Original Research Article

Group B streptococcus colonization in pregnancy: Prevalence, determinants and antibacterial susceptibility pattern in Sagamu, Nigeria

ABSTRACT

Aims: To establish the prevalence, determinants and the antibiotic susceptibility pattern of Group B streptococcus in pregnant women in Sagamu, Ogun State, Nigeria.
Study design: Prospective cross-sectional study

Place and Duration of Study: The study was carried out at the antenatal clinic at Olabisi Onabanjo University Teaching Hospital, Sagamu, Ogun State, Nigeria, between July 2017 and December 2017.
Methodology: The study involved 184 pregnant women attending antenatal clinic. Lower vaginal and rectal swabs were collected under aseptic condition and immediately sent to the laboratory for processing. The samples were assayed for the presence of group B streptococcus using conventional methods. Information on the socio demographic characteristics and details of delivery were recorded on a data capture sheet.

Results: The prevalence of Group B streptococcus was 27.7%. The odds of Group B streptococcus colonization was significantly higher among women of low parity (≤ 2) and binary logistic regression analysis showed that parity was predictive of Group B streptococcus colonization (OR 3.7; 95% CI = 1.03-13.46; *P*=.045). Younger women (age ≤ 30 years) and women carrying term pregnancies had a non significant trend towards higher odds of Group B streptococcus colonization [(OR= 1.22, 95% CI: 0.6-2.3, *P* = .54) and (OR=1.6, CI: 0.8-3.2; *P* = .15) respectively]. The resistance of group B streptococcus isolates to penicillin and ampicillin was 39.2% and 37.3% respectively.
Conclusion: The group B streptococcus colonization rate in this study is high. Factors such as low parity, young maternal age and term pregnancies are associated with increased odds of colonization. The emergence of resistance to the commonly prescribed antibiotics calls for re-evaluation of the current recommendations regarding the antibiotics prophylaxis.

Keywords: Determinants, Group B streptococcus, Pregnancy, Prevalence, Risk factors

1. INTRODUCTION

Group B streptococcus (GBS) also known as *Streptococcus agalactiae* is a facultative anaerobic Gram positive cocci [1]. The bacteria commonly populate the gastrointestinal tract

and female genital tract, and colonization of these regions is a risk factor for subsequent infection in pregnant women and newborns.

The rate of GBS colonization in the vagina and rectum of pregnant women varies with ethnicity and geographical area. In Nigeria, prevalence values ranging from 4% - 18% have been reported [1-4]. Reports from some other countries have revealed prevalence rates such as 20.9% in Ethiopia [5], 19% in Saudi Arabia [6] and14% in Brazil [7].

GBS colonization of the birth canal during pregnancy has been noted to result in miscarriage, stillbirths, prematurity and neonatal sepsis [1,3]. Maternal GBS infections may be associated with urinary tract infections, chorioamnionitis, endometritis, puerperal sepsis, bacteremia, meningitis and wound infections [1,3]. However, the main clinical interest in this bacterium relates to its ability to cause serious neonatal illness such as pneumonia, meningitis, osteomyelitis and septic arthritis [1,8]. Vertical transmission of GBS from mother to neonate is the most recognized mode of transmission; however horizontal spread in form of nosocomial or community acquisition has also been reported [9,10]. There have also been reports of GBS transmission via breast milk especially for late onset neonatal infection [10].

There is evidence to suggest that intrapartum antibiotic treatment of women colonized with group B streptococcus reduces the incidence of early-onset neonatal GBS infection by up to 80% [11]. Penicillin G and ampicillin are the drugs most commonly recommended for prophylaxis and treatment of GBS [12]. In women with allergy to penicillin, clindamycin, erythromycin and vancomycin are recommended as alternatives [12]. There are two main strategies recommended for GBS chemoprophylaxis in pregnancy: risk-based strategy and screening-based strategy [13]. In the risk-based approach, intrapartum prophylaxis is offered to all women with risk factors for GBS. Such risk factors include: previous delivery of an infant with invasive GBS disease, preterm labor, preterm prelabor rupture of membranes, intrapartum fever (>38°C), ruptured membranes >18 hours prior to delivery. In the screening-based approach, all pregnant women are offered microbiological screening at 35-37 weeks of gestation and culture-positive women are treated. This approach has been found to be more effective at identifying intrapartum GBS colonization than the risk-based approach but necessitates the treatment of more women and is also a more expensive option [14].

