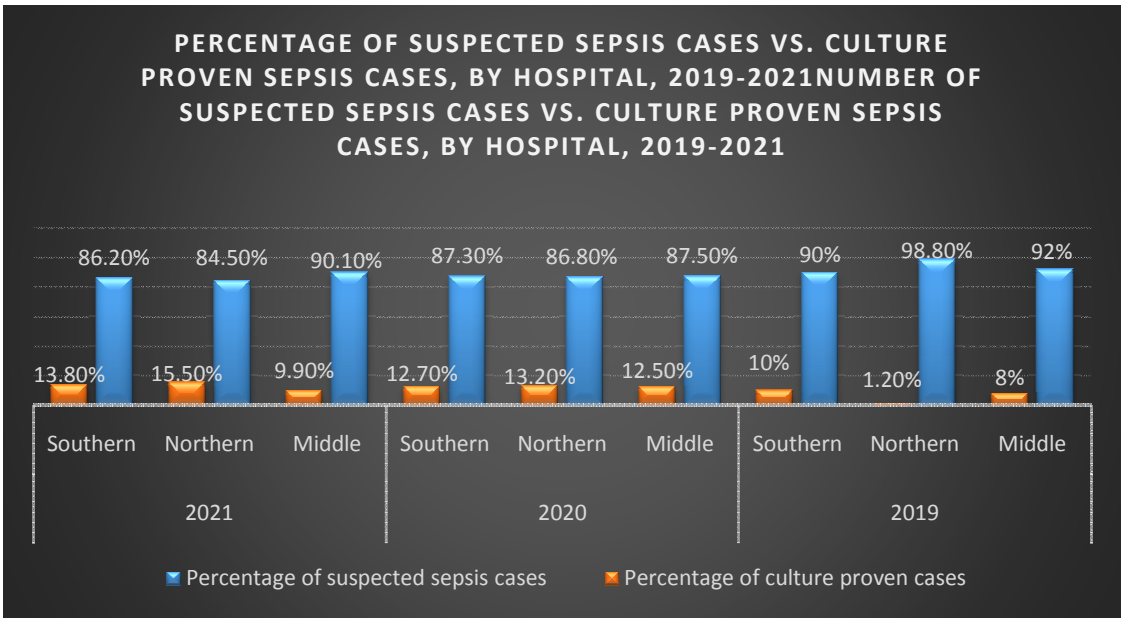


Figure 3 describes the distribution of the proven NS cases in the three hospitals during the three studied years. The percentages of culture proven cases range from (1.2-15%) among the suspected NS cases.

Table 1 described the demographic and perinatal variables of the neonates with culture proven sepsis admitted to NICU in the studied hospitals. Of the total 884 culture proven NS cases, 517 (58.5%) were males, 367 (41.5%) were females, 232(26.2%) were preterm neonates, 652(73.8%) were term neonates, 470(53.2%) were 0-7 days old, and 414(46.8%) were 8-28 days old.

In addition, the distribution of the 884 NS cases per hospital was 151(17.1%) from middle region hospital, 209(23.6%) from southern region hospital, and 524(59.3%) from northern region hospital. Increased NS cases over years were observed as following: 30%, 34%, and 36% for 2019, 2020, and 2021, respectively (Table1).

Table 1

Background descriptive demographic, perinatal variables of neonates with culture proven sepsis admitted to the NICU of three tertiary care hospitals in Palestine, January 2019 to December 2021, N = 884

Variable	Category	n (%)
Gender	Male	517(58.5)
	Female	367(41.5)
Gestational age	Term	652(73.8)
	Preterm	232(26.2)
Postnatal age	0-7 days old	470(53.2)
	8-28 days old	414(46.8)
Year	2019	267(30.2)
	2020	300(33.9)
	2021	317(35.9)
Hospital	Middle region hospital	151(17.1)
	Southern region hospital	209(23.6)
	Northern region hospital	524(59.3)

Table 2 describes the clinical characteristics of the neonates with culture proven sepsis admitted to the NICU of the studied hospitals. The clinical characteristics of participating neonates (N=884) were found to be as following: 293(33.1%) EOS cases and 591(66.9%) LOS cases, and 644(74.1%) Gram-positive bacterial, 225(25.9%) gram negative bacteria, 225(25.5%) suspected contaminated cultures with gram positive skin normal flora bacteria. MDROs (other than CoNS) contributed to 21.6% of the total cultures, and non MDROs contributed to 78.4%. Out of 884 culture proven NS cases, 152(17.2%) neonates died.

Table 2

Descritpitve clinical variables of the neonates with culture proven sepsis admitted to the NICU of three tertiary care hospitals in Palestine, January 2019 to December 2021.

Variable	Category	n (%)
Sepsis class	EOS ≤ 72 hours of life	293(33.1)
	LOS> 72 hours of life	591(66.9)
Gram stain	Gram-positive	644(74.1)
	Gram-negative	225(25.9)
MDR	Yes	191(21.6)
	No	693(78.4)
Resistance Pattern	ESBL	93(10.5)
	CRE	35(4)
	MRSA	26(2.9)
	Acinetobacter baumannii	23(2.6)
	VRE	1(0.1)
	Pseudomonas MDR	1(0.1)
Suspected contamination of blood culture	Yes	225(25.5)
	No	659(74.5)
Mortality	Lived	732(82.8)
	Died	152(17.2)
General matching status to WHO empiric antibiotics regimens	Matched with both regimens.	141(16)
	Matched to one regimen.	39(4.4)
	Matched to non-regimen.	704(79.6)
Specific matching status to WHO empiric antibiotics regimens	Matched to ampicillin-gentamicin.	179(20.2)
	Matched to ampicillin–cefotaxiem.	142(16.1)

3.3 Bacteriological profile

The majority of bacterial isolates overall the study period were Gram-positive bacteria which was 644 isolates (74 %), while Gram-negative bacteria were 225(26%). The results yielded a large contribution of Gram-positive bacteria in EOS and LOS where they found to be 82% and 70%, respectively.

Figure 4

Gram stain distribution by year, hospital, and sepsis class

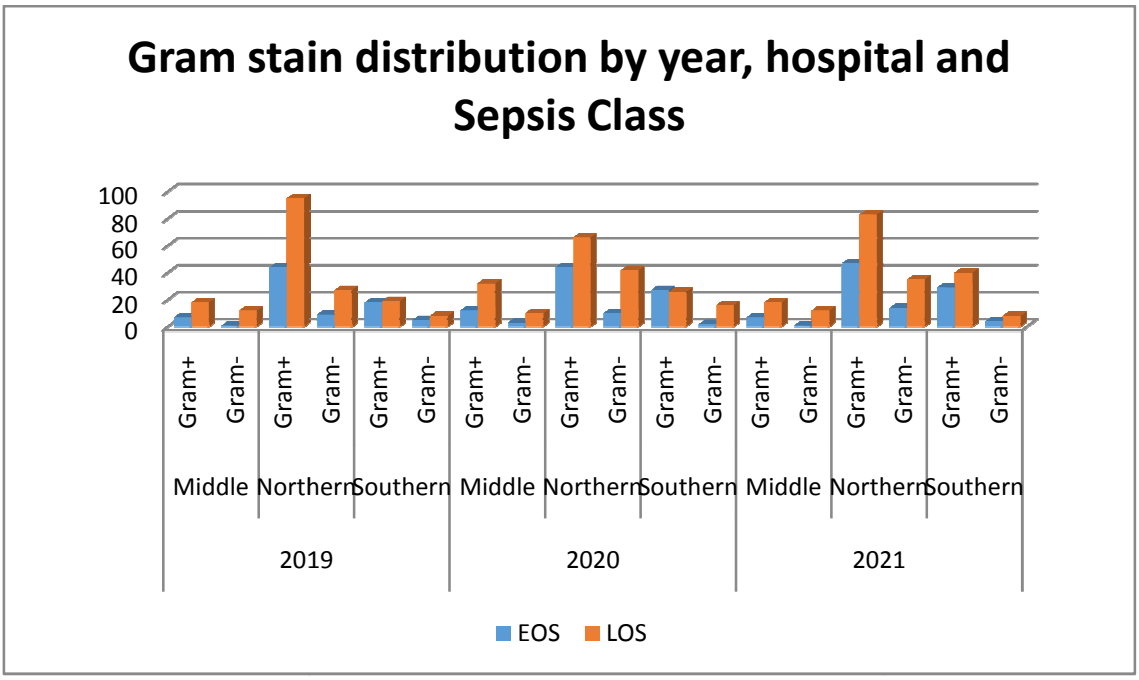


Figure 4 shows the distribution of bacteria based on gram stain type over years and per hospitals, where gram positive were predominant despite that 25.5% of them where suspected to be contamination.

Among all organisms CoNS (56.4%), *Klebsiella species* (13.5%) and *Streptococcus* species (7.5%) were the most common pathogens (Table 3). On the other hand, ten cases (1%) had polymicrobial sepsis and 16 cases (1.8%) had yeast, where all yeast isolates were identified in LOS except one case was EOS NS in a preterm neonate.

Table 3

Organisms distribution among overall culture proven NS cases

Organism name	Category	n (%)
Gram-positive bacteria	CoNS	499(56.4)
	Staphylococcus aureus	38(4.3)
	Enterococcus spp.	21(2.4)
	Streptococcus spp.	66(7.5)
	Bacillus spp.	11(1.2)
	Micrococcus spp.	9(1)
	Total	644 (74 %)
Gram-negative bacteria	*Klebsiella* spp.	119(13.5)
	Escherichia coli	42(4.8)
	Pseudomonas spp.	11(1.2)
	Acinetobacter spp.	29(3.3)
	Stenotrophomonas spp	11(1.2)
	Enterobacter spp.	4(0.5)
	Serratia spp	4(0.5)
	Hemophillus influenza	4(.5)
	Total	225(26%)
Fungus	Yeast	16(1.8)

Distribution of Gram-positive and Gram-negative causative organisms and their contribution by hospitals, years and sepsis class are presented in (Appendix B: Supplementary Table S1& S2).

However, there is an overestimation of Gram- positive bacteria numbers in general and specially CoNS isolates because of the increased likelihood of contamination during culturing procedure (normal skin flora), where the suspected contaminated blood cultures (mainly by CoNS) were 225 (25.5%) of all bacterial isolates.

3.4 Matching status to WHO empiric antibiotic regimens

Out of 884 NS cases, only 141(16%) cases were matched with both WHO regimens (ampicillin-gentamicin & ampicillin-cefotaxime), and 704(79.6%) cases that did not match to any of the WHO regimens**.**

Despite the results showed higher matching prevalence to the ampicillin-gentamicin 179 (20%) cases when compared to ampicillin-cefotaxime 142 (16%), the prevalence is still very low for both regimens (Table 2).

3.4.1 Differences in matching status to WHO empiric antibiotic regimens and in neonatal demographic and clinical factors

Table 4 shows the univariate analysis for differences between matching status based on neonatal demographic and clinical factors. Although most of the Gram-positive bacteria did not match with any regimen (79.1%), most of the bacteria that matched with both regimens (65.2%) were Gram-positive bacteria. On the other hand, most of the bacteria that matched to one regimen were Gram-negative bacteria (82%). The analysis suggested a highly significant difference for matching status and the postnatal age (p-value <0.001). Among those who matched to both empiric antibiotics regimes (66.7%) were 0-7 days old neonates and (33.6%) were 8-28 days old neonates. There were significant differences between species class and matching status (P-value<0.001). Most of the EOS cases were among those who matched for both regimens (53.2%) while most of the cases that matched for one regimen were LOS cases (71.8%).

