PREVALENCE OF GROUP B STREPTOCOCCUS (GBS) COLONIZATION IN ANTENATAL WOMEN AT KENYATTA NATIONAL HOSPITAL (KNH)

A RESEARCH STUDY

SUBMITTED BY

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DEDICATION

To my lovely wife Huda, my precious sons Fuqurdin and Kamaldin and beautiful daughter Zahida for their love, support and inspiration.

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DECLARATION

I hereby declare that this research in part fulfillment of M.Med degree in Obstetrics and Gynaecology is my original work under the guidance of my supervisors and has not been presented before for a degree course to any other University.

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CERTIFICATE OF SUPERVISION

This is to certify that this research study has been conducted and written by **Dr. Salat Girad Mohamed** under my guidance and supervision, and is submitted with my approval for M.Med in Obstetrics and Gynaecology.

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LIST OF ABBREVIATIONS

AAP	:	American Academy of Paediatrics
ACOG	:	American College of Obstetrics & Gynecology
CDC	:	Centre for Disease Control
FDA	:	Food and Drug Agency
GBS	:	Group B Streptococcus
HB	:	Hemoglobin
HIV	:	Human Immunodeficiency Virus
IAP	:	Intrapartum Antibiotic Prophylaxis
KNH	:	Kenyatta National Hospital
KEMRI	:	Kenya Medical Research Institute
NBU	:	New Born Unit
NCCL	:	National Council of Clinical Laboratory
PCR	:	Polymerase Chain Reaction
PROM	:	Premature rupture of membranes
UON	:	University of Nairobi
USA	:	United States of America
VDRL	:	Venereal Disease Research Laboratory

ABSTRACT

Background Group B streptococcus (GBS; Streptococcus agalactiae) is a gram-positive coccus and approximately 10-30% of women worldwide carry it in their urogenital or lower gastrointestinal tract. It is a leading cause of perinatal morbidity and mortality and a common cause of maternal peripartal infections in some developed countries. In Kenya, however, the spectrum of GBS disease remains a largely under-recognised problem. Prophylactic treatment is easily achieved with penicillin G which is accessible at low cost at Kenyatta National Hospital (KNH) with excellent gains in neonatal and maternal health.

***Objective*:** To determine the prevalence of GBS among antenatal women at KNH and assess associated risk factors.

***Study Deign*:** A cross-sectional descriptive study.

Setting: KNH antenatal clinic.

***Study Population*:** Pregnant women attending the antenatal clinic at KNH.

***Methodology*:** A total of 322 consenting pregnant women ≥35 weeks gestation by dates with no history of antibiotic use within the two weeks prior to the study and no current per vaginal bleeding were recruited into the study. Swabs for culture of group B streptococcus (GBS) were obtained from the lower vagina and the anorectal canal. A second smear was taken from the walls of the lower vagina for microscopic examination of bacterial vaginosis. Antenatal profile data such as HIV, VDRL, and HB were extracted from patients' files. Data entry and analysis was carried out using EPI-Info 3.3 and SPSS Version 13 respectively. A p-value of less than 0.05 was considered statistically significant.

Results

The mean age of the mothers was 28.2 (±5) with a range of 18 to 45 years. Majority of the women were married (90.3%), had at least secondary level of education (81%) and were employed (62.3%). Slightly over 60% were multigravid. About 29% of those who were multi-gravid reported history of still birth. GBS was isolated in 81 (25.2%) of the participants. The anorectal colonization was greater (21.2%) than vaginal colonization (14%), however, isolation of GBS in the vaginal cavity was significantly associated with isolation in the anorectal canal, p= <0.0001. GBS colonization was significantly associated with History of still birth, (p = 0.011). No significant association was found between GBS colonization and age, parity, employment and level of education. Similarly no significant association was found between GBS colonization and HIV status, bacterial vaginosis, and history of other bad pregnancy outcomes such as preterm delivery, early neonatal death, early neonatal sepsis, premature rupture of membranes (PROM) and fever in previous pregnancy.

Conclusions and recommendations

A quarter of the mothers attending antenatal clinic at KNH are GBS colonized a figure that is comparable to findings in other parts of the world. GBS colonization is significantly associated with history of still birth. We therefore advocate for access to GBS screening for high risk mothers in our unit. We recommend a follow up study to determine the impact of GBS colonization on maternal and perinatal morbidity and mortality and the cost effectiveness of universal antenatal screening in our set-up.

INTRODUCTION

Group B streptococcus (GBS; Streptococcus agalactiae) is a gram-positive coccus and although, it has no clinical manifestations, approximately 10-30% of women worldwide carry it in their urogenital or lower gastrointestinal tract *(1, 2)*. It is recognized as a leading cause of perinatal morbidity and mortality and a common cause of maternal peripartal infections in the developed world *(3, 4)*. In Kenya, however, the spectrum of GBS disease is largely under-recognized. GBS is associated with preterm birth *(5)*, a leading cause of neonatal mortality at KNH *(6)*. Maternal intrapartum GBS colonization is a major risk factor for early-onset disease in infants and vertical transmission of GBS from mother to fetus primarily occurs after the onset of labor or membrane rupture (8). Intrauterine fetal infection results from ascending infection from the vagina of a colonized woman who is typically asymptomatic *(8)*. Approximately 50 - 65% of neonates born to colonized mothers are carriers while up-to 2% of these neonates develops invasive GBS disease (7). Before the wide-spread use of intra-partum antibiotic prophylaxis (IAP), the incidence of invasive neonatal GBS disease in the United States (US) ranged from 1.5 to 2 cases per 1,000 live births with case fatality ratios as high as 50% *(8)*.

Black race and low socioeconomic status have repeatedly been identified as risk factors for GBS colonization *(5)*. As the treatment for GBS by both chemoprophylaxis and vaccine becomes more refined more GBS data is critical for effective public health planning. This study therefore seeks to determine the prevalence of GBS colonization among pregnant women attending ANC at KNH and assess associated risk factors.

LITERATURE REVIEW

GBS emerged as the leading infectious cause of neonatal morbidity and mortality in the USA in the 1970s and initial case series reported case-fatality ratios as high as 50 % *(8)*. During the 1980s and early 1990s, early-onset neonatal GBS disease incidence remained fairly constant in the USA ranging between 1.5 to 2 cases per 1,000 live births (8). As prevention implementation increased in the mid 1990s, disease incidence declined by 70% to a rate of 0.5 cases per 1,000 live births *(8)*. In Africa only Zimbabwe has an active research programme on GBS colonization and burden of disease *(9)*.

GBS epidemiology

GBS prevalence among pregnant women worldwide ranges between 10 and 30% *(1, 2)*. GBS isolation rate depends on the culture technique and the number of sites swabbed. In the USA: 10 – 35% of pregnant women are asymptomatic carriers of GBS and approximately 50 - 65% of neonates born to colonized mothers are carriers while up-to 2% of these neonates develops invasive GBS disease (7). In Malawi T Dzowella reported a colonization rate of 16.5% and found a significant correlation between history of bad pregnancy outcomes and GBS colonization *(10)*.

GBS PREVALENCE IN SOME PARTS OF THE WORLD

COUNTRY	AUTHOR/JOURNAL	METHOD OF ISOLATION AND SITES SWABBED	PREVALENCE %
USA	Regan JA,1991 (7)	RECTO-VAGINAL CULTURE	10-35
AUSTRALIA	RCOG AUST & NZ 2007(13)	"	15-25
UK	Easmon CS, 1986 (14)	"	25
ZIMBABWE	Moyo et al (4)	"	30.6
MALAWI	T Dzowella (10)	"	16.5
NIGERIA	Dawodu et al (11)	Vaginal culture	19.5
KENYA	E WERE (12)	Recto-vaginal PCR	30.7

The gastrointestinal tract serves as the natural reservoir for GBS and is the likely source of vaginal colonization (7). Vaginal colonization is unusual in childhood but becomes more common in late adolescence *(15).* Studies in college-aged women found new acquisition was frequent among sexually active women *(16)* and in Trinidad colonization was significantly greater in multigravid than in the primigravid women while recovery of GBS from women older than 24 years of age was slightly higher (36.6%) than from women younger than 24 years (26.9%) *(17).* The prevalence of GBS among black pregnant women both in South Africa and the United states has been shown *(8, 18, 19)* to be higher than in women of other racial groups.

