

Maternal colonization of group B streptococcus: prevalence, associated factors and antimicrobial resistance

Mubashir Ahmad Khan,^a Aftab Faiz,^b Ahmad Mohammad Ashshi^a

From the ^aDepartment of Laboratory Medicine, Umm Al Qura University, Makkah, Saudi Arabia; ^bDepartment of Microbiology, Maternity and Children Hospital, Makkah, Saudi Arabia

Correspondence: Prof. Mubashir Ahmad Khan · Department of Laboratory Medicine, Umm Al Qura University, PO Box 7607, Makkah 21955, Saudi Arabia · T: 966-509-010-825 · mubashirpmrc@yahoo.com

Ann Saudi Med 2015; 35(6): 423-427

DOI: 10.5144/0256-4947.2015.423

BACKGROUND AND OBJECTIVES: Group B streptococcus (GBS, *Streptococcus agalactiae*) can be transferred during delivery to neonates from mothers who are colonized with GBS in the genital tract. GBS can cause sepsis and meningitis in newborns. This study was conducted to determine GBS colonization rates among pregnant women and the antibiotic sensitivity patterns.

DESIGN AND SETTING: Prospective descriptive study at the Maternity and Children Hospital, Makkah.

PATIENTS AND METHODS: Vaginal swabs from 1328 pregnant women (≥ 35 weeks of gestation) attending antenatal clinic were cultured in Todd-Hewitt broth supplemented with gentamicin and nalidixic acid. After 36 hours of incubation, subculture was made onto sheep blood agar and incubated in 5% carbon dioxide for 18 to 24 hours. A Microscan Walk Away system was used for the identification and antibiotic susceptibility of GBS isolates. Each isolate was also tested for group B by using latex slide agglutination test. Information such as maternal age, gestational age and parity was collected using a predesigned questionnaire.

RESULTS: The study population ranged between ages 17-47 years. The GBS colonization in all age groups was found to be 13.4%. A higher colonization rate was seen in pregnant women >40 years of age (27.4%). Women with gestational age >42 weeks were colonized (25%) more frequently than women with a gestational age from 41-42 weeks (20.2%). An increased rate of colonization was found in women who delivered >5 times and no colonization in women who delivered once. All GBS isolates were 100% sensitive to penicillin G, ampicillin and vancomycin. Erythromycin and clindamycin showed resistance—15.7% and 5.1%, respectively.

CONCLUSION: The high prevalence of GBS colonization in pregnant women demands for screening in women attending an antenatal clinic so that intrapartum antimicrobial prophylaxis can be offered to all women who are colonized with GBS, thus preventing its transfer to the newborn.

Group B streptococcus (GBS), also known as *Streptococcus agalactiae* is the normal flora of the female urogenital tract and rectum. Its chief clinical importance is that it can be transferred during delivery to a neonate from their mothers, who are colonized with GBS in the genital tract, and cause sepsis and meningitis in newborns.¹ Group B streptococcus, even when it is asymptomatic, has been associated with adverse pregnancy outcomes such as low birth weight, pre-term delivery, and premature rupture of the membranes.^{2,3}

There is a large geographical variation in GBS colonization rates with about 15% to 40% of pregnant women colonized by GBS in different regions of the

world.³ Variable rates of GBS colonization (9.2% to 31.6%) among pregnant women were also reported from different parts of Saudi Arabia.⁴⁻⁹

Intrapartum antibiotics given to women colonized with GBS reduce the incidence of early onset GBS neonatal sepsis.¹⁰ Various strategies for the use of intrapartum antibiotics (IPA) are used globally, with risk-based IPA administration in the UK¹¹ and a universal culture-based screening program in the US.¹² The introduction of intrapartum antibiotics in the developing world could reduce the number of infants who die from neonatal sepsis each year. However, to determine the likely impact and the optimal strategy of antibiotic administration, GBS colonization prevalence and the antibiotic

susceptibility pattern of clinical isolates in the target population must be known. Although, variable rates of GBS colonization among pregnant women were reported from different parts of Saudi Arabia,⁴⁻⁹ no study has been reported from Makkah, Saudi Arabia. Therefore, the aim of this study was to determine the GBS colonization rate and antimicrobial resistance pattern in the pregnant women in Makkah, Saudi Arabia so that an effective prophylactic regimen can be developed.

PATIENTS AND METHODS

This prospective descriptive study was carried out from September 2013 to December 2014 at the Microbiology Laboratory of the Department of Laboratory Medicine, Faculty of Applied Medical Sciences, Umm Al Qura University, Makkah, Saudi Arabia, after the approval of the institutional bioethical committee. Pregnant women ≥ 35 weeks of gestation attending the antenatal clinic at Maternity and Children Hospital, Makkah were included in the study. Relevant information such as maternal age, gestational age and parity was collected using a predesigned questionnaire.

