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Article

Group B Streptococci Recto-Vaginal Colonization, Antimicrobial Susceptibility Pattern and Associated factors among Pregnant Women at Selected Health Facilities of Wolaita Sodo Town, Southern Ethiopia

Abera Kumalo Shano [1,*], Biruk Gebre Takele [2], Shimelis Shiferaw Jejaw [1], Wokil Wolde Dana [3] and Tamirayehu Shonde Jagiso [4]

[1] Affiliation 1: Abera Kumalo; Departement of Medical Laboratory Science, College of Health Sciences and Medicine, Wolaita Sodo University, Ethiopia; aberak2000@gmail.com Shimelis Shiferaw; Departement of Medical Laboratory Science, College of Health Sciences and Medicine, Wolaita Sodo University, Ethiopia; shimelis1shiferaw@gmail.com

[2] Affiliation 2: Biruk Gebre; Departement of Medical Laboratory Science, Wolaita Sodo University Comprehensive Specialized Hospital, Ethiopia; birukgebre10@gmail.com

[3] Dr. Wokil Wolde; Departement of Obstetrics & Gynecology, School of Medicine, College of Health Sciences and Medicine, Wolaita Sodo University, Ethiopia; wokilwolde@yahoo.com

[4] Dr. Tamirayehu Shonde, School of Medicine, College of Health Sciences and Medicine, Wolaita Sodo University, Ethiopia; tamirayehushonde4@gmail.com

* Correspondence: Abera Kumalo; Departement of Medical Laboratory Science, College of Health Sciences and Medicine, Wolaita Sodo University, Ethiopia; aberak2000@gmail.com / Mobile Phone: +251911807270.

Abstract: Background: *Streptococcus agalactiae* or *Group B Streptococcal* colonization of the gastrointestinal and genital tracts of pregnant women usually remains asymptomatic; even if it is the critical determinant of infection in neonates and young infants. It causes early and late onset of invasive *Group B Streptococcus (GBS)* disease manifesting as septicemia, meningitis and pneumonia. Now it is recognized as an important cause of maternal and neonatal morbidity and mortality in many parts of the world including Ethiopia where the magnitude of the problem has been little studied. The aim of this study was to assess the prevalence of *GBS* colonization, to identify associated risk factors and antimicrobial susceptibility pattern among pregnant women at selected health facilities of Wolaita Sodo Town, Southern Ethiopia. **Methodology:** A health facility based cross-sectional study design was conducted at WSUCSH & Wolaita Sodo Health Center from June to August, 2022. A total of 279 pregnant women who were in ANC follow up with at 35-37 weeks of gestation were included. For *GBS* isolation, recto-vaginal swabs were inoculated in 1ml Todd-Hewitt broth medium supplemented with 10µg/ml colistin and 15µg/ml nalidixic acid and followed by identification of isolates based on colonial morphology, gram stains, catalase reaction and CAMP tests. Antimicrobial susceptibility testing was performed using modified Kirby–Bauer disk diffusion method. All collected data were organized in Epi info 4.6.0.2, then transfer tabulated using SPSS version 20. Logistic regression analysis was used to see the association between variables. Finally, the p-value < 0.05 were considered statistically significant. **Results:** In present study, 279 pregnant mothers included and their age was between 15 to 38 years with a mean of 26.5 ± 4.5 years. Of all participants, the highest 120 (43.01%) were housewives. The overall carriage rate of *GBS* was 67(24.0%). *GBS* colonization showed a statistically significant association with college and above levels of maternal education [AOR= 6.610, 95% CI (1.724 - 25.349), P=0.01]. High susceptibility of *GBS* isolate was seen to Penicillin G & Chloramphenicol (92.5%) for each, Ampicillin, and Ceftriaxone (89.6%) each, following Vancomycin (74.62%), and Erythromycin (77%). Relatively, *GBS* showed high resistance to Tetracycline (88 %). **Conclusion:** In this study, the overall prevalence of *GBS* colonization was 24.0%. College and above educational level was statistically significant with *GBS* colonization. This study used to give attention to the management of pregnant women by making *GBS* culture one of the routine diagnoses during ANC follow-up and to prevent infection by early detection.

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Keywords: *GBS*; Antibiotic susceptibility pattern; pregnant women; Wolaita Sodo; Ethiopian

1. Introduction

Streptococcus agalactiae (GBS) is typical microbiota of healthy adults' female genital tracts and anal areas, with the gastrointestinal tract acting as a natural reservoir and source of vaginal colonization. Pregnancy-related diseases such as urinary tract infection, bacteremia, chorioamnionitis, postpartum endometritis, preterm labor, preterm rupture of membranes, and perinatal transfer of the organism are all possible outcomes of maternal *GBS colonization. GBS's* ability to rise from the lower genital tract and colonize the upper genital tract has been linked to intrauterine infection [1–4].

