Incidence, bacteriological profile and antibiotic sensitivity pattern of neonatal sepsis in a tertiary health facility in Abuja, North-central Nigeria.

Olutunde Oluyinka
West Suffolk Hospital
Kareem I. Airede
University of Abuja Teaching Hospital
Kudi E. Olateju
University of Abuja Teaching Hospital
Obaro K. Stephen
University of Nebraska Medical Center
Nosakhare Izevbigie
University of Abuja
Kenechukwu K. Iloh
University of Nigeria Teaching Hospital
Obianuju Igbokwe
University of Nigeria Teaching Hospital
Chidiebere Donatus Ignatius Osuorah (chidi.osuorah@gmail.com)
MRC Laboratories The Gambia

Research article

Keywords: neonatal sepsis, mortality, cross-sectional study, antibiotic sensitivity

DOI: https://doi.org/10.21203/rs.2.21673/v2

Abstract

Background: Neonatal sepsis is commonly caused by bacteria in the first 28 days of life. Due to diagnostic limitations in developing settings, prompt laboratory identification of causative organisms is usually a challenge. To prevent mortality, clear knowledge of bacteria and their antibiotic sensitivity patterns are important for prompt empirical treatment.

Methods: This prospective study enrolled 339 newborns with signs and symptoms suggestive of neonatal sepsis out of 645 that were admitted into the special care unit of the University of Teaching Hospital during the study period. Socio-demographic and clinical profiles of the newborns were obtained using a questionnaire and blood culture was done from every enrolled newborn (339 newborns) using BACTEC 9050. The bacteriological profile and antibiotic sensitivity pattern of newborns with confirmed neonatal sepsis were documented.

Results: A total of 339 newborn were admitted for probable sepsis out of a total admission of 645 newborns during the study period based on clinical features and initial laboratory work-up. Forty-six of the 645 newborns (46/645) had culture proven sepsis resulting in a neonatal sepsis incidence rate of 71.3 (95%CI 50.7-91.9) per 1000 admitted newborns. Seventeen of the 46 confirmed sepsis cases were among the 1322 newborns delivered within the study facility during the study period giving an in-hospital neonatal sepsis incidence rate of 12.9 (95% CI 6.7-19.0) per 1000 live births. Amongst the 46 babies with positive blood culture, 27/46 (58.7%) had normal white cell count while the remaining 19/46 (41.3%) had abnormal results. Fifty-two (52) counts of bacteria categorized into 11 bacteria species were isolated from the 46 positive blood cultures. Enterococcus spp and streptococcus species were the commonest gram-positive while Escherichia coli and Pseudomonas luteola were the commonest gram-negative bacteria isolates. Imipenem, amoxicillin/clavulanic acid, Vancomycin, and ofloxacin had the widest coverage of bacteria isolated from newborn with sepsis.

Conclusion: Neonatal sepsis is still prevalent in our environment and compared to previous documented isolates and sensitivity pattern, the bacteria causes, and their antibiotic sensitivity patterns appears to be changing.

Background

Neonatal sepsis is defined as signs of systemic inflammatory syndrome response caused by any toxin or organisms (bacteria, virus or fungi) in the first twenty eight days of life.[1] It is a life threatening emergency and any delay in treatment may result to septic shock and death.[2] According to a 2006 report of the World Health Organization, it is estimated that 1.6 million deaths occur globally every year due to neonatal sepsis and it is responsible for 30 to 40% of all neonatal deaths occurring in developing countries.[3-5] Nigeria ranks number one in annual newborns death in Africa and second highest in the world.[6] The neonatal mortality in Nigeria stands at 33/1000 live births.[7] In developing countries, bacterial pathogens are the most common cause of neonatal sepsis,[8] causing a wide variety of infections including meningitis, pneumonia, urinary tract infection and sepsis.[4,6] These infections can run a rapid course with death occurring in less than 24 hours, if prompt effective empirical treatment is not instituted.[4, 9] The development of effective empirical antibiotic protocols depends on the knowledge of the prevailing bacterial pathogens in that locality. In the neonatal period, it has been documented that the commonest blood-culture isolates in many low income countries were *Staphylococcus aureus*, *Streptococcus pyogenes*, and *Escherichia coli*.[8, 10] Ogunlesi et al in Sagamu and Olateju et al in Gwagwalada, described Klebsiella specie and coagulase-negative *Staphylococci* featuring prominently in Nigeria.[11,12] The spectrum of organisms that cause neonatal sepsis changes over time and varies from region to region, even in the same center from time to time.[4,11] These organisms have also developed increasing multi-drug resistance over the last two decades.[4] Therefore, current knowledge of center specific patterns of bacterial isolates and their antimicrobial susceptibility profiles is useful for prompt management of patients. The present study was undertaken to highlight the local patterns of bacterial isolates in neonates and their antimicrobial sensitivity profiles in a tertiary care hospital in Abuja.

Methods

Study design and area

This prospective study was conducted over an 8-month period, from August to March 2014 at the Special Care Baby Unit (SCBU) of the University of Abuja Teaching Hospital (UATH). The UATH is a 350-bed tertiary hospital that runs primary and secondary in addition to tertiary health care services. The hospital is in Gwagwalada Area Council of the Federal Capital Territory (FCT), Abuja; North Central region of Nigeria.[12] It is the main referral centre for neonatal care in north-central region. UATH has annual deliveries of 2,500 babies and an average annual SCBU admission of 968 babies using the year 2012 records. The SCBU is a 30-bed unit with separate inborn and outborn sections manned by a neonatologist, four senior and junior residents each, six house officers and twenty neonatal nurses. One thousand, three hundred and twenty-two (1322) newborns were delivered in the center during the study period.