For maternal colonization, vaginal swabs from 1328 pregnant women were processed in the microbiology laboratory following standard microbiological procedures; no urine cultures were processed. From each pregnant woman, one vaginal swab was collected and inoculated into Todd-Hewitt broth supplemented with gentamicin and nalidixic acid (BBL). After 36 hours incubation, the broth was subcultured onto sheep blood agar and incubated in 5% carbon dioxide for 18-24 hours. The isolates were identified as gram-positive cocci in a chain, catalase-negative and by an appropriate reaction (Group B) with commercial latex-group-specific streptococcal typing system, MASTASTREP (Mast, UK) and Microscan Walk Away system (40SI, Siemens).

The Microscan gram-positive identification and susceptibility panel (PBC-28) was used in the Microscan Walk Away system for the identification of each GBS isolate and its antibiotic susceptibility pattern. The Microscan microtiter plate for a gram-positive panel contained separate wells for biochemical agents for identification and separate wells in the same plate for antimicrobial agents with a different concentration in double dilutions for sensitivity testing. The test was performed by touching five freshly grown colonies of the test organism using specific prompts. These colonies were suspended in 25 mL of pluronic suspension fluid. The inoculated fluid was dispensed in special trays and transferred to dehydrated substrates in the microtiter plate by RENOK system. The inoculated plates were

then placed in the Microscan Walk Away system for identification and antibiotic susceptibility testing. The results were read automatically between 16-24 hours. The data of all isolates was entered and analyzed using Microsoft Excel 2007.

RESULTS

Among 1328 pregnant women enrolled in the study, 178 (13.4%) had vaginal colonization with GBS. The age of the women ranged from 17-47 years. There was an association between maternal age, gestational age, parity and GBS colonization among pregnant women (Table 1). The study found a higher colonization rate in 28 pregnant women (27.4%) belonging to the age group ≥ 40 years and no GBS colonization in the age group < 20 years. A similar rate of GBS colonization was observed in 47 pregnant women (15.2%) of age groups 20-24 years and 56 pregnant women (14.3%) of 25-29 years of age. In age groups 30-34 years and 35-40 years similar rates of GBS colonization were observed, i.e., 28 (9.7%) and 19 (9.5%) pregnant women, respectively.

Pregnant women with gestational age > 42 weeks were found to have higher colonization ($n=9$, 25%), followed by those with gestational ages of 41-42 weeks ($n=19$, 20.2%), 39-40 weeks ($n=84$, 14.3%), and 35-36 weeks ($n=19$, 13.3%). A comparatively low rate of colonization was found for patients with a gestational age between 37-38 weeks ($n=47$, 10%). Parity ranged from zero to 12 with a high colonization in the women ($n=38$, 25%) who had delivered > 5 times followed by those who delivered 5 times ($n=10$, 20%) and then who delivered 2 times ($n=28$, 17.6%). The women who delivered 4 times were more often colonized ($n=9$, 8.3%) than those who delivered 3 times ($n=9$, 6.3%). The colonization in the women with zero parity was ($n=84$, 16.3%). No colonization was found in women who delivered once.

The antimicrobial susceptibility pattern of GBS isolates showed that all were 100% sensitive to penicillin G, ampicillin and vancomycin. Resistance was shown both to erythromycin 28 (15.7%) and clindamycin 9 (5.1%) (Table 2).

DISCUSSION

To our knowledge, this is the first study conducted in Makkah to determine GBS colonization among pregnant women. The colonization rate of 13.4% obtained in this study is within the range of 15% to 40% reported previously both from developed and developing countries^{3,13} and is similar to rates of 10% to 17% reported from Iran,¹⁴ Hong Kong,¹⁵ United Arab Emirates,¹⁶ Tunisia,¹⁷ Kuwait¹⁸ and Eastern province of Saudi

Arabia.⁵ However, higher GBS colonization rates (>23-31.6%) were reported from Tanzania,¹⁹ Egypt,²⁰ Iran²¹ and different parts of Saudi Arabia.⁶⁻⁹ These differences could be due to geographic, ethnic, and socioeconomic factors.

On the association of colonization and maternal age, in the present study GBS was more frequently isolated from women of age >40 years (27.4%), which is similar to study reported from Netherland where they found that about 25% women of age >40 years were carriers of GBS but differences were not statistically significant.²² Conversely, a study from Tanzania¹⁹ reported that GBS were more commonly isolated from women of age group between 30-34 years (32.1%), whereas in our study women of this age group were less colonized with GBS (9.7%). These differences are difficult to explain, but possibly underscore the fact that GBS colonization might be subjected to multiple factors which may vary from one geographical location to another.

