# The Prevalence of Streptococcus agalactiae (Group B) Colonization in Pregnant Women at Thammasat Hospital

Siripen Tor-Udom MSc\*, Pharuhat Tor-Udom MD\*\*, Wanwarang Hiriote MSc\*

\* Department of Microbiology, Faculty of Medicine, Thammasat University \*\* Department of Obstetrics and Gynecology, Faculty of Medicine, Thammasat University

**Background:** Group B Streptococci (GBS) is responsible for serious infections in newborns such as septicemia and meningitis.

**Objective:** The present study was carried out to find the prevalence of GBS colonization in pregnant women and to determine the pattern of antibiotic resistance of the isolates.

*Material and Method:* From November 2004 to February 2005, 406 pregnant women were examined for GBS. *Results:* GBS colonization rate was 16% in pregnant women, receiving antenatal care at Thammasat Hospital. *Conclusion:* All the isolates were sensitive to Ampicillin, Penicillin, Vancomycin and Cephazolin. Resistant was seen with Clindamycin (3%) and Erythromicin (1.5%).

Keywords: Streptococcus agalactiae, Group B streptococci, GBS colonization, Pregnancy, Prevalence

J Med Assoc Thai 2006; 89 (4): 411-4 Full text. e-Journal: http://www.medassocthai.org/journal

Group B streptococcus (GBS) is one of the most common causes of neonatal sepsis throughout the world<sup>(1)</sup>. In the United States, 5-10 percent of neonatal deaths are caused by this organism<sup>(2)</sup>. In December 2002, the Centers for Disease Control and Prevention (CDC) developed a strategy to reduce the morbidity and mortality of GBS - associated neonatal disease<sup>(3)</sup>. They recommended culturing all pregnant women at 35 to 37 weeks of gestation for GBS colonization using vaginal-rectal specimens. For maximum sensitivity of GBS detection, the CDC recommends the use of a selective broth containing antibiotics, such as Todd-Hewitt broth supplemented with both colistin and nalidixic acid. Specimens were inoculated into selective broth, and after 18 to 24 hours of inoculation, the broth was subcultured onto a sheep blood agar plate (SBA) and was incubated for 18 to 24 hours. Colonies of suspected GBS were confirmed either by serological methods or by CAMP testing. If colonies resembling GBS are absent after 18 to 24 hours, reincubation for an additional to 24 hours and reexaminations are recommended. The purpose of the present study was to find out the prevalence of GBS colonization in mothers using the method recommended by CDC and to determine the pattern of antibiotic resistance

#### **Material and Method**

From September 2004 to February 2005, 406 women at the 35 to 37 weeks of gestation, receiving antenatal care at Thammasart Hospital, who met the inclusion and exclusion criteria were enrolled in the present study. The inclusion criteria were 1) Pregnant women who planned vaginal delivery. 2) No evidence of GBS infection 3) Signed consent forms after the informed consent process. The exclusion criteria were 1) Pregnant women who had received antibiotics within one week before coming to the antenatal clinic. 2) Had elective caesarean section and 3) Refused to enroll in the present study.

#### Specimen Collection

Pregnant women at the 35-37 weeks of gestation were instructed to collect specimen by themselves. Two swabs were collected separately, one from the vagina and another from the rectum. Two different

Correspondence to : Tor-Udom S, Department of Microbiology, Faculty of Medicine, Thammasat University, Klongluang, Pathumthani 12121, Thailand. Phone: 0-2992-9710, Fax: 0-2992-9711, E-mail: siripent@tu.ac.th

swabs were placed into the same container of Todd-Hewitt broth with 10 mg of colistin per ml and 5 mg of oxolinic acid per ml (GBS supplement, Oxoid). Vaginal-rectal specimens were delivered to the laboratory within 4 hours.

# **Culture Method**

The specimens in the recommended selective broth were incubated at 37 C in ambient air of 5% CO<sub>2</sub>. After 18-24 hours of incubation subcultured the broth onto a sheep blood agar plate (tryptic soy agar with 5% defibrinated sheep blood) and incubated at 37 C in 5% CO<sub>2</sub>. Both beta-hemolytic and non-hemolytic colonies morphologically resembling GBS were tested with 1) Gram's stain, 2) Catalase test, 3) CAMP test and 4) streptococcal grouping by latex agglutination test (Oxoid, UK). All negative subcuture plates were reincubated for an additional 18-24 hours and reexamined.

## Susceptibility testing of isolates

All of GBS isolates were tested for drug susceptibility as recommended by the CDC (1996). GBS colonies were inoculated in Mueller-Hinton broth and incubated at 37 C in 5% CO<sub>2</sub>. After 18-24 hours, adjusted turbidity to match a 0.5 McFarland standard. Within 15 minutes of adjusting the turbidity, disk diffusion test was performed with antibiotic-containing disks (Oxiod, UK). The antimicrobial agents tested included clindamycin (2 mg), Erythromycin (15 mg), Penicillin G (10 mg), Ampicillin (10 mg) and Vancomycin (30 mg). The diameter of the zone of inhibition was measured using a ruler, interplet according to NCCLS guide lines. The results were described by frequency number and percentage.

