PATTERN OF MATERNAL GROUP B STREPTOCOCCUS (GBS) COLONIZATION IN RELATION TO CD4 COUNT AMONG HIV POSITIVE WOMEN IN JOS

ABSTRACT:

Aim

The aim of this study was to determine the prevalence of GBS colonization among HIV positive and HIV negative pregnant women in relation to CD4 cell counts

Materials and Methodology

This was a hospital based descriptive cross-sectional study of 200 pregnant women (100 HIV positive and 100 HIV negative) and 100 non-pregnant women (50 HIV positive and 50 HIV negative) obtaining health care at the Jos University Teaching Hospital between July 2017 and November 2017. Systematic sampling technique and written informed consent were used in recruiting subjects for this study. High vaginal and anorectal swabs were collected from each subject after filling a structured questionnaire. CD4 cell count was also done for all the HIV positive patients at Aids Prevention Initiative in Nigeria (APIN) of Jos University Teaching Hospital (JUTH). The results from the laboratory analysis of the specimens were computed using SPSS version 21.

Results

A colonization rate of 7.3% was observed in HIV positive patients compare to 5.3% in HIV negative. The different in colonization rate between the two groups was not statistically significant ($X2 = 0.507$; $P = 0.477$) (table 1). In pregnant women living with HIV, colonization rate was 8.0% compare to 5.0% observed in non-pregnant women living with HIV. This however, was not statistically significant (Table 2) ($\chi2 = 0.013$; $P = 0.908$). HIV positive subjects with low CD4 counts (<200cells/µl) were observed to have high colonization

rate (20.0%) than patients with high CD4 counts (≥500 cells/µl). Those with CD4 counts between 200-499 cells/µl had 8.1% colonization rate. These findings, though not statistically significant (table 4) ($\chi 2 = 1.3814$; $P = 0.2399$), the increased in colonization rate in low CD4 cell counts may be due to inability of the patient to mount immune response against the organism.

Conclusion

There was no statistically significant difference in GBS colonization among HIV positive patients. A higher colonization rate was observed in HIV patients among the age group 21-25 years; ager was not significantly risk factor for GBS colonization in HIV patients. CD4 cell counts seem not to play any significant role in GBS colonization rate. Although, it was observed to be higher in patients with low CD4 cell counts; the different was not statistically significant.

INTRODUCTION

Streptococcus agalactiae (Group B *Streptococcus*, GBS) has been documented as the leading infectious agent responsible for neonatal morbidity and mortality[1]. The primary risk factor for newborn disease is colonization of the maternal gastrointestinal tract and vagina by GBS[2]. The newborn acquired the infection by vertical transmission of GBS from a vagina-colonized mother upon rupture of membrane or after the onset of labour. This can lead to life-threatening infections such as sepsis and meningitis[3].

Maternal vaginal colonization by GBS in late pregnancy or at delivery is the main factor associated with both early onset neonatal diseases (EOD) and late onset neonatal diseases (LOD)[4]. GBS colonizes the human gastrointestinal and genital tract of 20-30% of healthy humans. The colonization of the vagina can be transient, intermittent or chronic and serve as potential source of infection to the newborn[5,6].

The colonization in pregnant women may remain asymptomatic or may be associated with spontaneous abortion[7], chorioamnionitis[8], premature rupture of membrane which may result in serious neonatal and maternal morbidity and mortality[9,10]. This link has motivated the recommendation of universal antenatal screening of pregnant women for GBS at 35-37 weeks of gestation and the administration of intrapartum antibiotic prophylaxis (IAP) in patients with colonization result[11]. This strategy has been associated with a significantly decreased in incidence of EOD but has limited impact on the incidence of LOD[12].

Many factors have been established to influence the vaginal colonization by GBS[13]. However, conflicting reports have emerged as to the role of HIV infection as risk factor for GBS colonization. While Cutland and his colleagues in 2012 reported that GBS colonization is lower in HIV infected than HIV uninfected women[14], Gray et al 2011[15] and Shah et al 2011[16] have earlier documented in separate studies that HIV infection is not independently associated with GBS. Nevertheless, the proportion of women colonized with GBS is significantly higher in HIV-infected women with a CD4 cell count higher than 500cells/mm^3 when compared to women with a CD4 cell count lower than 200 cells/mm^3 [14,15]. It has also being demonstrated that GBS colonization is significantly higher in HIV infected women with higher CD4 cell counts than HIV negative women [15]. The increased colonization rate in HIV-positive women with high CD4 cell count might be biased by the presence of other risk factors for GBS colonization like diabetes or obesity[4]. On the other hand, HIV-infected women with low CD4 cell count are known to have increased prevalence of bacterial vaginosis that could compete with GBS and are more likely to take cotrimoxazole prophylaxis resulting in lower GBS carriage rates[17].