HIV infected women and especially those with low CD4 cell count would be expected to have higher colonization with GBS. However, in one study by P. El Beitune et al found no significant increase in GBS colonization in HIV-1-infected pregnant women and GBS colonization was not associated with their immunological status (20).

Maternal intrapartum GBS colonization is a major risk factor for early-onset disease in infants, and vertical transmission of GBS from mother to fetus primarily occurs after the onset of labor or membrane rupture (8). GBS colonization can be transient, chronic, or intermittent, therefore culture status can vary between pregnancies, and screening during each subsequent pregnancy is advised.

Incidence rates of neonatal sepsis is about 10 – 50 per 1000 live births in developing countries and about 5 times that in developed countries *(21).* At KNH in the general paediatric ward, the Neonatal Mortality Rate is 31.5% and suspected neonatal sepsis accounts for 71% of morbidity (8.4% confirmed by blood cultures) *(22).* GBS was noted to be the second commonest bacterial agent in neonatal meningitis in KNH new born unit (NBU) accounting for 26.7% *(23).* Sequelae of acute meningitis can be severe, ranging from hearing loss to global brain damage, and occur in as many as 30% of survivors *(1).* At the Queen Elizabeth Central Hospital, Blantyre in Malawi the commonest causes of neonatal sepsis were Group B *Streptococcus* (17%) and non-typhoidal *Salmonella* (14%). In-hospital case-fatality rate for neonatal sepsis was high at 48% *(24).*

THE GROUP B STREPTOCOCCI

In 1933, Lancefield reported her classic taxonomic classification of ß-hemolytic streptococci. The most common groups causing human infection are: A (*Streptococcus pyogenes*), B (*S. agalactiae*), and D, which includes enterococci. Groups C and G are occasional causes of infection in humans. GBS are facultative, gram-positive diplococci, with approximately 99% of strains showing ß (complete) hemolysis on blood agar plates. She characterised two polysaccharide antigens from GBS: the group specific polysaccharide common to all strains, and the type specific polysaccharide that distinguishes serotypes. The predominant types causing disease are Ia, Ib, II, III, and more recently, types V (USA) *(25)* and VIII (Japan, Denmark) *(26)* being clinically important.

Virulence factors — GBS produces many extra cellular substances, some of which have a role either in virulence or as protective antigens *(3).* The best characterized are the capsular polysaccharides, which confer serotype specificity to GBS *(27).* The capsule confers virulence to the organism, at least in part, by inhibiting the deposition of complement components on the surface of the organism in the absence of serotype-specific antibody.

Protective immunity is related to antibodies directed at these serotype-specific capsular polysaccharide structures *(28).* Candidate group B streptococcal capsular polysaccharide–protein conjugate vaccines for types Ia, Ib, and III have been developed and have been tested for safety and immunogenicity in healthy adults *(29).* Conjugate vaccines for serotype II and the newly emerged type V also have been developed and have undergone phase 1 clinical trials (30).

MATERNAL DISEASE

GBS causes bacteremia, urinary tract infection, chorioamnionitis, postpartum bacteremia or septicemia, and endometritis. It is the second most common cause of bacteremia in obstetric patients *(31).*

Urinary Tract — GBS is a cause of asymptomatic bacteriuria, cystitis, and pyelonephritis during pregnancy. GBS bacteriuria in any concentration in a pregnant woman is a marker of heavy genital colonization with GBS *(8)* and

bacteriuria and pyelonephritis are associated with preterm labor and premature birth. GBS is isolated in 5 to 29 percent of cases of asymptomatic bacteriuria during pregnancy *(32)*. Treatment is initially with broad spectrum antibiotics but when GBS is isolated as the single causative agent treatment should be narrowed to penicillin G alone.

Chorioamnionitis — Chorioamnionitis, a clinical syndrome characterized by fever, leukocytosis, maternal and fetal tachycardia, and uterine tenderness, is a significant cause of preterm labor and premature birth. There is increasing evidence that subclinical GBS infection also is a cause of preterm delivery *(33, 34)*. Chorioamnionitis affects 0.5 to 2 percent of pregnancies and is associated with poor nutrition, low socioeconomic status, invasive procedures (ie, amniocentesis or cerclage placement), coinfection with pathogens such as *Neisseria gonorrhoeae* and Chlamydia, and heavy colonization with GBS in the second trimester *(35)*. Causative agents include E. coli and anaerobic organisms as well as GBS. Treatment includes broad-spectrum antibiotics and delivery of the infant regardless of gestational age.

Puerperal Sepsis — puerperal sepsis usually occurs within 12 hours of delivery and is characterized by fever, tachycardia, abdominal distention and endomyometritis. It is treated empirically with broad-spectrum antibiotics and supportive care. One large study in the USA of bacteremic obstetric patients found that GBS accounted for 17 percent of bloodstream isolates *(31)*

Postpartum Endometritis — Endometritis is a more common complication of cesarean than vaginal deliveries (up to 30 versus 3 percent) *(35)*. Risk factors for the development of endometritis include internal fetal monitoring, frequent cervical examinations, preterm labor, premature rupture of membranes, low socioeconomic status, infection with N. gonorrhoeae and Chlamydia, and colonization with GBS.

Colonization with GBS significantly increases the risk of developing postpartum endometritis *(36)*. The immediate cause of infection often is polymicrobial however and diagnostic cultures rarely are performed. In studies of endometritis, GBS has been identified as a single pathogen in 2 to

14 percent of cases but is more commonly a component of polymicrobial infections *(37)*. Endometritis is treated with broad-spectrum antibiotics including anaerobic coverage (clindamycin plus gentamicin or cefoxitin alone).

NEONATAL DISEASE

In the neonate, the most severe form of GBS is characterized by a fulminant sepsis, pneumonia, and meningitis with a high rate of fatality and residual damage. Routes of infection include intrauterine fetal infection, ascending infection from the vagina of a colonized woman who is typically asymptomatic; contamination during passage through the birth canal and aspiration of infected amniotic fluid which can also lead to stillbirth *(8)*.

GBS infection in neonates and young infants is classified by age at onset into early- and late-onset infection. Early-onset disease generally presents at or within 12 hours of birth but can occur through day six of life *(34)*. Late-onset disease occurs at 7 to 89 days of age.

Early-onset GBS disease manifests as neonatal bacteraemia, septicaemia, pneumonia, meningitis. The clinical signs usually are apparent in the first 24 hours of life.

Risk factors for early-onset disease Risk factors for early-onset neonatal GBS disease include maternal GBS colonization, prolonged rupture of membranes, preterm delivery, GBS bacteriuria during pregnancy, birth of a previous infant with invasive GBS disease, maternal chorioamnionitis as evidenced by intrapartum fever, young maternal age, African-American race, Hispanic ethnicity, and low levels of antibody to type-specific capsular polysaccharide antigens *(5)*. Although many of these factors occur in concert, multivariate analyses have demonstrated that African-American race, maternal age, and gestational age are independent predictors of early-onset disease risk *(5)*.

Maternal GBS colonization is also a strong risk factor for neonatal GBS disease in the late-onset period *(38)*.Because meningitis can occur in up to one third of late-onset cases, risk of long-term neurological sequelae seems to be higher among survivors of late-onset disease compared with early-onset.

INVESTIGATIONS, DIAGNOSIS & TREATMENT

Evaluation of the ill infant suspected of GBS infection includes a complete blood count (CBC) with differential, blood culture, chest radiograph, and lumbar puncture for cerebrospinal fluid (CSF) cell count, protein and glucose concentration, Gram stain and culture. Isolation of GBS from a normally sterile body site (e.g., blood, CSF, pleural fluid, and bone) confirms the diagnosis of GBS infection.