The knowledge of the epidemiological situation of GBS in a defined area is crucial in deciding on the need for a screening programme, the strategy to be adopted for chemoprophylaxis and to evaluate the cost-effectiveness of such a strategy. In Olabisi Onabanjo University Teaching Hospital (OOUTH) Sagamu, there is no data on the prevalence of GBS colonization in pregnant women to guide the approach to management of the condition. Hence, this study was designed to provide data on the prevalence, determinants and the antibiotic susceptibility pattern of GBS in pregnant women in Sagamu, Ogun State Nigeria. The determinants of GBS may be used to identify a subset of the general obstetric population that will benefit more from screening.

2. MATERIAL AND METHODS

This was a cross-sectional study carried out at the obstetric unit of Olabisi Onabanjo University Teaching Hospital (OOUTH), Sagamu, Ogun State, Nigeria. The patients who receive care in this hospital are of mixed ethnic and socioeconomic background.

The study participants were pregnant women within the gestational age of 36 and 40 weeks. These women were recruited at the antenatal clinic of the hospital. The sample size (n) for the study was determined using the Leslie-Kish formula [15] for single proportion: $n = Z^2pq/d^2$; where n is the desired sample size; Z is the standard normal deviate corresponding to

95% confidence level set as 1.96; *p* is the prevalence of GBS; *q* = 1-*p*; and *d* is the degree of accuracy desired, set at 0.05. In a previous study carried out in Ile-Ife, the prevalence rate of group B streptococcal colonization in late pregnancy was found to be 11.3% [4]. The sample size *n* = 1.962 x 0.11 x 0.89 / 0.052 = 150. To correct for attrition, 10% of calculated sample size was added to give a minimum sample size of 165. However, a final sample size of 184 was used for the study.

The inclusion criteria were: pregnant women without apparent signs and symptoms of bacterial infections, pregnant women who had not taken antibiotics within two weeks of recruitment, and pregnant women without obvious sign of cervical or vaginal erosion. The exclusion criteria included: pregnant women with diabetes mellitus, pregnant women who had used antibiotics within two weeks of recruitment, and pregnant women who refused to give informed consent. Women who matched the inclusion criteria were approached and given verbal and written explanation of the study and invited to participate. For those willing to participate, a written informed consent was obtained. These women were recruited consecutively until the desired sample size was reached.

Data was collected with the aid of a structured proforma designed based on the study objectives. This proforma was administered to the pregnant women before specimen collection. Information on the socio-demographic data such as age, parity, last menstrual period, estimated gestational age at recruitment, level of education, tribe and religion were recorded. This information was obtained through a review of the subject's antenatal records. Data on labor characteristics such as gestational age at delivery, duration of membrane rupture, use of oxytocin for augmentation during course of labor, mode of delivery, fetal birth weight and APGAR scores were recorded from the delivery records. Information on maternal and neonatal complications, and swab culture results were also recorded. The participants were reassured of the confidentiality of data obtained from them.

2.1 Sample collection

Lower vaginal and rectal swabs were collected from the participants using sterile cotton swabs incorporated with Amies transport medium within a sterile container. The women were placed in the dorsal position and a sterile bivalve speculum was introduced to less than 4cm from the fourchette of the vaginal. The swab stick was inserted and rolled in 360 degree twice to take the sample from the lateral vaginal wall. The sample was immediately placed in the transport medium. Another swab stick was inserted through the anal sphincter and rotated twice to collect the rectal sample, and placed into a separate container of transport medium. The samples were taken to the microbiology laboratory of Olabisi Onabanjo University Teaching Hospital where they were processed.