Approximately 10–30% of pregnant women are colonized with *GBS* in the vaginal area, and 60% of their babies are infected through the birth canal. The main risk factor for early-onset invasive *GBS* disease is maternal *GBS* colonization in the genitourinary or gastrointestinal tract and transfer to the newborn during labor and delivery. Antibiotic resistance among *GBS* isolates has been a concern due to the extensive use of intrapartum antibiotic prophylaxis to avoid early-onset *GBS* illness [2,3,5]. One of the contributing reasons is asymptomatic *GBS* recto-vaginal colonization in women. Although little is known about its epidemiology and risk in resource-poor nations, it is the most important pathogen of newborn illnesses as it accounts for a considerable part of all mortality.

Group B streptococcus (GBS) infections are major public health problems in humans worldwide. Worldwide mortality is decreasing from 12.7 million in 1990 to 6.3 million in 2013, but continuous effective measures should be made to decrease the mortality of newborns in developing countries [6]. In Africa, the mortality rate is 4 times higher compared to America and Europe. So strategies for the prevention of *GBS* have a crucial role in mortality [6,7].

Group B streptococcus colonization of pregnancy is different; in different study of countries, in the Americas (19.7 %) [7] and Europe (19.0 %) [6]. Reduced by almost 80% in the United States, cases fell from 1.8 cases per 1,000 live births in the early 1990s to 0.23 cases per 1,000 live births in 2015 [2]. In the United States and Europe, *GBS* is the major cause of mortality and morbidity. It can be found in the vaginal microbiota of up to 30% of pregnant women and can be transmitted to the infant via a perinatal transmission [8]. Southeast Asia shows the lowest mean prevalence around (11.1%) [7]. The estimated mean prevalence of *GBS* colonization shows 17.9 % overall and in Africa high rate recorded

GBS has the potential to thrive in a variety of diverse host environments [9]. The most common cause of early-onset newborn sepsis followed by *S. agalactiae* became the main infectious cause of early newborn morbidity and mortality.

In sub-Saharan Africa, the prevalence of Kampala in central Uganda was 3.9% prevalence of *GBS* [10] and Democratic Republic of Congo, women at 35 - 37 weeks of gestation that were enrolled in the study, 24 (23.07%) were found to be *GBS* carriers[1]. In another study of sub-Saharan Africa Province, Sri Lanka *GBS* vaginal colonization in the 100 specimens was 18% (18 vaginal and 0 rectal) in another study 49% [11] (37 vaginal and 27 rectal) from Hospitals in Kenya were positive for *GBS* [12]. Overall *GBS* was identified in 60/292 (20.5%) of participants among the positive isolates from Kenya hospital [12].

The problem is particularly immense in developing countries like Ethiopia that do not have quality microbiological laboratory facilities to isolate pathogens and determine their antimicrobial susceptibility pattern, in addition to the presence of fake drugs in circulation, and misuse of antimicrobials by health care providers, unskilled practitioners, and patients

Effectively use of intrapartum antibiotic prophylaxis (IAP) has direct impact around 80% reductions in early onset *GBS* disease. So a strategy on IAP evaluation for prevention of EOD should be done in developed countries and to decrease burden of GBS disease, development of vaccine or other preventive plan should be considered [13].

There are many studies conducted in different cities of Ethiopia that show a high prevalence of the disease in mothers. However, there is no strategic plan developed to minimize the disease.

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Different studies conducted in Ethiopia indicated Addis Ababa's prevalence of *GBS* colonization among pregnant women was 14.6% [14]. Jimma prevalence of *GBS* colonization among pregnant women was 19.0 % [2]. Gondar prevalence of *GBS* colonization among pregnant women was 7.0% [15]. Nekemte prevalence of *GBS* colonization among pregnant women was 12% [3].

GBS infection is one of the challenging problems; much research has been done in the world this research showed the prevalence of *GBS*, and their antimicrobial pattern has changed from place to place and from time to time. So it needs to update epidemiological data for a given place and time [16]. Therefore, the main aim of this study will be to determine the prevalence of *GBS* bacteria in pregnant women and an antimicrobial susceptibility test in Wolaita Sodo town, Southern Ethiopia. The result of this study may show the currently updated burden of the disease and the importance of the possible findings of the study, we evaluated the prevalence of *GBS* in pregnant mothers in Wolaita Sodo town, Southern Ethiopia. And as provide updated information for responsible bodies to formulate policies, to implement prevention plans by universal screening for *GBS* in ANC units; and also effective use of prophylaxis to prevent early *GBS* infection.

2. Materials and Methods

2.1. Study Area

The study was conducted in Wolaita Sodo town, which is located 327 km from Addis Ababa and 129 km from Hawassa. There are two general public hospitals, one governmental specialized hospital (WSUCSH), and three government health centers. In this study, two health facilities WSUCSH and Wolaita Sodo Health Center [17]

Wolaita Sodo Health Center under SNNPR Health Bureau and gives outpatient service (ANC) follow for adult OPD, pediatric OPD, delivery service, TB patient follow-up, HIV counseling, and screening unit, and Health package service.