Initial empiric antibiotic therapy includes broad coverage for organisms known to cause disease in these age groups (GBS and other streptococci, gram-negative enteric organisms, staphylococci, and rarely, *Listeria monocytogenes*) *(3)*. When GBS is identified as the sole causative organism, treatment should be narrowed to penicillin G alone. GBS is susceptible to penicillin G, ampicillin, extended-spectrum penicillins, first- and second-generation cephalosporins, and vancomycin, but penicillin G is the most active agent in vitro (39). Approximately 20 to 30 % of isolates are resistant to erythromycin and 10 to 20 percent to clindamycin, rates that appear to be increasing (40).

PREVENTIVE METHODS

In the early 1980s, clinical trials in the USA demonstrated that administering antibiotics during labour to women at risk of transmitting GBS to their newborns could prevent invasive disease in the first week of life (i.e., early-onset disease) (*8*). As a result of the collaborative efforts of clinicians, researchers, professional organizations, parent advocacy groups, and the public health community in the 1990s, recommendations for intrapartum prophylaxis to prevent perinatal GBS disease were issued in 1996 by the American College of Obstetricians and Gynecologists (ACOG) (*41*) and Centers for Disease Control and Prevention (CDC) (*2*), and in 1997 by the American Academy of Pediatrics (AAP) (*42*).

Those guidelines recommended the use of one of two prevention methods, a risk-based approach or a culture-based screening approach. Providers using the risk-based method identify candidates for intrapartum antibiotic

prophylaxis (IAP) according to the presence of any of the following intrapartum risk factors associated with early-onset disease: delivering at <37 weeks' gestation, having an intrapartum temperature ≥100.4°F (≥38.0°C), or rupture of membranes for ≥18 hours. The screening- based method recommends screening of all pregnant women for vaginal and rectal GBS colonization between 35 and 37 weeks' gestation. Colonized women are then offered intrapartum antibiotics at the time of labour. Under both strategies, women with GBS bacteriuria during their current pregnancy, or who previously gave birth to an infant with early-onset GBS disease are candidates for IAP. However a large, population-based retrospective cohort study of deliveries during 1998-1999 in the USA, demonstrated that routine screening and prophylaxis for carriers prevented more cases of early-onset disease than the risk-based method *(43)*.

In response to this finding, in 2002, CDC, ACOG, and AAP endorsed revised guidelines that discarded the risk-based approach in favour of universal screening of pregnant women for GBS carriage and administering prophylaxis to carriers *(8)*. Because colonization can be intermittent or transient, culture status can vary between pregnancies and colonization early in pregnancy is not predictive of neonatal sepsis (*33*). Culture screening of both the vagina and rectum for GBS late in gestation during prenatal care can detect women who are likely to be colonized with GBS at the time of delivery and are thus at higher risk of perinatal transmission of the organism (*44*). The predictive value of prenatal screening improves with shorter intervals between culture and delivery *(45)* so that prenatal screening at 35–37 weeks of gestation is currently recommended in the USA (IAP should be based upon the 35–37 week culture even if early cultures were obtained.) However, in one recent study, the rate of recurrence for GBS colonization (53%) was noted to be significantly higher when judged against women GBS negative in their index pregnancy (15%) *(46)*. Prior GBS colonization should be considered in the algorithm to treat women with unknown GBS status during term labor.

The Canadian (47) and Australian (13) Obstetricians and Gynecologists societies concur with CDC and recommend universal antenatal screening and IAP for all mothers who are noted to be GBS positive. However, the Royal College of Obstetricians and Gynecologists of the United Kingdom (UK) does not recommend universal screening citing some of the following reasons:

Firstly, the incidence of early-onset GBS disease in the UK is 0.5/1000 births (48), which is similar to that seen in the USA after universal screening and IAP, despite comparable vaginal carriage rates (14). Secondly, there have been no randomized controlled trials (RCTs), comparing antenatal screening, whether bacteriological or risk factor based, with no antenatal screening. No RCTs have compared the different screening strategies and estimates of the efficacy of the screening strategies are based on observational studies. The society however recommends IAP for those mothers with risk factors and if GBS is detected incidentally.

In Kenya, Aga Khan University and Teaching Hospital have largely adopted the UK recommendations but offer IAP only if GBS is detected incidentally (49).

There has been intense interest in tests for rapid identification of group B streptococci. Recently, a polymerase chain reaction (PCR) test reported excellent results, with sensitivity of 97%, specificity of 100%, positive predictive value of 100% and negative predictive value of 98.8% with conventional ano-vaginal cultures as the gold standard. The reported laboratory turnaround time was 40–100 minutes for these PCR methods *(50)*. The US Food and Drug Administration (FDA) recently approved intrapartum rapid PCR test (IDI-Strep B, Infectio Diagnostic, Inc., Quebec, Canada) that has strong sensitivity and specificity (compared with GBS culture methods) raising the possibility that some centres may consider shifting from a late antenatal screening policy to an intrapartum screening policy.

CDC Recommended regimens for Intrapartum Antimicrobial Prophylaxis for Perinatal Prevention of Group B Streptococcal Disease	
Recommended	Penicillin G, 5 million units IV initial dose, then 2.5 million units IV every 4 hours until delivery
Alternative	Ampicillin, 2 g IV initial dose, then 1 g IV every 4 hours or 2 g every 6 hours until delivery
If penicillin allergic[a]	
Patients **not** at high risk for anaphylaxis	Cefazolin, 2 g IV initial dose, then 1 g IV every 8 hours until delivery
Patients at high risk for anaphylaxis and with GBS susceptible to clindamycin and erythromycin	Clindamycin, 900 mg IV every 8 hours until delivery
	OR
	Erythromycin, 500 mg IV every 6 hours until delivery
GBS resistant to clindamycin or erythromycin or susceptibility unknown	Vancomycin, 1 g IV every 12 hours until delivery

GBS = group B streptococcus.

[a]History of penicillin allergy should be assessed to determine whether a high risk for anaphylaxis is present. Penicillin-allergic patients at high risk for anaphylaxis are those who have experienced immediate hypersensitivity to penicillin including a history of penicillin-related anaphylaxis. Other high-risk patients are those with asthma or other diseases that would make anaphylaxis more dangerous or difficult to treat, such as persons being treated with -adrenergic blocking agents.

Penicillin remains the agent of choice for intrapartum antibiotic prophylaxis (8). Ampicillin is an acceptable alternative, but penicillin is preferred because it has a narrower spectrum of antimicrobial activity and may be less likely to select for resistant organisms. GBS isolates with confirmed resistance to penicillin or ampicillin have not been observed to date (*8*). Intravenous administration is the only route of administration recommended for intra-partum chemoprophylaxis to prevent perinatal GBS disease, regardless of the antimicrobial agent used, because of the higher intra-amniotic concentrations achieved with this method (8)

In vitro studies have shown strong activity of some microbicides against GBS, and several clinical trials evaluating the disinfectant chlorhexidine have suggested that application to the birth canal during labour or wiping of the newborn at birth or both can reduce vertical transmission of GBS colonization from mother to newborn *(51).* A trial at a large hospital in Malawi found significant reductions in neonatal morbidity and mortality and maternal postpartum infections in the chlorhexidine arm of the trial compared to no treatment *(52).* However, a Cochrane review of randomized and quasi randomized trials comparing vaginal disinfection with chlorhexidine during labour to no treatment, found that vaginal chlorhexidine resulted in a statistically significant reduction in GBS colonization of neonates, but was not associated with reductions in early onset neonatal GBS disease (53).

