

International Journal of Research Publication and Reviews

Journal homepage: www.ijrpr.com ISSN 2582-7421

Incidence of Methicillin Resistant *Staphylococcus Aureus* in Mother-Neonates Pair in the Delivery Units of Three Secondary Healthcare Facilities in Zaria, Kaduna State, Nigeria

Ibiam Alunma Nkemdirim¹, Ibrahim Yakubu Kokori E.², Ehinmidu Joseph Olorunfemi ³, Shitta Nasisi, Namuma⁴, Achilus Francis⁵, Dr. Okoh Jocelyn Cletus⁶, Onoja Queen Mary⁷

1, 2 & 3 Department of Pharmaceutical Microbiology, Ahmadu Bello University, Nigeria

⁴ Department of Biology (Innovative Drug Design), Sechenov University Moscow, Russia

⁵ Department of Science Laboratory Technology, Taraba State Polytechnic, Jalingo, Nigeria

⁶ Clinical Medicine and Public Health, Sechenov University Moscow, Russia

⁷ Department of Medical Laboratory Science, Madonna University, Nigeria

Email address: alunmaibiam@gmail.com

Doi: https://doi.org/10.55248/gengpi.5.0924.2501

ABSTRACT

Neonatal mortality accounts for 47 % of deaths in children of 5 years and below across the World which is often as a result of infections with Methicillin Resistant *Staphylococcus aureus* (MRSA) being one of the most implicated. This study was aimed at determining the epidemiological status of MRSA among mother-neonates pair in the delivery units of three Secondary healthcare facilities in Zaria, Kaduna State, Nigeria between May to October 2023. Swab samples were collected from 83 mother – neonate pairs in three Secondary selected hospitals. Morphology Mannitol Salt Agar (MSA), Gram's reaction and biochemical tests were carried out to preliminarily identify characteristic yellow colonies on MSA. Confirmation of isolates as *S. aureus* was done using Liofilchem Integral System Stafilococchi®. Cefoxitin resistance test was done to confirm isolates as MRSA using Kirby-Bauer disc diffusion method. Antibiotics susceptibility pattern of MRSA isolates was determined by Kirby-Bauer disc diffusion method using antibiotics. Thirteen (13) isolates were confirmed to be *S. aureus* of which all were phenotypically MRSA. Prevalence of MRSA was 5.22 % of which 2.41 % were vaginal carriers in pregnant women in labour while 9.64 % originated from the nares of the neonates. Vertical transmission through vaginal delivery from mother to neonate was 50 %. There was 100 % resistance to cefoxitin and amoxicillin-clavulanic acid. Resistance of isolates to other antibiotics were slightly lower. Low resistance was recorded to gentamicin (23.08 %) and rifampicin (30.77 %). All isolates had Multiple Antibiotics Resistance Index greater than 0.2.

Keywords: Staphyloccoccus aureus, MRSA, Antibiotic resistance, Neonates.

INTRODUCTION

Nosocomial infections also called hospital-acquired infections (HAIs) refer to infections that are not present in patients at the time of admission into healthcare facilities and usually obtained in medical institutions two days or more after admission to the hospital, or within 30 days of discharge [5]. Nosocomial infection has been reported in all age group that visit hospital but with different level of occurrence and mortality.

Despite recent breakthroughs in neonatal care, sepsis remains a primary cause of infant morbidity and mortality worldwide [6]. Preterm neonates are at heightened risk of colonisation with *S. aureus* which often increases the risk of subsequent infection [7], [11].

Neonatal mortality rates (NMR) in Nigeria is estimated to be about 39 per 1000 live births [20], with infections being the major cause. Mortality is higher in extreme preterm and very low birth weight neonates [12]. South Asia and sub-Saharan Africa records about one fifth of the World's annual 2.7 million neonatal deaths as a result of infections and subsequent sepsis [19]. It amounted to nearly 47 % of all deaths in children under the age of 5 years globally with sub-Saharan African Children facing the greatest risk of dying as a neonate [31].

Methicillin-resistant Staphylococcus aureus (MRSA) has been reported as the most common cause of both early and late infections [6].

Methicillin-resistant *Staphylococcus aureus* has been linked to skin and soft tissue infections (SSTI) as well as respiratory tract infections in neonates [15].

The target of sustainable development goal number 3 which is the reduction of neonatal mortality to under 12 deaths per 1000 live births [29] have not yet been achieved.

About 25 % of healthy people are asymptotic carriers of one or more strains of *S. aureus* (Olorode & Nanighe, 2021). Epidemiological data of *S. aureus* shows that epidemical Methicillin-resistant *S. aureus* (MRSA) strains of certain phage types have increased in virulence and have rapidly spread in hospitals [24].

The difficulty in treatment of MRSA infection after its development places great importance on the prevention of MRSA outbreaks [21]. Early detection and prevention of infection is of importance in pre term, low birth weight and poor Appearance Pulse Grimace Activity and Response (APGAR) scoring babies [4] if MRSA outbreak in neonatal intensive care units would be curtailed. Most focus of several studies have been on MRSA infection in neonatal intensive care unit Worldwide with isolates taken from already septicaemic neonates. Works have also been conducted to demonstrate MRSA colonization in these NICU and on the handlers. However, little or no work has been done to study the colonization of MRSA in neonates that have not come down with septicaemia as well as to relate the susceptibility pattern of such isolates from the neonates with that of the mothers and their handlers in other to establish a relationship that will show possible origin of neonatal MRSA infection.

