Multidisciplinary Research and Development Journal Int'l
https://mdrdji.org
Vol 5 Issue 1. May 2024

ANTIBACTERIAL RESISTANCE PATTERN OF GROUP B STREPTOCOCCI INFECTION IN PREGNANT WOMEN ATTENDING ST JOHN'S HOSPITAL, OWERRI, NIGERIA

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ABSTRACT

Streptococcus agalactiae (group B streptococci, GBS) is one of the causes of maternal and neonatal morbidity and mortality in many parts of the world. It is associated with severe maternal and neonatal outcomes. This study investigated antibacterial resistance of group B streptococci infection in pregnant women attending St John's hospital, Owerri. A cross-sectional research was conducted for this study. A total of 50 pregnant women with a gestational period of ≥36 weeks were included. A structured questionnaire was used to collect data on the participants' demographic and clinical history. Vaginal-rectal samples were collected by brushing the lower vagina and rectum with a sterile cotton swab for bacteriological examination. Antibacterial susceptibility test was performed using the Kirby-Bauer disk diffusion method. Data were entered and analyzed using SPSS version 25. The results of this study showed that GBS colonization among pregnant women attending antenatal care was at 13.3% (4/30). The Antibacterial susceptibility test result showed that the majority of the isolates were sensitive to vancomycin (96.6%), chloramphenicol (96.6%), ampicillin (93.1%) azithromycin (89.7%), and penicillin (86.2%). In contrast, the isolates were found to be resistant to ceftriaxone, erythromycin, ciprofloxacin, clindamycin, and tetracycline at 17.2%, 20.7%, 27.6%, 27.6%, and 34.5%, respectively. Multidrug resistance (MDR) was noted in 4 isolates (13.79%). GBS colonization was significantly associated with history of preterm labor (18 hours). The colonization rate of GBS was considerably high among pregnant women attending antenatal care in the present study area. Based on the findings of this study, it is recommended that there should be administration of appropriate intrapartum antibiotic prophylaxis which has the function of reducing the risk of early-onset disease in newborns caused by GBS infection to all women whose vaginal–rectal cultures are positive for GBS.

Keywords: Group B Streptococcus, pregnant women, multidrug resistance, antenatal care, neonatal outcomes

INTRODUCTION

Streptococcus agalactiae, most times called group B streptococci (GBS), is part of the normal flora of gastrointestinal and genital tracts [1]. The bacteria use numerous adhesins and stress response apparatuses and immune evasion schemes for vaginal colonization [2]. The lower intestinal tract is the most likely reservoir of the bacteria with a secondary spread to the genitourinary system. The risk of colonization of GBS is greater among infants who are born from mother with GBS colonization. GBS transmission is vertical, transmitted throughout labor, or in utero through the transmission of the bacteria from the vaginal or the mucosa that is anorectally colonized [3].

During the time of pregnancy, the colonization of GBS could be brief, chronic, or intermittent [4]. Colonization commonly does not show any symptoms. However, at some point in pregnancy, GBS multiplication around the vaginal area can lead to maternal morbidity as likely as neonatal complications [5]. About 10 to 30% of women of childbearing age lift GBS in the recto-vaginal area without showing any symptoms. When untreated, nearly 50–75% of neonates born from a GBS infected mothers could be potentially colonized [6]. In Ethiopia, studies revealed that maternal colonization of GBS ranges from 7.2% to 20.9% [7].

Different factors are associated with GBS colonization. The history of premature rupture of membranes, gastrointestinal GBS colonization, increased maternal age, low vitamin D intake, poor personal hygiene, sexual intercourse, health care occupation, and illiteracy were reported as important associated factors with GBS vaginal colonization [8].

GBS is one of the leading cause of infections among neonates worldwide [9]. In 2017, the World Health Organization (WHO) stated that infection associated with GBS caused about 147,000 infant deaths worldwide, despite the use of intrapartum antibiotic prophylaxis (IAP). With 54% of estimated cases and 65% of stillbirths and infant deaths, Africa had the highest burden [10]. While majority of GBS infections could be detected during labor or delivery, women in their post-delivery period can also be at a greater risk for invasive GBS complications even in the absence of extra risk factors [2]. The early onset of GBS infection may cause severe neurological damages and other serious neonatal outcomes [11].