STUDY JUSTIFICATION

GBS is recognized as a leading cause of perinatal morbidity and mortality and it is also responsible for significant maternal peripartal disease in some developed countries *(3, 4)*. In Kenya, however, the spectrum of GBS disease is largely under-recognized problem. Approximately 10-30% of pregnant women worldwide are colonized *(1, 2)*. The primary risk factor for early-onset neonatal GBS infection is maternal GBS colonization (3). GBS is associated with preterm birth *(5)* a leading cause of neonatal mortality at KNH *(6)*. Neonatal Mortality Rate (NMR) At KNH general pediatric ward is 31.5% and suspected neonatal sepsis accounts for 71% of morbidity *(22)* while GBS was noted to be the second commonest bacterial agent in neonatal meningitis accounting for 26.7% *(23)*. The prevalence of GBS among black pregnant women both in South Africa and the United states has been shown *(8, 18, 19)* to be higher than in women of other racial groups.

While it is part of routine screening in some parts of the developed world we do not screen routinely for it in our antenatal clinic. Prophylactic treatment is easily achieved with penicillin G which is readily available and accessible at this institution with excellent gains in neonatal and maternal health. However there is paucity of local data to make recommendations. There is the need therefore to establish GBS disease burden in our set-up and asses associated risk factors. The result may be used to advocate for access to GBS screening at KNH and assist in further research.

RESEARCH QUESTION

1. What is the extent of GBS colonization among pregnant women attending ANC at KNH?
2. What factors are associated with GBS colonization?

BROAD OBJECTIVE:

To determine the prevalence of Group B Streptococcus colonization in ante-natal women at Kenyatta National Hospital

SPECIFIC OBJECTIVES:

1. To determine the extent of vaginal and anorectal GBS colonization in antenatal mothers.
2. To describe factors associated with GBS colonization.

MATERIALS AND METHODS

STUDY DESIGN

This was a cross-sectional descriptive study carried out over a period of three months (August to October, 2008).

STUDY AREA

The study was carried out at KNH antenatal care clinic. The hospital is the largest national referral center in the country. The antenatal clinics run every morning from Monday to Thursday. They are conducted by consultants and postgraduate doctors. The clinic primarily serves the peri-urban population of Nairobi. It also serves other neighbouring environs including Kiambu, Thika and Machakos.

STUDY POPULATION

Pregnant women $\geq$ 35 weeks gestation by dates (confirmed by clinical examination) attending ante-natal care clinic at KNH formed our study population. About 60 (both old and new) clients are seen on each clinic day. About 20 % (12) of the mothers were estimated to be $\geq$35 weeks gestation. Clients, both old and new, were recruited on Mondays, Tuesdays, Wednesdays and Thursdays.

INCLUSION CRITERIA

- Gestational age $\geq$35 weeks by dates (confirmed by clinical examination)
- Informed written consent

EXCLUSION CRITERIA

- Gestation < 35 weeks by dates
- Antibiotic treatment within the last two weeks prior to study
- Current history of per vaginal bleeding

SAMPLE SIZE DETERMINATION

The sample size was calculated using formula for cross-sectional studies (Woolson's formula, 1987) as follows:

$$n = \frac{Z^2PQ}{d^2}$$

Where:

n = Desired sample size

Z = the standard normal deviate, usually set at 1.96 which corresponds to 95% Confidence interval.

P= prevalence of GBS colonization in antenatal women.

GBS prevalence among pregnant women worldwide ranges between 10 and 30% *(1, 2)*. In Kenya, at Moi Teaching & Referral Hospital (MTRH) it was noted that 30.7% of antenatal women were colonized with GBS *(12)*.

Therefore taking the higher percentage (30%)

P = 0.3

Q = 1.0 – P = 0.7

d = Precision with which to measure prevalence, set at 0.05 (5%).

Substituting the above in the formulae we get;

$$n = \frac{1.96 \times 1.96 \times 0.3 \times 0.7}{0.05 \times 0.05}$$

n = 322 participants to be recruited in the study.

SAMPLING TECHNIQUE

Sampling was done by consecutive sampling until the sample size was achieved.

SCREENING AND RECRUITMENT

The Principal Investigator with the assistance of a trained study nurse reviewed the files of the antenatal clients as they came to the observation area for blood pressure and weight measurements. Those who satisfied the study criteria were then informed of the study and those willing to participate were considered eligible.

Eligible clients were called one by one into the consulting room; the screening provider went through the information on the consent form regarding relevance of the study, questionnaire, physical examination and specimen collection. Willing clients then signed or thumb printed the consent form. No study procedures were conducted before informed consent was provided by clients.

Clinical Procedures:

Participants were interviewed using a pre-tested coded questionnaire to gather socio-demographic and other relevant information such as history of current pregnancy, antibiotic use within the last two weeks, previous miscarriages and stillbirths. The study physician conducted general physical examination, obstetrical examination, collected study samples and completed the routine ANC visit for that day.

LABORATORY SPECIMENS

1. GENITAL SAMPLES

    a. GBS – lower vaginal and anorectal Dacron swab
    b. Bacterial Vaginosis –lower vaginal Dacron swab smeared on a glass slide.

2. BLOOD – data collected from routine ANC screening from patients' files

    HIV, VDRL, HB

PROCEDURE FOR SPECIMEN COLLECTION

The clients were provided with additional counseling to ensure they were comfortable prior to collection of genital samples.

The principal investigator adorned a pair of sterile latex gloves and in the presence of a female nurse as a chaperone the mother was requested to lie in the semi-lithotomy position. While at the foot of the bed the study physician

examined the external genitalia, and then parted the labia with the left hand to access the vaginal introitus. One sealed sterile Dacron swab was used to swab the lower vagina (without speculum placement) and a second sealed sterile Dacron swab was used to swab the anorectal canal. Both swabs were placed immediately into two separate labelled Stuart's transport media without charcoal (Biotec laboratory ltd, 38 Anson road martesham healths). The transport media maintains GBS viability for up to 4 days at room temperature or under refrigeration) *(8)*.

A second lower vaginal smear was taken from the walls of the vagina and smeared on a clean glass slide, air-dried and sent for microscopic examination of bacterial vaginosis.

Three of our study participants complained of drainage of liquor at the time of recruitment which was confirmed by speculum placement after taking the study specimens. These patients were subsequently admitted to labour ward for further assessment.

Specimen labels indicated participant's serial number. It also clearly identified that specimens were for group B streptococcal culture and microscopic examination of bacterial vaginosis. None of our study participants reported history of penicillin allergy and therefore sensitivity testing for Clindamycin and Erythromycin was not performed. The specimens were transferred within 12 hours to the KEMRI laboratory.