Studies focused on interventions plans for MRSA are limited, especially in developing countries [25]. Therefore, this prevention targeted study will be carried out to explore MRSA transmission and colonization trend in delivery units of secondary hospitals in Zaria. The aim of this study was to determine the incidence of Methicillin Resistant *Staphylococcus aureus* among Neonates in the delivery units of some Secondary healthcare facilities in Zaria.

MATERIALS AND METHOD

Equipment used included the following : Incubator (NAPCO 630, National Appliances Co. USA), Electronic weighing balance (AdventurerPro AR224CN, OHAUS Europe GmbH, Nänikon Switzerland), Hot-Air-Oven (DHG-9101-1SA, SearchTech Instruments), Autoclave (YX-280A), Microscope (0603405, Fisher Scientific Company, China), Micropipette (Merck KGaA, Darmstadt, Germany), Refrigerator, Micro-centrifuge (FC5515R, OHAUS CORPORATION USA, Germany), Vortexer (Scientific Industries Inc., Bohemia, USA), Thermocycler (Eppendorf AG 22331 Hamburg, Germany), Gel Electrophoresis Tank (NANOPAC-300P, Cleaver Scientific Ltd., Rugby, UK), Gel Documentation Imaging (Vilber, France), Microwave Oven (MX310TCSL, Hinari Lifestyle, UK). Bacteriological media used were: Muller Hinton Agar and Mannitol Salt Agar (Titan Biotech Ltd, India), while Nutrient Agar was from HiMedia Laboratories Pvt Ltd, India and Nutrient Broth product of Merck KGaA, Darmstadt, Germany. Various chemicals and reagents were utilized in this study. They are Crystal violet (May and Baker Ltd. Dagenham, England), Lugol's iodine (May and Baker Ltd. Dagenham, England), Acetone, Hydrogen peroxide (SKG Pharma Ltd. Ikeja, Nigeria), Sterile deionized water, Methyl-red, Carbol Fuschin, oil immersion, Serum Plasma, Physiological Saline, Resistant gene Primers, Liofilchem Integral System Stafilococchi® kit (LiofilChem Bacteriology Products, Italy), DNA extraction kit (Zymo ResearchTM, Germany), DNA Ladder (Solarbio Life Sciences, Beijing, China).

Primers		Sequence (5' – 3')	Amplicon Size	References
mecA	Forward	TCCAGATTACAACTTCACCAGG	162	[8] [28]
	Reverse	CCACTTCATATCTTGTAACG	-	
mecC	Forward	GAAAAAAAGGCTTAGAACGCCTC	138	[8] [28]
	Reverse	GAAGATCTTTTCCGTTTTCAGC	-	
blaTem	Forward	GAGACAATAACCCTGGTAAAT	459	[27]
	Reverse	AGAAGTAAGTTGGCAGCAGTG		

Table 3.1: List of Primers

Key: *mecA* and *mecC* (Methicillin resistance factor gene)

The hospitals included in this study were; Hajiya Gambo Sawaba General Hospital, Kofan Gaya, St. Luke's Anglican Hospital, Wusasa, and Ahmadu Bello University Medical Centre, ABU Main Campus, Samaru, Zaria, all located in Zaria and Sabon Gari LGAs of Kaduna State, Nigeria.

Ethical approval was sought and obtained for the recruitment of patients sampled in this study. Informed consent was also sought from patients before being included in the study.

The study population was made up women who were admitted to the delivery unit in first stage of labour. Their age was from 19 - 40 years old. They comprised of woman with various levels of education and career. The neonates born to these women were also sampled. The neonates were within 0 - 2 days of life.

High vaginal swabs were collected from mothers by the nursing staff on duty while nasal swabs were collected from the right and left nostrils of neonates by the researcher following laid down procedures and the supervision of the superintending nurse. Samples were collected using sterile swab sticks and immediately inoculated wholly into sterile nutrient broth.

A loop full of overnight culture of swabs in nutrient broth was inoculated on Mannitol Salt Agar (MSA) and incubated for 24 hours at 37°C. Yellow colonies with surrounding yellow zone were the distinct feature for sub-culturing on a nutrient agar to obtain pure colonies of *S. aureus*. In some cases, MSA culture was further incubated for another 24 hours to given room for slow growing inoculums before selecting positive plates or discarding negative ones. Preliminary identification was based on the morphological characteristics upon Gram staining such as the arrangement and shape of cells, biochemical tests (ability to ferment Mannitol in MSA cultures, Catalase and Coagulase production). Further confirmation as *S. aureus* was done using rapid identification kit for *S. aureus*.

Gram Staining: Procedure for the Gram Staining was as described by Cheesebrough (2006). A drop of distilled water was placed at the centre of a clean glass slide. Smears of presumptive Staphylococci colony was placed in the drop of distilled water and suspension made using a sterile wire-loop. It was allowed to air dry and then heat fixed by passing it over a flame. A primary stain (Crystal violet) was added to the smear and allowed for 30 seconds, and then washed with distilled water. A mordant (Lugol's iodine) was added for 1 minute and the stain washed off using clean water. Acetone was then applied and washed off immediately. A counter stain (carbol fuschin) was applied and allowed for 2 min and then rinsed with clean water. The stained slides were air dried and then observed under the microscope using ×100 objective lens with oil immersion. Purple cluster of cells was taken as positive Gram reaction.