2.2 Group B Streptococcus culture

The swabs were innoculated in Todd-Hewitts broth supplemented with gentamycin (8microgram/ml) and nalidixic acid (15microgram/ml) and incubated at 5% carbondioxide for 18-24hrs at 35-37°C, after which they were sub-cultured on sheep blood agar and incubated for 18-24 hours.

2.3 Group B Streptococcus identification

Typical GBS colonies were Gram positive, catalase negative cocci. The plates that did not grow were reincubated for another 24hrs. CAMP (Christie, Atkins, Munch, Peterson) test was done for presumptive identification of GBS. All CAMP test positive bacteria were subjected

to latex agglutination test using Group B Streptococcus reagent kit (Oxoid, United Kingdom, Batch code 2113431 REF:-DR0593G), for confirmation of GBS.

2.4 Antibiotics Susceptibility Testing

This was done according to Clinical Laboratory Standards Institute (CLSI) standards. The selected antibiotics include penicillin 10 units, ampicillin 10µg, erythromycin 15µg, clarithromycin 2µg, and ceftriaxone 30µg.

2.5 Follow up

Mothers that tested positive to GBS grouping latex test kit were assumed positive and were immediately contacted and placed on ampicillin capsules 500mg 6hrly for a period of five days according to Centers for Disease Control and Prevention (CDC) revised guidelines 2010 [11].

2.6 Data Analysis

Data analysis was done using IBM-SPSS statistics for windows version 21.0 (IBM Corp., Armonk, NY, USA). Categorical variables were summarized using frequencies and percentages. Continuous variables were summarized using descriptive statistics such as mean and standard variation at 95% confidence interval. The influence of risk factors on GBS colonization was determined by calculating the odds ratio at 95% confidence interval. *P* value < 0.05 was deemed statistically significant.

3. RESULTS AND DISCUSSION

3.1 Results

One hundred and eighty four pregnant women were recruited for the study. Table 1 shows the socio-demographic characteristics of the subjects. The mean age of the subjects was 29.81 years (SD 5.1) and age range of 15-41 years. The modal age group was 30-39yrs, accounting for (51.1%) of subjects. Majority of the subjects, (52.7%) had tertiary education, (41.8%) had up to secondary education while (5.4%) had only primary education. There were more Christians, (75.0%) than Muslims (25.0%) in the study participants. Most of the participants, (85.3%) were of Yoruba ethnicity. The parity of the subjects ranged from 0-4, with mean parity of 1.1(SD 1.1). Majority of study participants, (39.1%) were nulliparous. The mean gestational age at recruitment of subjects was 37.1 weeks (SD 1.1), majority (58.2%) were recruited at term.

Table 1. Sociodemographic characteristics of the study participants

Sociodemographic characteristics	**Frequency**	**Percentage**
Age (years)		
<20	5	2.7

20-29	80	43.5
30-39	94	51.1
≥40	5	2.7
Level of education		
Primary	10	5.4
Secondary	77	41.8
Tertiary	97	52.7
Religion		
Christianity	138	75.0
Islam	46	25.0
Parity		
0	72	39.1
1	53	28.8
2	31	16.8
3	23	12.5
4	5	2.7
Gestational age		
<37weeks	77	41.8
≥ 37 weeks	107	58.2

Table 2 shows the outcome of rectal and vaginal swab tests to determine GBS colonization using the Latex agglutination grouping kit test. Twelve subjects (6.5%) had only their rectal swabs positive for GBS, 23 subjects (12.5%) had only their vaginal swabs positive for GBS while 16 subjects (8.7%) had both vaginal and rectal swabs positive. In all, 51 women out of the total of 184 tested positive for Group B streptococcus giving a prevalence of 27.7%.