Different colonization rates during pregnancy may also be attributed to gestational age, parity and ethnicity. In our study, maternal colonization was higher in gestational age >42 weeks and between 41-42 weeks compared to women of gestational age between 39-40 weeks. This indicates that an increase in GBS carriage is related to an increase in gestational age. Similar findings were reported from a study conducted in Tanzania where they found the women of gestational age between 41-42 weeks were more colonized (46.7%).¹⁹ Findings of our study are also in agreement with the results of a study from Saudi Arabia⁷ in which they also found an increase in GBS carriage with increased gestational age.

There are variable results on the effect of parity on GBS colonization. In various studies no significant differences in colonization rates were noted on the basis of age or parity,^{23,24} but increasing age or parity has also sometimes been associated with increased GBS colonization.¹⁹ In the latter study they found higher GBS colonization (50%) in women who delivered 5 times or more vs fewer numbers of deliverues, but the difference was not statistically significant. In our study about 45% of the women were colonized in the same parity cluster. Studies from Tanzania¹⁹ and the Netherlands²² showed that the women with zero parity had quite a substantial rate of GBS colonization, (23.9% and 21%, respectively, not statistically significant for different levels of parity) whereas our study showed that about 16.3% of the women with zero parity were colonized with GBS and the difference between parity states was significant. In some studies women with a lower parity number are more colonized than women with a higher parity number, such as in a study reported from the

Table 1. Association between maternal age, gestational age and parity and Group B streptococcus colonization among pregnant women.

Variable	Total (n=1328)	Number of GBS isolated (n=178)	Percentage
Maternal age (years)			
<20	39	0	0
20-24	309	47	15.2
25-29	391	56	14.3
30-34	288	28	9.7
35-40	199	19	9.5
>40	102	28	27.4
Gestational age (weeks)			
35-36	143	19	13.3
37-38	468	47	10
39-40	587	84	14.3
41-42	94	19	20.2
>42	36	9	25
Parity			
0	514	84	16.3
1	202	0	0
2	159	28	17.6
3	143	9	6.3
4	108	9	8.3
5	50	10	20
>5	152	38	25

GBS: Group B streptococcus. Statistical tests for maternal age (Pearson chi-squared = 16.79563, df=4, P=.002), gestational age (Pearson chi-squared=9.494363, df=4, P=.05, Fisher exact test P=.045), parity (Pearson chi-squared=88.50628, df=5, P<.001).

Table 2. Antimicrobial susceptibility pattern of Group B streptococcus isolated from pregnant women.

Antimicrobial agent	GBS Isolates (n=178)	
	Sensitive No (%)	Resistant No (%)
Penicillin G	178 (100)	0 (0)
Ampicillin	178 (100)	0 (0)
Vancomycin	178 (100)	0 (0)
Erythromycin	150 (84.3)	28 (15.7)
Clindamycin	169 (94.9)	9 (5.1)

Netherlands,²² where it was reported that women who delivered two times were more colonized (29%) than women who delivered 3 or more times (21%). Similar findings were reported from Turkey,²³ where they found high GBS colonization in women who delivered once (52.1%) compared with women who delivered three times (15.2%). Our findings are also similar where the women who delivered two times were more colonized (17.6%) than the women who delivered three times (6.3%). The reasons for such variable colonization are unclear and need further study.

The GBS antibiotics susceptibility patterns obtained in this study are in agreement with those reported previously^{20,23,25} and confirm its predictable empiric susceptibility to penicillin and ampicillin. Thus all GBS isolates were 100% sensitive to penicillin, ampicillin, and vancomycin, implying their possible use for empiric prophylaxis. Resistance to erythromycin and clindamycin was 15.7% and 5.1% of the isolates, respectively, indicating there is still rational use of these antibiotics to some extent in Saudi Arabia.

The continued susceptibility of GBS to members of the penicillin family, plus the resistance of a few isolates to other antibiotics^{26,27} supports the first-line role of penicillin and ampicillin in intrapartum prophylactic treatment regimens against early-onset of neonatal GBS infection. Due to its narrow spectrum of activity there is less chance for the selection of resistant organisms and, therefore, penicillin G can be used parenterally whereas ampicillin can be used by both routes.