Management overview of neonatal sepsis in UATH

On presentation to the SCBU, historical assessment and detailed physical examination are done by the resident on duty. After collection of the relevant samples, the subjects are commenced on empiric antibiotics of amoxicillin/clavulanic acid at 15mg/kg body weight and gentamicin at 2.5mg /kg body weight per dose every 12 hours. Necessary changes are made based on clinical response, blood culture and sensitivity results. Sometimes, third generation cephalosporins mostly cefotaxime and ceftazidime are used for severe infections and in babies who present with relevant clinical features and/or clinical examination suggestive of meningitis. Other supportive measures where necessary are also initiated.

Sample size determination and sampling technique

Cochran's formula[13] was used to determine the sample size needed to estimate the neonatal incidence rate with a confidence interval of 95% and a level of precision estimated to be ± 5% of the assumed prevalence in addition to a non-response rate of 10%. The calculation was based on neonatal sepsis prevalence of 32/1000 live births from a previous study by Olateju et al.[12] The calculated minimum sample size was 331. Consecutive neonates who met the inclusion criteria were enrolled into the study**.** The inclusion criteria included neonates admitted to the SCBU with features suggestive of sepsis (such as fever, jaundice, lethargy, poor suck etc) and those whose parents/guardians gave consent for enrolment of their newborn in the study. Excluded from this study were newborns with major congenital malformations (this was to avoid mortality not directly linked to sepsis) and neonates who have had administration of antibiotics for more than 12 hours, prior to presentation to the hospital.

Blood and data collection method

All the neonates that were enrolled into the study had their blood specimens drawn for culture before treatment was initiated. The skin over the site of blood collection was cleaned thoroughly with 70% alcohol, allowed to dry and cleaned with Povidone iodine for two minutes before blood sample was collected. Two millilitres of blood were drawn by trained assistants from a peripheral vein. One millilitre was for blood culture, and the other for complete blood count (CBC). A structured interviewer administered questionnaire was used to collect relevant information for each newborn by one of the research assistants who was a doctor. The questionnaire was pre-tested at Federal Staff Hospital, Jabi in Abuja by another of the research assistant who was a doctor. It was then analysed for completeness and ease of completing the questionnaire. The neonatal information collected included age, sex, weight on admission, place of delivery, and specific clinical features such as fever, jaundice, hypothermia, hyperthermia, poor skin colour, respiratory distress, feed intolerance, bleeding diathesis, and abdominal distension etc.

Anthropometric measurements and definition of terms

i. Weight was measured using a standardized Bassinet scale, Salter ^{TM} Model 180, with a sensitivity of 0.05kg and calibrated in 0.1kg. Before placing the baby in the weighing scale, the scale was readjusted to zero each time for a new reading. All clothing was removed including the diaper, in order not to affect the weighing scale reading.
ii. Length of each baby enrolled in to the study, was measured using an inelastic tape measure, by placing the baby on a hard surface, lying supine and the lower limbs fully stretched.
iii. Occipito-frontal circumference (OFC) was measured using an inelastic tape measure. The frontal and occipital bony prominences were identified before measurements were taken using an inelastic tape measure.
iv. Gestational age of each baby was determined using modified Ballard score.[14]
v. Each newborns family's socio-economic status was determined using Olusanya's social economic classification.[15]
vi. Neonatal sepsis for the sake of this study was defined as blood stream infection with positive blood culture result in neonates with signs and symptoms suggestive of systemic inflammatory response (SIRS).

Laboratory methods

Blood specimen bottles collected from newborns admitted to the SCBU for probable sepsis were put within one hour of collection into BACTEC 9050 (Becton-Dickinson^{TM} New Jersey, USA) automated system, by a trained dedicated microbiologist to the BACTEC laboratory in UATH. It was then monitored for growth by the microbiologist, as flagged by the machine, every 24 hours, for maximum of five days, when the bottles were expelled by the automated machine.

For gram staining procedure, a single colony was picked and emulsified in a drop of normal saline on a clean glass slide and allowed to dry. The slide was then fixed with heat, stained with crystal violet for one minute, followed by procedure mordant with Lugo's iodine solution for 30 seconds. The slide was then washed with water. The process of decolourisation was carried out with acetone for one to two seconds before washing with water. Counter-staining was with safranin, added for two minutes and later washed with water and allowed to air dry. The slide was then examined microscopically. Gram positive bacteria (GPB) were presumed if the identified pathogen appeared dark purple, while a red colour suggested Gram negative bacteria (GNB).

Antimicrobial susceptibility testing was done using modified Kirby-Bauer disc diffusion method,[16] using multidisc antibiotics (Oxoid Ltd, Hampshire, UK), as described by the Clinical Laboratory Standards. The degree of inhibition of bacterial growth around the antibiotics in the impregnated discs was noted and the result was reported as sensitive or resistant. For determining sensitivity, the following anti-microbial discs were used; Ampicillin 10µg, Ceftazidime 30µg, Augmentin (Amoxicillin 20 µg; Clavulanic acid 10µg), 30µg, Vancomycin 30µg, Cefotaxime 30µg, Ceftriaxone 30µg, Gentamicin10µg, ofloxacin 5 µg and Imepenam10µgm.