With regard to the antimicrobial sensitivity profile of GBS, it is regarded as uniformly susceptible to penicillin, the first-line antibiotic for Intrapartum Antibiotics Prophylaxis (IAP) [2]. Macrolides (e.g., erythromycin) and lincosamides (e.g., clindamycin) are used as alternative drugs [2]. However, the global spread of antimicrobial resistance (AMR) has led to increased GBS infections among pregnant women and newborns that are difficult to treat and poses significant health concerns. Increased use of antimicrobials for prophylaxis without proper bacteriological screening is one factor that raises this concern [2]. GBS is becoming resistant to different antibiotics, including macrolides and lincosamides, and recently resistance to penicillin and fluoroquinolone was also reported [12]. A systematic review and meta-analysis conducted in China showed that significant rates of resistance to erythromycin, clindamycin, and tetracycline were observed; 0–86%, 4–84%, and 23–96%, respectively [13]. Another meta-analysis that summarized findings from 21 Africa countries also revealed that antibiotic resistance for GBS is a major concern for the whole continent. Higher antimicrobial resistance was observed against tetracycline at 82.6% and penicillin at 33.6% [14]. This research thus aimed at investigating the antibacterial resistance patterns of group b streptococci infections in pregnant women attending St John's Hospital, Owerri.

MATERIALS AND METHODS

Study Design and Sample Size

An institution-based cross-sectional study was conducted at St John's Hospital, Owerri. Owerri is the capital city of Imo state, Nigeria. The sample size used in this study was 50 pregnant women attending St John's Hospital, Owerri.

Data Collection
A pretested and structured questionnaire was used to collect data on the demographic and clinical history of the study participants. Demographic data such as maternal age, residence, marital status, occupation, and clinical data such as gravidity, prenatal care, urinary tract infection, outcomes of the previous delivery, prolonged rupture of membrane, and gestational age were collected.

Specimen Collection
Vaginal-rectal Swabs were collected during the antenatal care (ANC) follow-up by Lushing the lower vagina and rectum with a sterile cotton swab by trained nurses following universal precautions. The swabs were immediately transported to Imo State University Microbiology Laboratory for bacteriological analysis.

Bacterial Analysis
Samples were inoculated on MacConkey agar broth media supplemented with gentamycin and nalidixic acid and incubated at 37°C for 24 hours, then sub-cultured on blood agar plate at 37°C for 24 hrs. Colony characteristics, Gram-stain, and catalase test were used for presumptive identification. All Gram-positive cocci, beta-hemolytic, and catalase-negative isolates were further identified by Christie-Atkins-Munch- Petersen (CAMP) and bacitracin tests. The CAMP test was used to identify CAMP-positive GBS from other beta-hemolytic Streptococci. *Staphylococcus aureus* was inoculated onto a sheep blood agar plate by making a narrow streak down the center of the plate with a loop. Then, the test organism (GBS) was streaked in a straight-line inoculum at right angles to the *S. aureus* within 2 mm. The plates were incubated at 35°C for 24h. A positive CAMP test was indicated by an "arrowhead" shaped enhanced zone of beta-hemolysis in the area between the two cultures with the arrow point" toward the *S. aureus* streak. No enhanced zone of beta-hemolysis was observed in a CAMP negative reaction. In addition, the bacitracin test was used to differentiate GBS from group A streptococcus which are both beta-hemolytic.

Statistical Analysis
The data obtained was subjected to analysis of variance (AVOVA) test to determine the significant difference at 95% confidence limit.

RESULTS

Demographic Characteristics of the pregnant women
The demographic characteristics and clinical data of the respondents were presented in table 1. The result showed that 53% were between the ages of 18years to 35 years. About 83.3% were Christian while the rest were traditionalists. The clinical data showed that only 4 (13.3%) of the pregnant women were positive for streptococci infection.

Table 1. Demographic Characteristics of the Pregnant women

Variables	Frequency	Percentage (%)
Age		
18-35	16	53.3
36-45	14	46.7
Total	**30**	**100**
Religion		
Christianity	25	83.3
Islam	-	-
Traditional	5	16.7
Total	**30**	**100**
Present Status of Streptococci Infection		
Positive	4	13.3
Negative	26	86.7
Total	**30**	**100**

Antimicrobial sensitivity profile of GBS isolated from pregnant women attending St' John hospital

Table 2 showed that most of the GBS isolates were found to be sensitive to vancomycin and ampicillin. They showed moderate level of resistance to tetracycline 10 (34.5%), clindamycin 8 (27.6%), and ciprofloxacin 8 (27.6%). The resistance of both penicillin and ampicillin was at 13.8% and 6.9% respectively.

