

# Genital Colonization of Group B Streptococcus at term pregnancy in Calabar, Nigeria

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## Citation

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## Abstract

Two hundred (200) term pregnant females of different age groups and socio-economic status, seen at University of Calabar teaching Hospital, General Hospital and Faith Foundation in Calabar from June 2002 to October 2003, were recruited for this study. The subjects were screened for patterns of genital colonization by Group B Streptococcus (GBS). 18 (9.0%) of the female subjects screened were found to be colonized by GBS. Colonization pattern was as follows: 5 (27.8%) had heavy colonization, 8 (44.4%) had moderate colonization and 5 (27.8%) had light colonization. Heavy GBS colonization was observed more among the primigravidae. While moderate GBS colonization was observed more among the second gravitate. Light GBS colonization was uniformly distributed among all the parity levels studied. GBS colonization was more frequently seen among 83.3% pregnant women between 20 years to 30 years of age. Antimicrobial susceptibility pattern showed that GBS isolates were uniformly susceptible to Ampicillin (100%) and Penicillin G (100%).

## INTRODUCTION

Lancefield group B streptococcus -- *Streptococcus agalactiae* is a gram-positive coccus in chains that when cultured on sheep blood agar forms glistening gray-white colonies with a narrow zone of beta hemolysis. It is an invasive encapsulated organism capable of producing severe disease in immunocompromised patients (1). *S. agalactiae* is now best known as a causative agent of post-ovarian infection and as the most common cause of neonatal sepsis.

The major human reservoir of *S. agalactiae* is the vagina and the perianal regions. Other sites frequently colonized are the oropharynx and the external auditory meatus of neonates. An association has been shown between GBS colonization of the vagina and the maternal cervix in pregnancy and subsequent adverse outcome of the pregnancy such as premature birth (2). The carrier rate in earlier pregnancy is less than at term (3). Studies (2,4) have observed higher GBS colonization in women towards the end of their gestation period. Significantly, higher colony counts of the organism were obtained in primigravidae and second gravidae (5). This finding indicates that neonates born to primigravidae mothers who are likely to carry GBS in high concentrations may be at risk of developing GBS neonatal disease. The present paper records genital colonization of GBS at term pregnancy in Calabar, Nigeria.

## MATERIALS & METHODS

The samples were drawn from females outpatients at 36-40 weeks gestation. These were women of different age groups and social status attending the antenatal clinics in the University Teaching Hospital, Calabar (UCTH), General Hospital, and faith foundation clinic and maternity, Calabar. These hospitals are the three largest hospitals serving a large population of the people in Calabar and its environs.

Two hundred (200) patients were sampled from the three hospitals. The patients' consents were sought and gained by explaining to them the objectives of the study and its potential benefits to the patients. The volunteers were asked to fill a questionnaire, which contained information such as age, gestational age, educational background, occupation, marital status, and parity.

Method for collection and culturing of GBS from pregnant women, which was adapted by the prevention in 1996. without using a speculum, a single swab was used to sweep over the skin from the vaginal introitus to the anus (lower vagina).

One hundred non-pregnant females of childbearing age and of various social status were sampled as controls. The controls were those who were attending gynecological clinics, who showed no signs of obvious discharge, pelvic

infections, urinary, tract infection, and who were not on any antibiotics one month prior to the time of sampling. All swabs were placed in Ames transport medium and transported to the microbiology laboratory of the UCTH, Calabar. Specimens were inoculated onto 5% Neomycin sheep Blood agar plate. The plates were then incubated anaerobically at 37°C for 48–72 hours in an anaerobic jar with a gas pack.

Subcultures were inoculated onto new sheep blood agar plates and incubated as above. Isolate were identified based on the colonial morphology, catalase reaction, and the CAMP test.

Estimation of the density of colonization was performed. Semi quantitative cultural procedure was utilized to standardize inoculation of vaginal swabs on Neomycin sheep blood agar. The standardized plating involved a primary inoculum smear (2 cm in diameter) on Neomycin sheep Blood agar (NSBA), followed by interval of consecutive streaking over sectors on NSBA plates. Growth density score was on a scale of 0-4+:

- 0 = no GBS colonies detectable
- 1+ = ≥ 10 colonies only at primary inoculum smear (sector 1)
- 2+ = numerous colonies at sector 1 and 10 at sector 2.
- 3+ = numerous colonies at sector 1 and 2 10 at sector 3
- 4+ = numerous colonies at sector 1-3 and 10 at sector 4.

Growth scores of 2+, 3+, and 4+ were regarded as light, moderate and heavy colonization respectively while a score of 0 it was usually regarded as light colonization or ignored. The disc diffusion method was used to test the antibiotic susceptibility pattern of the isolates. A loopful of the 24-hour-old isolate was inoculated into 10ml peptone water and incubated at 37°C for 24 hours.