LABORATORY PROCEDURES

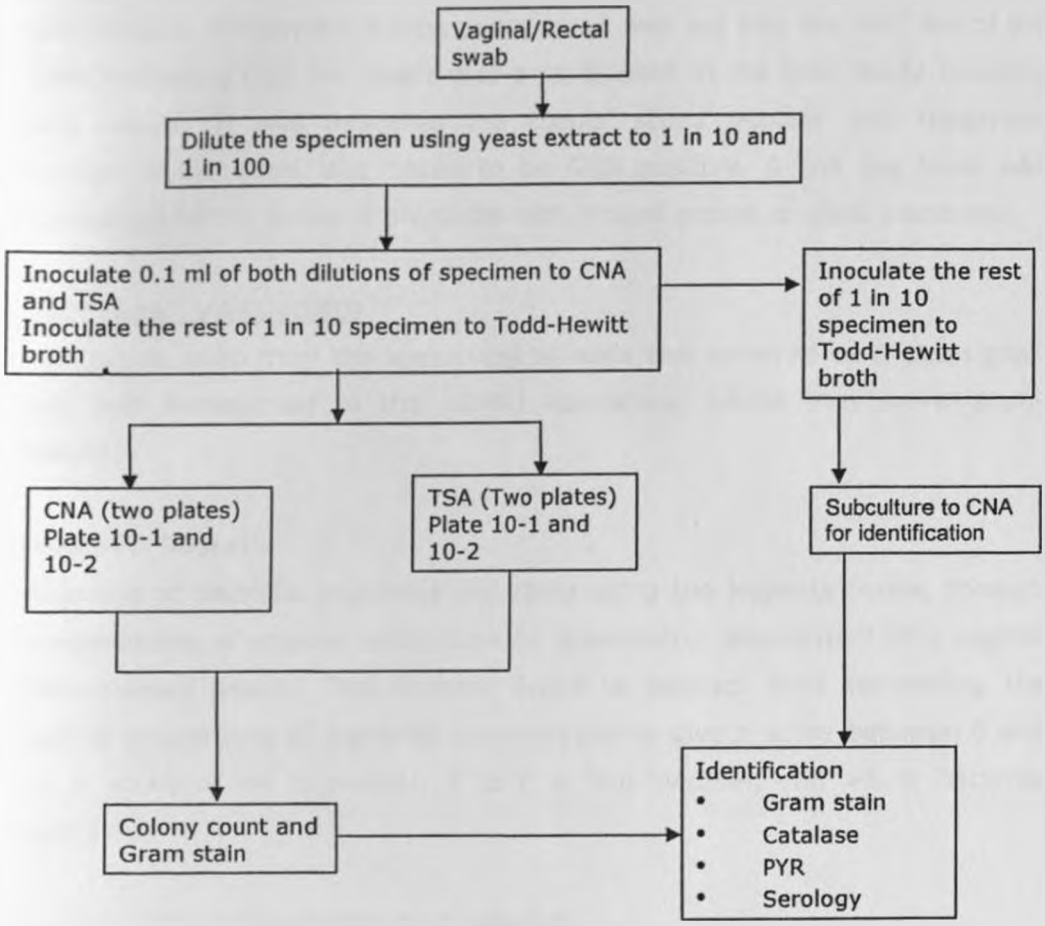
Procedure For Processing Clinical Specimens For Culture Of Group B Streptococcus

The specimens were received and processed at KEMRI microbiology laboratory by two accredited laboratory technologists. The laboratory was initially established in 1998 to undertake microbiological studies related to sexually transmitted infections (STI). It currently offers a wide variety of microbiological cultures including aerobic and anaerobic bacteriology. It is a national reference laboratory with rigorous external and internal quality control.

The specimens were processed as follows:

Rectal and vaginal swabs were separately inoculated in three different media. The inoculation was first done on two solid media (Colistin nalidixic acid Collumbia agar (CNA) with 5% sheep blood and Trpticase Soy Agar (TSA) with 5% sheep blood) after the sample was diluted using yeast extract to enable colony count. The rest of the fluid was inoculated into a selective broth medium (Todd-Hewitt supplemented with gentamycin (8 mg/ml) and nalidixid acid (15 mg/ml)). All the media were incubated at 35–37 °C for 18–24 h under 5% carbon dioxide circumstances. The broth was subcultured onto a blood agar and incubated at 35–37 °C for 18–24 h under 5% carbon dioxide circumstances. All colony types were counted and then Gram stained. A catalase test was performed for all Gram positive cocci. On all catalase negative colonies, a pyrrolidonyl aminopeptidase (PYR) (Key scientific) test was performed. On all PYR negative, a streptococcus grouping latex agglutination test (remel) was performed to identify GBS. Calculation of Colony Forming Units of GBS was done on all positive samples.

A flow-chart showing the procedure for processing clinical specimens for culture of Group B Streptococcus.



All clients were informed of their results during their subsequent ANC visit. To avoid double recruitment a blue paper script was put into the ANC file of the client indicating that the client was a participant in the GBS study awaiting GBS results. It also indicated the clients' study number and treatment regimen if the client was noted to be GBS positive. A link log book was maintained by the nurse or physician with limited access to other personnel.

BACTERIAL VAGINOSIS

The Dacron swab from the lower vaginal walls was smeared on a clean glass slide and transported to the KEMRI laboratory, where they were gram-stained.

Nugent's Score:

Diagnosis of bacterial vaginosis was done using the Nugents Score, through categorization of vaginal micro-flora on quantitative assessment of a vaginal Gram-stained smear. The Nugent Score is derived from estimating the relative proportions of bacterial morphotypes to give a score between 0 and 10. A score of <4 is normal, 4 to 6 is intermediate, and >6 is bacterial vaginosis.

Nugent criteria are defined as follows:

- Grade 1 (Normal): Lactobacillus morphotypes predominate.
- Grade 2 (Intermediate): Mixed flora with some Lactobacilli present, but *Gardnerella* or *Mobiluncus* morphotypes also present.
- Grade 3 (Bacterial Vaginosis): Predominantly *Gardnerella* and/or *Mobiluncus* morphotypes. Few or absent Lactobacilli.

DATA COLLECTION

Data was collected using a coded questionnaire to gather socio-demographic and other relevant history and findings on physical examination. The antenatal profiles such as HIV, VDRL and HB were collected from patients' files while microscopy and culture results of genital specimens were obtained from the laboratory request forms.

DATA MANAGEMENT AND ANALYSIS

Questionnaires were stored under lock and key before and after data entry. The database was restricted from access using a password. During entry data backed up daily on a DVD and kept by the principal investigator. Once data entry was complete, data cleaning was done by checking each data entry against the hard copy forms. Missing values, range checks and inconsistency in the data was done using an automated program run during data entry using EPI-Info and on Statistical Package for Social Scientists (SPSS – Version 13) syntax.

Data analysis was carried out using SPSS Version 13 after exporting the data from EPI-Info.

Prevalence of Group B Streptococcus (GBS) was estimated by calculating the proportion of study participants who are colonized as follows;

$$\text{Prevalence of GBS} = \frac{\text{number of GBS colonized clients}}{\text{Total sample size (n)}} \times 100$$

$$= (81/321) \times 100 = 25.2\%$$

Chi-square test was used to evaluate associations between GBS status and potential risk factors. A p-value of less than 0.5 was considered statistically significant. Kappa was used to assess the agreement in GBS results obtained from vaginal and rectal swabs.

STUDY LIMITATIONS

This was a cross-sectional survey; therefore we were unable to follow-up mothers and their babies to determine the impact of GBS colonization on maternal and perinatal morbidity and mortality. The CD4 cell counts for most of HIV positive mothers were not available at the time of study therefore it was not possible to evaluate the impact of immune-suppression on prevalence of GBS.

We relied on recall information and recall bias could have arisen where participants were asked about their past obstetric and medical history. Similarly clients' prior antibiotic use may not have been disclosed and this could have affected the GBS isolation rate if clients were on antibiotics to

which GBS was sensitive. Although inferences can be drawn, the results of this study may not be generalizable to the general population since data collected was hospital based and participants were from the periurban areas of Nairobi, thus not representative of pregnant women in Kenya.

BENEFIT TO SUBJECT

The clients did not pay for culture of specimens and intrapartum antibiotic prophylaxes were given to those who were noted to be GBS positive.

ETHICAL CONSIDERATIONS

Participation in the study was voluntary and no incentives were given. No client was denied care or their care prejudiced if they declined participation. Clients' were given a written and oral explanation of what the study entailed, its potential benefits and dangers by the principal investigator and research assistants and consent to participate in the study was sought.

A female nurse (chaperon) was present during physical examination and specimen collection. No name identifiers were placed on study materials in order to protect clients' privacy. All clients with positive cultures had their files retrieved, GBS status recorded and instructions on GBS treatment required during labour noted. The results were conveyed to the clients during their subsequent ANC visit by the attending physician. Ethical approval was obtained from the Scientific and Ethical Review Committee at Kenyatta National Hospital and the department of Obstetrics and Gynaecology of the University of Nairobi before commencing the study.

RESULTS

Over a period of three months (August to October 2008), 322 pregnant women ≥35 weeks gestation by date attending ANC at KNH were enrolled in the study and had swabs for culture of GBS taken from the lower vagina and anorectal canal. One participant had GBS culture specimen misplaced and therefore was not included in the analysis. The GBS culture results are shown in table 1.