Table 4

Univariate analysis for status matching to WHO empiric antibiotics with other factors

Variable	Category	Matched to both regimen n (%)	Matched to one regimen n (%)	Matched to non-regimen n (%)	P value
Gender	Female	55(39)	22(56.4)	290(41.2)	0.138
	Male	86(61)	17(43.6)	414(58.8)	
Postnatal age	0-7	94(66.7)	19(48.7)	357(50.7)	<0.001
	8-28	47(33.3)	20(51.3)	347(49.3)	
Gram stain	Negative	49(34.8)	32(82)	144(20.9)	<0.001
	Positive	92(65.2)	7(17.9)	545(79.1)	
MDR	Yes	1(0.7)	22(56.4)	168(23.9)	<0.001
	No	140(99.3)	17(43.6)	536(76.1)	
Sepsis class	EOS	75(53.2)	11(28.2)	207(29.4)	<0.001
	LOS	66(46.8)	28(71.8)	497(70.6)	
Mortality	Lived	117(83)	33(84.60	582(82.7)	0.968
	Died	24(17)	6(15.4)	122(17.3)	
Gestational age	Term	104(73.8)	23(59)	525(74.6)	0.097
	Preterm	37(26.2)	16(41)	179(25.4)	
Hospital	Northern	74(52.5)	21(53.8)	429(60.9)	0.301
	Southern	40(28.4)	9(23.1)	160(22.7)	
	Middle	27(19.1)	9(23.1)	115(16.3)	
Year	2019	46(32.6)	12(30.8)	209(29.7)	0.482
	2020	40(28.4)	16(41)	244(34.7)	
	2021	55(39)	11(28.2)	251(35.7)	

3.4.2 Multiple logistic regressions for the association between matching status to WHO empiric antibiotic regimens and other neonatal demographic and clinical factors

Table 5 describes the multiple logistic regressions for the association between the matching status and neonatal demographic and clinical factors. The results revealed that EOS is 2.4 times more likely than LOS to match with both empiric antibiotic regimens than not matching to any regimen [AOR=2.392 (95%CI: 1.361-4.205), p-value 0.002]. Moreover, MDRO blood cultures were 97.6% less likely than non-MDROs to match for both antibiotic regimen than not matching to any regimen [AOR=0.026 (95%CI: 0.004-0.187), p-value< 0.001]. and MDRO blood cultures were 69.9% less likely than non-MDROs to match for one antibiotic regimen than not matching to any regimen [AOR=0.301 (95%CI: 1.947-8.922), p-value< 0.001].

Table 5

Multiple logistic regression for association between matching status to WHO empiric antibiotics regimens and other factors

Matching for both WHO antibiotic regimens*					
Covariate	**Covariate category**	**Covariate reference**	**Odds Ratio**	**95% Confidence Interval**	**P-value**
Gender	Female	Males	0.966	0.656-1.424	0.863
Gestational age	Term	Preterm	0.865	0.513-1.461	0.588
Sepsis class	EOS	LOS	2.392	1.361-4.205	0.002
Hospital	Northern	Middle	1.047	0.662-1.653	0.100
	Southern	Middle	0.706	0.706-0.424	0.324
MDR	Yes	No	0.026	0.004-0.187	<0.001
Year	2019	2021	0.635	0.532-43.952	0.845
	2020	2021	0.746	0.936-93.593	0.180
Postnatal age	0-7 days	8-28 days	0.967	0.541-1.730	0.911
Mortality	Lived	Died	0.916	0.541-1.550	0.743
Matching for one WHO antibiotic regimen					
Covariate	**Covariate category**	**Covariate reference**	**Odds Ratio**	**95% Confidence Interval**	**P-value**
Gender	Female	Males	1.766	0.900-3.465	0.098
Gestational age	Term	Preterm	0.597	0.263-1.355	0.218
Sepsis class	EOS	LOS	1.535	0.575-4.101	0.392
Hospital	Northern	Middle	0.892	0.369-2.150	0.799
	Southern	Middle	0.938	0.334-2.505	0.901
MDR	Yes	No	0.301	1.947-8.922	<0.001
Year	2019	2021	1.339	0.563-3.185	0.509
	2020	2021	1.087	0.447-2.644	0.854
Postnatal age	0-7 days	8-28 days	0.597	0.263-1.355	0.898
Mortality	Lived	Died	1.764	0.687-4.525	0.238

*Matched to non-regimen as reference

3.5 Antimicrobial susceptibility pattern

Supplementary Table S3 in Appendix B describes Sensitivity among major Gram-positive NS organisms divided by year and sepsis class. Supplementary Table S4 in Appendix B describes Sensitivity among major Gram-negative NS organisms divided by year and sepsis class.

Results showed a variation in antimicrobial susceptibility by years, sepsis class and organisms' type, however among Gram-positive isolates, vancomycin sensitivity was 99.8%. On the other hand, among Gram-negative isolates, colistin sensitivity – using broth micro dilution method -was 100% regardless of sepsis class.

Among Gram-positive bacteria and after excluding *Staphylococcus* spp., ampicillin sensitivity was high 81.4%. Moreover, GBS cultures were 100% sensitive to ampicillin.

On the other hand, among Gram-negative bacteria, the sensitivity of ampicillin, cefotaxime, gentamicin, amikacin, meropenem, piperacillin-tazobactam and ceftazidime were as following 7.1%, 21.8%,33%, 63%, 70%, 65.6% and 29% respectively.

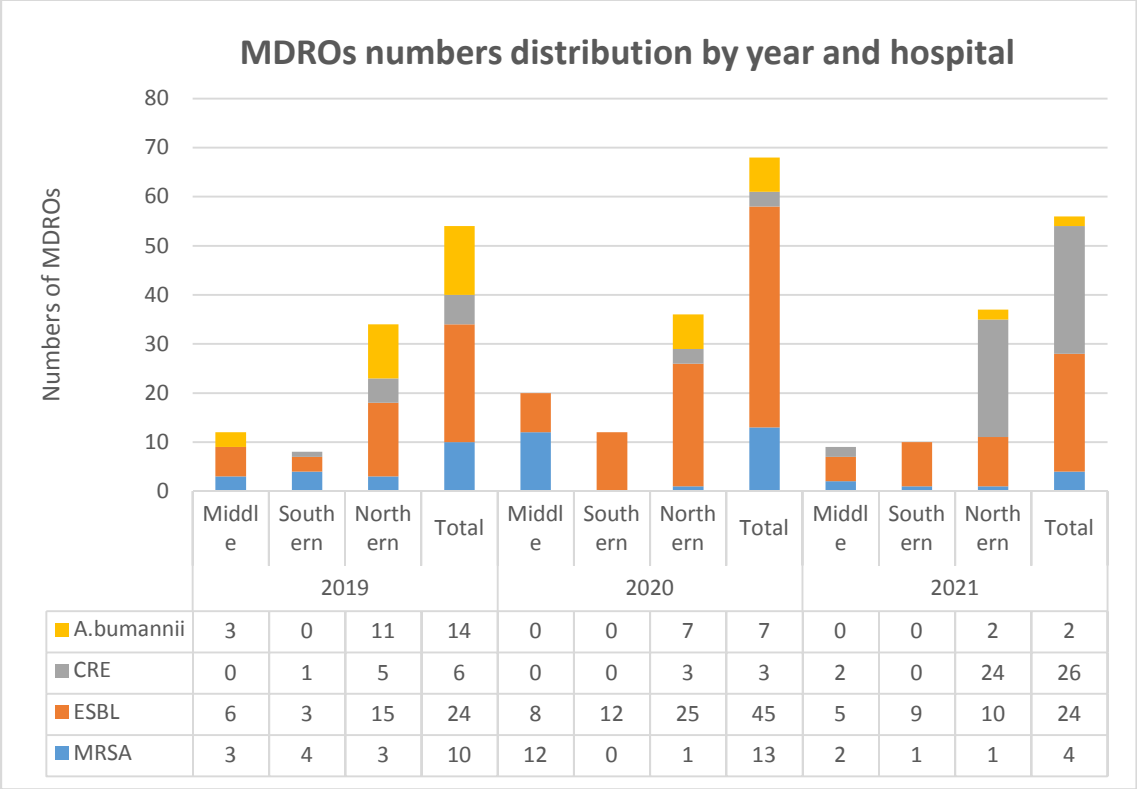
For *Klebsiella* spp. sensitivity was 5% for ampicillin, 32% for gentamicin, 12% for Cefotaxime,68% for Amikacin, 70% for Piperacillin-Tazobactam and 73% for Meropenem.

In comparison, *Staphylococcus.*spp. regardless of coagulase status (CoNS and *Staphylococcus aureus*) was not tested for ampicillin and cefotaxime since they are never used to treat septicemia with such organism, where vancomycin is considered the definitive treatment in such cases, however *Staphylococcus* spp. was found sensitive 100% to vancomycin, and 60% to gentamicin.

3.6 MDROs distribution and trends by hospitals and years

Figure 5

Distribution of MDROs numbers by years of three tertiary care hospitals in Palestine, January 2019 to December 2021.



	2019				2020				2021			
	Middle	Southern	Northern	Total	Middle	Southern	Northern	Total	Middle	Southern	Northern	Total
A.bumannii	3	0	11	14	0	0	7	7	0	0	2	2
CRE	0	1	5	6	0	0	3	3	2	0	24	26
ESBL	6	3	15	24	8	12	25	45	5	9	10	24
MRSA	3	4	3	10	12	0	1	13	2	1	1	4

Figure 5 describes the variation of distribution of MDROs from neonates' blood cultures among hospitals per years, where northern region hospital had the highest number of CRE isolates, and constituting 32 out of the 35 CRE (91.4%), which was increased from 5 isolates in 2019 to 24 isolates in 2021. On the other hand, middle region hospital in 2020 contributed to 12 out of 13 MRSA isolates (92.3%).

In general, MDROs (excluding CoNS) constituted 21.6% of all obtained blood culture organisms. ESBL, CRE, MRSA, *Acinetobacter baumannii*, VRE and *Pseudomonas* MDR contributed to 10.5%, 4%, 2.9%, 2.6%, 0.1% and 0.1% respectively of the overall obtained cultures.

Figure 6

Trend of MDROs numbers over years of three tertiary care hospitals in Palestine, January 2019 to December 2021

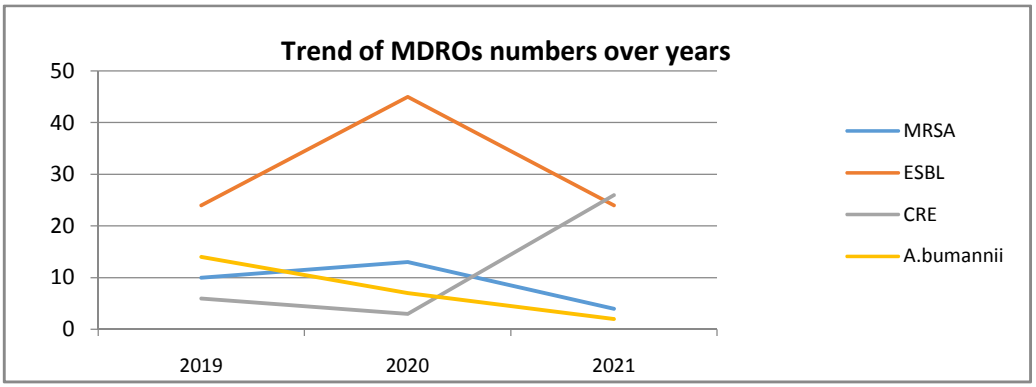


Figure 6 shows the trend of major MDROs numbers over years, where a noticed decline in ESBL pattern of resistance was found in alliance with a rising of CRE pattern. However, this variation where predominantly attributed to northern region hospital.

3.6.1 Association between MDROs prevalence and other neonatal demographic and clinical factors

Table 6 shows the relationship between MDROs and neonatal demographic and clinical factors. There is a significant association between the presence of MDROs in the obtained blood cultures and gestational age [COR=0.597 (95%CI: 0.422-0.843), p-value= 0.004], postnatal age [COR=0.951 (95%CI:0. 428-0.817), p-value=0.001], sepsis class [COR=0.333 (95%CI: 0.221-0.502), p-value< 0.001]. Additionally, Gram stain type [COR=60 (95%CI: 37.36-97.457), p-value< 0.001], and the mortality outcome [COR=0.432 (95%CI: 0.269-0.636), p-value<0.001].

However, there was a significant variation in distribution of MDROs among hospitals (p-value<0.001), where among all MDROs blood cultures, northern region hospital contributed to 60.7%, southern region hospital 17.3%, and middle region hospital 22%.

Table 6

Univariate analysis for multidrug resistant organisms with other factors

Variable	Category	NS Blood cultures with MDRO	NS Blood cultures with no-MDRO	OR	95%CI	P value
Gender	Female	82(42.9)	285(41.1)	1.077	0.779-1.489	0.679
	Male	109(57.1)	408(58.9)			
Postnatal age	0-7	82(42.9)	388(56)	0.591	0.428-0.817	0.001
	8-28	109(57.1)	305(44)			
Gram stain	Negative	163(85.8)	62(9.1)	60.078	37.36-97.457	<0.001
	Positive	27(14.2)	617(90.9)			
Sepsis class	EOS	32(16.8)	261(89.1)	0.333	0.221-0.502	<0.001
	LOS	159(83.2)	432(62.3)			
Mortality	Lived	138(72.3)	594(85.7)	0.432	0.296-0.636	<0.001
	Died	53(27.7)	99(14.3)			
Gestational age	Term	125(65.4)	527(76)	0.597	0.422-0.843	0.004
	Preterm	66(34.6)	166(24)			
Ampicillin-Genatimicin matching status	Matched	23(12)	156(22.5)	0.471	0.294-0.755	0.002
	Not matched	168(88)	537(77.5)			
Ampicilllin-Cefotaxime matching status	Matched	1(0.5)	141(20.3)	0.021	0.003-0.148	<0.001
	Not matched	190(99.5)	552(79.7)			
Hospital	Northern	116(60.7)	408(58.9)			<0.001
	Southern	33(17.3)	176(25.4)			
	Middle	42(22)	109(15.7)			
Year	2019	57(29.8)	210(30.3)			0.308
	2020	73(38.2)	227(32.8)			
	2021	61(31.9)	256(36.9)			

3.6.2 Adjusted binary logistic regression for the association between MDRO and neonatal demographic and clinical factors

Table 7 describes the binary logistic regression for the associations between MDRO and sepsis class and adjusted to other neonatal demographic and clinical factors. The results revealed that, EOS is 66.3% less likely to have a blood culture with MDRO than LOS [AOR=0.337 (95%CI: 0.117-0.972), p-value 0.04]. Moreover, Gram-negative bacteria are 852 times more likely than gram positive to be MDRO [AOR=852.101 (95%CI: 868,101-2708.218), p-value< 0.001].