Catalase Test: This was carried out to detect the presence of Catalase enzyme in the isolate. Procedure was as described by Cheesebrough (2006). On a clean slide, a drop of 3% hydrogen peroxide (H₂O₂) was placed onto which pure colonies from a 24-hour culture of Gram positive cocci isolate will be mixed using a sterile applicator stick. Formation of gas bubbles was taken as positive test.

Coagulase test: This was the distinguishing test for *S. aureus* from *S. epidermidis* and *S. saprophyticus* using their ability to produce coagulase. The slide agglutination test was employed. A drop or two distilled water was placed on a clean glass slide to which a colony of 24-hour catalase positive cocci cluster isolate was added and mixed using a sterile wire loop. Two drops of rabbit serum were added and rocked for 30 seconds. Test reaction was taken as positive if agglutination appeared on them, while negative isolates showed no agglutination in the test reaction.

Specific identification of *S. aureus* was done using Liofilchem Integral System Stafilococchi® rapid test kit. Catalase and Coagulase positive isolates was used for this test. Colonies were emulsified from an 18 - 24 hours culture in 5ml of sterile physiological saline water and mixed thoroughly and adjusted to 0.5 Mc Farland Standard. The lids on the top of the micro well test systems were removed and micropipette was used to transfer 0.2 ml of the bacterial suspension to the first 8 well of the system. Two (2) drops of Vaseline for microbiological use was used to overlap wells 1-ADC and 2-UR. The lids were replaced and the system incubated aerobically at 37 °C for 18 - 24 hours. The micro wells were read after 18-24 hours of incubation and interpreted using the tables provided in the manufacturer's manual.

The method for detection of isolates as MRSA was the disk diffusion method using Muller Hinton Agar as recorded in EUCAST guideline. *S. aureus* isolates was streaked on the agar, allowed for 15-30 minutes, cefoxitin (30 µg disk) was placed aseptically on the inoculated plates and then incubated at 37 °C for 18 hours. Within 16-18 hours, zones of inhibition were measured. Clear zones of diameter <22 was interpreted as Methicillin resistant.

The antibiotic susceptibility pattern of MRSA isolates was determined using the modified Kirby-Bauer disc diffusion method (EUCAST, 2023). Selected antibiotics used according to the EUCAST (2023) were: Cefoxitin (FOX 30 µg), Gentamicin (GEN 10 µg), Azithromycin (AZM 15 µg), Erythromycin (ERY 15 µg), Ciprofloxacin (CIP 5 µg), Amoxicillin/Clavulanic acid (AMC 30 µg), Rifampicin (RD 5 µg), Trimethoprim-sulfamethoxazole (1.25 µg-23.75 µg), and Tetracycline (TET, 30 µg).

Discrete colonies of *S. aureus* isolates on Mannitol salt agar plates were emulsified in 5 ml of sterile physiological saline and the turbidity adjusted to 0.5 McFarland standards (approximately a cell density of 1.5×10^8 cfu/ml). The standardized suspensions were inoculated onto the surface of sterile Mueller Hinton agar. The sensitivity discs of the various antibiotics were placed aseptically using a sterile forceps on the dried inoculated agar surface. The plates were allowed to stand for 15-30 minutes before they were incubated at 37°C for 18 hours. After incubation, the zones of inhibition were measured using metre rule and results interpreted according to [10]

RESULTS AND DISCUSSION

A total of 249 samples were collected from 83 mothers and their neonates, of which 80 (96.39 %) were live births. Figure 1 shows the distribution of samples across the 3 hospitals included in the study. Nasal swab was collected from the right and left nostril of 42 (50.6 %) male and 41 (49.4 %) female neonates. The mean weight of the MRSA positive neonates was 2.99 ± 0.54 kg while the mean APGAR score was 8.48 ± 2.16 .

A total of 40 different Staphylococcal isolates were isolated on Mannitol salt agar. It comprised 8 isolates from mothers, 15 from the right and 16 from the left nostril of the neonates. Upon specific identification using Integral System Staphylococchi from Liofilchem Diagnostici, 2 isolates came out positive from the mothers, 5 from the right and 6 from the left nostril of the neonates giving a total of 13 positive *Staphylococcus aureus* isolates from eight (8) participants. All the 13 isolates were resistant to Cefoxitin disc and hence considered as Methicillin Resistant *Staphylococcus aureus*. Neonates with 1 MRSA isolate are 5 while neonates with 2 MRSA isolates are 3. Figure 2 gives a description of MRSA isolates based on Sample type. The demography of the 8 neonates and the 2 mothers is presented in Table 1. while distribution of the isolates by hospital and sampling site is presented

in Tables 2 and 3. A 5.22 % incidence of MRSA obtained in this study implies that there is possibility of colonization of healthy and high APGAR scoring neonates and their mothers with opportunistic pathogens such as MRSA, which is often the genesis of infection. This therefore necessitates the need for infection surveillance even in healthy neonates and their mothers. The isolates were phenotypically identified as methicillin resistant from the cefoxitin disk screen test and this agrees with the work of [8] in Turkey who had all their 494 *S. aureus* phenotypically positive for methicillin using oxacilin and cefoxitin disc.