A complete blood count (CBC) with white blood cell differential count was done for each subject. The complete blood count was processed in the general laboratory of University of Abuja Teaching Hospital. The CBC sample bottle was placed in the BC- 3200 autolizer machine Mindray.^{TM} The machine extracts the required volume of blood for auto analysis and then displayed the result on the monitor. Other investigation done included serum bilirubin estimation for babies with visible jaundice, while serum electrolytes, urea, creatinine and glucose were also evaluated in babies with perinatal asphyxia and neonatal seizures. For the sake of this study, normal white cell count on CBC was defined as cell count between 5-30,000/mm^3 (or 5-30 x 10^9/L). Values above or below this range were considered abnormal.

Data analysis

The data was entered into an Excel spread sheet and analysed using the Statistical Package for Social Sciences (SPSS) version 20 Microsoft USA. Frequency tables were generated for simple proportions and descriptive analysis.

Results

Characteristics of newborns enrolled for study

Figure 1 shows a summary of the recruitment process. A total of 645 newborns were admitted to the SCBU during the study period. Four hundred and seventy-four (74%) of the total neonates were delivered at the study centre (inborns), whereas the remaining 177 neonates (26%) were referred from other centres (outborns). (Figure 1). Three hundred and thirty-nine (339) of the 645 admitted newborns met the inclusion criteria for the study and were enrolled into the study. Table 1 shows the socio-demographic characteristics and anthropometric indices of newborns enrolled. About one-third, 110 (32%) of the enrolled newborns were ≤ 1 day, 58 (17%) were 2-7 days old while 171(51%) were more than 7 days old. The mean age of enrolled neonates was 4.2 ± 1.6 days. There were more males 196 (58%) compared to females 143 (42%). A majority (81%) of the admitted newborns were discharged alive after full recovery, 15 (4%) left against medical advice following request by parent/caregiver while 49 (15%) of the 339 neonates died during hospital stay. Eight of the 49 recorded mortality had sepsis confirmed by blood culture. Other characteristics of enrolled newborns are shown in Table 1.

Table 1: Characteristics of newborns with probable sepsis admitted during the study period to the Special Care Baby unit of the University of Abuja Teaching Hospital, Nigeria

Newborn variables	Frequency n (%)	Newborn variables	Frequency n (%)
Age at Presentation	*n= 339*	*Gestational age at delivery*	*n= 339*
≤ 1 day	110 (32.4)	Pre-term (< 37 weeks)	109 (32.2)
2-7 days	58 (17.1)	Term (37-40 weeks)	211 (62.2)
> 7 days	171 (50.4)	Post-term (> 40 weeks)	19 (5.6)
Sex	*n= 339*	*Head circumference*	*n= 295*
Male	196 (57.8)	< 33cm	99 (33.6)
Female	143 (42.2)	33-37cm	169 (57.3)
		> 37cm	27 (9.2)
Place of delivery	*n= 339*		
Inborn	168 (49.6)	*Length at birth*	*n= 268*
Outborn	171 (50.4)	< 45 cm	118 (44.0)
		≥ 45 cm	150 (56.0)
Mode of delivery	*n=339*		
Spontaneous Vertex ± assisted	214 (63.1)	*Birth weight*	*n= 339*
Operational (Caesarean section)	125 (36.9)	< 2.5kg	148 (43.7)
		2.5-4.0kg	180(53.1)
Maternal education	*n=339*	> 4.0	10 (2.9)
≤ 6 years	67 (19.8)		
7-12 years	118 (34.8)	*Feeding history*	*n= 339*
> 12 years	114 (33.6)	Breastmilk only	235 (69.3)
		Infant formula ± breastmilk	104 (30.7)
Maternal socio-economic class	*n=339*		
Low	171 (50.4)	*Outcome*	*n= 339*
Middle	58 (17.1)	Survived	275 (81.1)
High	110 (32.4)	Died	49 (14.5)
		Left against medical advice	15 (4.4)

Incidence rate and clinico-laboratory presentation of neonates with sepsis

Forty-six of the 339 enrolled had bacterial isolates on blood culture. Since 645 newborns were admitted during this period, this gave a neonatal sepsis incidence rate of 71.3 (95% CI 50.7-91.9) *per 1000 admitted newborns*. Seventeen of the 46 confirmed sepsis cases were among the 1322 newborns delivered within the study center during the period of study giving an in-hospital neonatal sepsis incidence rate of 12.9 (95% CI 6.7-19.0) *per 1000 live births.* Twenty nine of the 46 confirmed sepsis were in outborns who had been referred to the study center. Twenty-seven of the 46 newborns with confirmed sepsis (58.7%) had white cell count within normal range while the remaining 19 (41.3%) had abnormal white cell count.