WSUCSH is a teaching and referral hospital of Wolaita Sodo University in the Health College sciences which started community service in 2009. It has about 500 beds and also it has more than 300 health workers and it gives service to an average of about 1000,000 patients annually [17].

2.2. Study Design and Period

A health facility-based cross-sectional design was conducted at the ANC clinic of Wolaita Sodo University Comprehensive Specialized Hospital and Wolaita Sodo Health Center from the period of June to August 2022.

2.3. Study Population

All pregnant mothers who attended ANC follow up at WSUCSH and Wolaita Sodo Health Center in sodo town and who were in their 35 to 37 weeks of gestational period.

2.4. Sample size determination

The sample size was calculated by using a single population proportion formula calculation considering the following assumptions.

$$n = \frac{Z\alpha^2 P (1- P)}{d^2}$$

By using a 95% confidence level, the Z value was 1.96, 5% margin of error (d)

The proportion from others =20.9% prevalence of *GBS colonization* among pregnant women previously done Hawassa by Musa Mohammed [18].

P= estimated prevalence rate= (20.9%) α = 0.05 (level of significance)

n= the required sample size no response rate = 10%

$\frac{(1.96)2 (0.209 [1-0.209])}{(0.05)^2}$ =254 + (10% contingency) = 279

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2.5. Sampling Method

The study participants were enrolled by using a systematic sampling technique until a sample size of 279 was achieved. The first participant was selected by the lottery method and by using the formula K=N/n (K= 462/279=2 from Wolaita Sodo Health Center and K=614/279=2.2 WSUCSH) in three months period. Therefore, every two individuals are taken from two data collection institution. Individual participants were selected randomly in every K^{th} interval during the study period.

2.6. Data Collection

The data on socio-demographic variables and other relevant information were collected by using a predesigned and pretested structured questionnaire and by reviewing medical records. The questionnaire was adapted from other similar studies and initially prepared in English and was translated to Amharic and then translated back to English by another translator to check for consistency. Informed consent was obtained from each study participant after explaining the purpose and procedure of the study. The questionnaire was administered by the attending midwives and nurses and pregnant women with gestational age from 35-37 weeks were interviewed.

Specimen Collection

Specimens were collected as per the ACOG committee opinion and American Society for Microbiology (ASM) protocols. A vaginal-rectal swab was sampled from the mother at the point of ANC and labor by trained midwives using a sterile cotton swab. Using an aseptic technique by applying sterile cotton-tipped swabs in separate sterile tubes at the site of the rectum and vagina, the vagina swab from the mucosal secretions of the lower-third part was obtained. Thereafter, the rectum swab was carefully inserted into the anal sphincter and gently rotated to touch the anal crypts. Within 30 minutes using sterile cotton swabs the vaginal swab was taken for samples placed in Amies transport media and was transported to the Microbiology Laboratory of WSUCSH within an hour of collection. Samples were transported in an ice box. All samples were cultured within an hour of arrival in the laboratory following standard bacteriological techniques (5).

2.7. Laboratory Procedures

2.7.1. Culture and Identification of Group B Streptococci

The Todd-Hewitt broth, an enrichment medium for *GBS* and swabs were inoculated in 1ml broth supplemented with 10µg/ml colistin and 15 µg/ml nalidixic acid to prevent contaminant growth and was incubated at 37°C aerobically for 18– 24 h then sub-cultured onto sheep blood agar plates and re-incubated at 37 °C . After 24 h, the culture inspected for growth and all negative culture plates re-incubated for an additional 18–24 h and then re-observed. Plates that show growth were identified by their characteristic appearance and biochemical tests such as catalase and CAMP testing; those with no growth was discarded or reported as negative.

CAMP testing was performed on sheep blood agar plate (SBAP) by streaking of *S.aureus* down the middle of SBAP and the test organism was then streaked perpendicular to the *Staphylococcal* streak. And the streaks did not touch. CAMP factor produced by *S. agalactiae* and β lysine produced by *S. aureus* act synergistically on SBAP to produce enhanced hemolysis. After incubation overnight under candle jar atmospheres, the SBAP was examined for an arrowhead shaped zone of enhanced lysis Christie, Atkins, and Munch-Petersen (CAMP) factors. Those are Gram-positive cocci in gram stain, catalase-negative in Biochemical tests and CAMP positive was identified as *S. agalact*iae [19].