Clindamycin and erythromycin are recommended currently as second-line antimicrobials for intrapartum chemoprophylaxis for GBS infections for women who are allergic to penicillin.¹² Resistance of GBS to clindamycin and/or erythromycin has already been reported, ranging from 0.7% to 51.3% for erythromycin and from 1.7 to 50% for clindamycin.^{17,26,28} The highest rate for erythromycin resistance (51.3%) in GBS strains was reported from Tunisia¹⁷ whereas a study from Brazil²⁹ reported the highest rate of clindamycin resistance (50%). This suggests the importance of ongoing population-based monitoring for trends in antimicrobial resistance for GBS.

In our study, resistance to erythromycin and clindamycin was 15.7% and 5.1%, respectively. This is similar to a recent study from Saudi Arabia in which GBS strains exhibited 10% resistance to erythromycin and 6% to clindamycin.⁹ Similar results of resistance to erythromycin and clindamycin were also reported from Turkey²⁵ and Kuwait.²⁸

Various strategies for the use of intrapartum antibiotics are used globally, with risk-based IPA administration in the UK¹¹ and a universal culture-based screening program in the USA.¹² However, in Saudi Arabia there is no uniform policy or program in place to screen pregnant women for GBS colonization. The high colonization rate found in this study emphasizes the need for implementation of a universal culture-based screening program for GBS colonization in all pregnant women attending antenatal clinics. Intrapartum antimicrobial prophylaxis would then be provided to women found colonized, which will prevent the transfer of GBS during delivery to neonates and avoid the development of sepsis and meningitis in newborns.

In conclusion, the high prevalence of GBS colonization in pregnant women indicates the need for screening in women attending antenatal clinics so that intrapartum antimicrobial prophylaxis can be offered to all women found colonized with GBS and in turn prevent transfer to the newborn.

Acknowledgments

The authors would like to thank Anwar Fahad Al-Abdali, Ameera Matooq Al-Sigami, Batool Hashim Al-Ghamdi and Ghadi Ismail Sindi for their excellent technical assistance. They all assisted in collection of samples and transportation from the hospital and its processing in the laboratory. This work was supported by research grant of Faculty of Applied Medical Sciences, Umm Al Qura University, Makkah, Saudi Arabia.

Conflict of interest

None to declare.

Statistical analysis by the scientific editor, Annals of Saudi Medicine using gmodels package in R.