Figure 3 shows a breakdown of the isolates gotten and the relationship between isolates from mothers and that of neonates. Of the 2 positive mothers, the right and left nasal swab of one of the mother's neonate produced positive isolates. Seven (7) other neonates whose mother's vaginal swabs were negative for MRSA were positive for MRSA of which 3 were already being breastfed as at the time of sample collection. The 2 (2.41 %) positive for MRSA in the vaginal swab is similar to the findings of [21] which recorded 1 % Vaginal MRSA carriage in Japan with a sample size of 898 pregnant women. But lower than the study of [17] that gave colonization among pregnant women in African as ranging from 4.36 – 15.34 % and the report of [22] that reported prevalence of MRSA vaginal carriage among pregnant women in Ado-Ekiti, Nigeria to be 14.3 % with a sample size of 350 pregnant women. The low value of vaginal MRSA carriers in this study may be due to the low sample size used and the fact that sampling was limited to pregnant women in labour whose labour has progressed to the point of admission to the delivery unit. Women whose labour was just beginning were not included as they were not admitted by the hospital and as such most of them don't often return back to the hospital and may likely give birth at home subsequently.

The finding of [8] who observed 8.1 % incidence among paediatrics patient in Turkey, is similar to the 9.64% obtained in this study. Our result is slightly higher than the findings of [15] who reported an annual incidence of MRSA among neonates to range from 5.66 - 7.66 % in China. It is higher than [30] who recorded 5.3 % among children aged 6 months – 16 years old in Ibadan, Nigeria. Other studies for example [16] recorded 7 % MRSA isolation in Northern Nigeria while [21] reported 0.8 % incidence in the umbilical cord or nasal swab of neonates taken as at the time of delivery in Japan. The disparity in these results may be due to the difference in sample sizes used and the routine hygiene practices obtainable the facilities sampled.

One neonate (50 %) of the MRSA positive mothers gave positives isolates in both nares which is similar to the findings of Ogura *et al.* 2021 which had 44.4 % and higher than the report of [18] which reported 27.8 %. This shows that it is possible for a Vaginal MRSA carrier mother to vertically transmit the pathogen to their new born during vaginal delivery. [23] demonstrated in their study that *S. aureus* is the only organism that was vertically transferred among other pathogenic organisms that cause neonatal septicaemia. The MRSA isolated from one of the nares of the neonate of the carrier mother exhibited similar antibiotics susceptibility profile as that isolated from the mother.

Three of the 7 neonates with MRSA negative mothers had begun breastfeeding before samples were taken from them and their mothers affirmed not washing their hands before commencing breastfeeding. Although this research did not collect nasal and hand swabs from mothers for examination, works by several authors including [23] and [21] suggests that vertical transmission can also take place from mothers who harbour MRSA in their nares or other location to their neonates when these neonates come in contact with them. This work agrees with this finding since appropriate hygiene measures were upheld in this particular hospital where the neonates were born ruling out the fact that the hospital equipment (such as penguin suckers, heaters, resuscitator, and so on) or the nurses could be a source of transmission of the pathogens.

One of the 7 MRSA positive neonates born to the no MRSA group mothers was delivered as an intra-uterine foetal death (IUFD) weighing 1.4 kg with gestational age of 29 weeks. This suggests that colonization with MRSA can also take place in the uterus since the baby had not come in contact with any hospital equipment as at the time of sample collection and the mother's vaginal swab was negative for MRSA. This agrees with the suggestion of [15] and [26] that colonisation with MRSA can take place in the womb too.

It is interesting to note the remaining 3 isolates were obtained from MRSA negative vaginal swabs who gave birth in the same delivery unit. Their samples were taken during dressing of the neonates by the hospital staff before contact with the mothers or relatives. This isolates also showed related antibiotics resistance pattern. This suggests that these pathogens could have been transferred from the hospital staff or equipment as [9] had opined. These sources were not sampled and hence it is a postulate.

Isolation of MRSA was recorded more in the left nares (46.16 %) of the neonates than in the right nares (38.46 %) of the neonates as shown in figure 4. Five (62.5 %) of the 8 positive neonates showed colonisation in only 1 naris, 3 in the left and 2 in the right. The isolates from the 3 (37.5 %) positive neonates that were positive in both nares had varying culture characteristics and antibiotics susceptibility profiles. This study shows that swabbing both nares separately is important for identifying colonization with MRSA in the nares and the specific site of colonisation contrary to [11] who only used one swab to collect specimen from both nares. Others studies [13] and [21] recommend additional sampling from other sites like the umbilical cord, stool, rectum, groin, throat and axilla for adequate representation of colonization.

Male neonates (14.3 %) were more predisposed to MRSA colonisation than the female neonates (4.9 %) as against the report of [11] who recorded more colonisation in female neonates than in males probably because more male neonates were born during the period of this study which may have increased the chances of isolation from male neonates than females.