Bacterial profile and antibiotic sensitivity pattern in newborns admitted for sepsis

In all, 52 counts of organism categorized into 11 bacteria species were isolated among the 46 newborns with blood culture confirmed sepsis. Some of the cultures grew more than one organisms. Of these, 28 (53.8%) were gram positive bacteria while the remainder, 24 (46.2%) were gram-negative organisms. Table 2 shows the species of bacteria isolates on blood culture. *Enterococcus feacalis* 14/52 (26.9%) and *streptococcus aureus* 9/52 (17.3%) were the commonest organisms isolated from the newborns. Other encountered organisms included *Escherichia coli* 6/52 (11.5%), *Pseudomonas luteola* 5/52 (9.6%), *Klebsiella species* 4/52 (7.7%) *Acinetobacter baumanii* 3/52 (5.8%) and *staphylococcus aureus* 3/52 (5.8%). Less commonly cultured bacteria included *Pseudomonas species* 2/52 (3.8%), *Citrobacter freundii* 2/52 (3.8%) and *Vibro fluvialis* 1/52 (1.9%). The different bacterial isolates stratified by selected newborn parameters are summarized in Table 2. Enterococcus species was the most common Gram positive organism in each group, including females (6/22, 27.3%), than males (8/30, 26.6%) late-onset (7/29, 24.1%) than early-onset (9/23, 39.1%), birth weight <2.5kg (8/23, 34.8%) than ≥2.5kg (8/30, 26.7%), outborns (6/21, 28.6%) than inborn (10/32, 31.3%). *Escherichia coli* as the most common Gram- negative organisms was isolated more frequently in males (4/30, 13.3%) than females (2/22, 9.1%), in early onset-sepsis (4/29, 13.8%) than late-onset sepsis (2/23, 8.7%), in newborns with birth weight <2.5kg (4/23, 17.4%) than those ≥2.5kg (2/30, 6.7%), and in outborns (4/32, 12.5%) than inborns (2/21, 9.5%)..

Table 2 Blood culture bacterial isolates stratified by some selected clinical and demographic parameters of newborns admitted for sepsis to the special care baby unit of the University of Abuja Teaching Hospital Gwagwalada, Abuja, Nigeria

ial isolates	Newborn parameters											
	Gender			Time of onset after birth			Birth weight			Place of birth		
	Male	Female	Total	< 72 hrs.	≥ 72 hrs.	Total	< 2.5 kg	≥ 2.5 kg	Total	Inborn	Outborn	Total
positive sms												
ococcus faecalis	8	6	14	5	9	14	7	7	14	5	9	14
tococcus species	4	5	9	6	3	9	4	5	9	3	6	9
ylococcus aureus	2	1	3	3	0	3	0	3	3	2	1	3
bacter cloacae	1	0	1	1	0	1	1	0	1	1	0	1
obacter akii	1	0	1	1	0	1	0	1	1	0	1	1
negative isms												
richia coli	4	2	6	4	2	6	4	2	6	2	4	6
omonas luteola	4	1	5	3	2	5	2	3	5	2	3	5
iella species	1	3	4	2	2	4	1	3	4	3	1	4
tobacter anii	1	2	3	2	1	3	0	3	3	0	3	3
omonas species	2	0	2	0	2	2	2	0	2	0	2	2
bacter freundii	0	2	2	1	1	2	1	1	2	1	1	2
fluvialis	1	0	1	0	1	1	1	0	1	0	1	1
xella catarrhalis	1	0	1	1	0	1	0	1	1	1	0	1
	30	**22**	**52**	**29**	**23**	**52**	**23**	**30**	**52**	**21**	**32**	**52**

Antibiotics sensitivity pattern of the isolated organisms are shown in Table 3. 90% of the *Streptococcus spp* tested were sensitivity to cefuroxime, 80% to ceftriaxone and 80% to amoxicillin/clavulanic acid. 60% were sensitive to each of clindamycin and imipenem; 40% to ofloxacin, 30% to

erythromycin, 20% to each of ceftazidime and chloramphenicol; and only 10% were sensitive to each of ampicillin and vancomycin.

Table 3: Bacteria isolates and patterns of antibiotic responses among newborns presenting with sepsis to the special care baby unit of the University of Abuja Teaching Hospital Gwagwalada, Abuja, Nigeria

a in	BACTERIAL ANTIBIOGRAM (%)																		
	AM	AC	CH	CL	CL_2	CO	CE	CE_2	CE_3	CE_4	CI	ER	GE	IM	LE	ME	OF	TE	VA
coccus	10.0	80.0	20.0	0.0	60.0	0.0	80.0	90.0	20.0	0.0	0.0	30.0	0.0	60.0	0.0	0.0	40.0	0.0	10.0
ococcus	0.0	40.0	60.0	20.0	60.0	0.0	0.0	0.0	0.0	20.0	0.0	40.0	20.0	60.0	0.0	0.0	20.0	20.0	20.0
chia	14.0	50.0	65.0	0.0	14.0	7.0	21.5	21.5	14.0	0.0	7.0	14.0	21.5	50.0	7.0	7.0	14.0	0.0	14.0
monas	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	100	0.0	100	0.0	0.0	100	0.0	0.0	100	0.0	0.0
occus	5.0	15.5	18.5	0.0	5.0	0.0	2.5	2.5	0.0	0.0	2.5	5.0	5.0	15.5	5.0	5.0	2.5	0.0	15.5
lla spp	0.0	25.0	50.0	0.0	0.0	0.0	75.0	75.0	25.0	0.0	0.0	0.0	25.0	100	0.0	0.0	50.0	0.0	0.0
lla alis	100	100	100	0.0	0.0	0.0	100	0.0	100	0.0	100	0.0	100	100	0.0	0.0	100	100	0.0
luvialis	0.0	0.0	100	0.0	0.0	100	0.0	100	0.0	0.0	0.0	0.0	100	0.0	0.0	0.0	0.0	100	0.0
bacter nii	0.0	33.3	0.0	0.0	0.0	0.0	0.0	0.0	66.7	0.0	100	0.0	66.7	66.7	33.3	33.3	33.3	0.0	0.0
cter i	0.0	0.0	100	0.0	0.0	0.0	100	100	100	0.0	0.0	0.0	0.0	100	0.0	0.0	0.0	0.0	0.0
monass	0.0	0.0	0.0	0.0	0.0	40.0	0.0	0.0	40.0	0.0	80.0	0.0	40.0	100	0.0	0.0	100	20.0	0.0