There is a significant difference between MDROs presence and matching probability to ampicillin-gentamicin and ampicillin-cefotaxime (p-value<0.001), where it found that MDROs presence in blood culture is 93.8% [AOR=0.062 (95%CI: 0.018-0.217)], and 99.8% [AOR=0.002 (95%CI: 0.000-0.021)] less likely to match to ampicillin-gentamicin and ampicillin-cefotaxime, respectively.

Table 7

Adjusted binary logistic regression for association between multidrug resistant organisms and other factors

Presence of MDRO in blood culture*					
Covariate	**Covariate category**	**Covariate reference**	**Odds Ratio**	**95%Confidence Interval**	**P-value**
Gender	Female	Males	1.025	0.449-2.104	0.946
Gestational age	Term	Preterm	0.668	0.297-1.502	0.329
Gram stain	Negative	Positive	852.101	268.101-2708.218	<0.001
Sepsis class	EOS	LOS	0.337	0.117-0.972	0.040
Hospital	Northern	Middle	0.720	0.246-2.108	0.549
	Southern	Middle	0.139	0.054-1.360	0.075
Suspected contamination	Yes	No	0.000	0.000-0.000	0.994
Postnatal age	0-7 days	8-28 days	0.967	0.541-1.730	0.329
Mortality	Lived	Died	0.558	0.541-1.550	0.165
Ampicillin-Genatimicin matching status	Matched	Not matched	0.062	0.018-0.217	<0.001
Ampicilllin-Cefotaxime matching status	Matched	Not matched	0.002	0.000-0.021	<0.001

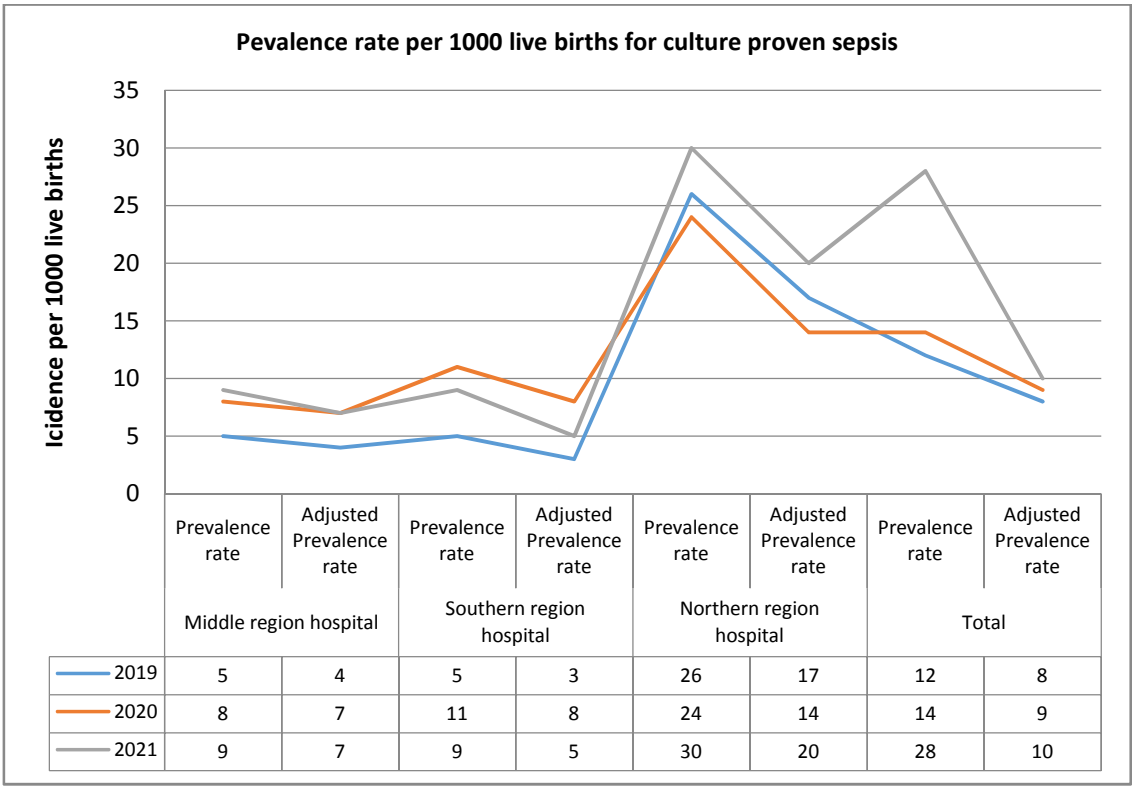
*Absence of MDROs in blood culture as reference.

3.7 Prevalence rate of culture proven NS

To avoid over or underestimation among suspected NS cases, the prevalence rate was calculated for culture proven NS cases.

Figure 7

Prevalence and adjusted prevalence rates per 1000 live births among hospitals per years for neonates with culture proven sepsis admitted to the NICU of three tertiary care hospitals in Palestine, January 2019 to December 2021.



	Middle region hospital		Southern region hospital		Northern region hospital		Total	
	Prevalence rate	Adjusted Prevalence rate	Prevalence rate	Adjusted Prevalence rate	Prevalence rate	Adjusted Prevalence rate	Prevalence rate	Adjusted Prevalence rate
2019	5	4	5	3	26	17	12	8
2020	8	7	11	8	24	14	14	9
2021	9	7	9	5	30	20	28	10

Figure 7 shows the trend of culture proven sepsis rates before and after excluding the suspected contaminated blood culture (Adjusted rate). It shows an increasing rate over years of 8, 9, and 10 per 1000 live births over the years of 2019, 2020, and 2021, respectively. Moreover, a variation in rate was noticed among hospitals especially northern region hospital where the rate was above 2 folds the rate of the other hospitals.

3.8 Prevalence rate of early onset NS

Table 8 describes the prevalence rate of EOS among hospitals and per years before and after excluding neonates with contaminated blood cultures. There is an increase in EOS prevalence rate over time from 2 per 1000 live births in 2019 to 4 per 1000 live births in 2021.

Table 8

Prevalence rate per 1000 live births for culture proven EOS classified before and after the adjustment, among hospitals and per years

hospital	Variable	2019	2020	2021
Middle region	Live births numbers	3510	4165	4145
	Prevalence rate per 1000 live births	2	4	3
	Adjusted Prevalence e rate per 1000 live births	1	3	2
Southern region	Live births numbers	5952	5728	5592
	Prevalence rate per 1000 live births	4	4	4
	Adjusted Prevalence rate per 1000 live births	2	2	3
Northern region	Live births numbers	5552	5099	4882
	Prevalence rate per 1000 live births	8	8	9
	Adjusted Prevalence rate per 1000 live births	4	4	5
Total	Live births numbers	15014	14992	14619
	Prevalence e rate per 1000 live births	5	8	8
	Adjusted Prevalence e rate per 1000 live births	2	3	4

However, there is a significant variation in prevalence of EOS among hospitals (p-value 0.001) (Supplementary table S5 in Appendix B), where table 9 of binary logistic regression for associations between sepsis class and other factors shows that compared to middle region hospital, Northern region hospital is 54 times more likely to have EOS than LOS [AOR=45.453 (95%CI: 4.336-683.861), p-value= 0.002].

Table 9

Adjusted Binary logistic regression for association between sepsis class and other factors

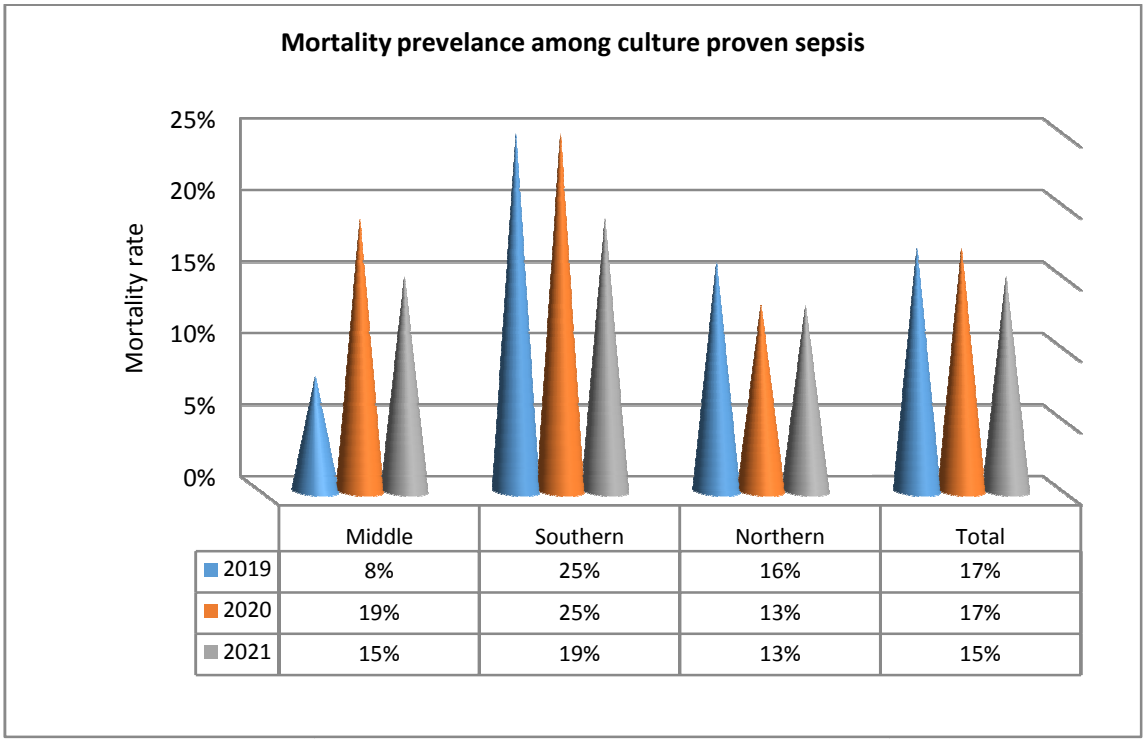
EOS*					
Covariate	Covariate category	Covariate reference	Odds Ratio	95% Confidence Interval	P-value
Gender	Female	Males	1.284	0.218-7.548	0.782
Gestational age	Term	Preterm	0.090	0.007-1.132	0.062
Gram stain	Negative	Positive	8.541	0,299-254.497	0.210
Hospital	Northern	Middle	54.453	4.336-683.861	0.002
	Southern	Middle	4.732	0.226-99.257	0.317
MDR	Yes	No	0.385	0.012-11.9937	0.586
Year	2019	2021	4.837	0.532-43.952	0.162
	2020	2021	9.359	0.936-93.593	0.057
Ampicillin-Genatimicin matching status	Matched	Not matched	0.624	0.010-40.230	0.825
Ampicilllin-Cefotaxime matching status	Matched	Not matched	0.377	0.003-43.698	0.687
Mortality	Lived	Died	0.329	0.032-3.410	0.351
Suspected contamination	Yes	No	0.989	0.139-3.434	0.991

*LOS as reference

3.9 Mortality among culture proven NS

Figure 8

Mortality prevalence among culture proven NS by hospital and year of three tertiary care hospitals in Palestine, January 2019 to December 2021



	Middle	Southern	Northern	Total
2019	8%	25%	16%	17%
2020	19%	25%	13%	17%
2021	15%	19%	13%	15%

Figure 8 shows the mortality prevalence among hospitals and per years of culture proven NS.

Out of the 884 culture proven sepsis cases 152 neonates were died (17.2%). Results showed that among the 152 neonates who died 60 neonates (42%) had EOS and 83 neonates (58%) had LOS over all the study period from January 2019 to December 2021. Moreover, (41%) had Gram-positive bacteria in their blood culture and (59%) had Gram-negative bacteria. Additionally, (36%) of neonates who died had a blood culture with MDROs in their blood stream. However, compared to those who died, those who survived sepsis where 56.8% less likely to have a blood culture with MDROs [COR=0.432 (95% CI: 0.269-0.636), p-value<0.001] (Table 6).