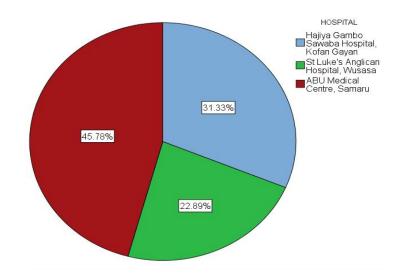


Figure 1: Distribution of Samples Across the 3 Hospitals

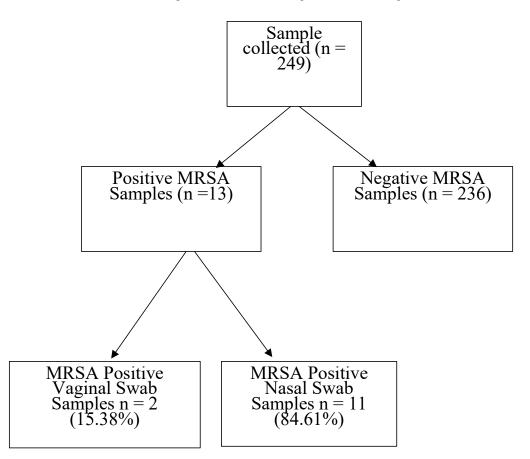


Figure 2: Distribution of MRSA Isolates Based on Sample Type

Table 1: Demography of Neonates with MRSA

S/N	Isolate	Gender	Weight (kg)	APGAR
1	A22R	Male	1.4	0.0
2	A22L	Male	1.4	0.0
3	B7L	Male	1.9	9.0
4	B8L	Male	3.1	10.0

5	B13	Female	-	-
6	B13R	Male	3.0	9.5
7	B13L	Male	3.0	9.5
8	B15R	Female	3.0	9.0
9	B15L	Female	3.0	9.0
10	C14R	Female	2.6	9.5
11	C19L	Male	3.3	9.0
12	C31	Female	-	-
13	C36R	Male	2.9	9.5

Key: APGAR = Appearance Pulse Grimace Activity and Response

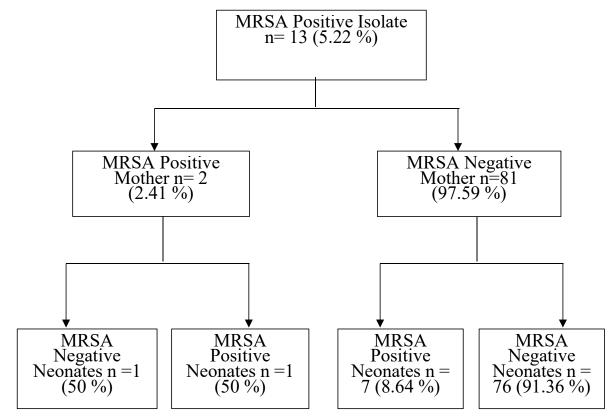


Figure 3: Flow Chart Showing Vertical Transmission of MRSA from Mother to Neonate

Table 2: Percentage Distribution of S. aureus Isolates Base	ed on Sampling Sources (Hospitals)
---	------------------------------------

Hospital	No. of MRSA Samples	% Frequency
Hajiya Gambo Sawaba General Hospital, Kofan Gaya. (n = 78)	2	2.56
St. Luke's Anglican Hospital, Wusasa. (n = 57)	7	12.28
ABU Medical Centre, Samaru (n = 114)	4	3.51

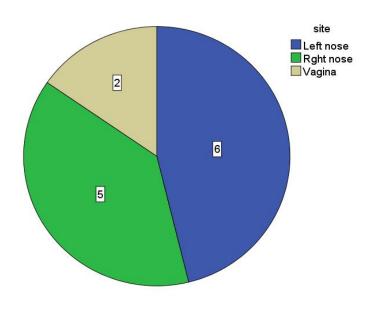


Figure 4: Distribution of Isolates by Sampling Site (n = 13)

From the antibiotics selected according to [10], high resistance was exhibited by the isolates against Cefoxitin, Amoxicillin/Clavulanic acid, Azithromycin and Trimethoprim-sulfamethoxazole, moderate resistance was recorded to Erythromycin. In contrast, percentage resistance was much lower against Gentamicin, Ciprofloxacin, Rifampicin and Tetracycline ranging from 22 - 38 %. As the report of susceptibility to antibiotics shows in figure 5, a large proportion of the isolates were resistant to commonly used drugs which is in consonance with reports of several other researches [8], [1], [3] and [32]. However, it differs from the findings of [33], [5] and [2] that reported lower levels of resistance to these antibiotics. The high resistance to Amoxicillin-clavulanic acid could be due to the fact that it was the drug of choice prescribed to the pregnant women that had infection before delivery in the study area.

Classification of isolates was based on their resistance pattern. From the [10] Antibiotics Susceptibility Test (AST) breakpoint, 46.15 % of the MRSA isolates were multidrug resistant (MDR), 23.08 % were extensively drug resistant (XDR) and 15.38 % were Pan drug resistant (PDR) (Figure 6). Figure 7 shows the distribution of resistance types according to [10] selected antibiotics.

The ratio of the number of antibiotics isolates were resistant to, to the total number of antibiotics isolates were exposed to was used to determine the Multiple Antibiotic Resistance Index (MARI). The range was from 0.3 to 1 as shown in Table 5. All isolates had MAR index greater than 0.25, which suggests that isolates sources were environment with heavy and perhaps indiscriminate use of antibiotics.