Table 2. Swab test results

Variables		**Frequency**	**Percentage**
Positive	Rectal swabs only	12	6.5

	Vaginal swabs only	23	12.5
	Both rectal and vaginal swabs	16	8.7
Negative		133	72.3

Table 3 shows the risk factors for GBS colonization. Younger women (age ≤ 30 years) had a slightly increased odds of GBS colonization when compared to women > 30 years of age (OR= 1.22, 95% CI: 0.6-2.3); this was however not statistically significant (*P* =.*54*). Similarly, women carrying term pregnancies had slightly higher odds of GBS colonization when compared to those who were preterm. This finding was also not statistically significant (OR=1.6, CI: 0.8-3.2; *P* =.15). The odds of GBS colonization was significantly higher among women of low parity (≤ 2) when compared to women of higher parity (OR= 3.7, 95% CI: 1.1-12.8; *P* =.03). After controlling for age, binary logistic regression analysis showed that parity was predictive of GBS colonization (OR= 3.7; 95% CI: 1.03-13.46; *P* =.045). Women with primary level education had reduced odds of GBS colonization when compared to women with post primary education (OR=0.3, 95% CI: 0.0-2.2); this was not statistically significant *P* =.2). Christian women also had reduced odds of GBS colonization when compared to Muslim women (OR=0.8, 95% CI: 0.4-1.7); this was also not statistically significant (*P* =.64).

Table 3. Risk factors for Group B Streptococcus colonization

Variables	GBS Positive n(%)	GBS Negative n(%)	Odds Ratio	95% CI	P value
Age					
≤ 30	29(29.6)	69(70.4)	1.22	0.6-2.3	0.544
>30	22(25.6)	64(74.4)			
Level of education					
Primary	1(10.0)	9(90.0)	0.3	0.0-2.2	0.198
Post primary	50(28.7)	124(71.3)			
Religion					
Christianity	37(26.8)	101(73.2)	0.8	0.4-1.7	0.634
Islam	14(30.4)	32(69.6)			
Parity					

≤ 2	48(30.8)	108(69.2)	3.7	1.1-12.8	0.029*
>2	3(10.7)	25(89.3)			
Gestational age					
≥ 37 weeks	34(31.8)	73(68.2)	1.6	0.8-3.2	0.149
<37 weeks	17(22.1)	60(77.9)			

*statistically significant

Table 4 shows the antimicrobial sensitivity pattern of GBS positive swabs. Erythromycin and ampicillin had the highest sensitivity (66.7%, 62.7% respectively). Penicillin, clarithromycin and ceftriaxone had the least sensitivity to the GBS isolates (60.8%).

Table 4. Antimicrobial sensitivity pattern of GBS positive swabs

Antibiotics	Resistant (%)	Sensitive (%)
Penicillin	20(39.2)	31(60.8)
Ampicillin	19(37.3)	32(62.7)
Erythromycin	17(33.3)	34(66.7)
Clarithromycin	20(39.2)	31(60.8)
Ceftriaxone	20(39.2)	31(60.8)

3.2 Discussion

This study shows the prevalence of GBS colonization in Sagamu to be 27.7%. This prevalence rate is much higher than that the reported prevalence rates in other Nigerian cities such as Uyo (4%), Maiduguri (9.8%), Ile-Ife (11.3%) and Enugu (18%) [1-4]. A likely factor that may have contributed to the disparity in the prevalence values is the sample collection site [4,16]. In Maiduguri and Ile-Ife, only lower vaginal swabs were collected for culture. In this study, both vaginal and rectal samples were taken for culture and the detection rate of GBS was found to be higher in vaginal samples than in rectal samples. Other authors [16] have also reported similar findings regarding detection rates of GBS in these different sites. This study thus suggests that it is preferable to use both anorectal and vaginal samples to improve detection rate of GBS colonization.

