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### ANTIBIOTIC SUSCEPTIBILITY PROFILE OF Staphylococcus aureus AND Pseudomonas aeruginosa FROM NIPPLE OF BREASTFEEDING MOTHERS ATTENDING POSTNATAL CARE AT GENERAL HOSPITAL KATSINA

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ABSTRACT

Several microorganisms may contaminate nipples of breastfeeding mothers and become potential to cause mastitis. The aim of this study is to investigate the antibiotic susceptibility profile of Staphylococcus aureus and Pseudomonas aeruginosa from nipples of breastfeeding mothers. Fourty (40) samples were collected using sterile oneuse swab sticks from breast nipples of breast-feeding mothers at postnatal and immunization unit of General Hospital Katsina. The samples collected were inoculated on Nutrient agar and Mannitol salt agar by streak plate method. The cultured plates were then incubated for 24 hours at 37°C after which the plates were observed for growth. Biochemical tests were carried out on the isolates to identify the bacteria. Antimicrobial susceptibility test was carried out for the bacterial isolates using Disc diffusion method. The zones of inhibition observed were measured in millimetres. All the Fourty samples collected were culture positive. Staphylococcus aureus 34 (85%) was the most prevalent bacterium associated with nipple contamination compared to Pseudomonas aeruginosa 6 (15%). Breast-feeding mothers of 18 to 25 years, (75%) have the highest number of contaminated nipples with the bacteria. The antibiotics with highest sensitivity to Staphylococcus aureus was ciprofloxacin, while the ones with the least sensitivity were gentamicin and streptomycin. Staphylococcus aureus was completely resistant to Ampiclox and Cefuroxine. Similarly, Pseudomonas aeruginosa was highly sensitive to ciprofloxacin, while the organism has least sensitivity to amoxicillin. The antibiotics with the highest sensitivity to both organisms are ciprofloxacin, followed by Perfloxacin and amoxicillin.

Keywords: Breast nipple, Staphylococcus aureus, Pseudomonas aeruginosa, antibiotic

#### INTRODUCTION

The microorganisms associated with human nipple contamination and potential to cause mastitis are Staphylococcus aureus, Pseudomonas aeruginosa, Candida albicans, Escherichia coli, Lactobacillus spp, Enterococcus faecalis and Staphylococcus epidermidis (Gerber et al., 2003). The most common bacteria causing lactational mastitis is Staphylococcus aureus (Wambach and Riordan, 2014). Mastitis simply means an inflammation of the breast tissue. This inflammation may or may not involve bacterial infection' (Amir and ABM, 2014). It usually occurs in the second and third weeks after delivery, and rarely after the twelfth week (WHO, 2000). Initially, the intraductal pressure rises due to milk stasis, with consequent flattening of the alveolar cells and development of spaces between the cells. In mastitis, the affected portion of the breast is

painful, reddish, swollen and warm. If infection is present, systemic manifestations may occur such as malaise, high-grade fever (body temperature  $\geq$  38 °C) and chills. Mastitis is commonly unilateral but may be bilateral as well. Sodium and chloride levels are elevated in the milk whereas lactose levels are low, which makes the milk taste saltier and may be rejected by the infant. (Wambach and Riordan, 2016). It appears to develop from engorgement of noninfective mastitis to infective mastitis and finally to breast abscess as a complicated disease (Amir et al, 2014). Non-infective mastitis presents with inflammation and complication of the disease. It may arise from milk stasis due to poor removal, sudden changes in the feeding pattern of babies, trauma and from pressure of clothing or pressure from holding the breast, resulting in a plugged or blocked ducts (Wambach and Riordan, 2016).

Special Conference Edition, November, 2019

Infective mastitis is caused by infections either in the outer skin of the breast or within the glandular tissue of the breast. The incidence of lactational mastitis varies from 2.6% to 33%, according to World Health Organization. The prevalence was about 10% among breastfeeding women across the globe. Most breast infections occur within the first or second month after delivery or at the time of weaning (Amir *et al.*, 2014). The aim of this study is to investigate the antibiotic susceptibility profile of *Staphylococcus aureus* and *Pseudomonas aeruginosa* from nipples of breastfeeding mothers attending postnatal care at General Hospital Katsina.

#### MATERIALS AND METHODS Study Area

The study was conducted at General Hospital Katsina, Katsina state northern Nigeria. The state has a total land area of about 3,370 square kilometres with a total population of 5,801,584 according to 2006 census. Katsina state is bounded to Niger republic to the north, Kano state to the east, Kaduna state to the south and Zamfara state to the west. Katsina is an agricultural land generating grains such as maize, millet, and cotton. It is a state comprising of the Hausa-Fulani with 3 senatorial zones namely; Katsina zone (Katsina central), Funtua zone (Katsina south) and Daura zone (Katsina north).