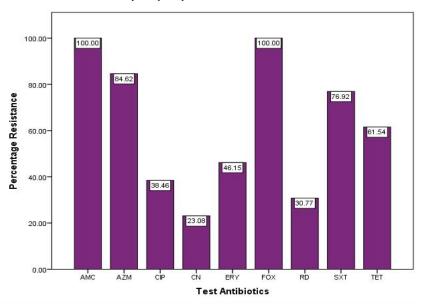


Figure 5: Antibiotics Resistant Pattern of MRSA Isolates (n = 13) with Antibiotics Selected from EUCAST (2023)

Key: Amoxicillin/Clavulanic acid (AMC), Azithromycin (AZM), Ciprofloxacin (CIP), Gentamicin (CN), Erythromycin (ERY), Cefoxitin (FOX), Rifampicin (RD), Trimethoprim-sulfamethoxazole (SXT), and Tetracycline (TET).

Isolate	Resistance Phenotypes	Number of antibiotics
C36R	FOX,CN,AZM,TET,AMC,SXT,RD,CIP,ERY	9
B13	FOX,CN,AZM,TET,AMC,SXT,RD,CIP,ERY	9
C31	FOX,CN,AZM,TET,AMC,SXT,RD,ERY	8
B13L	FOX,AZM,TET,AMC,SXT,CIP,ERY,	7
B8L	FOX,AZM,AMC,SXT,RD,CIP,ERY	7
B15L	FOX,AZM,TET,AMC,SXT,CIP	6
B15R	FOX,AZM,TET,AMC,SXT	5
C14R	FOX,AZM,TET,AMC,SXT	5
C19L	FOX,TET,AMC,ERY	4
A22R	FOX,AZM,AMC,SXT	4
B7L	FOX,AZM,AMC,SXT	4
A22L	FOX,AZM,AMC	3
B13R	FOX,AMC	2

TABLE 3: Phenotypic Distribution of the Resistance Profile of MRSA Isolates (n = 13) Based on EUCAST 2023

Key: Amoxicilin/Clavulanic acid (AMC), Azithromycin (AZM), Ciprofloxicin (CIP), Gentamicin (CN), Erythromycin (ERY), Cefoxitin (FOX), Rifampicin (RD), Trimethropin-sulfamethoxazole (SXT), and Tetracycline (TET).

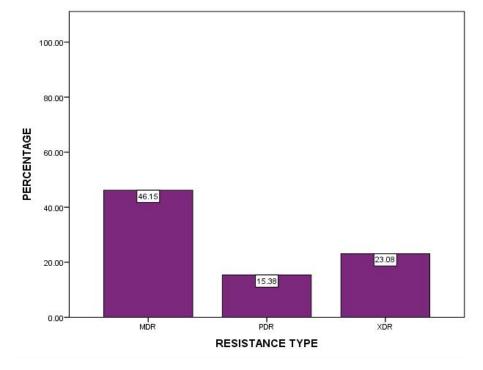


Figure 7: Percentage Distribution of Antibiotic Resistance Types of MRSA Isolates to EUCAST 2023 Selected Antibiotics Key: Multidrug resistant (MDR), Pan drug resistant (PDR), Extensively drug resistant (XDR)

MARI	MARI	No. of Isolates	Percentage %
1	0	0	0.0
0.89	0.1	0	0.0
0.78	0.3	2	15.4
0.78	0.4	2	15.4
0.67	0.5	3	23.1
0.55	0.6	2	15.4
0.44	0.7	0	0.0
0.44	0.8	2	15.4
0.33	0.9	1	7.7
0.22	1.0	1	7.7

Table 5: Multiple Antibiotic Resistance Indices (MARI) of the MRSA Isolates (n = 13)

CONCLUSION

Incidence of *S. aureus* was 5.22 % in this study with 9.64 % of neonates being nasal carrier and 2.41 % of mothers were vaginal carrier. All *S. aureus* isolates were phenotypically MRSA. All isolates showed resistance to more than 1 class of antibiotics. Majority of the isolates were MDR (84.62 %) with MAR indices greater than 0.2. The percentage of MRSA in healthy neonates within 0 - 2 days of birth is worrisome and should be a source of concern.