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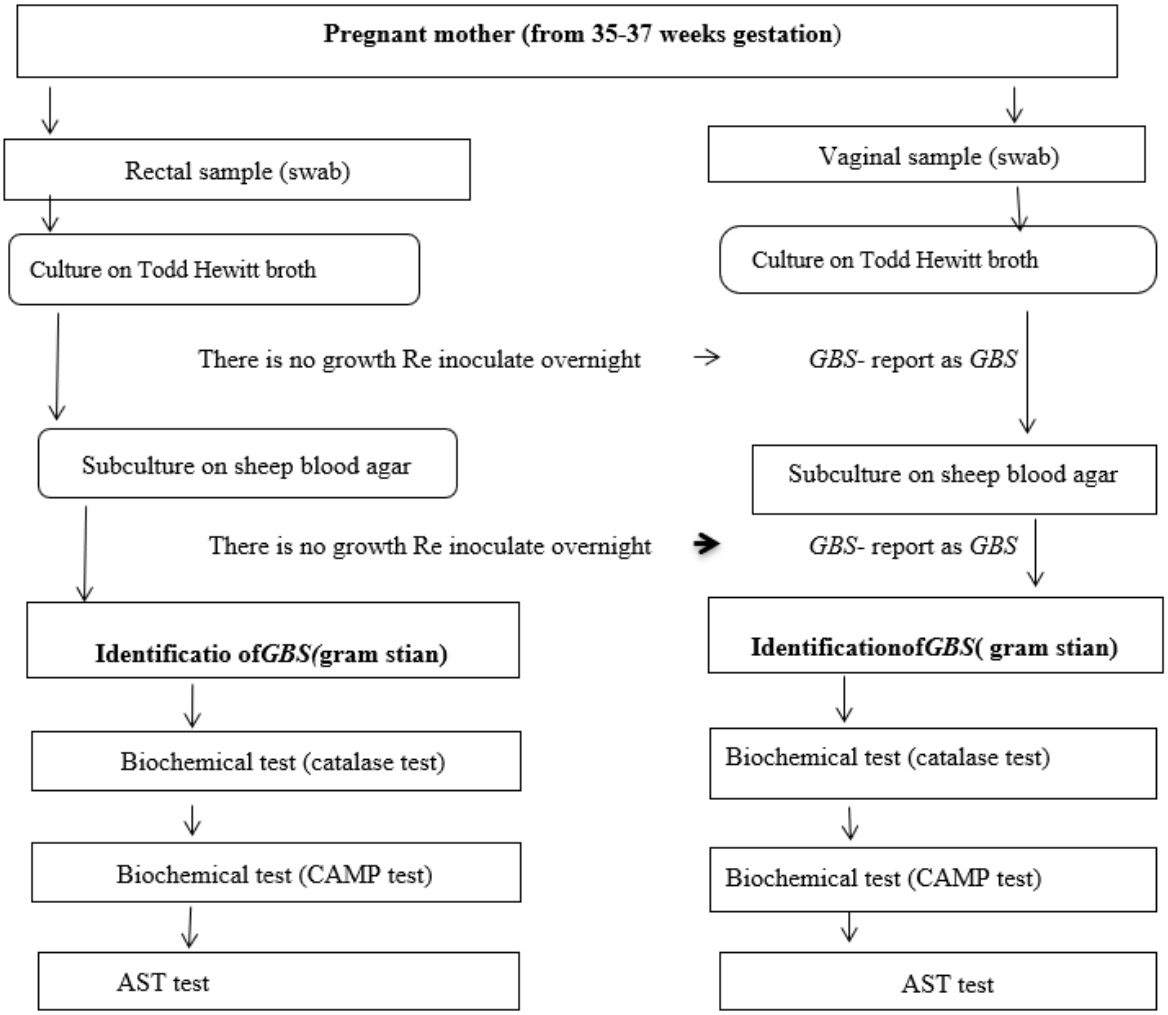

Figure 1. Flow chart diagram showing Culture isolation and laboratory identification of *GBS.*

2.7.2. Antimicrobial Susceptibility Testing (AST)

Kirby Bauer's disc diffusion technique was used to test the Antibiotic susceptibility test (AST). The media used was Muller Hinton agar (MHA) supplemented with 5 % sheep blood. From a fresh non-selective agar plate pure colonies were selected and transferred to 5 mL Sterile normal saline and thoroughly mixed to make the suspension homogeneous Turbidity was adjusted using a McFarland densitometer to match with a 0.5 McFarland Standard, then inoculated following the standard over the entire surface of an MHA plate using a sterile swab. Then using sterile forceps, the antibiotic discs were placed on MHA by considering the 24 mm distance between each disk and 15 mm from the border, zone of Inhibition was measured by the metric scale and reported as susceptible (S), intermediate (I), or resistance (R)**.** Using the updated guideline (CLSI 2021), [20] the following antibiotics disks were used for *Group B streptococcus susceptibility;* Penicillin G 10IU, Ampicillin 10, Erythromycin 15 Clindamycin 2, Ceftriaxone 30, Ciprofloxacin 5, chloramphenicol 30, Clindamycin 2, Vancomycin 30 and Tetracycline 30.