AM-ampicillin; **AC**-amoxicillin/clavulanic acid; **CH**- chloramphenicol; **CL**-cloxacillin; **CL_2**-clindamycin; **CO**-clotrimazole; **CE**-ceftriaxone; **CE_2**- cefuroxime; **CE_3**- ceftazidime; **CE_4**-cefixine; **CI**- ciprofloxacin; **ER**- erythromycin; **GE**- gentamicin; **IM**- imipenem; **LE**- levofloxacin; **ME**- meropenem; **OF**- ofloxacin; **TE**- tetracycline; **VA**- vancomycin (© DIC Osuorah)

Sixty percent (60%) of the isolated staphylococcus species were sensitive to each of clindamycin and imipenem, 20% were each sensitive to cloxacillin, cefixime, gentimicin, ofloxacin and tetracycline. For *Escherichia spp,* 65% of the species were sensitive to chloramphenicol while 50% were eac sensitive to amoxicillin/clavulanic acid and imipenem. . One hundred percent (100%) of the isolated *Pseudomonas spp* were sensitivity to ceftazidime, ciprofloxacin , ofloxacin and imipenem , similarly 100% *Moraxella catarrhalis* were sensitive to ampicillin, amoxicillin/clavulanic acid, chloramphenicol, cefuroxime, ceftazidime, ciprofloxacin, gentamicin, imipenem, ofloxacin and tetracycline. Finally, 100% of isolated *Klebsiella spp* were sensitive to imipenem, 75% was sensitive to each of ceftriaxone and cefuroxime, 50% sensitivity to chloramphenicol and ofloxacin and a 25% sensitivity to amoxicillin/clavulanic acid, gentamicin and ceftazidime. Sensitivity pattern of other less common organisms isolated on blood culture are shown in Table 3.

Table 4 shows in summary the antibiotics coverage pattern of bacteria isolates based on their gram stain property. Ninety two percent and 100% of all the gram positive and gram negative organisms respectively were sensitive to imipenem. None of the gram positive organism was sensitive to ampicillin, while 33% of the gram positive organisms were sensitive to ampicillin.

Table 4: Antimicrobial sensitivity pattern of blood culture isolates against commonly available antibiotics based on gram-staining properties of bacteria

S/N	Antibiotics	Sensitivity		
		Gram positive	Gram negative	Average Sensitivity
1.	Imipenem	92	100	96.0
2.	Ofloxacin	64	46	55.0
3.	Augmentin	62	50	56.0
4.	Vancomycin	60	75	67.5
5.	Ceftriaxone	40	60	50.0
6.	Cefotaxime	40	60	50.0
7.	Ceftazidime	7	61	34.0
8.	Cefuroxime	40	53	46.5
9	Gentamicin	20	42	31.0
10	Ampicillin	33	0	16.5

Discussion

In this study, bacteria were isolated in 46 of the 339 (13.6%) newborns enrolled with presumptive diagnosis of neonatal sepsis. This is lower than what was reported in studies within and outside the country. In Ilorin, a bacteria isolation rate of 30%,[17] was reported and 34% in India.[18] The difference in rates of positive samples can be explained by the differences in criteria for determining neonatal sepsis, blood culture system, location and health situation in the region, capacity of hospital, variability between microbiological techniques.

Our study found that more than half (53.9%) of the bacteria isolates were Gram positive organisms, while Gram negative organisms made up 46.1%. This pattern was found in Qatar[19] where a predominance of Gram-positive bacteria was noted (66%) and 16.2% for Gram negative bacteria, in Karachi (54.1%)[20] for Gram positive bacteria and 45.9% for GNB. In Gwagwalada, a report of 58% for GPB and 42% for GNB was reported.[17] Some other studies also revealed preponderance of GPB over GNB.[21,22] Unlike our study, another study in Pakistan[23] showed GNB of 54.6% and GPB of 45.4%. However, Iregbu et al[24] in Abuja, found equal occurrence of both Gram positive and negative organisms.

Enterococcus faecalis was the most common gram-positive organism isolated in this current study, while *Escherichia coli* were the commonest gram-negative organisms. This is a departure from findings from a similar study that was carried out in the unit 10 years earlier, where Staphylococcus *aureus* was found to be the commonest organism and *Klebsiella* was the leading Gram negative organism.[17] This further corroborates the fact that, the organisms responsible for neonatal sepsis vary from time to time, even in the same unit, and possible changing pattern of neonatal sepsis. .