Another important factor that may be responsible for the disparity in prevalence rate for GBS is the culture medium used and the technique for GBS identification [4,16]. The Maiduguri study [1] used only blood agar as culture medium while the Ile-Ife study [4] used in addition the streptococcus grouping kit, but not CAMP for GBS identification. In this study, both the CAMP test and streptococcal grouping kit were used for GBS identification. Evidence

suggests that using both GBS identification methods increased the number of GBS positive cases compared to using either of the two [17]. The use of other specialized methods such as Polymerase Chain Reaction (PCR) has also been associated with higher detection rate of GBS [18].

Studies have indicated that the risks of maternal GBS colonization may be influenced by factors such as age, parity, socioeconomic status, geographical location, race and ethnicity. Other factors such as sexual behavior, personal hygiene and diet also affect the risk of GBS colonization [4,19,20]. In this study, women with low parity had significantly higher odds of GBS colonization. Other authors [19] have reported similar findings. Young maternal factors age was associated with a non significant trend towards higher odds of GBS colonization. GBS was isolated more frequently in young women (aged ≤ 30 years) compared to older women. Similar findings were reported by other authors [20,21]. The implications of these findings are unclear but it is possible that younger maternal age and low parity may be proxy indicators for some yet to be identified risk factors. Moreover, after controlling for age, parity was found to be predictive of GBS colonization.

There was also increased rate of GBS colonization at term compared with preterm pregnancies. Other authors have also demonstrated increased GBS carriage with advancing gestational age [4,16]. This finding may indicate a dynamic nature of GBS colonization in pregnant women and will have implication for timing of screening. Evidence suggests that the results of screening done more than five weeks prior to delivery are less predictive of carrier status at delivery when compared to those done later [22]. Proper timing of screening should be an important consideration in centers where the screening-based approach to GBS prophylaxis has been adopted. This will ensure optimum timing of treatment and better outcome for the neonate.

Women who had primary level of education had reduced odds of GBS colonization when compared to those that had post primary education. This finding contrasts reports from other studies which have indicated that women with low educational level have higher odds of GBS colonization due to the likelihood of them having poor personal hygiene compared to women with higher educational level [16]. In this study however, only few women (5.4%) had low educational level; this may have been responsible for the conflicting finding.

The antimicrobial sensitivity pattern in this study showed a high resistance to most commonly administered antibiotics. The resistance to penicillin and ampicillin was 39.2% and 37.3% respectively. This is in keeping with a study done Ile-Ife, Nigeria where a high level of resistance was also observed for penicillin and ampicillin [4]. This finding is disturbing because penicillin and ampicillin are the drugs commonly recommended for GBS prophylaxis [11]. The high level of antibiotic resistance seen in this study could be due to antibiotic abuse and self medication which are prevalent in Nigeria. This study suggests that empirical use of antibiotics will not be effective for GBS prophylaxis and treatment in Sagamu, Nigeria; and justifies the need for routine screening and antimicrobial susceptibility testing prior to treatment.

The limitation of this study is the inability to determine the neonatal transmission rate in GBS positive pregnant women. For ethical reasons, all GBS positive women were treated with suitable antibiotics thus making it impossible to detect the neonatal infection rate in women who were not treated with antibiotics.

4. CONCLUSION

The group B streptococcus colonization rate in this study is high. Low parity, young maternal age and term pregnancies are associated with increased odds of GBS colonization; these factors could be used to identify a subset of the general obstetric population that will benefit more from GBS screening. There is an emerging trend indicating high resistance of group B streptococcus to the commonly recommended antibiotics. This finding indicates the need for reevaluation of the current recommendations regarding the antibiotics for GBS prophylaxis.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

CONSENT

All study participants were given full information on all aspects of the study and then asked to sign an informed consent form. The study participants were assured of the confidentiality of data obtained from them.

ETHICAL APPROVAL

Ethical approval for the study was obtained from the health research ethics committee of Olabisi Onabanjo University Teaching Hospital (Reference Number: OOUTH/HREC/29/2015). The research was performed in accordance with ethical standards laid down in the 1964 Declaration of Helsinki.

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