Table 1: GBS culture results for the participants, N=321

A mother was considered positive for GBS colonies when either or both of the swabs (Vaginal or Rectal) grew GBS.

	N=321
GBS isolated from the vagina; count (%)	45 (14%)
GBS isolated from the rectum; count (%)	68 (21.2%)
GBS isolated in both canals; count (%)	32 (10%)
GBS isolated in either of the canals; count (%)	81 (25.2%)

The prevalence of GBS colonization among our study population was 25.2%

Table 2: The socio-demographic characteristics of the study participants, (N=321)

	Total population (N=321)	GBS+ (N=81)	GBS- (N=240)
Age; mean(SD)	28.2(5.0)	27.4(4.7)	28.5(5.1)
Marital Status; count (%)			
Single	31(9.6)	6(7.4)	25(10.4)
Married	290(90.3)	75(92.6)	215(89.6)
Education; count (%)			
Primary	61(19)	19(23.5)	42(17.6)
Secondary	137(42.7)	33(40.7)	103(43.1)
College/University	123(38.3)	29(35.8)	94(39.3)
Occupation; count (%)			
Salaried Job	132(41.1)	33(40.7)	99(41.2)
Self-employed	68(21.2)	21(25.9)	47(19.6)
Housewife	96(29.9)	21(25.9)	75(31.3)
Unemployed	25(7.8)	6(7.5)	19(7.9)

As shown in table 2, the mean age of the study participants was 28.2 (± 5.34) with a range of 18 to 45 years. Majority were married (90.3%), had at least secondary level of education (81%) and were employed (62.3%).

Table 3: Obstetric and Clinical findings among Study Participants

	ALL(N=321)	GBS+ (N=81)	GBS- (N=240)
Parity; count(%)			
Primigravida (pregnancies=0)	121(37.7)	33(40.7)	88(36.7)
Mulitigravida (pregnancies>0)	200(62.3)	48(59.3)	152(63.3)
Previous pregnancies; count (%)*			
History of still births	58(18.0)	21(25.9)	37(15.4)
History of births before 37 weeks	17(5.3)	11(4.6)	6(7.4)
History of rupture of membranes≥18 hrs	15(4.7)	3(3.7)	12(5)
History of early neonatal death	22(6.8)	4(4.9)	18(7.5)
History of early neonatal sepsis	13(4.0)	2(2.5)	11(4.6)
History of fever in previous pregnancy	12(3.7)	1(1.23)	11(4.6)
Ever use of contraceptive; count (%)**	186(57.8)	48(59.3)	138(57.5)
IUCD	17(9.1)	6(12.5)	11(7.97)
Injectab	65(34.9)	19(39.5)	46(33.3)
CONDOMS	10(5.37)	2(4.16)	8(3.3)
OCPs	109(33.9)	28(34.6)	81(33.8)
Norplant	11(3.4)	3(3.7)	8(5.79)
Natural	0(0)	0(0)	0(0)
Current clinical Symptoms; count (%)			
Pain when passing urine	21(6.5)	3(3.7)	18(7.5)
Lower abdominal pain	90 (28.0)	15(18.5)	75(31.2)
Abnormal vaginal discharge	115(35.7)	34(4.2)	80(33.3)
Yellow discharge	34(10.6)	9(11.1)	25(10.4)
Smelly discharge	42(13.0)	12(14.8)	29(12.1)
Discharge stains underwear	80(24.8)	21(25.9)	58(24.2)
Vulval itching	59(18.3)	20(24.7)	39(16.3)
Vaginal discharge increased in volume	31(9.6)	5(6.2)	26(10.8)
Other vaginal discharge***	25(7.7)	8(9.9)	17(7.1)
Rupture of membranes	3(.9)	0(0)	3(1.3)
General examination; count(%)			
Pallor	3(0.9)	1(1.2)	2(0.8)
Jaundice	1(0.3)	0(0)	1(0.4)
Oedema	44(13.7)	13(16.0)	31(12.9)
Obstetrical: Uterine tenderness; count (%)	3(0.9)	0	3(1.25)
Colour of discharge of swab#			
Yellow	40(13.9)	12(17.7)	28(12.8)
Brown	2(0.6)	0(0)	2(.9)
White	177(61.5)	39(57.3)	138(63.0)
Clear	69(24.0)	17(25.0)	51(23.3)

*These proportions were based on 200 women who had data on previous pregnancies.

** The percentages for various contraceptive methods were calculated against the number of participants who reported ever use of contraceptives (186).

*** White, brown,

34 participants had missing data on colour of discharge of swab.

As shown in table 3, slightly over 60% of the participants were multi-gravid and close to 60% had reported a history of ever use of contraceptive. About 29% of the participants reported history of still births, 35% complained of abnormal vaginal discharge and 28% reported lower abdominal pain.

Table 4: Laboratory findings among study participants

		ALL(N=321)	GBS+ (N=81)	GBS- (N=240)
Bacterial Vaginosis				
	Normal	244(76.0)	58(71.6)	186(77.5)
•	Intermediate	42(13.1)	12(14.8)	30(12.5)
	BV	35(10.9)	11(13.6)	24(10.0)
HIV+		22(6.8)	3(3.8)	19(8.1)
HB level				
	<=10 g/dl	26(8.7)	4(5.3)	22(9.9)
	>10 g/dl	274(91.3)	72(94.74)	201(90.1)

As shown in table 4, 10.9% of the participants were positive for bacterial vaginosis, majority had haemoglobin level > 10g/dl (91.3%) and HIV prevalence was 6.8%.

Table 5: Associations between GBS status and Potential Risk factors
A p-value of less than 0.05 was considered statistically significant.

	GBS+ (N=81)	GBS- (N=240)	OR	P-value
Age				
<=24	21(25.9)	52(21.7)	Ref	0.445
>24	60(74.1)	188(78.3)	0.790	
Marital Status; count (%)				
Single	6(7.4)	25(10.4)	Ref	0.428
Married	75(92.6)	215(89.6)	1.453	
Education				
Primary	17(21.5)	42(17.6)	Ref	
Secondary	33(41.8)	103(43.1)	0.708	0.730
College/University	29(36.7)	94(39.33)	0.682	
Parity; count (%)				
Primigravida (pregnancies=0)	33(40.7)	88(36.7)	Ref	0.513
Mulitigravida (pregnancies>0)	48(59.3)	152(63.3)	0.842	
Previous pregnancies; count (%)				
History of still births	21(25.9)	37(15.4)	1.920	0.034
History of births before 37 weeks	11(4.6)	6(7.4)	1.665	0.237
History of rupture of membranes≥18 hrs	3(3.7)	12(5)	0.731	0.769
History of early neonatal death	4(4.9)	18(7.5)	0.641	0.612
History of early neonatal sepsis	2(2.5)	11(4.6)	0.527	0.529
History of fever in previous pregnancy	1(1.23)	11(4.6)	0.260	0.307
Current clinical Symptoms; count (%)				
Pain when passing urine	3(3.7)	18(7.5)	0.474	0.304
Lower abdominal pain	15(18.5)	75(31.2)	0.5	0.032
Abnormal vaginal discharge	34(4.2)	80(33.3)	1.447	0.180
Yellow discharge	9(11.1)	25(10.4)	1.075	0.837
Smelly discharge	12(14.8)	29(12.1)	1.265	0.564
Discharge stains underwear	21(25.9)	58(24.2)	1.098	0.767
Vulval itching	20(24.7)	39(16.3)	1.690	0.099
Vaginal discharge increased in volume	5(6.2)	26(10.8)	0.541	0.279
Other vaginal discharge	8(9.9)	17(7.1)	1.438	0.472
Rupture of membranes	0(0)	3(1.3)	-	0.575
BV: Negative	70(86.4)	216(90)	Ref	
Positive	11(13.6)	24(10)	1.414	0.371
HIV+	3(3.8)	19(8.1)	0.447	0.205

As shown in table 5, history of still birth was significantly associated with GBS colonization (p=0.034). There was no significant association between GBS colonization and age, parity, marital status, level of education, bacterial vaginosis, HIV status and history of other bad pregnancy outcomes such as preterm delivery, early neonatal death, early neonatal sepsis, premature rupture of membranes (PROM) and fever in previous pregnancy.