Figure 9

Mortality prevalence of EOS vs. LOS over years of three tertiary care hospitals in Palestine, January 2019 to December 2021

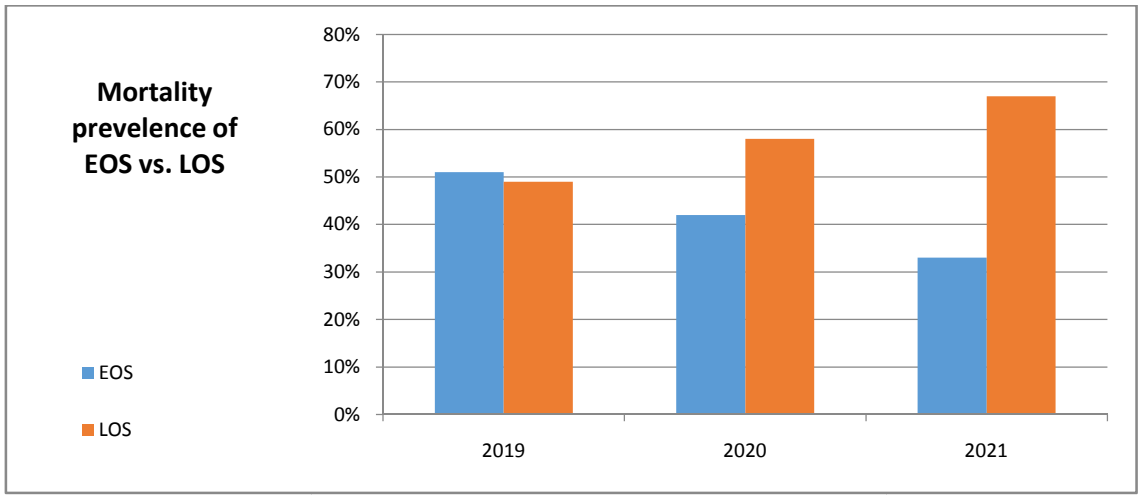


Figure 9 shows the Mortality prevalence of EOS vs. LOS over years, it was noticed that mortality rate for EOS was declining over years; while in the opposite direction mortality rate was rising for LOS despite it was statistically not significant.

Figure 10

Number of deaths among Culture Proven Sepsis cases, by selected Risk factors

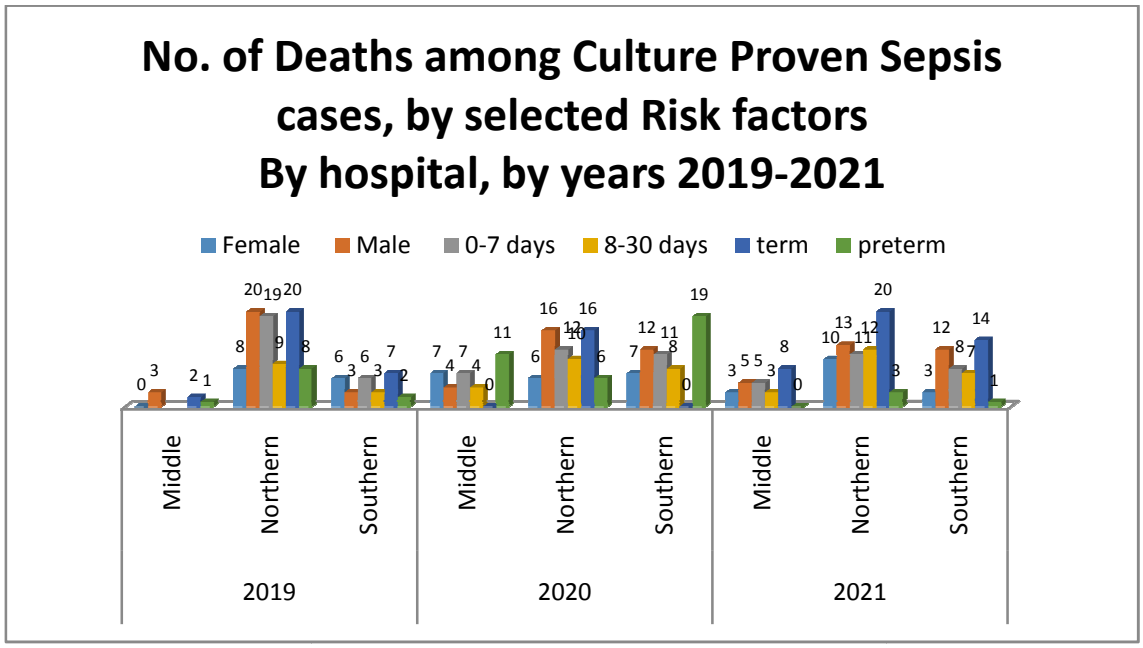


Figure 10 shows the deaths numbers among culture proven NS, by selected risk factors like gender, postnatal age, and gestational age.

Table 10 describes the mortality prevalence among major organisms obtained from the blood cultures of neonates, where it was found that near half the neonates (43%) who had a CRE pattern of resistance died, and (34%) of those who had ESBL resistance pattern died. Moreover (23%) of those who had MRSA sepsis died.

Table 10

Selected organisms associated mortality divided by sub-species and their recognized pattern of resistance of three tertiary care hospitals in Palestine, January 2019 to December 2021.

Organism	Total Isolates N = number	Number of neonates who died n (% n/N)
CoNS	**499**	**66(13.2)**
S. aureus	**38**	**8(21**)
- MSSA	11	2(18)
- MRSA	27	6(22.2)
***Streptococcus* spp.**	**66**	**8(10.6)**
- GBS	15	3(20)
- *Enterococcus* spp	16	1(6.2)
○ *Enterococcus* VRE	1	1(100)
- Other groups	34	3(8.8)
***Enterobacteriaceae* family**	**159**	**49(26)**
Escherichia coli	**42**	**15(36)**
- *E.coli*	20	0(0)
- *E.coli* ESBL	20	13(65)
- *E.coli* CRE	2	2(100)
Serratia marcescens	**4**	**1(25)**
- *Serratia.* CRE	2	1(50)
***Klebsiella* spp.**	**119**	**33(28)**
- *K.oxytoca*	4	2(50)
- *K. pneumonia*	11	0(0)
- *K.pneumonai* ESBL	73	19(26)
- *K.pneumonia* CRE	31	12(39)
Enterobacter cloacae	**4**	**0(0)**
Total CRE resistance pattern	**35**	**15(43)**
Total ESBL resistance pattern	**93**	**32(34)**
***Pseudomonas* spp**.	**11**	**1(9)**
Pseudomonas spp. non MDR	10	0(0)
P.aeruginosa MDR	1	1(100)
***Acinetobacter* spp**	**29**	**10(34)**
Acinetobacter spp non MDR	6	2(33)
A.baumannii MDR	23	8(35)
Stenotrophomonas maltifolia	**11**	**2(18)**

Chapter Four

Discussions and Conclusions

To the best of our knowledge, in the context of NS, this is the first study evaluating the matching status of used WHO empiric antibiotic regimens to the local bacteriologic profile and sensitivity pattern, growing trend and distribution of antibiotic resistance and associated risk factors; it is also the first study that calculated the prevalence rates of NS in general and EOS in particular, in addition to the mortality rate among neonates with culture-proven sepsis in Palestine.

Data suggests that for each 1000 live births, there are 100 neonates who were cultured (blood) at the NICU due to suspicion of NS. This result was in the range of a study in Iran, where the rate of suspected sepsis was 91 per 1000 live births (24); another study, BARNARDS, including 12 clinical sites in 7 LMICs, found that the rate was 166 per 1000 live births (23).

However, this incidence of suspected NS may not be precise due to the lack of clinical variables in the study, which is considered an important aspect for the diagnosis of NS. Furthermore, a lack of clarity with NS definitions locally and internationally may bias and cloud data comparisons, with researchers stating the need for harmonizing NS definitions in order to optimize diagnosis accuracy and limit measurement variations (3).

Therefore, in alliance with many other studies worldwide, calculations based on culture proven sepsis were performed in this study to calculate the most relevant incidence or prevalence rates of NS and its associated mortality in order to standardize the NS measurement tools (28, 35, 62, 63).

Among 6090 suspected NS cases, the percentage of culture-proven NS was 14.5%, which was similar to a study conducted in India in 2019 where culture-proven NS regardless of sepsis class constituted 14.8% of the suspected NS cases (25).

However, a higher percentage of culture-proven NS of 25% among suspected cases compared to our finding was found in the BARNARDS study of 7 LMIC countries (23); this might be due to either the increased number of suspected cases in our study, where the culturing threshold of admitted neonates was high, or the withdrawn blood quantity (minimum 1 mL) not being achieved for organism identification (27).

Regarding culture-proven NS, the prevalence rate of NS regardless of sepsis class and after excluding the suspected contaminated cultures was 8, 9, and 10 per 1000 live births over the years of 2019, 2020, and 2021, respectively.

The rate was very close to the incidence rate of culture-proven NS in Egypt, which was 8.6 per 1,000 live births in the period 2010–2014 (28), and to the incidence rate of culture-proven NS in India, which was also 8.9 per 1,000 live births in the 2019 year (25).

However, it was within the range of NS worldwide, which is 1–10 cases per 1000 live births, but was closer to the upper limit, similar to LMIC rates (3). A combination of factors may influence our rate, such as the country's lack of routine prenatal screening to rule out infections or pathogenic bacteria colonization of maternal urine and vagina, where maternal screening is critical for preventing sepsis (29).

Furthermore, there is poor practice and implementation of infection control policies, which is the gold standard preventive measure of hospital acquired infections, one of the leading causes of LOS, where the data indicated an outbreak of MDRO (CRE) that was not properly controlled (30).

On the other hand, the prevalence rate of EOS was calculated separately due to its clinical significance in practice, where the AAP implemented a calculator for predicting EOS depending on the local incidence of EOS (14).

In practice, and because no previous studies looked for an incidence rate in Palestine, we used to put in the EOS rate of 0.4 per 1000 live births, but the data suggests a prevalence rate of 4 per 1000 live births in 2021.

The prevalence rate of EOS after excluding the suspected contaminated cultures was found to be 2, 3, and 4 per 1,000 live births over the years of 2019, 2020, and 2021, respectively. The calculated prevalence rate was very high and equaled 10 folds the EOS incidence rate of HICs, which was 0.4 per 1000 live births (31).

Alternatively, when compared to the LMIC rate of 3.5–21 per 1,000 live births, our EOS rate was in the lower range (33). The reduction in EOS rates at HICs is due to the implementation of preventive strategies that are currently lacking in Palestine, such as maternal screening (for urinary and genital infections and colonization), strong infection control practices prior to and during delivery, minimizing frequent vaginal examinations, ensuring low-risk environmental factors (use of resuscitation tools, medical and nursing staff), and increasing parental education on the importance of breastfeeding and the early signs of EOS (29).

The mortality outcome was 17.2% of all the 884 culture-proven NS cases, which was in range with the global mortality prevalence of 11%–19% among 12 countries (9 HICs and 3 LMICs) (3). Where we are close to LMIC rates, it could be due to increased resistance to empiric antibiotics, slower NS identification, increased load and reduced staff for resuscitation, and higher virulence of MDROs (35).

However, the available mortality data was limited to neonates with culture-positive NS who had died at the time of culture result notification; as a result, we could not definitely make sepsis the only cause of mortality, and we also did not have data on those who died after the culture result was obtained.

However, there is a drop-in mortality over years that might be attributed to enhanced newborn care and resuscitation tools. Moreover, northern region hospital noticed to

have the lowest rate of mortality over years, where it was interpreted due to the fact of higher mortality among LOS compared to LOS because of the highest EOS cases in it compared to other hospitals in middle and southern regions, where they have less EOS case due to lack of places, where neonates are more frequently referred to other hospitals.

Data support the hypothesis of a decreased matching probability of WHO empiric antibiotic regimens and NS pathogens in Palestine, where the matching chance of organisms among culture-proven NS to both WHO empiric regimens was found to be only (16%).

Alternatively, 79.6% of the organisms did not match to any of the empiric antibiotic regimens, in a similar way to the results of the BARNARD study, which showed a significant decrease in the matching probability of empiric antibiotic regimens, where they found the target attainment probability of ampicillin-gentamicin was 33.7%, which is relatively higher when compared to our study's matching probability of 20% for ampicillin-gentamicin (23).

Ampicillin-cefotaxime was found to be less likely to match an organism with a matching percentage of 16%, despite the fact that many previous studies favored gentamicin over cefotaxime for NS not associated with meningitis, owing to better coverage and lower mortality (64).

Moreover, in the Gaza study, they recommended a change in empiric therapy for neonatal sepsis for vancomycin- meropenem after taking into consideration the hospital-specific antibiogram (58).