2.8. Data Quality Control

To assure the quality of the data a pre-test was done and 5% of the total sample was out of the study area. The training was given for the data collectors on an interview and recto-vaginal swab sample collection instruction for two days, and on how to clean, sterilize, and reusable laboratory materials for laboratory attendants for two days by the investigator participants was oriented on how to collect recto-vaginal swab samples by trained data collectors. The specimens were transported to

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the WSUCSH Central Laboratory within 30 minutes of collection in a cold chain (ice-box at 4°C) and immediately processed and inoculums density for bacterial suspension for the antimicrobial susceptibility testing was standardized to 0.5 McFarlane Supervision was undertaken during the whole phase of the study period by the investigator and Medical Microbiologist. All culture media was prepared following the manufacturer's instructions. All media was checked for sterility and performance.

Reference Strain *S. aureus* (ATCC-25923) was used as quality control throughout the study for culture, antimicrobial susceptibility testing, and a CAMP test. *E. fecalis* (ATCC -25212) and *S. pyogenes* (ATCC 19615), was used as a negative control for CAMP testing. Was used to check the quality of the culture media and antimicrobial disks, which were obtained from the EPHI Sample, were collected and processed aseptically by using a standard operating procedure

2.9. Methods of Data Analysis

Data were entered, cleaned, and processed into Epi 4.6.0.2, to transfer tabulated using SPSS version 20, Logistic regression analysis was used to see the association between variables finally the 'P' value was less than 0.05 which was considered statistically significant.

3. Results

3.1. Socio-Demographic Characteristics

A total of two hundred seventy-nine (279) pregnant women (from 35 to 37 weeks of gestation) were enrolled from June to August 2022 with a total response rate of 100%. The age of the study participants was between 15 to 38 years with a mean age with SD of 26.5 (± 4.5) years. Most of the study participants were in age categories from 25-29 years 135 (48.4%). The majority of the study participants were married 249 (89.2%) and the largest proportion of study subjects was urban residents 260 (93.2%). Of all participants, the highest proportion 120 (43.01%) were housewives followed by civil servants 64(22.9%), student 50(17.9%) and merchants 45(16.0%). A socio-demographic characteristic of study participants were summarized in Table 1 below.

Table 1. Socio-demographic characteristics among pregnant women at selected Health facility of Wolaita Sodo Town, Southern Ethiopia from June to August 2022 **(**n=279).

Socio-demographic characteristics	Categories	Frequency	Percentage (%)
Health Institutions			
WSU Comprehensive Specialized Hospital	--	169	60.6%
Sodo Health Center	--	110	39.4%
Age groups	15-19	23	8.2%
	20-24	53	19.0%
	25-29	135	48.8%
	30-34	48	17.2%
	≥ 35	20	7.1%
Residence	Urban	260	93.2%
	Rural	19	6.8%
Educational status	Primary	166	59.5%
	Secondary	55	19.7%
	College& above	58	20.8%
Marital status	Married	249	89.2%
	Divorced	6	2.2
	Single	21	7.5%
	Widowed	3	1.1
Occupational status	Housewife	120	43.01

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	Civil servant	64	22.9
	Student	50	17.9
	Merchant	45	16.0
Monthly income	1300-3000	221	79.2
	3100-5000	35	12.5
	5100-1000	20	20
	≥ 10000	3	1.1

3.2. Obstetric and Clinical Characteristics

Regarding obstetric and clinical characteristics of study participants; 173 (62.0%) were primigravida and the remaining 106 (38%) were multigravida. Of the total of 279 study participants, 51 (18.3%) had a history of abortion and 13(4.7%) women had a history of preterm labor. Among the 279 pregnant women included in the study 58 (20.8%), were at a gestational age of 35, 146 (52.3%) were at the gestational age of 36 and the rest 75 (26.9%) were at 37 weeks of gestational age. History of hormonal contraceptive usage had reported by 223 (79.9%) of the study participants.

Table 2. Obstetric and clinical characteristics among pregnant women at a selected facility of Wolaita Sodo Town, Southern Ethiopia from June to August 2022 **(**n=279).

Variables	Categories	Frequency	Percent (%)
Number of gravidity	primigravida	173	62.0
	Multigravida	106	38
Gestational age (in weeks)	35	58	20.8
	36	146	52.3
	37	75	26.9
History of Contraceptive usage	Yes	223	79.9
	No	56	20.1
History of abortion	Yes	51	18.3
	No	228	81.7
History of preterm labor	Yes	13	4.7
	No	266	95.3
History of Preterm prolonged rupture of membranes	Yes	-----	-----
	No	279	100
Diagnosis of UTI during pregnancy	Yes	33	11.8
	No	246	88.2
Diagnosis of STI during pregnancy	Yes	3	1.1
	No	276	98.9
History of ANC visit	Yes	279	100
	No	0	0
History of any antibiotic use	Yes	0	0
	No	279	100
History of any chronic medical illness	Yes	16	5.7
	No	263	94.3

PROM: Prolonged Rupture of Membrane, ANC: Antenatal care, CMI: Chronic Medical Illness, UTI: Urinary Tract Infection, STI: Sexually Transmitted Infections.