REFERENCES

- AbdulAziz Z. A., Onaolapo, J. A., Ibrahim, Y. K. E., Olayinka, B., & Abdulaziz, M. M. (2022). Prevalence and Antimicrobial Resistance Profile of Methicillin Resistant Staphylococcus aureus isolates from Wound Infections in Zaria, Nigeria. *Journal of Current Biomedical Research* 2(5), 475 - 489 https://doi.org/10.54117/jcbr.v2i5.6
- Akpudo, M. O., Jimoh, O., Adeshina, G. O., & Olayinka, B. O. (2023). Biofilm Formation and Antimicrobial Susceptibility Pattern of *Staphylococcus aureus* Clinical Isolates from Two Healthcare Facilities in Zaria. *Nig. J. Pharm. Res.* 2023, 19 (1),59-70 https://dx.doi.org/10.4314/njpr.v19i1.6
- Almohammady, M. N., Eltahlawy, E. M., & Reda, N. M. (2020). Pattern of bacterial profile and antibiotic susceptibility among neonatal sepsis cases at Cairo University Children Hospital. *Journal of Taibah University Medical Sciences*, 15(1), 39–47. https://doi.org/10.1016/j.jtumed.2019.12.005
- Aseffa, A., & Abathun, T. (2020). Prevalence of Neonatal Sepsis and Associated Factors amongst Neonates Admitted in Arbaminch General Hospital, Arbaminch, Southern Ethiopia, 2019. 10(1), 1–7. https://doi.org/10.15406/jpnc.2020.10.00404
- Asinobi, I., Ekwochi, U., Edelu, B., Iheji, C., Onu, N., & Ndu, I. (2021). Bacterial Isolates Of Surfaces In The Neonatal Intensive Care Unit Of Enugu State University Teaching Hospital, Parklane, Enugu, and Their Antibiotic Susceptibility Patterns. *Nigerian Journal of Medicine*, 30(2), 171. https://doi.org/10.4103/njm.njm_157_20
- Braima, O., Ali, M., & Abdulla, E. (2021). Bacteriological profile and antibiotic resistance in newborn infants with possible communityacquired neonatal sepsis in Khartoum State, Sudan. Sudanese Journal of Paediatrics, 21(1), 13–22. https://doi.org/10.24911/sjp.106-1601909519
- <u>Cailes B, Kortsalioudaki C, Buttery J, Pattnayak S, Greenough A. Epidemiology of UK neonatal infec- tions: the neonIN infection</u> surveillance network. 2017. https://doi.org/10.1136/archdischild-2017- 313203 PMID: 29208666.
- Cikman, A., Aydin, M., Gulhan, B., Karakecili, F., Cicek, A. C., Bilman, F. B., Ciftci, I. H., Kara, M., & Atmaca, S. (2019). Journal of Infection and Public Health Absence of the mecC gene in methicillin-resistant Staphylococcus aureus isolated from various clinical samples: The first multi-centered study in Turkey. *Journal of Infection and Public Health*, *12*(4), 528–533. <u>https://doi.org/10.1016/j.jiph.2019.01.063</u>
- Dramowski A., Madide A., & Bekker A. (2015), "Neonatal nosocomial bloodstream infections at a referral hospital in a middle income country: burden, pathogens, antimicrobial resistance and mortality," Paediatrics and International Child Health, 35(3), 265–272.