However, too many studies worldwide (NeoAMR network) studied the same topic and all stated a conclusion about the need to re-evaluate the WHO empirical regimens due to increased MDR bacteria and to further specify treatment based on institutional antibiograms, especially among LOS (42).

Data from multiple logistic regression associations for matching status to WHO empiric antibiotics shows that compared to LOS, EOS is 2.4 times more likely to match with both empiric antibiotic regimens than not matching to any regimen, which is similar to studies conducted worldwide where they contributed their findings due to the increased prevalence of MDROs in late infections where the primary source is the surrounding environment of the neonates, whether it's a community or hospital (9, 23, 42, 51, 63).

Data from multiple logistic regressions found that blood cultures with MDROs are 97.6% less likely to match for any of the antibiotic regimens compared to blood cultures with non-MDROs [AOR = 0.026 (95%CI: 0.004-0.187), p-value < 0.001].

MDROs constituted 21.6% of all 884 obtained cultures, despite the fact that all CoNS were excluded from the calculated prevalence of MDROs because of suspected contamination from skin, which if included may falsify the prevalence of MDROs and make it difficult to compare with other prospective studies assured contamination-free samples due to the availability of clinical pictures and/or adherence to contamination-free measures, e.g., multiple sets of blood culture (9, 50, 65). However, the prevalence of contamination differs among hospitals and depends mainly on human technique and hospital culturing policy. Whereas, compared to the Gaza study of NS, their contamination rate was 12% (58), and our contamination rate was higher and reached 25%.

Data suggest a significant association between the presence of MDROs NS and LOS, preterm status, 8–28 days postnatal age, and Gram-negative bacteria, similar to several studies worldwide, and one of them was conducted in a neighboring area focusing on the EOS bacteriologic profile, where they stated preterm status and gram-negative bacteria as risk factors for MDRO NS (35, 36).

Preterm infants are at higher risk for infection with MDROs due to the combination of an immature immune system, prolonged hospitalization, and frequent use of invasive devices (36).

The increased prevalence of MDROs among LOS and late postnatal age has been attributed to the acquisition of resistant bacteria from hospital environments, which are heavily influenced by infection control policies and antibiotic exposure (36, 37).

Data indicated a significant difference in MDROs distribution among hospitals (p-value <0.001), where northern region Hospital had the majority of CRE isolates compared with other hospitals, which suggested an uncontrolled outbreak (Figure S1).

Data shows that those who survived sepsis were 56.8% less likely to have a blood culture with MDROs compared to those who died [COR = 0.432 (95% CI: 0.269-0.636), p-value<0.001], which was similar to an Indian study in which neonates with MDROs NS had a 50% higher mortality rate than those without MDROs (40).

It was found that nearly half the neonates (43%) who had a CRE pattern of resistance had died, similar to a study in India that reported a mortality greater than 50% among CRE neonates (41).

Among all 884 cases of culture-proven sepsis, CoNS (56.4%), *Klebsiella species* (13.5%), and *Streptococcus* species (7.5%) were the most common pathogens (Table 3), despite the fact that CoNS percentage was overestimated due to the suspected contamination from skin flora. Results were close to those of the Gaza study, which found CoNS (57.3%), *E. coli* (10.4%), *Klebsiella* spp. (8.0%), and (7%) of *Streptococcus* spp. were the most common organisms (58).

Results showed a variation in antimicrobial susceptibility by years, sepsis class, and organism type, as presented in Tables S3 and S4.

However, all hospitals have standardized their susceptibility testing since May 2019 by using the Vitek2 compat device, and the suspected variation in results due to different sensitivity methods was limited to the first four months of 2019, where middle and southern region hospitals used traditional culturing methods for identification and sensitivity testing.

Among Gram-positive isolates, vancomycin sensitivity was 99.8%; on the other hand, among Gram-negative isolates, colistin sensitivity was 100% regardless of sepsis class; similar findings were obtained by the Gaza study and many studies worldwide, where colistin-resistant strains are still rare among neonatal populations (23, 42, 58).

Among Gram-positive bacteria, and after excluding *Staphylococcus* spp., ampicillin sensitivity was high 81.4%. Moreover, GBS cultures were 100% sensitive to ampicillin, similar to many other studies (58, 66, 67).

On the other hand, among Gram-negative bacteria, the sensitivity of ampicillin, cefotaxime, gentamicin, amikacin, meropenem, piperacillin-tazobactam, and ceftazidime were as follows: 7.1%, 21.8%, 33%, 63%, 70%, 65.6%, and 29%, respectively.

This indicated a reduced sensitivity among ampicillin, cephalosporin, and gentamicin and good coverage for amikacin, piperacillin-tazobactam, and meropenem, similar to many studies conducted among LMICs (9, 42–45, 53–56, 65).

4.1 Recommendation

1. Concerted efforts should be made by the stakeholders to implement strategies to decrease the burden of neonatal sepsis in Palestine, especially since the EOS rate was found to be 10 times higher than the rate at HICs.

2. Infection control policies and dedicated specialized teams should be available at each institution and work cooperatively with the ministry of health to assure proper routine practices and the control of any outbreaks.

3. Antimicrobial stewardship programs should be implemented at NICUs to prevent the rapid emergence of MDROs among the neonatal population.

4. WHO empiric antibiotic regimens of (ampicillin-gentamicin) and (Ampicillin-cefotaxime) are not effective any more for treating neonatal sepsis due to decreased susceptibility in Palestine, where amikacin-based regimens would be a good option.

5. There is an urgent need for developing institutional-specific protocols for treating neonatal sepsis depending on the local antibiogram for NICUs and according to sepsis class, site of infection, and co-morbidities.

4.2 Conclusion

In Palestine, the prevalence of NS was found to be 10 times higher than that of HICs for early onset sepsis. Data from culture-proven NS showed that CoNS (56.4%), *Klebsiella species* (13.5%), and *Streptococcus* species (7.5%) are the most common pathogens isolated. Despite the fact that around (25.5%) of the gram-positive bacteria predominantly CoNS were suspected to be contamination not true infection.

Antimicrobial resistance among the causative organisms of neonatal sepsis is rising rapidly and contributing to a significant mortality rate among the neonatal population, where the matching probability to WHO empiric antibiotic regimens was found to be very low.

However, amikacin-based empiric regimens are advised to be implemented after the evaluation of institution-specific antibiograms.

List of Abbreviations

Abbreviation	Meaning
AAP	American Academy of Pediatrics
AOR	Adjusted Odds Ratio
CI	Confidence Interval
CoNS	Coagulase Negative Staphylococcus
COR	Crude Odds Ratio
CRE	Carbapenem Resistant Enterobacteriaceae
EOS	Early Onset Sepsis
ESBL	Extended Spectrum Beta Lactamase
GBS	Group B streptococcus
HIC	High Income Country
LMIC	Low-Middle Income Country
LOS	Late Onset Sepsis
MDR	Multi Drug Resistant
MDRO	Multi Drug Resistant Organism
MRSA	Methicillin Resistant Staphylococcus Aureus
MSSA	Methicillin Sensitive Staphylococcus Aureus
NS	Neonatal Sepsis
PSBI	Possible Significant Bacterial Infection
VRE	Vancomycin Resistant Enterococcus
WHO	World Health Organization

References

1. Global, regional, and national under-5 mortality, adult mortality, age-specific mortality, and life expectancy, 1970-2016: a systematic analysis for the Global Burden of Disease Study 2016. Lancet (London, England). 2017;390(10100):1084-150.

2. Reinhart K, Daniels R, Kissoon N, Machado FR, Schachter RD, Finfer S. Recognizing Sepsis as a Global Health Priority — A WHO Resolution. New England Journal of Medicine. 2017;377(5):414-7.

3. Fleischmann-Struzek C, Goldfarb DM, Schlattmann P, Schlapbach LJ, Reinhart K, Kissoon N. The global burden of paediatric and neonatal sepsis: a systematic review. The Lancet Respiratory medicine. 2018;6(3):223-30.

4. Global, regional, and national age-sex specific mortality for 264 causes of death, 1980-2016: a systematic analysis for the Global Burden of Disease Study 2016. Lancet (London, England). 2017;390(10100):1151-210.

5. ATLAS WD. Palestine Infant mortality rate 1960-2020 [cited 2022 15 November 2021]. Available from: https://knoema.com/atlas/Palestine/Infant-mortality-rate.

6. Wynn JL. Defining neonatal sepsis. Current opinion in pediatrics. 2016;28(2):135-40.

7. Zea-Vera A, Ochoa TJ. Challenges in the diagnosis and management of neonatal sepsis. Journal of tropical pediatrics. 2015;61(1):1-13.

8. Wynn JL, Wong HR, Shanley TP, Bizzarro MJ, Saiman L, Polin RA. Time for a neonatal-specific consensus definition for sepsis. Pediatric critical care medicine: a journal of the Society of Critical Care Medicine and the World Federation of Pediatric Intensive and Critical Care Societies. 2014;15(6):523-8.

9. Jatsho J, Nishizawa Y, Pelzom D, Sharma R. Clinical and Bacteriological Profile of Neonatal Sepsis: A Prospective Hospital-Based Study. International journal of pediatrics. 2020;2020.

10. Clinical signs that predict severe illness in children under age 2 months: a multicentre study. Lancet (London, England). 2008;371(9607):135-42.

11. Fuchs A, Bielicki J, Mathur S, Sharland M, Van Den Anker JN. Reviewing the WHO guidelines for antibiotic use for sepsis in neonates and children. Paediatr Int Child Health. 2018;38(sup1):S3-S15.

12. Simonsen KA, Anderson-Berry AL, Delair SF, Davies HD. Early-onset neonatal sepsis. Clinical microbiology reviews. 2014;27(1):21-47.

13. Duber HC, Hartford EA, Schaefer AM, Johanns CK, Colombara DV, Iriarte E, et al. Appropriate and timely antibiotic administration for neonatal sepsis in Mesoamérica. BMJ Glob Health. 2018;3(3):e000650-e.

14. Fowler NT, Garcia M, Hankins C. Impact of Integrating a Neonatal Early-Onset Sepsis Risk Calculator into the Electronic Health Record. Pediatric quality & safety. 2019;4(6):e235.

15. Riskin A, Bryskin S, Zaitoon H, Toropine A, Iofe A, Zoabi-Safadi R, et al. Evaluation of Implementation of Early-Onset Sepsis Calculator in Newborns in Israel. The Journal of pediatrics. 2021;234:71-6.e2.

16. van der Weijden BM, Achten NB, Bekhof J, Evers EE, van Dongen O, Rijpert M, et al. Neonatal early-onset sepsis calculator recommended significantly less empiric antibiotic treatment than national guidelines. 2020;109(12):2549-51.

17. Singh M, Alsaleem M, Gray CP. Neonatal Sepsis. StatPearls. Treasure Island (FL): StatPearls Publishing Copyright © 2022, StatPearls Publishing LLC.; 2022.

18. Urzúa S, Ferrés M, García P, Sánchez A, Luco M. [Strategies to reduce infections and antibiotic use and its effects in a neonatal care unit]. Rev Chilena Infectol. 2017;34(2):99-107.

19. Dramowski A, Aucamp M, Beales E, Bekker A, Cotton MF, Fitzgerald FC, et al. Healthcare-Associated Infection Prevention Interventions for Neonates in Resource-Limited Settings. Front Pediatr. 2022;10:919403.

20. Organization WH. Pocket book of hospital care for children: guidelines for the management of common illnesses with limited resources: World Health Organization; 2005.

21. Kim HI, Park S. Sepsis: Early Recognition and Optimized Treatment. 2019;82(1):6-14.

22. Thomson KM, Dyer C, Liu F, Sands K, Portal E, Carvalho MJ, et al. Effects of antibiotic resistance, drug target attainment, bacterial pathogenicity and virulence, and antibiotic access and affordability on outcomes in neonatal sepsis: an international microbiology and drug evaluation prospective substudy (BARNARDS). The Lancet Infectious diseases. 2021;21(12):1677-88.

23. Thomson KM, Dyer C, Liu F, Sands K, Portal E, Carvalho MJ, et al. Effects of antibiotic resistance, drug target attainment, bacterial pathogenicity and virulence, and antibiotic access and affordability on outcomes in neonatal sepsis: an international microbiology and drug evaluation prospective substudy (BARNARDS). The Lancet Infectious Diseases. 2021;21(12):1677-88.

24. Afsharpaiman S, Torkaman M, Saburi A, Farzaampur A, Amirsalari S, Kavehmanesh Z. Trends in incidence of neonatal sepsis and antibiotic susceptibility of causative agents in two neonatal intensive care units in tehran, I.R iran. Journal of clinical neonatology. 2012;1(3):124-30.