Table 6: Assessing agreement between GBS results based on vaginal Swabs and Rectal Swabs

		Result based on Rectal Swab		
		Negative	Positive	Total
Result based on Vaginal Swab	Negative	240	36	276
	Positive	13	32	45
	Total	253	68	321

Agreement	Expected Agreement	Kappa	P
84.7%	70.81%	0.4787	<0.0001

As shown in table 6, isolation of GBS in the vaginal cavity was significantly associated with isolation in the anorectal canal, p<0.0001.

DISCUSSION

The prevalence of GBS colonization among antenatal mothers in this study was 25.2% and is comparable to findings in other parts of the world. In the USA, Regan et al reported GBS colonization rate of 10 - 35% *(7)* and in Malawi T, Dzowela reported a colonization rate of 16.5% *(10)* while in a local Kenyan study E. Were et al (using PCR method), found a colonization rate of 30.7% *(12)*. The differences in these prevalence levels can probably be explained by the different populations studied, different gestational ages at culturing, differences in culture site, and in the use of different culture techniques.

Cross-sectional studies in the United States have also found GBS colonization rates to be higher in African-American women compared with Caucasians or Asians, *(2)* and international reports confirm racial or ethnic differences are likely evident after accounting for methodological differences *(54)*

The anorectal GBS carriage rate in the present study was higher (21.2%) than vaginal colonization (14%), however, there was a statistically significant estimated 84.7% agreement between the GBS results obtained based on swabs from the vaginal canal and rectal canal, $p<0.0001$. Similar ratios of GBS isolation rates from both sites have been reported. In one study by Badri *et al.*, rectal and vaginal carriages were 18 and 10% respectively *(55)*. In contrast, a study by Moyo et al in Zimbabwe reported rectal and vaginal GBS carriage rates of 6.3% and 12.6% respectively *(4)*. The gastrointestinal tract serves as the natural reservoir for GBS and its close proximity to the vaginal canal may account for the significant association between GBS coloization in the anorectal and vaginal canal. Despite the high level of agreement, over 15% of the mothers who were GBS colonized would have been missed by swabbing either the vaginal or rectal canal alone. Swabbing both the lower vagina and rectum increases the yield substantially compared with sampling of either alone (55).

In our study there was significant association between history of still birth and GBS colonization, ($p = 0.011$). A recent review identified GBS as a "common cause of stillbirth." *(56)*. It is also known that women with GBS colonization are at an increased risk of GBS colonization in a subsequent pregnancy. In

one recent study, the rate of recurrence for GBS colonization (53%) was noted to be significantly higher when judged against women GBS negative in their index pregnancy (15%) *(46)*. Though there were no documentary evidence to show that GBS infection was responsible for the high still birth rate (29%) reported by our study participants GBS as a probable cause cannot be ignored given the high prevalence of GBS among our antenatal mothers (25.2%).

Our data shows no significant association between GBS colonization and history of other bad pregnancy outcomes such as preterm birth/s, PROM >18 hours, .Early neonatal death, Early neonatal sepsis and fever in previous pregnancy. In contrast, in Malawi, T Dzowela et al reported a significant correlation between history of bad pregnancy outcomes and GBS colonization *(10)*. Possible explanation for this could be the fact that there were no records of previous pregnancy outcomes to verify the reports given by the mothers and there might have been recall bias as well during the interview which resulted in under-reporting of such events.

GBS is a known cause of chorioamnionitis and premature rupture of membranes *(31)* is a risk factor for early onset neonatal GBS sepsis *(8)*. Three (0.9%) of our study participants reported rupture of membranes at the time of interview which was confirmed by speculum placement. However none were GBS positive and the cause of the PROM can probably be attributed to organisms such as E-coli and other anaerobic bacteria.

Reported risk factors for neonatal GBS infection include maternal age younger than 20 years *(57)*. However in the present study there was no significant association between maternal age and GBS colonization. There was also no significant association between parity, marital status, education, employment, and GBS colonization. E. Were et al in a study at Moi Teaching & Referral Hospital Eldoret, Kenya, also found no significant association between socio-demographic and obstetrical characteristics and GBS colonization *(12)*. In contrast in Trinidad colonization was significantly greater in multigravid than in the primigravid women while recovery of GBS from women older than 24 years of age was slightly higher than from women younger than 24 years

(17). Regan et al also found that GBS was less common among women with a higher level of education *(7)*.

The HIV prevalence level among our study population was 6.8% which approximates the Kenya Aids Indicator Survey (KAIS), 2007 which reported a prevalence level of 9.2% among women of child bearing age *(58)*. HIV infected women and especially those with low CD4 cell count would be expected to have higher colonization with GBS. However, in this present study no significant association was found between HIV and GBS colonization. In a similar study, P. El Beitune et al found no significant increase in GBS colonization in HIV-1-infected pregnant women and GBS colonization was not associated with their immunological status (20). The CD4 cell counts results for our HIV positive mothers were not available at the time of undertaking the study and therefore we were not able to assess the association between immuno-suppression and GBS colonization.

About 11% of our study participants were positive for bacterial vaginosis however, there was no significant association between GBS colonization and bacterial vaginosis.

Prematurity has been found to be a significant cause of neonatal morbidity and mortality at KNH *(6)*. It has also been identified as a risk factor for early onset GBS neonatal sepsis *(5)*. However to our knowledge only two maternal carriage studies have been performed in Kenya (12, 59). As the treatment for GBS by both chemoprophylaxis and vaccine becomes more refined more GBS data is critical for effective public health planning.

CONCLUSION

A quarter of the mothers attending antenatal clinic at KNH are GBS colonized and isolation of GBS in one canal appears to be associated with identification in the other – likely due to the anatomical proximity. GBS colonization is significantly associated with history of still birth. No significant associations were found between GBS colonization and infection with HIV and bacterial vaginosis. Similarly no significant associations were found between GBS colonization and maternal age, parity, marital status, education, employment, and history of other bad pregnancy outcomes such as preterm birth/s, PROM >18 hours, Early neonatal death, Early neonatal sepsis and fever in previous pregnancy.

RECOMMENDATIONS

In view of the high prevalence of GBS among our antenatal mothers which is comparable to findings in other parts of the world, we advocate for access to GBS screening for high risk mothers in our unit. However, a follow up study to determine the impact of GBS on maternal and perinatal morbidity and mortality and the cost effectiveness of universal antenatal screening in our set-up is recommended.

Further analysis using the GBS colony count may identify other risk factors hidden by generalizing the group.

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APPENDIX I: PATIENT INFORMATION AND CONSENT FORM

PREVALENCE OF GROUB B STREPTOCOCCUS COLONIZATION IN ANTENATAL WOMEN AT KENYATTA NATIONAL HOSPITAL

INVESTIGATORS

Investigator	Title	Institutions
Dr. Salat G. Mohamed	Principal Investigator	UoN
Prof. S.B.O. Ojwang	Supervisor	UoN
Dr. Nelly F. Mugo	Supervisor	KNH/UoN
Dr. Kudoyi W.	Supervisor	KNH/UoN

Principal investigators contact: 0720-453354

Investigator's statement

We are asking you to be in a research study. The purpose of this consent form is to give you the information you will need to help you decide whether to be in this study. Please read this form carefully. You may ask questions about what we ask you to do, if there is any risk, your benefits, your rights as a volunteer, or anything else about the research or this form that is not clear. When all your questions have been answered, you can decide if you want to be in this study or not. This process is called 'informed consent'.