Table 2. Antimicrobial sensitivity profile of GBS isolated from pregnant women attending St' John hospital

Antimicrobial	Disk potency (μg)	Sensitive *N* (%)	Intermediate *N* (%)	Resistant *N* (%)
Penicillin G	10	(86.2)	—	(13.8)
Ampicillin	10	(93.1)	—	(6.9)
Clindamycin	2	(62.1)	(10.3)	(27.6)
Erythromycin	15	(79.3)	0	(20.7)
Chloramphenicol	30	(90.6)	0	(3.4)
Ciprofloxacin	5	(72.4)	0	(27.6)
Ceftriaxone	30	(82.8)	—	(17.2)
Vancomycin	30	(96.6)	—	(3.4)
Azithromycin	15	(89.7)	0	(10.3)
Tetracycline	30	(51.7)	(13.8)	(34.5)

Multidrug resistance (MDR) profile of GBS isolates from pregnant women attending St' John Hospital

Multidrug resistance was observed in 4 isolates (13.79%) as presented in Table 3.

Table 3. Multidrug resistance (MDR) profile of GBS isolates from pregnant women attending St' John Hospital

Antibiotic combination	Frequency (%)	Remark
ERY: CD	3 (10.34)	—
P: TE	1 (3.44)	—
CTX: TE	1 (3.44)	—
CD:TE	2 (6.9)	—
CD: CTX	1 (3.44)	—
P: CTX	1 (3.44)	—
ERY: CD: CIP	2 (6.9)	MDR
AMP: TE: CIP	1 (3.44)	MDR
ERY: CD: CIP:TE	1 (3.44)	MDR

*ERY: Erythromycin CD: Clindamycin P: Penicillin TE: Tetracycline CTX: Ceftriaxone CIP: Ciprofloxacin AMP: Ampicillin.

DISCUSSION

The results of the demographic characteristics and clinical data of the respondents showed that 53% were between the ages of 18years to 35 years. About 83.3% were Christian while the rest were traditionalists. The clinical data showed that only 4 (13.3%) of the pregnant women were positive for streptococci infection. Most of the GBS isolates were found to be sensitive to vancomycin and ampicillin. They showed moderate level of resistance to tetracycline 10 (34.5%), clindamycin 8 (27.6%), and ciprofloxacin 8 (27.6%). Penicillin is the first choice for intrapartum antibiotic prophylaxis (IAP) and ampicillin as an alternative [15]. Studies conducted around the world, including reports in Ethiopia showed that these drugs have better action against GBS than other antibiotics [15].

In this study, the resistance to both penicillin and ampicillin was at 13.8% and 6.9%, respectively, which implies that these drugs are still good as a first choice to manage pregnant women colonized with GBS. Clindamycin and erythromycin could also be considered for patients who are allergic to penicillin once AST is performed [16]. The finding of this study was comparable to the studies conducted by [2].

High sensitivity of GBS was observed to chloramphenicol and azithromycin at 96.6% and 89.7% respectively. The finding of this study also concurs with similar studies in Ethiopia [17]. Vancomycin was also one of the commonly utilized drugs for patients who poorly responded for clindamycin and for the cases with a risk of high anaphylaxis. In this study, GBS isolates were found with 96.6% sensitivity to vancomycin which was similar with other studies conducted in Iran, Brazil, Cameron, Egypt, and Ethiopia where GBS isolates were found 100% sensitive [2].

Multidrug resistance was observed in 4 isolates (13.79%). This finding is similar with other studies in Ethiopia that reported 15.8–43.9% multidrug resistance GBS isolates. In general, the high level of antimicrobial resistance reported in our

study might be associated with different factors including over and misuse of drugs in the study area where there is a weak drug regulatory practice and scarce bacteriological surveillance system because of lack of routine AST testing [18]. Most of the antimicrobials listed above are available in the local market and individuals usually use these drugs without physician prescription.

CONCLUSION

This study investigated the antibacterial resistance of group B *Streptococcus* infection in pregnant women attending St John's Hospital, Owerri. The colonization rate of GBS was considerably high among pregnant women attending antenatal care in the present study area. In this study, most GBS isolates were found sensitive to vancomycin, chloramphenicol, ampicillin, azithromycin, and penicillin. However, the resistance to tetracycline, clindamycin, ciprofloxacin, erythromycin, and ceftriaxone was significant. Based on the findings, it is recommended that administration of appropriate intrapartum antibiotic prophylaxis which has the function of reducing the risk of early-onset disease in newborns caused by group B *Streptococcus* (GBS) infection to all women whose vaginal–rectal cultures are positive for GBS.

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