25. Tayal M, Sharma AK, Sharma R. Incidence of culture proven neonatal sepsis, pattern of antibiotic sensitivity and clinical course in neonatal intensive care unit in tertiary care center in North India. 2019. 2019;6(6):9.

26. Tumuhamye J, Sommerfelt H, Bwanga F, Ndeezi G, Mukunya D, Napyo A, et al. Neonatal sepsis at Mulago national referral hospital in Uganda: Etiology, antimicrobial resistance, associated factors and case fatality risk. PloS one. 2020;15(8):e0237085.

27. Lin PC, Chang CL, Chung YH, Chang CC, Chu FY. Revisiting factors associated with blood culture positivity: Critical factors after the introduction of automated continuous monitoring blood culture systems. 2022;101(30):e29693.

28. Medhat H, Khashana A, El kalioby M. Incidence of Neonatal Infection in South Sinai, Egypt. 2017;4(1):e36615.

29. Chan GJ, Lee AC, Baqui AH, Tan J, Black RE. Risk of early-onset neonatal infection with maternal infection or colonization: a global systematic review and meta-analysis. PLoS medicine. 2013;10(8):e1001502.

30. Downey LC, Smith PB, Benjamin DK, Jr. Risk factors and prevention of late-onset sepsis in premature infants. Early human development. 2010;86 Suppl 1(Suppl 1):7-12.

31. Giannoni E, Dimopoulou V, Klingenberg C, Navér L, Nordberg V, Berardi A, et al. Analysis of Antibiotic Exposure and Early-Onset Neonatal Sepsis in Europe, North America, and Australia. JAMA Network Open. 2022;5(11):e2243691-e.

32. Sands K, Spiller OB, Thomson K, Portal EAR, Iregbu KC, Walsh TR. Early-Onset Neonatal Sepsis in Low- and Middle-Income Countries: Current Challenges and Future Opportunities. Infection and drug resistance. 2022;15:933-46.

33. Schlapbach LJ, Hagmann C, Giannoni E. Time to tackle early-onset sepsis in low-income and middle-income countries. The Lancet Global health. 2022;10(5):e592-e3.

34. Milton R, Gillespie D, Dyer C, Taiyari K, Carvalho MJ, Thomson K, et al. Neonatal sepsis and mortality in low-income and middle-income countries from a facility-based birth cohort: an international multisite prospective observational study. The Lancet Global health. 2022;10(5):e661-e72.

35. Glikman D, Curiel N, Glatman-Freedman A, Megged O, Youngster I, Marom R. Nationwide epidemiology of early-onset sepsis in Israel 2010-2015, time to re-evaluate empiric treatment. 2019;108(12):2192-8.

36. Flannery DD, Chiotos K, Gerber JS, Puopolo KM. Neonatal multidrug-resistant gram-negative infection: epidemiology, mechanisms of resistance, and management. Pediatric research. 2022;91(2):380-91.

37. Martínez JL, Baquero F. Emergence and spread of antibiotic resistance: setting a parameter space. Upsala journal of medical sciences. 2014;119(2):68-77.

38. Flokas ME, Karanika S, Alevizakos M, Mylonakis E. Prevalence of ESBL-Producing Enterobacteriaceae in Pediatric Bloodstream Infections: A Systematic Review and Meta-Analysis. PloS one. 2017;12(1):e0171216.

39. Ballot DE, Bandini R, Nana T, Bosman N, Thomas T, Davies VA, et al. A review of -multidrug-resistant Enterobacteriaceae in a neonatal unit in Johannesburg, South Africa. BMC Pediatr. 2019;19(1):320.

40. Wattal C, Kler N, Oberoi JK, Fursule A, Kumar A, Thakur A. Neonatal Sepsis: Mortality and Morbidity in Neonatal Sepsis due to Multidrug-Resistant (MDR) Organisms: Part 1. Indian journal of pediatrics. 2020;87(2):117-21.

41. Smith A, Anandan S, Veeraraghavan B, Thomas N. Colonization of the Preterm Neonatal Gut with Carbapenem-resistant Enterobacteriaceae and Its Association with Neonatal Sepsis and Maternal Gut Flora. Journal of global infectious diseases. 2020;12(2):101-4.

42. Li G, Bielicki JA, Ahmed ANU, Islam MS, Berezin EN, Gallacci CB, et al. Towards understanding global patterns of antimicrobial use and resistance in neonatal sepsis: insights from the NeoAMR network. Archives of disease in childhood. 2020;105(1):26

43. Thomson KM, Dyer C, Liu F, Sands K, Portal E, Carvalho MJ, et al. Effects of antibiotic resistance, drug target attainment, bacterial pathogenicity and virulence, and antibiotic access and affordability on outcomes in neonatal sepsis: an international microbiology and drug evaluation prospective substudy (BARNARDS). The Lancet Infectious Diseases. 2021;21(12):1677-88.

44. Braima OA, Ali MA, Abdulla EM. Bacteriological profile and antibiotic resistance in newborn infants with possible community-acquired neonatal sepsis in Khartoum State, Sudan. Sudanese journal of paediatrics. 2021;21(1):13.

45. Oluyinka O, Airede KI, Olateju KE, Stephen OK, Izevbigie N, Iloh KK, et al. Incidence, bacteriological profile and antibiotic sensitivity pattern of neonatal sepsis in a tertiary health facility in Abuja, North-central Nigeria. 2021.

46. Ehtizaz L, Haider S, Fayyaz M, Ansar A, Nazir S, Rafique S. Neonatal sepsis and resistance pattern of isolates in Tertiary level neonatal unit: Time to evaluate the empirical antibiotics selection. Journal of Rawalpindi Medical College. 2021;25(2):264

47. Oo NAT, Edwards JK, Pyakurel P, Thekkur P, Maung TM, Aye NSS, et al. Neonatal Sepsis, Antibiotic Susceptibility Pattern, and Treatment Outcomes among Neonates Treated in Two Tertiary Care Hospitals of Yangon, Myanmar from 2017 to 2019. Tropical medicine and infectious disease. 2021;6(2):62.

48. Khan A, Farhat A, Ghaffar MS, Sajid NK, Jamil M, Mahmood T. Neonatal Sepsis: Determine the Etiology and Antimicrobial Patterns in Pediatrics. positivity. 2021;16:17.

49. Ahmad A, Sarwar N, Aslam R, Ali S, Aslam B, Arshad MA, et al. Pattern of clinical drug resistance and occurrence of Gram negative bacterial neonatal sepsis at a tertiary care hospital. Pakistan Journal of Pharmaceutical Sciences. 2021;34.

50. Sah R, Bhattarai S, Basnet S, Pokharel B, Shah NP, Sah S, et al. A Prospective Study on Neonatal Sepsis in a Tertiary Hospital, Nepal. Journal of Pure and Applied Microbiology. 2021;15.

51. Hussain SQ, Qayoom I, Ramzan A. TO STUDY THE BACTERIOLOGICAL PROFILE OF COMMUNITY ACQUIRED AND HOSPITAL ACQUIRED NEONATAL SEPSIS AND TO STUDY THE SENSITIVITY OF THE CAUSATIVE ORGANISMS TO ANTIBIOTICS. 2021.

52. Li X, Ding X, Shi P, Zhu Y, Huang Y, Li Q, et al. Clinical features and antimicrobial susceptibility profiles of culture-proven neonatal sepsis in a tertiary children's hospital, 2013 to 2017. Medicine. 2019;98(12).

53. Pokhrel B, Koirala T, Shah G, Joshi S, Baral P. Bacteriological profile and antibiotic susceptibility of neonatal sepsis in neonatal intensive care unit of a tertiary hospital in Nepal. BMC pediatrics. 2018;18(1):1-8.

54. Aku FY, Akweongo P, Nyarko K, Sackey S, Wurapa F, Afari EA, et al. Bacteriological profile and antibiotic susceptibility pattern of common isolates of neonatal sepsis, Ho Municipality, Ghana-2016. Maternal health, neonatology and perinatology. 2018;4(1):1-8.

55. Yadav NS, Sharma S, Chaudhary DK, Panthi P, Pokhrel P, Shrestha A, et al. Bacteriological profile of neonatal sepsis and antibiotic susceptibility pattern of isolates admitted at Kanti Children's Hospital, Kathmandu, Nepal. BMC research notes. 2018;11(1):301.

56. Kumar DVP, Mohan J, Rakesh P, Prasad J, Joseph L. Bacteriological profile of neonatal sepsis in a secondary care hospital in rural Tamil Nadu, Southern India. Journal of family medicine and primary care. 2017;6(4):735.

57. Haque KN. Definitions of bloodstream infection in the newborn. Pediatric critical care medicine: a journal of the Society of Critical Care Medicine and the World Federation of Pediatric Intensive and Critical Care Societies. 2005;6(3 Suppl):S45-9.

58. Jadba AEHN, Yazji MS. Neonatal septicemia in Gaza City hospitals. Pakistan Journal of Medical Sciences. 2009;25:226-31.

59. Magiorakos AP, Srinivasan A, Carey RB, Carmeli Y, Falagas ME, Giske CG, et al. Multidrug-resistant, extensively drug-resistant and pandrug-resistant bacteria: an international expert proposal for interim standard definitions for acquired resistance. Clinical Microbiology and Infection. 2012;18(3):268-81.

60. Tappero E, Johnson P. Laboratory Evaluation of Neonatal Sepsis. Newborn and Infant Nursing Reviews. 2010;10(4):209-17.

61. Murni IK, Duke T, Daley AJ, Kinney S, Soenarto Y. True Pathogen or Contamination: Validation of Blood Cultures for the Diagnosis of Nosocomial

Infections in a Developing Country. Journal of tropical pediatrics. 2018;64(5):389-94.

62. Hammoud MS, Al-Taiar A, Al-Abdi SY, Bozaid H, Khan A, AlMuhairi LM, et al. Culture-proven early-onset neonatal sepsis in Arab states in the Gulf region: two-year prospective study. International journal of infectious diseases: IJID: official publication of the International Society for Infectious Diseases. 2017;55:11-5.

63. Gaballah AH, Shawky S, Amer AN. Microbiological profiles of neonatal sepsis in northern Egypt. Microbes and Infectious Diseases. 2022;3(3):645-56.

64. Clark RH, Bloom BT, Spitzer AR, Gerstmann DR. Empiric use of ampicillin and cefotaxime, compared with ampicillin and gentamicin, for neonates at risk for sepsis is associated with an increased risk of neonatal death. Pediatrics. 2006;117(1):67-74.

65. Jatsho J, Nishizawa Y, Pelzom D, Sharma R. Clinical and Bacteriological Profile of Neonatal Sepsis: A Prospective Hospital-Based Study. Int J Pediatr. 2020;2020:1835945.

66. Akkala V, Sudhakar B. A study of bacteriological profile in neonatal sepsis in neonatal intensive care unit in a tertiary care centre.

67. Chen T, Ma HJ, Yang J, Liu W, Dai XT, Tong H. [An analysis of relative factors, bacteriological profile and antibiogram of neonatal sepsis]. Zhonghua yu fang yi xue za zhi [Chinese journal of preventive medicine]. 2018;52(10):1023-8.

Appendices

Appendix A

Supplementary Tables

Supplementary Table S1:

Distribution of Gram-positive causative bacteria, by hospitals, year and sepsis class of three tertiary care hospitals in Palestine, January 2019 to December 2021.