Staphylococcus aureus was the third commonest Gram-positive bacteria after *Enterococcus faecalis* and *Streptococcus* specie in the current study. Iregbu et al[24] in Abuja, found *Enterococcus faecalis* as a major cause of neonatal sepsis second only to Staphylococcus aureus among the Gram-positive bacteria. It was also found to be the commonest Gram-positive bacteria causing neonatal sepsis in Accra,[25] accounting for 14.4% of the organisms isolated. This study corroborates o the emergence of *Enterococcus* faecalisas an important cause of neonatal sepsis. Similarly, *Escherichia coli* were the commonest GNB identified as causes of neonatal sepsis in studies within Nigeria and outside.[26-31] It was also a leading cause of Gram-negative neonatal sepsis in some other studies, like the ones from Portharcourt[27] and Ife.[21]

Klebsiella species and *Staphylococcus aureus* were the third commonest Gram-negative bacteria and Gram-positive bacteria respectively in this study. This was a sharp departure from many studies both locally and internationally, where these organisms were identified as the leading causes of neonatal sepsis.[12,20,27,29,32-34] The reason for this may not be immediately deduced, but it could be due to the method of isolation, as there is no reported study in the Northern part of the country where BACTEC machine was used for bacteria isolation yet. This could also be an emerging pattern of neonatal sepsis in the country, particularly in the North central region. *Citrobacter spp* accounted for 3.8% of neonatal sepsis in our study, which was comparable to the 3.1% reported in a study done by Ramesh et al.[34] This organism was however not reported in other similar studies.[12, 17]

Ella et al in Kaduna,[35] reported similar antimicrobial sensitivity pattern of gram-positive organisms as reported in our study. Studies in India,[22, 36] reported 80% of isolated GPB were sensitive to vancomycin, whereas Bode Thomas et al[29] in Jos reported 67% of GPB were sensitive to gentamicin. Iruegbu et al[24] showed an 89% of GPB were sensitive to amoxicillin/clavulanic acid. . Studies in Calabar, India and Uganda,[28,36-37] observed a high sensitivity to gentamicin. Studies in South West Nigeria,[21,39] reported a high sensitivity to ofloxacin which is comparable with the finding of this study. The varied sensitivity pattern of Gram-positive organism across the country supports the theory that organisms

associated with neonatal sepsis and their sensitivity varies from place to place and region to region.[2,8] Use of empiric antibiotics, could be another reason why the sensitivity of these organisms to the antibiotics differs.

In this study, 100% of the Gram-negative bacteria (GNB) were sensitive to imipenem similar to findings in Abuja[24] and Baghdad,[26] where 100% sensitivity of Gram-negative bacteria to imipenem and meropenem was reported. We found that 53% to 60% of the GNB were sensitive to cephalosporins tested. In contrast, sensitivity to GNB was reported as 86.7% for cefotaxime and 81.3% for ceftazidime in Sagamu.[34] Mugalu et al in Uganda[37] reported a 94.1% sensitivity of *Escherichia coli* to ceftriaxone while Iregbu[24] in Abuja observed 94% sensitivity to cefotaxime. Though much higher than what was found in this study, it shows that cephalosporins are highly active against Gram negative bacteria. Therefore, caution should be applied in the use of these cephalosporins as empirical antibiotics, so resistance is not developed to this important class of antibiotics as was reported for gentamicin in the current study.

Limitation

One major limitation we had during this study was inability of some babies who presented over the weekend to be immediately screened for sepsis because the BACTEC laboratory was closed on weekends. This may have affected culture of organisms since their blood after collection had to be stored over the weekends.

Conclusion

Neonatal sepsis is prevalent in our setting. *Enterococcus* and *streptococcus species* were the commonest gram-positive organisms while *Escherichia coli* and *Pseudomonas luteola* were the commonest gram-negative organism. Varying antibiotics susceptibility pattern was noted in the current study compared to earlier studies done in the region and other places, there is the need for review of the antibiogram in the different units to determine the choice of empiric antibiotics.

List Of Abbreviations

EOS	Early onset sepsis
ESR	Erythrocyte sedimentation rate
GBS	*Group B Streptococcus*
GNB	Gram negative bacteria
GPB	Gram positive bacteria
LOS	Late onset sepsis
OFC	Occipito-frontal circumference
SCBU	Special care baby unit
UATH	University of Abuja Teaching Hospital
WBC	White blood cells

Declarations

Ethics approval and consent to participate

Ethical clearance was obtained from the Medical Ethics Committee of the University of Abuja Teaching Hospital, Gwagwalada for review and approval was obtained before the commencement of the research (reference number FCT/UATH/HREC/PR/304). Written informed consent was obtained from a parent and/or guardian for all participants. Participants were informed that voluntary withdrawal at any stage of the study was guaranteed without any adverse effect to their newborn. All information was handled with strict confidentiality.

Consent for publication

Not applicable

Availability of data and material

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

Competing interests

The authors declare that they have no competing interests

Funding

Funding for this study was from equal contributions from all authors. No External funding was received for this study.

Authors' contributions

OO conceptualized the study and together with KIA and KEO developed the study methodology. ODIC analysed the study data. Result and discussion were written by ODIC KKI and OI. Supervision of data collection and laboratory sample analysis was done by OK and NI. KIA and KEO supervised the entire work. All authors read and approved the final manuscript.

Acknowledgements

We are also grateful to the highly dedicated staff nurses SCBU for their tireless efforts in some study related documentations and heroic efforts in salvaging sick babies.