Background Information

Group B *Streptococcus* (GBS) is a bacterium that may be found in the vagina (birth canal). Women who have GBS in their vagina often do not show any signs or symptoms of disease. This bacterium is not a sexually transmitted infection. One third of all pregnant women have been found to carry these bacteria in their vagina during pregnancy. When present during pregnancy, GBS may increase the chances of a woman giving birth before the expected day of delivery. It also increases the chances of the water breaking before labor and infection of the womb after birth. Half the babies born to women, who have GBS in their vagina during pregnancy, get infection. These infections include pneumonia and blood infections.

Why is this study being done?

This study will help us find out how common GBS is among pregnant women attending clinic at the Kenyatta National Hospital. If we find that GBS is common amongst our pregnant mothers, we will advocate to make it part of routine testing for pregnant women at this hospital.

Study procedure

If you agree to participate, a medical history will be taken and physical examination done. This will entail inquiry about age, history of current pregnancy, previous miscarriages and still births.

In the presence of a female nurse (chaperon) the doctor will collect three swab specimens from you (two from the lower vagina and one from the anorectal region). The swabs for collecting specimens are sterile and non-traumatic.

You will be informed about the result of the test during your next antenatal visit and it will be shared with your primary care physician(s) for intervention where appropriate.

Confidentiality

All information obtained will be strictly confidential and will not be revealed to other persons, other than your primary care physician(s).

The quality of care given to you in this hospital will not be compromised by your refusal to participate in this study. Participation in this study is voluntary (at your own will) and you are free not to participate or to withdraw at anytime. More than 300 pregnant women will take part in this study. One visit will be required for research and the second visit will be for you to get your results. These results will be ready after one week.

Participant's benefits and risks

Benefits.

- I will inform you of your result during your next antenatal clinic visit.
- You will not pay for any laboratory charges
- If we find that you have a positive test for GBS, we will record it in your file so that you are given treatment during delivery

to prevent your baby from getting the infection during the process of birth. This is the treatment advised by the Centers for Disease Control and Prevention (CDC) in the United States of America. The treatment will include: Intravenous injection of Benzyl Penicillin (Penicillin G) 5 million units start as soon as possible after the onset of active labour and 2.5 units every 4 hours until delivery. Alternatively; Intravenous Ampicillin 2 grams start followed by 1 gram every 4 hours. For women who are allergic to penicillin, they will receive Intravenous erythromycin 500mg every 6 hours during labor or intravenous clindamycin 900mg every 8 hours till delivery.

- A separate prescription indicating the drug, dosage, frequency and route of administration will be given to you in case you deliver at another centre.

Risks

- By participating in this study no risks to you or to your baby are anticipated.

This study has been approved by the University of Nairobi Department Of Obstetrics and Gynecology and the Kenyatta National Hospital and Ethics Board.

Signatures

_____

Investigators Signature Date

Subject Statement

I .....of ..... Understand the above (purpose, procedures, risks and benefits of this study) and give my consent to participate in this study.

_____

Subject's Signature or (Thumbprint) Date

APPENDIX II

PREVALENCE OF GROUP B STREPTOCOCCUS COLONIZATION IN ANTENATAL WOMEN AT KENYATTA NATIONAL HOSPITAL

MATERNAL QUESTIONAIRE

Study number __ __ __ Hospital No.__ __ __ __ __ __

Date of interview (day/month/year) ___ ___/___ ___/___ ___

A: SOCIODEMOGRAPHIC DATA

1. Age _____ years

2. What is highest education level you have completed?

1. None
2. Primary
3. Secondary
4. College/University
5. Don't know

3. Marital status

1. Married
2. Single
3. Divorced/Separated
4. Widowed

4. Employment

1. Salaried job
2. Self-employed
3. Housewife
4. Unemployed

B: OBSTETRICS QUESTIONS

5. Estimated gestational age in weeks ☐☐

6. Have you ever used any form of contraception? 1. Yes 2. No

If yes, which one/s did you use?

1. Injectable 3.IUCD 5. Natural
2. Condoms 4.OCP 6. Norplant

7. Number of times pregnant ☐☐

8. Number of Live births ☐☐

9. Number of still births ☐☐

10. Abortions/Ectopic ☐☐

11. Number of premature births less than 37 weeks gestation ☐☐

12. Have you ever had your water break more than 18 hours before the baby was actually born? 1. Yes 2. No 3. N/A

13. Have any of your babies died within the first 7 days of birth?

1. Yes 2. No 3. N/A

14. Have you ever been told that your baby was born with infection after delivery?

1. Yes 2. No 3. N/A

15. Have you had a high temperature during a previous pregnancy?

1. Yes 2. No 3. N/A

C: PRESENT SYMPTOMS:

16. Do you have pain when passing Urine? 1. Yes 2. No
17. Do you have lower abdominal pains? 1. Yes 2. No
    a) When passing urine 1.Yes 2. No
    b) All the time 1. Yes 2. No
18. Do you have abnormal vaginal discharge? 1. Yes 2. No
    Yellow in colour 1. Yes 2. No
    Smelly 1. Yes 2. No
    Stains underwear 1. Yes 2. No
    Vulva itching 1. Yes 2. No
    Increased in volume 1. Yes 2. No
    Others specify _____
19. Have your fore water broken? 1. Yes 2. No
20. Did you use antibiotics within the last two weeks? 1. Yes 2. No
21. Are you allergic to penicillin? 1. Yes 2. No
    3.Don't know

Clinical Examination

22. Pallor 1.Yes 2. No
23. Jaundice 1.Yes 2. No
24. Oedema 1.Yes 2. No
25. Gestation by fundal height ☐☐
26. Uterine tenderness 1.Yes 2. No
27. Colour of discharge of swab
    1. Yellow
    2. Brown
    3. White
    4. Clear
    Others specify _____
28. Antenatal profile as per ANC card: HIV 1.Positive 2. Negative
    Syphilis 1.Positive 2.Negative
    Hb ☐☐.☐

APPENDIX III: LABORATORY FORM

PREVALENCE OF GROUP B STREPTOCOCCUS COLONIZATION IN ANTENATAL WOMEN AT KENYATTA NATIONAL HOSPITAL

Study number__ __ __ Hospital number__ __ __ __ __ __

Date specimen collected__ __/ __ __ /__ __ __ __

Specimen site_____

(Vaginal/Rectal)

Group B Streptococcus status 1. Positive 2. Negative

Sensitivity pattern (if client penicillin allergic):

Erythromycin: 1. Sensitive

2. Not sensitive

Clindamycin: 1. Sensitive

2. Not sensitive

BACTERIAL VAGINOSIS

1. Normal 2. Intermediate 3. Bacterial Vaginosis

NAME OF LAB.TECHNOLOGIST:.....

SIGNATURE:.....

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Ref: KNH-ERC/ 01/ 282

1st April 2008

Dr. Salat G. Mohamed
Dept. of Obs/Gynae
School of Medicine
University of Nairobi

Dear Dr. Mohamed

RESEARCH PROPOSAL: "PREVALENCE O F GROUP B STREPTOCOCCUS COLONIZATION IN ANTENATAL WOMEN AT KENYATTA NATIONAL HOSPITAL" (P351/11/2007)

This is to inform you that the Kenyatta National Hospital Ethics and Research Committee has reviewed and **approved** your above revised research proposal for the period 1st April 2008 – 31st March 2009.

You will be required to request for a renewal of the approval if you intend to continue with the study beyond the deadline given. Clearance for export of biological specimen must also be obtained from KNH-ERC for each batch.

On behalf of the Committee, I wish you fruitful research and look forward to receiving a summary of the research findings upon completion of the study.

This information will form part of database that will be consulted in future when processing related research study so as to minimize chances of study duplication.

Yours sincerely

PROF A N GUANTAI
SECRETARY, KNH-ERC

c.c. Prof. K.M. Bhatt, Chairperson, KNH-ERC
The Deputy Director CS, KNH
The Dean, School of Medicine, UON
The Chairman, Dept. of Obs/Gynae, UON
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