	CONS total Number=499 total %= 56.4	***Staph. Aureus*** Total Number=38 total %=4.3	***Enterococcus.*spp** Total Number=21 total %=2.4	***Strep.*spp** Total Number=64 total %=7.5	***Bacillus* spp.** Total Number=12 total %= 1.2	***Micrococcus*** Total Number=11 total %=1
	Organism contribution among culture proven sepsis n (% n/N of each year)					
2019						
Middle N=38	19(50)	5(13)	0(0)	3(8)	3(8)	2(5)
Southern N=51	27(51)	6(12)	0(0)	3(6)	1(2)	1(2)
Northern N=178	109(61)	5(3)	6(3)	18(10)	3(1.6)	1(1)
Total N =267	155(58)	16(6)	6(2)	24(9)	7(2.6)	4(1)
2020						
Middle N=60	31(52)	12(20)	0(0)	1(2)	0(0)	0(0)
Southern N=77	45(58)	0(0)	3(3.7)	7(9)	1(1)	0(0)
Northern N=163	92(56)	2(1.2)	5(3)	13(8	1(1)	2(1)
Total N= 300	168(55)	14(5)	8(2)	21(7)	2(1)	2(0.3)
2021						
Middle N=53	25(47)	1(1.5)	3(5.6)	2(4)	0(0)	0(0)
Southern N=81	53(65)	2(2.4)	4(5)	9(11)	1(1)	0(0)
Northern =183	98(54)	5(3)	0(0)	8(4.3)	2(1)	5(3)
Total N=317	176(56)	8(2.5)	7(1)	19(6)	3(1)	5(1.5)

Sepsis class	EOS n (%)	LOS n (%)	EOS n (%)	LOS n (%)	EOS n (%)	LOS n (%)	EOS n (%)	LOS n (%)	EOS n (%)	LOS n (%)	EOS n (%)	LOS n (%)
2019												
Middle	4(21)	15(79)	0(0)	5(100)	0(0)	0(0)	3(100)	0(0)	3(100)	0(0)	1(50)	1(50)
Southern	11(41)	16(59)	3(50)	3(50)	0(0)	0(0)	3(100)	0(0)	1(100)	0(0)	1(100)	0(0)
Northern	32(92)	77(71)	0(0)	5(100)	2(33)	4(67)	8(48)	9(52)	0(0)	3(100)	0(0)	1(100)
Total	**47(30)**	**108(70)**	**3(19)**	**13(81)**	**2(33)**	**4(67)**	**14(61)**	**9(39)**	**4(55)**	**3(45)**	**2(50)**	**2(50)**
2020												
Middle	10(32)	21(68)	1 (8)	11(92)	0(0)	0(0)	1(100)	0(0)	0(0)	0(0)	0(0)	0(0)
Southern	18(40)	27(60)	0(0)	0(0)	1(100)	0(0)	7(100)	0(0)	1(100)	0(0)	0(0)	0(0)
Northern	36(40)	56(60)	1(25)	4(75)	0(0)	5(100)	8(61)	5(39)	1(100)	0(0)	0(0)	1(100)
Total	**64(38)**	**104(62)**	**2(12)**	**15(88)**	**1(17)**	**5(83)**	**16(76)**	**5(24)**	**2(100)**	**0(0)**	**0(0)**	**1 (100)**
2021												
Middle	5(20)	20(80)	1(100)	0(0)	1(33)	2(67)	1(50)	1(50)	0(0)	0(0)	0(0)	0(0)
Southern	20(38)	33(62)	1(50)	1(50)	2(50)	2(50)	4(44)	5(56)	1(100)	0(0)	0(0)	0(0)
Northern	33(34)	65(66)	0(0)	5(100)	0(0)	0(0)	4(50)	4(50)	1 (50)	1(50)	3(60)	2(40)
Total	**58(33)**	**118(67)**	**2(40)**	**6(60)**	**3(43)**	**4(57)**	**9(47)**	**10(53)**	**2(67)**	**1(33)**	**3(60)**	**2(40)**

Supplementary Table S2:

Distribution of Gram-negative causative bacteria, by hospitals, year and sepsis class of three tertiary care hospitals in Palestine, January 2019 to December 2021.

	***Klebsiella* spp** Total number=119 Total % = 13.5	***E.coli*** Total number=47 Total %=4.8	***Pseudomons.sp*** Total number=10 Total %=1.2	***Acinetobacter*** Total number=29 Total %=33	***Stenotrophomons*** Total number=11 Total %=1.2	***Enterobacter*** Total number=4 Total%=0.5	***Serratia*** Total number=4 Total%=0.5
	Organism contribution among culture proven sepsis n (% n/N)						
2019							
Middle N=38	5(13)	2(5)	0(0)	3(8)	1(3)	1(3)	0(0)
Southern N=51	5(10)	3(6)	0(0)	0(0)	0(0)	0(0)	0(0)
NorthernN=178	15(8)	10(5.6)	2(1)	12(7)	2(1)	0(0)	1(0.6)
Total N =267	**25(7)**	**15(6)**	**2(1)**	**15(6)**	**3(1)**	**1(0.4)**	**1(0.3)**
2020							
Middle N=60	6(10)	5(8)	1(2)	0(0)	0(0)	1(2)	0(0)
Southern N=77	15(19)	1(1)	1(1)	1(1)	0(0)	0(0)	0(0)
NorthernN=163	26(16)	8(5)	4(2)	10(6)	3(2)	0(0)	1(0.6)
Total N= 300	**47(16)**	**14(5)**	**6(2)**	**11(4)**	**3(1)**	**1(0.3)**	**1(0.3)**
2021							
Middle N=53	6(11)	11(21)	0(0)	0(0)	0(0)	1(2)	0(0)
Southern N=81	8(10)	2(2)	0(0)	0(0)	2(2)	0(0)	0(0)
NorthernN=183	33(18)	5(3)	2(1)	3(2)	3(2)	1(0.5)	2(1)
Total N=317	**47(8)**	**18(6)**	**2(1)**	**3(1)**	**5(2)**	**2(0.6)**	**2(0.6)**

Sepsis class	EOS n (%)	LOS n (%)	EOS n (%)	LOS n (%)	EOS n (%)	LOS n (%)	EOS n (%)	LOS n (%)	EOS n (%)	LOS n (%)	EOS n(%)	LOS n(%)	EOS n (%)	LOS n (%)
2019														
Middle	0(0)	5(100)	0(0)	2(100)	0(0)	0(0)	0(0)	3(100)	0(0)	1(100)	0(0)	1(100)	0(0)	0(0)
Southern	0(0)	5(100)	1(33)	2(67)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Northern	2(13)	13(87)	3(40)	5(60)	1(50)	1(50)	3(25)	9(75)	1(50)	1(50)	0(0)	0(0)	0(0)	1(100)
Total	**2(8)**	**23(92)**	**4(31)**	**9(69)**	**1(50)**	**1(50)**	**3(20)**	**12(80)**	**1(33)**	**2(67)**	**0(0)**	**1(100)**	**0(0)**	**1(100)**
2020														
Middle	1(17)	5(83)	2(40)	3(60)	0(0)	1(100)	0(0)	0(0)	0(0)	0(0)	0(0)	1(100)	0(0)	0(0)
Southern	2(13)	13(87)	0(0)	1(100)	0(0)	1(100)	0(0)	1(100)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Northern	2(8)	24(92)	2(25)	6(75)	1(25)	3(75)	4(40)	6(60)	1(33)	2(67)	0(0)	0(0)	0(0)	1(100)
Total	**5(11)**	**42(89)**	**4(29)**	**10(71)**	**1(17)**	**5(83)**	**4(36)**	**7(64)**	**1(33)**	**2(67)**	**0(0)**	**1(100)**	**0(0)**	**1(100)**
2021														
Middle	0(0)	6(100)	2(8)	9(82)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	1(100)	0(0)	0(0)	0(0)
Southern	2(25)	6(75)	1(50)	1(50)	0(0)	0(0)	0(0)	0(0)	1(50)	1(50)	0(0)	0(0)	0(0)	0(0)
Northern	11(33)	22(67)	1(20)	4(80)	1(50)	1(50)	0(0)	3(100)	1(33)	2(67)	0(0)	1(100)	0(0)	2(100)
Total	**13(28)**	**34(72)**	**4(22)**	**14(78)**	**1(50)**	**1(50)**	**0(0)**	**3(100)**	**2(40)**	**3(60)**	**1(50)**	**1(50)**	**0(0)**	**2(100)**

Supplementary Table S3:

Sensitivity of major Gram-positive bacteria isolates

Gram-positive major bacterial isolates sensitivity N= number of tested isolates / n= number of sensitive isolates n(%n/N)												
Antibiotic	**CoNS**			***Staphylococcus aureus***			***Strep.*spp.**			***Enterococcus* spp.**		
	EOS	**LOS**	**Total**	**EOS**	**LOS**	**Total**	**EOS**	**LOS**	**Total**	**EOS**	**LOS**	**Total**
Ampicillin												
2019	NT	NT	NT	NT	NT	NT	14(93)	4(57)	18(82)	2(100)	2(50)	4(66)
2020	NT	NT	NT	NT	NT	NT	14(88)	3(100)	17(89)	1(100)	6(86)	7(87)
2021	NT	NT	NT	NT	NT	NT	15(94)	10(71)	25(86)	3(75)	3(60)	6(75)
Gentamicin												
2019	27(59)	86(57)	113(58)	5(83)	8(67)	13(72)	NT	NT	NT	2(100)	4(100)	6(100)
2020	32(50)	47(45)	79(48)	0(0)	4(27)	4(24)	NT	NT	NT	1(100)	5(71)	6(75)
2021	36(61)	35(29)	71(40)	2(67)	4(80)	6(74)	NT	NT	NT	3(75)	3(60)	6(75)
Cefoxitine												
2019	24(52)	51(33)	75(38)	5(83)	4(33)	9(50)	NT	NT	NT	NT	NT	NT
2020	38(58)	18(17)	56(33)	0(0)	1(30)	1(6)	NT	NT	NT	NT	NT	NT
2021	34(58)	14(11)	48(27)	2(67)	1(20)	3(37)	NT	NT	NT	NT	NT	NT
Vancomycin												
2019	46(100)	152(100)	198(100)	6(100)	12(100)	18(100)	15(100)	7(100)	22(100)	2(100)	4(100)	6(100)
2020	65(100)	104(100)	169(100)	2(100)	15(100)	17(100)	16(100)	3(100)	19(100)	1(100)	7(100)	8(100)
2021	59(100)	122(100)	181(100)	3(100)	5(100)	8(100)	16(100)	14(100)	30(100)	4(100)	4(80)	8(89)
TMP-SMX												
2019	25(54)	77(51)	102(52)	6(100)	6(50)	12(66)	14(93)	4(57)	18(81)	NT	NT	NT
2020	29(45)	45(43)	74(44)	1(50)	6(40)	7(41)	11(69)	3(100)	14(74)	NT	NT	NT
2021	41(69)	54(44)	95(52)	2(67)	2(40)	4(50)	10(71)	10(70)	20(70)	NT	NT	NT

Amoxi-Clav												
2019	NT	NT	NT	NT	NT	NT	15(100)	5(71)	20(86)	NT	NT	NT
2020	NT	NT	NT	NT	NT	NT	15(94)	2(67)	17(81)	NT	NT	NT
2021	NT	NT	NT	NT	NT	NT	16(100)	11(79)	27(90)	NT	NT	NT
Cefotaxime												
2019	NT	NT	NT	NT	NT	NT	14(93)	4(57)	18(75)	NT	NT	NT
2020	NT	NT	NT	NT	NT	NT	15(94)	3(100)	18(97)	NT	NT	NT
2021	NT	NT	NT	NT	NT	NT	15(94)	11(79)	16(87)	NT	NT	NT
NT: not tested												

Supplementary Table S4:

Sensitivity of major Gram-negative bacteria isolates

Gram negative major bacterial isolates sensitivity N= number of tested isolates / n= number of sensitive isolates n(%n/N)												
Antibiotic	***Klebsiella* spp.**			***Escherichia coli***			***Pseudomonas* spp.**			***Acinetobacter* spp.**		
	EOS	**LOS**	**Total**	**EOS**	**LOS**	**Total**	**EOS**	**LOS**	**Total**	**EOS**	**LOS**	**Total**
Ampicillin												
2019	1(33)	1(4)	2(9)	1(33)	0(0)	1(9)	NT	NT	NT	NT	NT	NT
2020	0(0)	0(0)	0(0)	1(25)	2(18)	3(20)	NT	NT	NT	NT	NT	NT
2021	1(7)	0(0)	1(2)	1(25)	5(33)	6(32)	NT	NT	NT	NT	NT	NT
Gentamicin												
2019	1(33)	9(39)	10(36)	1(33)	4(50)	5(45)	1(100)	2(100)	3(100)	0(0)	1(8)	1(6)
2020	2(40)	9(40)	11(61)	3(75)	5(45)	8(53)	1(100)	6(100)	7(100)	2(50)	1(13)	3(25)
2021	4(29)	6(18)	10(21)	2(50)	7(47)	9(32)	1(100)	1(100)	2(100)	-	1(33)	1(33)
Amikacin												
2019	3(100)	8(78)	21(81)	3(100)	7(88)	10(91)	1(100)	2(100)	3(100)	0(0)	1(8)	1(6)
2020	5(100)	33(77)	38(79)	4(100)	9(82)	13(87)	1(100)	6(100)	7(100)	2(50)	1(13)	3(25)
2021	6(43)	15(44)	21(44)	3(75)	12(80)	15(79)	1(100)	1(100)	2(100)	-	1(33)	1(33)
Cefotaxime												
2019	1(33)	3(13)	4(15)	1(33)	2(25)	3(27)	NT	NT	NT	0(0)	0(0)	0(0)
2020	2(40)	3(7)	5(10)	3(75)	5(45)	8(53)	NT	NT	NT	2(50)	1(13)	3(25)
2021	3(21)	2(6)	5(10)	2(50)	6(40)	8(42)	NT	NT	NT	-	0(0)	0(0)
Ceftazidime												
2019	1(33)	2(7)	3(12)	1(33)	2(25)	3(27)	1(100)	2(100)	3(100)	0(0)	1(8)	1(6)
2020	3(60)	3(7)	6(13)	3(75)	5(45)	8(53)	1 (100)	6(100)	7(100)	2(50)	1(13)	3(25)
2021	3(21)	2(6)	5(10)	2(50)	8(53)	10(53)	1(100)	1(100)	2(100)	-	0(0)	0(0)