MATERIALS AND METHODS:

Study Area

The study was carried out in Jos University Teaching Hospital (JUTH). JUTH is located in Jos the Plateau State capital. The hospital is a tertiary health institution with a 600 beds capacity serving Plateau State and majority of the states in the North-central and part of North-east geopolitical zones of Nigeria. JUTH is also a centre for AIDS Prevention Initiative in Nigeria (APIN) that cater for most people leaving with HIV (PLHIV) from within and the bordering states. The main occupation of the people is farming with majority of them in the city being civil servants and businessmen and women.

Study Population

The study population included HIV positive and HIV negative women attending antenatal clinic at the Jos University Teaching Hospital between July 2017 and November 2017.

Study Design

The study was a hospital based descriptive, cross-sectional study that recruited 300 consenting pregnant and non-pregnant women attending antenatal and gynaecology clinics at the Jos University teaching Hospital.

Ethical Consideration

This study was approved by the research ethical committee of Jos University Teaching Hospital with reference number JUTH/DCS/ADM/127/XIX/6583. Written informed consents were also signed by all subjects before enrollment in the study.

Sample Collection

Anorectal and vaginal swabs were carefully and aseptically collected from 150 HIV positive and 150 HIV negative women using sterile swab sticks by the attending physicians after given them appropriate instructions on how the sample should be collected (CDC, 2010).

Specimen Transport

The collected specimens were immediately inoculated into a selective enrichment broth, Todd - Hewitt broth (Oxoid LTD) supplemented with gentamycin (8µg/ml), nalidixic acid (15µg/ml) and 5% sheep blood to increase the recovery rate of GBS[18,19]. These were transported to the laboratory within three hours of inoculation.

Culture and Incubation

The tubes of inoculated Todd-Hewitt broth were incubated aerobically at 37^{O}C for 18 to 24 hours. After an overnight incubation, the broths were subcultured onto 10% sheep blood agar and chromatic Strepto B agar (Liofilchem, Italy), a selective medium for GBS.

The inoculated 10% sheep blood agar plates were incubated aerobically in 5-10% CO2 (candle extinction jar) at 37^{O}C for 18 to 24 hours, while the inoculated chromatic Strepto B agar plates were incubated aerobically at 37^{O}C for 18 to 24 hours[20]. The *Streptococcus agalactiae* control strain was also inoculated onto 10% sheep blood agar and chromatic Strepto B agar and incubated as stated above respectively.

Identification of GBS Isolates

GBS isolates were identified by their beta haemolytic pattern on 5% sheep blood agar and blue-green colour on chromatic Strepto B agar. The isolates were further subjected to Gram staining, catalase test, and Serogrouping using streptococcal grouping kit (DR0585A OXOID) from Oxoid

Data Analysis

The data obtained from the study were analyzed using Statistical Package for Social Sciences (SPSS) version 21 (IBM SPSS Inc, USA). Proportions were compared using Chi-square with confidence limit (p-value) of < 0.05 considered significant.

RESULTS

Of the 150 HIV positive participants, 100 were pregnant women while 50 were non-pregnant women. The overall GBS colonization among the HIV positive women was 7.3%. The colonization rate of 8.0% was observed in pregnant women with HIV compare to 5.0% non-pregnant women with HIV (Table 1).

In addition, GBS colonization did not appear to be influenced by maternal age in HIV infected women. The study revealed a higher colonization rate of 15.4% among the age group 21-25 years followed by age group 31-35 years. Result from age group 16-20 years was negative as no GBS was isolated among this age group (Table 3) ($\chi 2 = 1.3814$; $P = .24$).