3.3. Group B Streptococcus Colonization

The overall prevalence of *GBS* colonization among pregnant women at 35–37 weeks of gestation was 24% (67/279). The prevalence of *GBS* in the two health institutions was 37 (55.2%) from WSUCSH and 30 (44.8%) from Sodo Health Center respectively.

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3.4. Factors Associated with Maternal Group B Streptococci Colonization

The assessment of the association of the socio-demographic, obstetric, and clinical characteristics with *GBS* colonization's was demonstrated in (Table 3) below. During the study period, a total of 279 mothers were screened for *GBS* colonization and *GBS* was confirmed in 67 (24.0 %) of the study participants. In this study, the highest prevalence of GBS observed in the age group between 25-35 years 32.4/135 (48.4%), housewife 32/120(26.67%), and college and above education status 15/58 (25.9%) based on History of contraceptive usage high rate of *GBS* observed in those with contraceptive usage history 53/223(23.8%) compared with non-users. Out of 67, *GBS*-colonized pregnant women 34(19.7%) were Primigravida and 33 (31.1%) were multigravida. On the other hand, mothers who were at 37 weeks of gestation were 25 (33.33%) %) culture positive for *GBS* and those women with no history of abortion had an 11.5% rate of colonization as compared to those women with a recent history of abortion which were (33.3%). Variables candidate for multivariate logistics regression were selected by considering p<0.25 from the bivariate model. Multivariable logistic regression analysis showed that mothers whose educational status was at the college level and above had a significant association with an increased risk of *GBS* colonization (P=0.01). In this study educational status, gravidity, maternal age and gestational age showed an association with *GBS* colonization in binary logistic regression but not in multi-logistic regression.

Table 3. Bivariate and Multivariate analysis of socio-demographic & obstetric factors among pregnant women at selected health facility for Wolaita Sodo town, Southern Ethiopian from June to August 2022 (n=279).

Characteristics	***GBS***			**COR 95% CI**	**AOR 95% CI**	**P-value**
Variables	**Categories**	**Positive (%)**	**Negative (%)**			
Age group	15-19	6(30.4)	16(69.6)	0.403(0.089,1.835	0.380(0.078,1.861)	0.240
	20-24	13(24.5)	40(75.5)	0.543(0.137,2.153)		0.385
	25-29	32(15.6)	114(84.4)	0.9589(0.258,3.560		0.949
	30-34	12(47.9)	25(52.1)	0.192(0.050,0.741)	0.300(0.072,1.254)	0.099
	>35	4 (15.0)	17(85.0)	1	1	
Educational Status	Primary	49(30.7)	117(69.3)	1	1	
	Secondary	3(3.6)	52(96.4)	0.812(0.413,1.596)		0.545
	College & above	15(25.9)	43(74.1)	6.047(1.642,22.370	6.610(1.724, 25.349)	0.01
Occupational Status	Housewife	32(26.7)	88(73.3)	0.770(0.376,1.578)		0.475
	Civil servant	14(21.9)	50(78.1)	1.276(0.501,3.245)		0.609
	Student	10(20.0)	40(80.0)	0.770(0.317,1.870		0.564
	Merchant	11(24.4)	34(75.6	1	1	
Gravidity	Primigravida	34(19.7)	139(80.3)	1	1	
	Multigravida	33(31.1)	73(68.9)	0.499(0.286,0.871)	1.761(0.941,3.296)	0.077
Gestational age	35	14(24.1)	44(75.9)	1	1	
	36	28(19.2)	118(80.8)	1.433(0.671,3.061)		0.352
	37	25(33.3)	50(66.7)	2.204(1.166,4.164)	1.509 (0.700,3.254)	0.15
Contraception	Yes	53(23.8)	170(76.5)	0.946(0.474,1.890)		0.875
	No	14(25.0)	42(75.0)	1	1	
Abortion	Yes	12(23.5)	39(76.5)	1.184(0.569,2.462)		0.651
	No	55(24.1)	173(75.9)	1		

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Preterm-labor	Yes	5(38.5)	8(61.5)	0.698(0.208,2.345)		0.561
	No	62(23.3)	204(76.7)	1	1	
UTI	Yes	6(18.2)	26(81.8)			
	No	61(24.8)	186(75.2)	0.835(0.345,2.020)		0.689

UTI: Unitary Tract Infection, COD: Crude Odds Ratio, **AOR:** Adjusted Odds Ratio.

3.5. Antimicrobial Susceptibility pattern of GBS Isolates

The susceptibility patterns of *GBS* (n = 67) isolated from pregnant women were tested against nine antimicrobial agents are presented in Table 4. A high susceptibility rate of *GBS* isolate was seen for Penicillin G & Chloramphenicol (92.5%) for each, Ampicillin and Ceftriaxone (89.6%) following, Vancomycin (74.62%) and Erythromycin (77%). Relatively, *GBS* showed high resistance to Tetracycline (88%), Ciprofloxacin (55.22 %), and Clindamycin (23.9%). respectively. Regarding, the antibiogram of *GBS* isolates revealed that Susceptibility to all antibiotics was observed in 2 (2.98%) of *GBS* isolates and one or more antibiotic resistance was observed in 65 (97.01%) tested *GBS* isolates. According to the study, the most active drugs for *GBS* isolates were Penicillin, Chloramphenicol, Ampicillin, and Ceftriaxone with susceptibility results of 92.5%, 92.5%, 89.6%, and 74.62%, respectively. Moreover, 5(7.46%) isolates of *GBS* showed intermediate susceptibility to Erythromycins and Chloramphenicol, Vancomycin 6(8.9%) for each and 2(2.98%) showed for Tetracycline.