References

1. Khalid N. H. Definitions of bloodstream infection in the newborn. *Pediatr Crit Care Med* 2005; **6:** S45–9.
2. Premalatha D.E, Mallikarjun K, Halesh L.H, Siddesh K.C, Prakash N. The bacterial profile and antibiogram of neonatal septicaemia in a tertiary care hospital. International Journal of recent trends in Science and technology. 2014;10: 451–55.
3. Leah Y, Rajam R. Assessing and applying evidence-based interventions at the community level in India as a model policy to reduce neonatal mortality rates in Nigeria. Available at www. jglobalhealth.org | -, last accessed on 2nd July 2015.
4. Bambala P Z, Vishnu B, Belgode N H, Thirunavukkarasu A B, Noyal M J. Neonatal sepsis in a tertiary care hospital in South India: bacteriological profile and antibiotic sensitivity pattern. Indian J Pediatr DOI 10.1007/s12098-010-0314-8.
5. Zaidi A K M, Ganatra H A, Syed S, Cousens S, Lee A C C, Black R et al. Effect of case management on neonatal mortality due to sepsis and pneumonia. BMC Pub Hlth 2011; 11: 1–15.
6. Federal Ministry of Health. Saving newborn lives in Nigeria: Newborn health in the context of the Integrated Maternal, Newborn and Child Health Strategy. 2nd edition. Abuja: Federal Ministry of Health, Save the children, Jhpiego; 2011.
7. Child Mortality Estimates. Country-specific neonatal mortality rate Estimates generated by the UN Inter-agency Group for Child Mortality Estimation (UN IGME) in 2019. Downloaded from http://data.unicef.org
8. Osrin D, Vergnano S, Costello A. Serious bacterial infections in newborn infants in developing countries. Curr Opin Infect Dis 2004; 17:217–24.
9. Masood H R, Sadia K, Tooba , Sabah N, Sadaf S. Sepsis in infants: analysis of bacterial pathogens and their antibiotic susceptibility, a study at Government Tertiary Care Hospital, Karachi. J Dow Univ Hlth Sc Karachi 2013; 7: 35-40.
10. Vergnano S, Sharland M, Kazembe P et al. Neonatal sepsis: an international perspective. Arch Dis Child Fetal Neonatal 2005; 90: 220–224.
11. Ogunlesi AT, Olusoga BO. Predictors of mortality in neonatal septicemia in an under-resourced setting. J Natl Med Assoc 2010; 102: 915-21.
12. Olateju EK, Okechukwu AA, Mokuolu OA. Neonatal septicaemia at the University of Abuja Teaching Hospital, Gwagwalada, Nigeria. New Nig J Clin Res 2011; 1: 95-103
13. Lwanga SK, Lemeshow S. Sample size determination in health studies. WHO 1991: 1-30
14. New Ballard score. Available at ballardscore.com/files/Ballardscore_scoresheet.pdf, last accessed on 26th May 2015.
15. Olusanya O. Okpere E, Ezimokhai M. The importance of Social class in voluntary fertility control in a developing country. W Afr J Med 1985; 4: 205-212.
16. Clinical and Laboratory Standard Institute; Performance Standards for Antimicrobial Susceptibility Testing; Twenty second Informational Supplement. 2012 (M100 S22) 32:1- 139.
17. Mokuolu AO, Jiya N, Adesiyun OO. Neonatal septicaemia in Ilorin: bacterial pathogens and antibiotic sensitivity pattern. *Afr J Med Sci* 2002; 31: 127-30.
18. Chacko B and Sohi I. Early onset neonatal sepsis. Indian journal of paediatrics; 2005; 72: 23-26.
19. Afif A, Samawal L, Abdul Rouf PV, Moza A, Sajjad R, Wessam El Kassim, *et al*. Incidence of bacterial isolates from blood culture in the neonatal intensive care unit of Tertiary Care Hospital. *Int J Drug Dev and Res* 2012; 4: 359-67.
20. Khetam HRA, Deia KK and Lamia A. The bacterial profile and C - reactive protein of suspected septic neonates admitted to the Al-Kadyemia teaching hospital. *Internat J Recent Scientific Res* 2013; 11: 1723-27.