The correlation between GBS colonization and CD4 cell counts was also analyzed. It was observed that CD4 cell counts were not independently associated with GBS colonization. The result showed that women with CD4 cell counts of <200cells/µl were colonized in 20.0%. Those with CD4 counts between 200-499 cells/µl were positive in 8.1%. Based on our findings, the colonization rate was lower in those with CD4 counts ≥500 cells/µl as only 5.1% were positive for GBS (Table 4). This study has demonstrated that though colonization is higher in HIV positive women with low CD4 cell count, the difference is not statistically significant ($\chi 2 = 1.702$; $P = .43$). .

Table 1

Group B Streptococcal carriage rates among HIV positive and HIV negative women in Jos University Teaching Hospital

HIV Status	No. Tested	No. Positive	Percentage positive
Positive	150	11	7.3%
Negative	150	8	5.3%
Total	300	19	6.3%

$\chi 2 = 0.506$ P = .48 df = 1

Table 2

Group B Streptococcal carriage rates among HIV positive and HIV negative women in Jos University Teaching Hospital

	Pregnant women	Non-Pregnant women

HIV Status	No. Tested	No. Positive	No. Tested	No. Positive (%)
Positive	100	8		8%
Negative	100	5		5%
Total	200	13		6.5%

$\chi 2 = 0.013$ P = .91 df = 1

Table 3

Carriage of *Streptococcus agalactiae* among HIV positive women according to maternal age in Jos University teaching Hospital

Age Group (years)	HIV Positive	
	No. Tested	No. Positive (%)
16-20	1	0(0.0)
21-25	13	2(15.4)
26-30	33	1(3.0)
31-35	40	4(10.0)
36-40	41	3(7.3)
≥40	22	1(4.5)
Total	150	11(7.3)

$\chi 2 = 1.3814$ P = .24 df = 5

Table 4

Streptococcus agalactiae colonization in relation to CD4 cells count in HIV positive women in Jos University Teaching Hospital

CD4 Cell Counts (cells/µl)	No. Tested	No. Positive	% Positive
<200	5	1	20.0
200-499	86	7	8.1
≥ 500	59	3	5.1
Total	150	11	7.3

$\chi 2 = 1.702$ P = .43 df = 2

DISCUSSION

This study recruited 150 HIV positive and 150 HIV negative women. The result has shown that HIV infection was not independently associated with GBS colonization among women. This is in contrast to report by Lekala and his colleagues in South Africa that HIV infection is significantly associated with GBS colonization than in HIV negative women[21]. We also discovered that HIV infected pregnant women had a carriage rate of 8.0%, though higher than the rate of 5.0% in HIV negative pregnant women, it was not statistically significant. This means that HIV infection is not independently associated with GBS colonization among pregnant women. This finding is comparable with the report in Malawi where the correlation of GBS and HIV was analyzed but no association was found between HIV seropositive and GBS colonization in pregnancy[15].

This study also revealed that the age group 21-25 years was associated with the higher in HIV infected women colonization rate of GBS followed by the age group 31-35 years, though, this was not statistically significant. This is a sexually active age group and it has been stated that vaginal colonization by GBS is associated with sexual intercourse[22,23]. However, this research did not corellate the importance of sexual intercourse in this age group to GBS colonization. Ezeonu and his colleagues in 2012 in Enugu state of Nigeria also documented that GBS carriage rate was highest in women between age group 21-25 years[24]. This is also similar to the report of Dzowela *et al* (2005) and Lekala *et al* (2015) but in contrast with some other research findings that stated that colonization rate increases with advanced maternal age rather that this age group although no reason was stated[15,25].

It was also observed that the rate was higher in HIV positive patients with low CD4 counts and less frequent in those with CD4 counts ≥ 500 cells/µl though the different was not statistically significant. This is contrary to the report by Gray *et al* (2011) and Shah et al (2011) that GBS carriage rate significantly increased at higher CD4 counts[15,16].

CONCLUSION

In conclusion, this research has demonstrated that HIV infection is not a risk factor associated with GBS colonization. Even though colonization rate was more in HIV positives compare to HIV negative patients,, it was not statistically significant. Also, this study was able to establish that CD4 cells count is not significantly associated with GBS colonization. The rate was higher in patients with low CD4 cell counts with no statistical difference. In addition, colonization rate was observed to be higher in HIV patients among the age group 21-25 years; age was not found to be a contributor to GBS carriage rate.

Consent for publication

All the authors reviewed and gave their consent for this article to be submitted for publication.

Competing of interest

There are no conflicts of interests among the authors.

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