Table 4. Antimicrobial susceptibility patterns of *GBS* isolated from pregnant women at selected Health facility of Wolaita Sodo Town, Southern Ethiopia (n=67) .

Antibiotics with disc potency	Susceptible (%)	Intermediate (%)	Resistant (%)
Penicillin G (10 IU)	62(92.5%)	-----	5 (7.46%)
Ampicillin (10µg)	60 (89.6%)	-----	7 (10.4%)
Erythromycin (15µg)	52/ (77%)	5/67 (7.46%)	10/ (14.92%)
Clindamycin (2µg)	51/ (76.11%)	----	16/ (23.88%)
Vancomycin (30µg)	50/ (74.62%)	6 (8.9%)	11 (16.41%)
Ceftriazone (30µg)	60/ (89.6%)	-----	7/ (10.4%)
Ciprofloxacin (5µg)	30/ (44.77%)	-----	37 (55.22%)
Chloramphenicol (30µg)	62/ (92.5%)	5/ (7.5%)	------
Tetracycline (2µg)	6/ (8.9%)	2/ (2.98%)	59/ (88%)

4. Discussion

The overall prevalence of *Group B streptococcus (GBS)* in the present study among pregnant women was 24.0%. Such a result in this study is comparable with studies worldwide ranging from 10-30% in the USA, [7] 6.5-36% in Europe [6] 7.1-16% in Asia [8], and 11.9-31.6% in Africa[16]. This study is also relatively similar to studies conducted in different parts of Ethiopia; 20.9% in Hawassa Health Centers [21], 19% in Jimma Hospital [22], and 14.6% in different health centers in Addis Ababa [14]. The rate of *GBS* colonization in this study is lower than the study conducted in Brazil at 28.4 % [23] and South Africa at 30.9 % [24].

The rate of *GBS* found in this study and some countries of Europe is comparable, for example in Italy two studies done and reported *GBS* rates as 17.9% [23], and 18% [25]. In Switzerland and Poland, positivity rates were 21% [26] and 17.2% [27] respectively, the study done in the Netherlands shows 21% [28]. However, a Lower *GBS* colonization rate was recorded from Istanbul and Elazin in Turkey giving 8% [29] and 8.7% [14], respectively, a study in Northern Greece reported the lowest rate of 6.6% [30].

GBS colonization is an important cause of infection in pregnant women and is associated with adverse outcomes in their newborns; however, there have been limited studies available in Ethiopia [14]. It also has variable prevalence and susceptibility against commonly prescribed drugs in different geographic locations.

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Providing adequate knowledge for pregnant women on *GBS* risk factors plays a crucial role in decreasing the morbidity and mortality related to maternal *GBS* infections The geographical differences, variability in the sample size, and methods employed for *GBS* detection might be possibly explained the disparities.

In this study socio-demography (age, residence, education status, marital status, income, and occupation); Obstetrics and clinical characteristics (gravidity; gestational age; history of Preterm PROM, preterm labor, contraceptive use; history of abortion; UTI pregnancy STI pregnancy and any antibiotic) has no relation to the *GBS* colonization. Similar finding was reported from studies done Italy and reported *GBS* rate as 17.9% [23], Poland positivity rate was 17.2% [27]. But college and the above level were significantly associated with maternal colonization (p=0.01) as the study done in Poland [31] and Bangladesh [27]. Maternal age and, gestational weeks, were identified as risk factors for *GBS* colonization [23,25,27,31] in studies done before but no association was seen in the current study Thailand researchers reported that lower maternal age and lower gestational age were risks for colonization by *GBS* [29].

The relationship between these factors and *GBS* colonization however showed marked inconsistencies. In some studies, colonization increased with age reported [26], while other reports confirmed younger age groups showed the highest [14,29]. The possible reason for this difference seems to be seasonal differences globally, the availability of laboratory facilities for detecting *GBS*, and also the shorter study period mentioned.

In this study maternal age, Gravid and gestational weeks, showed association with *GBS* colonization in binary logistic regression, and college and above the educational level on multi-logistic regression to show.

The susceptibility pattern of *GBS* isolates to Penicillin (92.5%), Vancomycin (74.62%), ampicillin (89.6%), Ceftriaxone (89.6%), Chloramphenicol (92.5.%), Erythromycin (77%), Clindamycin (76.11%) and is comparable with previous studies conducted in different countries in which similar records were found from Tanzania [32], USA, (72), Canada [33] and Lebanon [34] in this study.