REFERENCES

1. Schrag S, Gorwitz R, Fultz-Butts K, Schuchat A. Prevention of perinatal group B streptococcal disease: revised guidelines from CDC. *MMWR Recomm Rep.* 2002; 51(RR-11): 1–22
2. Baker CJ, Edwards NS. Group B streptococcal infections. In Remington JS, Klein JK, editors. *Infectious diseases of the fetus and newborn infant* (4th ed). Philadelphia: WB Saunders. 1995. p. 980-1054.
3. Edward MS, Nizet V. Group B streptococcal infections. In Jack S, Remington JS, Klein JO, Wilson CB, Nizet V, Maldonado YA, editors (7th ed). *Infectious diseases of the fetus and newborn infant*. Philadelphia: WB Saunders. 2011. p. 417-467.
4. Uduman SA, Chatterjee TK, Al-Mouzan MI, Al-Suleiman S. Group B streptococci colonization among Saudi women in labor and neonatal acquisition. *Int J Gynaecol Obstet.* 1985; 23: 21-4.
5. Al-Suleiman SA, Farrag I, Kingsley TD, Uduman SA., Al-Mouzan MI. Third trimester colonization and treatment of group B ?-haemolytic streptococcus among obstetric patients in the Eastern province of Saudi Arabia. *J Obs Gynae.* 1991; 11: 409-413.
6. Madani TA, Harding GK, Helewa M, Alfa MJ. Screening pregnant women for group B streptococcal colonization. *Infection.* 1998; 26: 288-91.
7. El-Kersh TA, Al-Nuaim LA, Kharfy TA, Al-Shammary FJ, Al-Saleh SS, Al-Zamel FA. Detection of genital colonization of group B streptococci during late pregnancy. *Saudi Med J.* 2002; 23: 56-61.
8. Zamzami TY, Marzouki AM, Nasrat HA. Prevalence rate of group B streptococcal colonization among women in labor at King Abdul-Aziz University hospital. *Arch Gynecol Obstet.* 2011; 284: 677-679.
9. El-Kersh TA, Neyazi SM, Al-sheikh, YA, Niazy AA. Phenotypic traits and comparative detection methods of vaginal carriage of Group B streptococci and its associated micro-biota in term pregnant Saudi women. *African J Microbiol Res.* 2012; 6: 403-413.
10. Schrag SJ, Zywicki S, Farley MM, et al. Group B streptococcal disease in the era of intrapartum antibiotic prophylaxis. *New Engl J Med.* 2000; 342: 15-20.
11. Royal College of Obstetrician and Gynaecology. Prevention of early onset neonatal group B streptococcal disease. London (UK): Royal College of Obstetricians and Gynaecologists (RCOG), Green-top guideline no. 36 (2 Ed): 2012; 1-13.
12. Centers for Disease Control and Prevention. Prevention of perinatal group B streptococcal disease-revised guidelines from CDC. *MMWR Recomm Rep.* 2010; 59 (RR-10):1-36.
13. Stoll B, Schuchat A. Maternal carriage of group B streptococci in developing countries. *Pediatric Infect Dis J.* 1998; 17: 499-503.
14. Hassanzadeh P, Motamedifar M, Gharaghani MN. Carriage rate of Group B Streptococci in pregnant women in three teaching hospitals in Shiraz, Iran. *Med Princ Prac.* 2011; 20: 277–282.
15. Tsui, MH, Margret Ip, Ng PC, Sahota DS, Leung TN, Lau TK. Change in prevalence of group B streptococcus maternal colonization in Hong Kong. *Hong Kong Med J.* 2009; 15: 414-419.
16. Amin A, Abdulrazzaq YM, Uduman S. Group B streptococcal serotype distribution of isolates from colonized pregnant women at the time of delivery in United Arab Emirates. *J Infect.* 2002; 45: 42-46.
17. Ferjani A, Ben Abdallah H, Ben Saida N, Gozzi C, Boukadida J. Vaginal colonization of the Streptococcus agalactiae in pregnant woman in Tunisia: risk factors and susceptibility of isolates to antibiotics [abstract]. *Bull Soc Pathol Exot.* 2006; 99: 99-102.
18. Al-Sweih N, Maiyegun S, Diejomaoh M, et al. Streptococcus agalactiae (Group B streptococci) carriage in late pregnancy in Kuwait. *Med Princ Pract.* 2004; 13:10–14.
19. Joachim A, Matee MI, Massawe FA, Lyamuya EF. Maternal and neonatal colonisation of group B streptococcus at Muhimbili National Hospital in Dar es Salaam, Tanzania: prevalence, risk factors and antimicrobial resistance. *BMC Public Health.* 2009; 9: 437-943.
20. Shabayeka SAAE, Abdallaa SM, Abouzeid AMH. Vaginal carriage and antibiotic susceptibility profile of group B Streptococcus during late pregnancy in Ismailia, Egypt. *J Infect Pub Hlth.* 2009; 2: 86-90.
21. Javanmanesh F, Eshraghi N. Prevalence of positive recto-vaginal culture for Group B streptococcus in pregnant women at 35-37 weeks of gestation. *Med J Islam Repub Iran.* 2013; 27:7-11.
22. Valkenburg-van den Berg AV, Sprij AJ, Oostvogel PM, et al. Prevalence of colonization with group B Streptococci in pregnant women of a multi-ethnic population in The Netherlands. *Europ J Obstet Gyn Rep Biol.* 2006; 124: 78–183.
23. Eren A, Kuçukercan M, Oguzoglu N, Unal N, Karateken A. The carriage of group B streptococci in Turkish pregnant women and its transmission rate in newborns and serotype distribution. *Turk J Pediatr.* 2005; 47: 28-33.
24. Mansouri S, Ghasami E, Najad NS. Vaginal colonization of group B streptococci during late pregnancy in Southern Iran: incidence, serotype distribution and susceptibility to antibiotics. *J Med Sci.* 2008; 8: 574-577.
25. Dilek AR, Kesbiç H, Dilek N. Antibiotic susceptibilities of group B streptococci. *J Expert Clin Med.* 2011; 28: 1-3.
26. Al-Sweih N, Jamal M, Kurdia M, Abduljabar R, Rotimi V. Antibiotic susceptibility profile of Group B streptococcus (*Streptococcus agalactiae*) at the Maternity hospital, Kuwait. *Med Princ Pract.* 2005; 14:260–263.
27. Garland SM, Cottrill E, Markowski L, et al. Antimicrobial resistance in group B streptococcus: the Australian experience. *J Med Microbiol.* 2011; 60: 230–235.
28. Boswihi SS, Udo EE, Al-Sweih N. Serotypes and antibiotic resistance in Group B streptococcus isolated from patients at the Maternity hospital, Kuwait. *J Med Micro.* 2012; 61: 126–131.
29. Castellano-Filho D, Da Silva V, Nascimento T, Vieira M, Diniz C. Detection of group B streptococcus in Brazilian pregnant women and antimicrobial susceptibility patterns. *Braz J Microbiol.* 2010; 41: 1047-1055.