#### Study Population

The population involved in this study were breastfeeding Mothers attending postnatal care at maternity unit of General Hospital Katsina and whose ages ranged from 18-35 years during their postnatal visit.

#### **Ethical Clearance and Consent**

Ethical clearance was obtained from katsina state ministry of health with the approval number (MOH/ADM/SUB/1152/1/221). Informed consent was obtained from the study subjects before the sample collection.

#### Sample Collection

Fourty (40) samples were collected using simple random sampling method. Sterile one-use swab sticks were used to collect the samples from the nipples of breast feeding mothers attending postnatal care at General Hospital Katsina. The specimens were labelled based on age and number of the participants and then immediately transported to the microbiology laboratory of Umaru Musa Yar'adua University Katsina in a cooler containing ice blocks for analysis. The samples were enriched in Nutrient Broth and Incubated at  $37^{0}$  C for 24 hours prior to analysis.

#### **Bacterial Detection**

The 40 samples collected were inoculated on Nutrient agar and Mannitol salt agar by streak

plate method. The cultured plates were then incubated for 24 hours at 37°C after which the plates were observed for bacterial growth (Ewansiha *et al.*, 2015). Identification of the bacteria was carried out using biochemical tests such as Gram staining, coagulase test, catalase test, oxidase test, indole, motility test, and Vogas Proskauer. Antibacterial sensitivity test was performed using disc diffusion method. Sensitivity of the isolates was determined by taking the average diameter of each zone of inhibition around each disc and the values obtained were compared with that of the clinical laboratory standards institute (Cheesbrough, 2006).

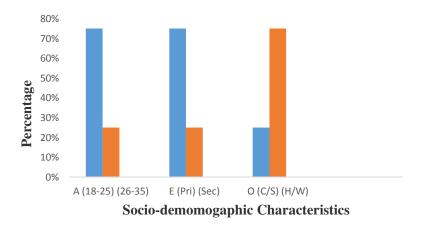
#### **Data Analysis**

The data obtained from demographic characteristics of the study subjects and the pathogenic bacteria detected were analyzed using SPSS version 16.0 and presented by descriptive statistics using frequency and percentage.

#### RESULTS

Culture results from fourty (40) samples of the nursing mothers were positive for Staphylococcus aureus and Pseudomonas aeruginosa. Breast-feeding mothers within the age of 18-25 years (75%) have the highest number of contaminated breast nipple compared to those aged 26-35 years (25%). More than half of the mothers 30 (75%) have primary school certificates. Majority of the clients are full house wives having 30 (75%), while the remaining 10 (25%) are civil servants (Figure 1). The most frequently detected bacterium from breast nipple swab was Staphylococcus aureus 34(85%) while the least detected bacterium was Pseudomonas aeruginosa 6 (15%) (Table 1).

The Staphylococcus aureus isolated was sensitive to ciprofloxacin, Perfloxacin, erythromycin, cotrimaxazole, ceftriaxone, amoxicillin, gentamycin and streptomycin while resistant to ampiclox and ceftriaxone. The antibiotic to which the bacterium had highest sensitivity is ciprofloxacin (18.5mm), while the ones with the least sensitivities are gentamycin (13.0mm) and streptomycin (13.00mm) (Table Pseudomonas aeruginosa isolates were 2). sensitive to ciprofloxacin, ofloxacillin, Perfloxacin, gentamycin, streptomycin, Sparfloxacin amoxicillin cotrimixazole, and gentamycin whereas the organism was completely resistant to amoxicillin clavulanic acid and Chloramphenicol (Table 3). The antibiotic sensitivity the highest to with both Staphylococcus aureus and Pseudomonas aeruginosa is ciprofloxacin with zone of inhibition 18.5mm and 20.5mm respectively.



#### Figure1: Socio-demographic Characteristics of the Study Subjects.

Key: A: Age (yrs)

E: Education (Pri; Primary; Sec; Secondary)

O: Occupation (C/S; civil servants; H/W; house wives)

# Table 1: Prevalence of the bacteria detected from the nipples of breastfeeding mothers.Bacteria isolatedNumber (%)

Staphylococcus	34 (85%)
aureus Pseudomonas	6 (15%)
<i>aeruginosa</i> Total	40 (100%)

#### Table 2: Antibiogram against S.aureus isolated from Breast Nipple of lactating mothers.

Antibiotics Used	Zone of inhibition(mm)	Organisms inhibited
Amoxicillin	14.5mm	S. aureus
Ampiclox	0.0mm	S. aureus
Septrin	15.5mm	S. aureus
Gentamycin	13.0mm	S. aureus
Zinnacef	0.0mm	S. aureus
Ciprofloxacin	18.5mm	S. aureus
Streptomycin	13.0mm	S. aureus
Erythromycin	17.0mm	S. aureus
Perfloxacin	17.0mm	S. aureus
Rocephin	15.5mm	S. aureus