However high resistance was observed in Tetracycline (88. %), Ciprofloxacin (55.22%), and Clindamycin (23.88%), in which similar records were found from Lebanon [34], USA [35,36], Tanzania [32] Canada [33] and in this study. Erythromycin (14.92%), Vancomycin (16.41%), Ceftriaxone and Ampicillin (10.4%), and, *GBS* resistant with reduced Penicillin susceptibility have been detected. Penicillin is the first agent for the prevention and treatment of *GBS* infections; however, nowadays *GBS* strains with reduced susceptibility to Penicillin have been reported periodically as seen in this study.

To prevent *GBS* Erythromycin and Clindamycin are the alternative antibiotics for Penicillin allergic pregnant women with a high risk of anaphylaxis.

The rising of *GBS* strains resistant to Erythromycin and Clindamycin from time to time is complicating the management of pregnant women who are allergic to Penicillin [30]. In contrast to reports from many other countries, the highest susceptibility in the present study was seen to Erythromycin (77%) and Clindamycin (76.11%) and only a few isolates were resistant to Erythromycin and Clindamycin. In this study, 15% Erythromycin and 24% Clindamycin resistance was reported which is similar to studies done in Ethiopia Gondar showing that 22.7% Erythromycin is resistant and 17.6% to 18.2% resistant to Clindamycin [37]. Also in South Africa, 17.2% Clindamycin and 21.1% Erythromycin resistance were reported [38]. In Tanzania, 17.6% [39] and USA 21.0% [40] of Clindamycin resistance was reported which is comparable to our study.

Generally, in contrast to this study, worldwide studies reported a high resistance rate to Erythromycin which ranges from 18 to 54% [27]. Absence or low antibiotic resistance of *GBS* strains in the present study may indicate the suitability of Penicillin, Ampicillin, and Chloramphenicol for Ethiopia to prevent *GBS* until the vaccine is available on the market.

Similar to the present study low level of resistance to Erythromycin was reported in Australia (6.4%) [41], Brazil (4.1%) [42], Thai-Myanmar border (8.5%) [31], and France (4%) [27]. No resistance to Chloramphenicol is observed in this study and 2 (7.5%) and 3(10.4%) of the isolates showed resistance to Penicillin and Ampicillin respectively.

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High resistance to Tetracycline (88%) in this study was reported and similar reports from other countries, Brazil (83%) [30], Australia (85.9%) [39], Kuwait (89.5%) [36], Canada (89%) [33], and Island (85%) [43] also reported.

CDC approved patients can take Penicillin or Ampicillin if they are not allergic to Penicillin. Clindamycin or Vancomycin is the drug of choice for those who had a major Penicillin allergy and Ceftriaxone for a minor allergy to Penicillin [44]. Because, it is difficult for developing a vaccine for *GBS* due to multiple serotypes are found and vary in geographical location [45]. Nowadays Clindamycin and Erythromycin resistance which are first-line drugs for those with a penicillin allergy, increased rapidly [45].

Resistance to Erythromycin ranged from 7 to 40 % and Clindamycin from 3 to 26.4 % and related to some serotypes [29,40,46]. Inappropriate use of antimicrobial drugs leads to the high resistance of drugs. In Ethiopia, peoples easily go to pharmacy shops without a prescription to buy antibiotics and this type of use of antibiotics might responsible for the high drug resistance rates observed currently.

4.1. The Strengths of This Study

In this studies more valid method used to identify *GBS* colonization, which is culture, presence of THB, primary selective broth media for isolation of *GBS*, 10µg/ml colistin and 15 µg/ml nalidixic, one of the antibiotics which make the primary media to be selective for isolation of the bacteria, makes our isolation adequate to indicate maximum carriage rate.

4.2. Limitations of the Study

- No serotyping was done and using only disc diffusion for antibiotic susceptibility test was conducted.
- Failure to assess the outcome on neonates whose mother detected to be colonized by *GBS* on the study.

5. Conclusions

The prevalence of *GBS* in the current study was 24.0%. Among 279 pregnant women the carriage rate of *GBS* was high among those aged 25-29 years. College and above level of maternal education were significantly associated factors to maternal colonization in the current study [AOR= 6.610, 95% CI (1.724 - 25.349), P=0.01]. The highest susceptibility was shown for penicillin and chloramphenicol each (92.5%). High resistance was observed against Tetracycline (88%), Ciprofloxacin (55.22%). The overall prevalence of *GBS* in the current study is rationally high enough. Therefore, there were a need for screening of pregnant mothers near term delivery and to determine their antibiotic susceptibility so as to set appropriate intervention mechanisms, like early diagnosis, proper management and treatment is very important to reduce *GBS* infection of neonates and newborns.

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Informed Consent Statement: The objective of the study was explained to clients, and written consent was obtained.

Data Availability Statement: All relevant data are within the article, but any additional data required are available from the corresponding author upon request.

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