21. Adejuyigbe E.A, Adeodu O.O, Ako-nai K. A, Taiwo O, Owa J.A. Septicaemia in high risk neonates at a teaching hospital in Ile-Ife, Nigeria. *East Afr Med J* 2001; 78: 540-43.
22. Tallur SS, Kasturi AV, Shobha D. Nadgir, Krishna BVS. Clinico-bacteriological study of neonatal septicemia in Hubli. *Indian J Pediatr* 2000; 67: 169 -74.
23. Muhammad Z, Ahmed A, Hayat U, Wazir MS, Rafiyatulla, Waqas H. Neonatal sepsis: causative bacteria and their resistance to antibiotics. JAMC; 2010; 22(4): 33-36
24. Iregbu KC, Elegba OY, Babaniyi IB. Bacteriological profile of neonatal septicaemia in a tertiary hospital in Nigeria. *Afr Hlth Sc* 2006; 6: 151-4.
25. Anyebuno M, Newman M. Common causes of neonatal bacteraemia in Accra, Ghana. *East Afr Med J* 1995; 72: 805-8.
26. Tosson AM, Speer CP. Microbial pathogens causative of neonatal sepsis in Arabic countries. *J Matern Fetal Neonatal Med* 2011; 24: 990-4.
27. Awoala WB and Tabansi PN. Clinico-bacteriological profile of early and late onset sepsis in a tertiary hospital in Nigeria. *J Medicine Med Sc* 2012; 3: 107-11.
28. Neonatal sepsis: Self-learning packet 2004. Available at www.orlandohealth.com/pdf folder/neonatal sepsis.pdf, last accessed on 25th May 2015.
29. Bode-Thomas F, Ikeh EI, Pam SD, Ejeliogu EU. Current aetiology of neonatal sepsis in Jos University Teaching Hospital. *Niger J Medicine: J National Assoc Resident Doctors Nigeria* 2004; 13:130-135
30. Antia-Obong OE, Utsalo SJ, Udo JJ, Udo KT. Neonatal septicaemia in Calabar, Nigeria. *Central Afr J Med* 1992; 38: 161-5.
31. Ghiorghis B. Neonatal sepsis in Addis Ababa, Ethiopia: a review of 151 bacteremic neonates. *Ethiop Med Journal* 1997; 35: 169-76.
32. Ojewumi TK, Ojewumi JS. Trends in infant and child mortality in Nigeria: a wakeup call assessment for intervention towards achieving the 2015 MDGs. *Sc J Sociol Anthropol* 2012;
33. Ogunlesi TA, Ogunfowora OB, Osinupebi O, Olanrewaju DM. Changing trends in newborn sepsis in Sagamu, Nigeria: bacterial aetiology, risk factors and antibiotic susceptibility. *J Paediatr Child Hlth* 2011; 47: 5-11.
34. Ramesh BY, Leslie ESL, Vandana KE. Bacterial isolates of early-onset neonatal sepsis and their antibiotic susceptibility pattern between 1998 and 2004: an audit from a center in India. *Ital J Pediatr* 2011; 37: 32
35. Ella EE, Ahmad AA, Ogala WN, Umoh VJ Aliyu-Zubair R. Bacteriology and sensitivity profile bacterial agents responsible for neonatal septicaemia in a Tertiary Hospital of Kaduna Metropolis. *J pure applied microbiol* 2008; 2: 103-8.
36. Ghanshyam DK, Ramachandran VG, Gupta P. Bacteriological analysis of blood culture isolates from neonates in a Tertiary Care Hospital in India. *J Health Popul Nutr* 2002; 20: 343-47.
37. Mugalu J, Nakakeeto MK, Kiguli S, Kaddu – Mulindwa DH. Aetiology, risk factors and immediate outcome of bacteriologically confirmed neonatal septicaemia in Mulago Hospital, Uganda. *Afr Hlth Sc* 2006; 6: 120-6.
38. Awoniyi O, Udo SJ, Oguntibeju OO. An epidemiological survey of neonatal sepsis in a hospital in western Nigeria. *Afr J Microbiol Res* 2009; 3: 385-9.

Figures

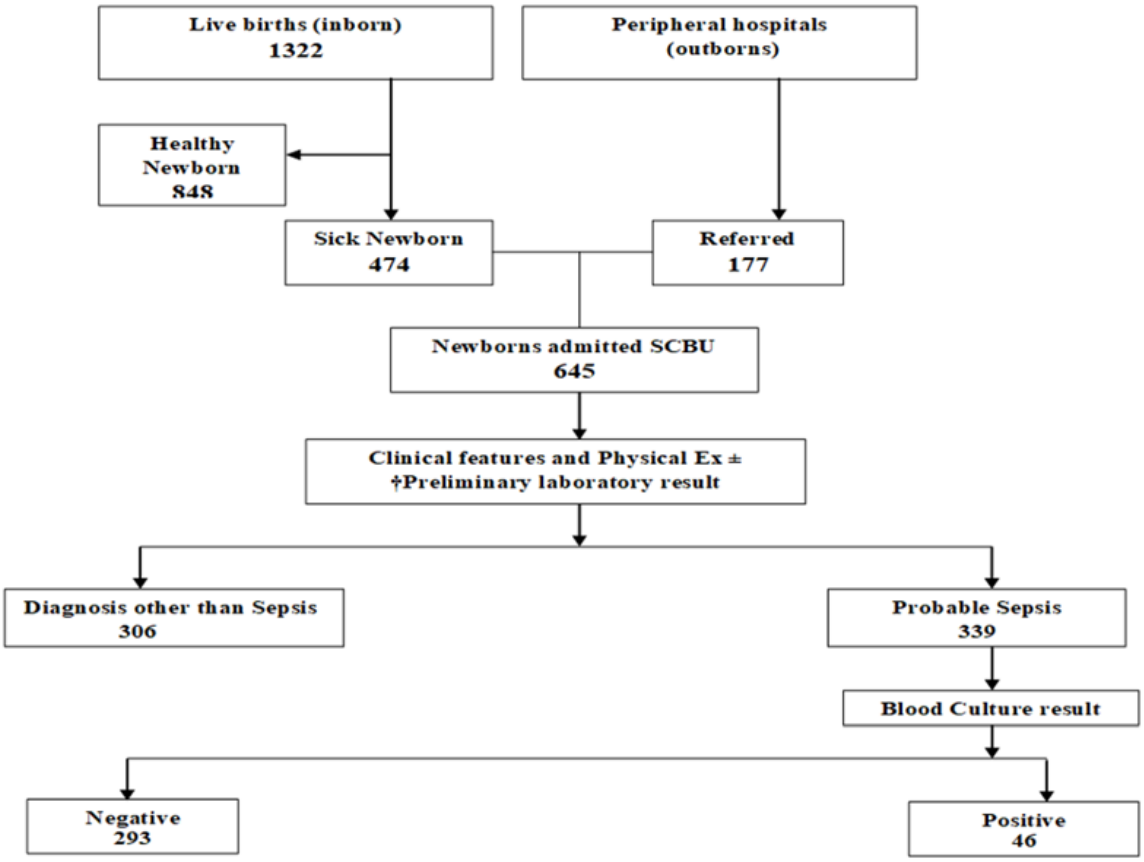

Figure 1

Overview of recruitment processes of surveyed newborns in the SCBU between September 2013 to April 2014 (†Complete blood count, urinalysis and gram stain)

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- supplementaryfile.docx