0.2ml of this growth was dropped on sheep blood agar plates using sterile Pasteur pipette and spread on the plate using sterile hockey stick. The multiple commercial antibiotic disc containing Ampicillin (25mc g/e), Penicillin (1.5u), erythromycin )10 mcg/e), Gentamicin (10 mcg/e), Chloramphenicol (10mcg/e), Streptomycin (10 mcg/e), Cloxacillin (5 mcg/e) was incorporated and incubated at 37°C overnight after which the plates were read for zones of inhibition.

**RESULTS**

Eighteen (18) of 200 pregnant women (9.0%) screened were

colonized by *S. agalactae*. Of the 18 (9.0%) pregnant women colonized by GBS 5 (27.8%) were heavily colonized while 8 (44.4%), were moderately colonized. Light GBS colonization in 5 (27.8%) women was also observed. (Table 1). Non –pregnant females sampled showed lower colonization rates (Table 1). The rates of colonization between pregnant and non-pregnant females were significantly different ( $\chi^2=2.82$ ,  $p < 0.01$ ).

**Figure 2**

Table 3: GBS colonization rates in pregnant and non-pregnant Subjects in Calabar (June 2002 – Oct. 2003)

Subject status	No. of subjects	Degree of colonization			Total (%)
		Heavy	Moderate	Light	
Primigravidae	72	4(5.5)	1(1.4)	1(1.4)	6 (8.3)
Second gravidae	57	1(1.8)	5(8.7)	1(1.8)	7 (12.3)
3rd gravidae	24	0(0.0)	2(8.3)	1(4.1)	3 (12.5)
Multigravidae	47	0(0.0)	0(0.0)	2(4.3)	2 (4.3)
Total	200	5(27.8)	8(44.4)	5(27.8)	18(9.0)
Controls (non-pregnant women)	100	2(2.0)	1(1.0)	0(0.0)	3(3.0)

When the degree of GBS colonization was studied in relation to the number of pregnancies, heavy colonization was obtained more (5.5%) among the primi gravidae, moderate GBS colonization (8.7%) was observed more among the second gravidae. Light GBS colonization was uniformly distributed among all the parity levels studied. (Table 2) these differences were however not statistically significant ( $p < 0.05$ ). Overall, 88.9% of women with GBS had three or fewer pregnancies. GBS colonization rates were higher in women aged 20-25 years and 26-30 years, when compared with younger women less than 19 years and older women 31-36 years

**Figure 3**

Table 4: Antibiotic susceptibility pattern of GBS isolates in pregnant subjects in Calabar (June 2002 – Oct. 2003)

Antibiotic	Concentration mcg/ml	Disc Sensitivity	(%)
Ampicillin	25	18	100
Penicillin	1.5µ	18	100
Erythromycin	10	14	77.77
Gentamicin	10	0	0.00
Chloramphenicol	10	12	66.66
Streptomycin	10	0	0.00
Tetracycline	10	10	55.55
Cloxacillin	5	5	27.77

Antimicrobial susceptibility pattern of 18 GBS isolates showed uniform sensitivity to Ampicillin and Penicillin G (Table 4).

{image:3}

**DISCUSSION**

GBS colonization rate of 9% in term pregnancies obtained in

this study is higher than that reported by (2) in Jos (6.6%) and lowest than that reported by (6) in 1980 in Ibadan (17.6%) the reason for the varying results may be attributed to the fact that GBS maternal colonization varies from place to place. Other factors that may have contributed to this variation include socio-economic factors, variation in clinical practices of samples collected and the techniques used for the sampling (6) has also suggested that ethnic and genetic factors might play a role in variation of the rates of infection with GBS. Heavy maternal GBS colonization rate was obtained more among the primigravidae. This finding is in line with the study done by (3) and (5), who found 19% and 20% prevalence respectively. GBS colonization was observed more among the primigravidae and second gravitatas. This was consistent with the findings of (5) who also suggested that this greater GBS colonization often observed among the primigyavidae and the second gravidal had epidemiological implications.

However, the reason for this parity – specific group B streptococcal; susceptibility is not clearly understood. Overall, 88.9% of the women with GBS had three or fewer pregnancies. This finding was consistent with the findings of (7) who found that 74.3% of women with GBS they studied had three or fewer pregnancies.

The degree of colonization has been significantly correlated with the risk of perinatal infection (8). CDC in 1996 also found that mothers who are carriers of GBS have 50% chance of infecting their babies before or during birth. The findings therefore is this study indicates that the infants born to the primigravidae and second gravidae mothers who are carrying GBS in high concentrations may be at risk of developing GBS neonatal disease of the GBS positive cases studied 83.3% were women of 20-30 years of age. This corresponds with age of increasing sexual activity and GBS organisms are known to be sexually transmitted.

The GBS organisms were all susceptible in vitro to Ampicillin and Penicillin G. this is consistent with observations by other investigations (1,2,9). Ampicillin has

been preferred by numerous investigators as a drug of choice because it is safe and has a broader spectrum and antimicrobial activity than penicillin G. Screening for genital colonization of GBS at term, pregnancy is a very important bacteriologic factor for predicting the infant at increased risk of infection. Therefore, all term pregnant mothers should be screened for GBS Colonization and this screening exercise is incorporated tin antenatal care programs in the nations health care delivery.

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