**Key:** *S.aureus* = *Staphylococcus aureus* 

## Table 3: Antibiogram against *P.aeruginosa* isolated from Breast Nipple of breastfeeding mothers.

Antibiotics used	Zones of inhibition(mm)	Organism inhibited
Ciprofloxacin	20.5mm	P.aeruginosa
Augmentin	0mm	P.aeruginosa
Perfloxacin	16.0mm	P.aeruginosa
Septrin	10.0mm	P.aeruginosa
Tarivid	19.5mm	P.aeruginosa
Gentamycin	15.0mm	P.aeruginosa
Streptomycin	14.0mm	P.aeruginosa
Chloramphenicol	0.0mm	P.aeruginosa
Sperfloxacin	10.0mm	P.aeruginosa
Amoxicillin	10.0mm	P.aeruginosa

**Keys:** *P.aeruginosa* = *Pseudomonas aeruginosa* 

### Special Conference Edition, November, 2019 DISCUSSION

Contamination of nipple with bacteria might be a potential risk for mastitis. Mastitis is one of the common breast infection in breastfeeding mothers. A devastating disease that leads to cessation or interruption of breastfeeding among the affected mothers. It causes decrease breast milk production significantly due to the inflammatory processes occurring within the nipple or entire breast breast tissue (Angelopoulou et al., 2018). This study identified Staphylococcus aureus and Pseudomonas aeruginosa from nipples of breastfeeding mothers potential to cause mastitis. The most vulnerable mothers are identified from this study are those aged 18 to 25 years (75%) because they have the highest number of contaminated breast nipples with these bacteria. This may be related to their low level of education.

In this study, the most frequently isolated bacterium from the contaminated nipples of breast feeding mothers is Staphylococcus aureus followed by *Pseudomonas aeruginosa*. The Staphylococcus aureus was sensitive to ciprofloxacin, Perfloxacin, erythromycin, ceftriaxone, cotrimaxazole, amoxicillin, gentamycin and streptomycin but resistant to ampiclox and Cefuroxime. This is comparable to the findings of Ewansiha et al. (2015) in which Staphylococcus aureus and Staphylococcus epidermidis were the most frequently isolated bacteria from the breast nipple of nursing mothers. These bacteria were sensitive to ciprofloxacin, chloramphenicol, amoxicillin and levofloxacin but resistant to ampicillin. The similarities in both findings are Staphylococcus aureus was the most commonly isolated bacterium isolated from breast nipples of lactating mothers and was sensitive to ciprofloxacin and amoxicillin but resistant to ampicillin/ampiclox. Although Pseudomonas aeruginosa was the least isolated bacterium in this study, it was found to be sensitive to ciprofloxacin, ofloxacillin. Perfloxacin, cotrimaxazole, streptomycin, Sparfloxacin, amoxicillin and gentamycin while completely resistant to Amoxicillin clavulanic acid and Chloramphenicol.

Moreover, it was observed that mastitis caused by *Staphylococcus aureus* in breastfeeding mothers should be treated with cephalexin and

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dicloxacillin (Spencer, 2008). However, this is contrary to our findings in which ciprofloxacin is most effective for treatment of the Staphylococcus aureus potential to cause mastitis in lactating mothers as evidenced by highest sensitivity of 18.5 mm than other antibiotics. Since there is a dilemma of using ciprofloxacin during breastfeeding in terms of safety, it can be replaced by amoxicillin considering its sensitivity to both organisms. Staphylococcus aureus is also resistant to Cefuroxine, which is in the same class with cephalexin as both are second-generation Livingstone et al. (1996) cephalosporins. reported that majority of the breastfeeding mothers with secondary nipple infection were tested positive for Staphylococcus aureus.

#### CONCLUSION

The colonisation of breast nipples with bacteria during breast-feeding can cause secondary breast nipple contamination, which may subsequently lead to infections. This study revealed that *Staphylococcus aureus* and *Pseudomonas aeruginosa* are the commonest bacteria contaminating nipples of breastfeeding mothers and may cause mastitis. The antibiotic with the highest sensitivity to both the two organisms was ciprofloxacin, followed by Perfloxacin and amoxicillin. The most vulnerable age group to breast nipple contamination with the two bacteria are breast-feeding mothers aged 18 to 25 years.

#### RECOMMENDATIONS

- Ciprofloxacin, Perfloxacin, erythromycin, Cefuroxine and amoxicillin should be considered during empirical treatment for acute mastitis in lactating mothers attending postnatal care at general hospital katsina.
- Cefuroxine and ampiclox should not be considered as first drugs of choice during empirical treatment for mastitis in breastfeeding mothers at the same hospital.
- Nursing mothers should improve their Personnel hygiene including breast care to avoid breast nipple contamination, which can cause infection and disruption of breastfeeding.
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