- 10. European Committee on Antimicrobial Susceptibility Testing EUCAST 2023.
- Geng, W., Qi, Y., Li, W., McConville, T. H., Hill-Ricciuti, A., Grohs, E. C., Saiman, L., & Uhlemann, A. C. (2020). Epidemiology of Staphylococcus aureus in neonates on admission to a Chinese neonatal intensive care unit. *PLoS ONE*, 15(2), 1–17. <u>https://doi.org/10.1371/journal.pone.0211845</u>
- 12. Haider, S., Nazir, S., Sehar, K., & Saleem, M. (2018). Mortality associated with isolated pathogens in neonatal sepsis at Izzat Ali Shah Hospital, Wah Cantt. *Medical Forum Monthly*, 29(9), 36–39.
- Heinrich, N., Mueller, A., Bartmann, P., Simon, A., Bierbaum, G., & Engelhart, S. (2011). Successful management of an MRSA outbreak in a neonatal intensive care unit. *European Journal of Clinical Microbiology and Infectious Diseases*, 30(7), 909–913. https://doi.org/10.1007/s10096-011-1175-4
- 14. Huang YC, Chou YH, Su LH, Lien RI, Lin TY. (2006) Methicillin-resistant Staphylococcus aureus colonization and its association with infection among infants hospitalized in neonatal intensive care units. *Pediatrics*. 118(2):469–474
- Huang, H., Ran, J., Yang, J., Li, P., & Zhuang, G. (2019). Impact of MRSA Transmission and Infection in a Neonatal Intensive Care Unit in China: A Bundle Intervention Study during 2014-2017. *BioMed Research International*, 2019. <u>https://doi.org/10.1155/2019/5490413</u>
- John-Ugwuanya, G. A., Obaro, S. K., Olayinka, B. O., Onaolapo J. A., Hassan-hanga, F., Munir, H., & Fey, P. D. (2018). *Staphylococcus aureus* Bacteraemic Infection in Blood Culture of Children Attending Different Hospitals in Northern Nigeria. 7(6), 2013.
- Maliihe, N.S., Mahdi, S., Abolfazl, M., Nayereh, A., Soghra, K., Zohreh, S., Mahmonir, D., Sahar, E., Ali, H.M., & Ali, R. (2022). Worldwide Prevalence of Maternal Methicillin-resistant *Staphylococcus aureus* Colonization: A systematic review and Meta-analysis. ScienceDirect. 171. https://doi.org/10.1016/j.micpath.2022.105743.
- Matok, L. A., Azrad, M., Leshem, T., Abuzahya, A., Khamaisi, T., Smolkin, T., & Peretz, A. (2021). Mother-to-neonate transmission of antibiotic-resistant bacteria: A cross-sectional study. *Microorganisms*, 9(6), 1–18. <u>https://doi.org/10.3390/microorganisms9061245</u>.
- 19. Mitra, D.K., Mullany, L.C., Harrison, M. (2018). Incidence and risk factors of neonatal infections in a rural Bangladeshi population: a community-based prospective study. *J Health Popul Nutr.* 37(6):1–11.
- 20. National Population Commision (NPC) [Nigeria] and ICF International. (2019). Nigerian Demographic and Health Survey 2018. Abuja Nigeria and Rockville, Maryland, USA: NPC and ICF International
- Ogura, J., Inayama, Y., Sasamoto, N., Hirayama, T., Ohara, T., Sakai, M., Suzuki, H., Yasumoto, K., Suginami, K., & Yamanoi, K. (2021). Vertical transmission of methicillin-resistant Staphylococcus aureus at delivery and its clinical impact: An observational, prospective cohort study. *Acta Obstetricia et Gynecologica Scandinavica*, 100(11), 1986–1994. https://doi.org/10.1111/aogs.14240
- Okiki, P. A., Eromosele, E. S., & Sobajo, O. A. (2020). Occurrence of mecA and blaZ genes in methicillin-resistant *Staphylococcus aureus* associated with vaginitis among pregnant women in Ado-Ekiti, Nigeria. *New Microbes and New Infections*, 38, 100772. https://doi.org/10.1016/j.nmni.2020.100772
- Okomo, U. A., Darboe, S., Bah, S. Y., Ayorinde, A., Jarju, S., Sesay, A. K., Kebbeh, N., Gai, A., Dibbasey, T., Grey-Johnson, M., Le Doare, K., Holt, K. E., Lawn, J. E., & Kampmann, B. (2023). Maternal colonization and early-onset neonatal bacterial sepsis in the Gambia, West Africa: a genomic analysis of vertical transmission. *Clinical Microbiology and Infection*, 29(3), 386.e1-386.e9. https://doi.org/10.1016/j.cmi.2022.10.012
- Olorode, O. A., & Nanighe, S. O. (2021). Molecular Detection of Methicillin Resistance Genes (mecA; Pvl) in Methicillin Resistant Staphylococcus aureus Isolates from Federal Medical Centre, Yenagoa, Bayelsa State, Nigeria. 04(10).
- Pierce, R., Lessler, J., Popoola, V. O., & Milstone, A. M. (2017). Meticillin-resistant *Staphylococcus aureus* (MRSA) acquisition risk in an endemic neonatal intensive care unit with an active surveillance culture and decolonization programme. *Journal of Hospital Infection 95*(1), 91–97. https://doi.org/10.1016/j.jhin.2016.10.022
- Sakaki H, Nishioka M, Kanda K (2009). An investigation of the risk factors for infection with methicillin-resistant Staphylococcus aureus among patients in a neonatal intensive care unit. Am J Infect Control. 2009; 37:580–586. [PubMed: 19535174]
- Sharma, M., Pathak, S., & Srivastava, P. (2013). Prevalence and Antibiogram of Extended Spectrum β-Lactamase (ESBL) producing Gram negative bacilli and further molecular characterization of ESBL producing Escherichia coli and Klebsiella spp. *Journal of clinical and diagnostic research* 7(10), 2173-2177.
- Sule, I., Omokanye, M., Adekunle, O., Abdulsalam, Z., & Elemi, F. (2022). Molecular Detection of spa, mecA, and mecC genes in *Staphylococcus aureus* Isolated from Soils and Wooden Table Surfaces from Abattoirs in Ilorin, Kwara, Nigeria. *Alexandria Journal of Veterinary Sciences*, 74(1), 52. <u>https://doi.org/10.5455/ajvs.100451</u>

- Tumuhamye, J., Sommerfelt, H., Bwanga, F., Ndeezi, G., Mukunya, D., Napyo, A., Nankabirwa, V., & Tumwine, J. K. (2020). Neonatal sepsis at Mulago national referral hospital in Uganda: Etiology, antimicrobial resistance, associated factors and case fatality risk. *PLoS ONE* 15(8), 1–14. <u>https://doi.org/10.1371/journal.pone.0237085</u>
- Tuta, K. E., Okesola, A. O., & Umeokonkwo, C. D. (2019). The Prevalence and Risk Factors Associated with Nasal Methicillin-Resistant Staphylococcus aureus Colonization among Children in a Tertiary Hospital in Nigeria. Ethiopian Journal of Health Sciences, 29(4), 487– 494. https://doi.org/10.4314/ejhs.v29i4.10
- 31. UN IGME. Levels and trends in child mortality report 2022. New York, NY: UN Inter-agency Group for Child Mortality Estimation, 2022.
- Wu, X., Wang, C., He, L., Xu, H., Jing, C., Chen, Y., Deng, J., Lin, A., Deng, H., Cai, H., Chen, Y., Yang, J., Zhang, T., Cao, Q., Hao, J., Huang, Y., & Yu, H. (2023). Clinical characteristics and antibiotic resistance profile of invasive MRSA infections in new-born inpatients: a retrospective multicenter study from China. *BMC Pediatrics* 23(1), 1–8. <u>https://doi.org/10.1186/s12887-023-04084-0</u>
- Zamarano, H., Musinguzi, B., Kabajulizi, I., Manirakiza, G., Guti, W., Muhwezi, I., Hussein, A. A., Baweera, A., Kabahinda, B., Itabangi, H., & Bazira, J. (2021). Bacteriological profile, antibiotic susceptibility and factors associated with neonatal Septicaemia at Kilembe mines hospital, Kasese District Western Uganda. *BMC* Microbiology, 1–11. https://doi.org/10.1186/s12866-021-02367-z