Meropenem												
2019	3(100)	18(78)	21(81)	3(100)	8(100)	11(100)	1(100)	2(100)	3(100)	0(0)	1(8)	1(6)
2020	5(100)	39(91)	44(92)	11(100)	15(100)	15(100)	1(100)	6(100)	7(100)	2(50)	2(25)	4(33)
2021	8(87)	18(53)	26(54)	3(75)	14(93)	17(89)	1(100)	1(100)	2(100)	-	1(33)	1(33)
Pip-Tazo												
2019	3(100)	17(73)	20(78)	3(100)	8(100)	11(100)	1(100)	2(100)	3(100)	0(0)	1(8)	1(6)
2020	5(100)	38(88)	43(90)	4(100)	10(91)	14(93)	1(100)	6(100)	7(100)	2(50)	2(25)	4(33)
2021	7(50)	17(50)	24(50)	3(75)	14(93)	17(51)	1(100)	1(100)	2(100)	-	1(33)	1(33)
Amoxi-Clav												
2019	1(33)	1(4)	2(8)	0(0)	2(25)	2(18)	NT	NT	NT	0(0)	0(0)	0(0)
2020	2(20)	2(5)	4(6)	2(50)	3(27)	5(33)	NT	NT	NT	0(0)	0(0)	0(0)
2021	3(21)	3(9)	6(13)	1(25)	5(33)	6(32)	NT	NT	NT	-	0(0)	0(0)
TMP-SMX												
2019	1(33)	3(13)	4(15)	1(33)	1(13)	2(18)	NT	NT	NT	0(0)	0(0)	0(0)
2020	2(40)	2(5)	4(8)	2(50)	3(27)	5(33)	NT	NT	NT	1(25)	0(0)	1(8)
2021	1(7)	2(6)	3(6)	1(25)	5(33)	6(32)	NT	NT	NT	-	0(0)	0(0)
Colistin												
2019	NT	NT	NT	NT	NT	NT	NT	NT	NT	3(100)	12(100)	15(100)
2020	NT	9(100)	9(100)	2(100)	3(100)	5(100)	NT	NT	NT	1(100)	8(100)	10(100)
2021	6(100)	16(100)	22(100)	2(100)	5(100)	7(100)	NT	NT	NT	-	1(100)	1(100)

Supplementary Table S5:

Univariate analysis for sepsis class and other factors

Variable	Category	EOS	LOS	OR	95%CI	P value
Gender	Female	109(37.2)	258(43.7)	0.765	0.574-1.019	0.070
	Male	184(62.8)	333(56.3)			
Gram stain	Negative	52(17.9)	173(29.9)	0.509	0.359-0.722	<0.001
	Positive	239(82.1)	405(70.1)			
MDR	Yes	32(10.9)	159(26.9)	0.333	0.221-0.502	<0.001
	No	261(89.1)	432(73.1)			
Mortality	lived	233(79.5)	499(84.4)	0.716	0.499-1.027	0.072
	Died	60(20.5)	92(15.6)			
Gestational age	Term	214(73)	438(74.1)	0.946	0.689-1.299	0.746
	Preterm	79(27)	153(25.9)			
Ampicillin-Genatimicin matching status	Matched	86(29.4)	93(15.7)	2.225	1.591-3.110	<0.001
	Not matched	2.7(70.6)	498(84.3)			
Ampicilllin-Cefotaxime matching status	Matched	75(25.6)	67(11.3)	2.691	1.867-3.877	<0.001
	Not matched	218(74.4)	524(88.7)			
Hospital	Northern	170(58)	354(59.9)			0.001
	Southern	88(30)	121(20.5)			
	Middle	35(11.9)	116(19.6)			
Year	2019	87(29.7)	180(30.5)			0.979
	2020	100(34.1)	200(33.8)			
	2021	106(36.2)	211(35.7)			

Appendix B

Approval from the Faculty of Higher education

An-Najah
National University
Faculty of Graduate Studies
Dean's Office

جامعة
النجـــاح الوطنية
كلية الدراسات العليا
مكتب العميد

التاريخ: 2021/2712

حضرة الدكتورة سعاد بكير المحترمة
منسقة برنامج ماجستير الامراض المعدية

تحية طيبة وبعد،

الموضوع: الموافقة على عنوان الاطروحة وتحديد المشرف

قرر مجلس كلية الدراسات العليا في جلسته رقم (413) المنعقدة بتاريخ 2021/12/6، الموافقة على مشروع الأطروحة المقدم من الطالبة/ بيان عاصم محمد ابراهيم، رقم التسجيل 12053391، تخصص ماجستير الامراض المعدية، عنوان الأطروحة:

نموذج ونوع البكتيريا المصاحبة لإلتهاب الدم في العناية الحثيثة لحديثي الولادة، فلسطين (2019-2021)
Bacteriological Profile and Antibiotic Susceptibility Pattern of Septicemia in Neonatal Intensive Care Units, Palestine (2019-2021)

بإشراف: د. محمد القادري

ملاحظة: لاعتماد الأطروحة وتسجيلها في الفصل الثاني 2022/2021.

يرجى اعلام المشرف والطالب بضرورة تسجيل الاطروحة خلال اسبوعين من تاريخ اصدار الكتاب، وفي حال عدم تسجيل الطالبة/ للاطروحة في الفترة المحددة لها/ ستقوم كلية الدراسات العليا بإلغاء اعتماد العنوان والمشرف

وتفضلوا بقبول وافر الاحترام ،،،

عميد كلية الدراسات العليا
أ.د. وليد صويلح

نسخة: د. رئيس قسم الدراسات العليا للعلوم الطبية والصحية المحترم
: عميد القبول والتسجيل المحترم
: مشرف الطالب

جامعة النجاح الوطنية من أفضل 500 جامعة على مستوى العالم في تصنيف التايمز البريطاني 2022

فلسطين، نابلس، ص.ب 7-707 هاتف: 2345115 ،2345114 ،2345113 (09)(972)+ فاكس: 2342907 (09)(972)+
Nablus, P. O. Box (7) 707 Tel. 972 9 2345113, 2345114, 2345115 هاتف داخلي (5) 3200
Facsimile 972 92342907 www.najah.edu - email fgs@najah.edu

Appendix C

IRB

An-Najah National University
Faculty of Medicine & Health Sciences
Institutional Review Board

جامعة النجاح الوطنية
كلية الطب وعلوم الصحة
لجنة اخلاقيات البحث العلمي

Ref: Mas. Dec. *2021/9*

IRB Approval Letter

Title of Research:
Bacteriological Profile and Antibiotic Susceptibility Pattern of Septicemia in Neonatal Intensive Care Units, Palestine (2019-2021)

Submitted by:
Bayan Asem

Supervisor:
Mohammad Qadi

Approved:
15th Dec. 2021

Your Study **Title " Bacteriological Profile and Antibiotic Susceptibility Pattern of Septicemia in Neonatal Intensive Care Units, Palestine (2019-2021) "** reviewed by An-Najah National University IRB committee and was approved on 15th Dec. 2021

Hasan Fitian, MD
IRB Committee Chairman

IRB

Nablus - P.O Box :7 or 707 | Tel (970) (09) 2342902/4/7/8/14 | Faximile (970) (09) 2342910| E-mail : IRB@najah.edu

Appendix D

Facilitation of a Researchers' Mission

An-Najah
National University
Faculty of Graduate Studies

جامعة
النجـــاح الوطنية
كلية الدراسات العليا

التاريخ: 2021/12/19

حضرة الدكتورة امل ابو عوض المحترمة
مدير عام التعليم الصحي
وزارة الصحة الفلسطينية

تحية طيبة وبعد ،

الموضوع : تسهيل مهمة الطالبة/ بيان عاصم محمد ابراهيم، رقم تسجيل 12053391،
تخصص ماجستير الأمراض المعدية

الطالبة / بيان عاصم محمد ابراهيم ، رقم تسجيل 12053391، تخصص ماجستير الأمراض المعدية، في كلية الدراسات العليا، بصدد إعداد الاطروحة الخاصة بها بعنوان:

نموذج ونوع البكتيريا المصاحبة لإلتهاب الدم في العناية الحثيثة لحديثي الولادة، فلسطين (2019-2021)
Bacteriological Profile and Antibiotic Susceptibility Pattern of Septicemia in Neonatal Intensive Care Units, Palestine (2019-2021)

يرجى من حضرتكم تسهيل مهمتها في قسم تكنولوجيا المعلومات في وزارتكم الموقرة للحصول على معلومات عن نموذج ونوع البكتيريا المصاحبة لتسمم الدم في العناية الحثيثة لحديثي الولادة، فلسطين للأعوام (2019-2021) لاستكمال مشروع البحث.

شاكرين لكم حسن تعاونكم.

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نموذج ونوع البكتيريا المصاحبة لإنتان الدم في العناية الحثيثة لحديثي الولادة، فلسطين (2019-2021)

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قدمت هذه الرسالة استكمالاً لمتطلبات الحصول على درجة الماجستير في مكافحة وضبط العدوى، من كلية الدراسات العليا في جامعة النجاح الوطنية، نابلس-فلسطين.

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الملخص

المقدمة: الإنتان الوليدي للدم هو مصدر قلق للصحة العامة في جميع أنحاء العالم بسبب ارتفاع معدلات الامراضية والوفيات، بالإضافة إلى وجود المقاومة المتزايدة تجاه المضادات الحيوية التي أوصت بها منظمة الصحة العالمية، حيث كان الهدف من الدراسة هو تقييم حالة مطابقة المضادات الحيوية الموصى بها مع الجراثيم المسببة للإنتان الوليدي في ثلاثة مستشفيات حكومية في فلسطين.

منهج الدراسة: تم اجراء الدراسة بأثر رجعي ولمدة ثلاث سنوات من يناير 2019 إلى ديسمبر 2021، حيث تم استخراج سجلات حديثي الولادة ممن تم الاشتباه بإصابتهم بإنتان الدم في وحدات العناية المركزة لحديثي الولادة في ثلاث مستشفيات حكومية من قاعدة البيانات الإلكترونية لوزارة الصحة، ثم تم إجراء التحليل الاحصائي للمقارنة مع العوامل المصاحبة لإنتان الدم.

النتائج: من بين 6090 من حديثي الولادة المشتبه بإصابتهم بالإنتان الوليدي، أظهر 884 (14.5٪) حالة وجود بكتيريا في زراعة الدم، حيث ان معدل انتشار الانتان الوليدي كان بمعدل 9 لكل 1000 ولادة حية و4 لكل 1000 مولود حي للإنتان المبكر الحصول، حيث كانت البكتيريا المعزولة السائدة هي *Staphylococcus* spp. (56.4%) و *Streptococcus* spp. (13.5%) و *Klebsiella pneumonia* (7.5%)، حيث ان معظم البكتيريا المعزولة (79.6%) لم تتوافق في الحساسية مع المضادات الحيوية المقترحة من منظمة الصحة العالمية. شكلت البكتيريا المقاومة للمضادات الحيوية من غير نوع ال *Staphylococcus* spp. نسبة 21.6% وكانت حساسية المضادات لبكتيريا سلبية جرام مرتفعة للمضادات التالية amikacin (63%), meropenem

(70%), piperacillin-tazobactam (65.5%), colistin (100%) وكانت بالمقابل منخفضة لل ampicillin (7.1%), cefotaxime (21.8%), ceftazidime (29%)، اما بالنسبة للبكتيريا موجبة جرام كانت حساسة بنسبة (99.8%) لل vancomycin وكانت حساسة بنسبة (81%) لل ampicillin بعد استثناء ال *Staphylococcus* spp.

الاستنتاجات: إن معدل انتشار الإنتان الوليدي المثبت مخبريا بالزراعة في فلسطين هو 10 أضعاف المعدل في البلدان ذات الدخل المرتفع، والبكتيريا المسببة للإنتان الوليدي كانت مقاومة للمضادات الحيوية الموصى بها من منظمة الصحة العالمية. وبالتالي، هناك ما يبرر الحاجة إلى مراجعة العلاج الاولي الموصى به من قبل وزارة الصحة للإنتان الوليدي وضرورة تنبي برامج مكافحة عدوى وترشيد لاستهلاك المضادات الحيوية لمحاولة تقليل سرعة انتشار البكتيريا المقاومة للمضادات الحيوية في حديثي الولادة.

الكلمات المفتاحية: الإنتان الوليدي، حساسية المضادات الحيوية، معدل انتشار الانتان الوليدي.