### Results

The results of the study are summarized in Table 1. 65 pregnant women from 406 cases or 16 percent at 35-37 weeks of gestation were colonized with *Streptococus agalactiae*. All isolates were susceptible to Ampiciillin (100%), Penicillin (100%), Vancomycin (100%), Cephazolin (100%), followed by Erythromycin (98.5%), and Clindamycin (96.9%)

# Discussion

The group B streptococci (GBS) are known to cause a wide variety of infections in adults, but clinical interest in these bacteria mainly relates to their ability to cause serious neonatal illness, especially meningitis and sepsis. In developed countries, these organisms are the leading cause of neonatal sepsis and meningitis with a case fatality rate of 40 to 80%<sup>(4)</sup>. However, in developing countries like Thailand, the problem has not been adequately studied and there are only a few reports available<sup>(5,6)</sup>. Since the transmission of GBS from the positive mother to the neonate occurred in 20%-50% of the case<sup>(4,7,8)</sup>, a preventive strategy has to be employed to avoid serious neonatal sepsis. An antenatal screening that provides vaginal-rectal swab culture at 35-37 weeks of gestation and a chemoprophylaxis intrapartum in the positive cases appears to be the most effective approach. In the present study, the authors used the appropriate culture method and specimen collection as recommended by the CDC. The results showed 65 pregnant women from 406 cases were positive for GBS. This result reflects a high prevalence of 16% of pregnant women receiving antenatal care at Thammasart Hospital. The prevalence was different from Werawatakul Y et al in Khon Kaen, Thailand<sup>(5)</sup> (6%), but similar to Jadsada T et al<sup>(6)</sup> (14.5% in Khon Kaen, 12% in Bangkok). Reports of low prevalence rates of GBS were 4% in Mexico City<sup>(9)</sup>, 6% in Peru<sup>(10)</sup>, 6.6% in Italy<sup>(7)</sup>, 8.7% in Turkey<sup>(11)</sup> and 9% in Ethiopia<sup>(12)</sup>. The high prevalence rate was found in the United States<sup>(2)</sup> (15- 25%), in Jordan<sup>(13)</sup> (30%) and in Trinidad<sup>(14)</sup> (31%).

For better isolation of GBS from clinical material, method of specimen collection, transport medium used and culture method are important. In the

**Table 1.** GBS colonization and the antimicrobial susceptibility of the isolates

Total number of	No. of GBS isolates	Prevalence of GBS colonization (%)	Antimicrobial susceptibility <sup>a</sup>					
at 35-37 weeks of gestation			Clin	Ery	Pen G	Amp	Van	Ceph
406	65	16.0	63 (96.9%)	64 (98.5%)	65 (100%)	65 (100%)	65 (100%)	65 (100%)

a Tested for Clindamycin (Clin), Erythromycin (Ery), Penicillin G (Pen G), Ampicillin (Amp), Vancomycin (Van), Cephazolin (Ceph)

present study, the authors collected specimens from both the vagina and rectum in which the detection of GBS carriage in pregnant women was increased<sup>(7,8)</sup>. According to the CDC procedure, all of the patients (406) were carefully instructed to take vaginal and rectal swabs by themselves, which lessened the physician's burden. Because the vaginal-rectal swab was kept in the GBS selective medium, the normal flora present in the perianal area and genital area would be inhibited and a contamination from other microorganism would also be prevented. According to the above reason, there should be no difference in the specimen collection by either patients or physicians. Since the selective broth [Todd-Hewitt broth with colistin (10 mg/ml), oxolinic acid (5 mg/ml)] was used as a transport medium, the GBS recovery rate was better than using the Amies transport medium (Data not shown). This finding was similar to Kulkarni AA et al<sup>(4)</sup> which demonstrated that the selective broth medium is the most effective transport medium for GBS isolation as it inhibits the normal vaginal and rectal flora without any inhibitory effect of GBS, and enhances the growth of GBS. To subculture the selective broth, the authors found that the recovery rate of GBS was higher when sheep blood agar was used instead of human blood agar (Data not shown). All of the isolates were sensitive to Ampicillin, Penicillin, Vancomycin and Cephazolin. Resistance was observed with Clindamycin (3%) and Erythromicin (1.5%).

In conclusion, the prevalence of GBS colonization at Thammasat Hospital was higher than that reported in other hospitals in Thailand. An antenatal screening at 35-37 weeks of gestation by vaginal-rectal swab culture and a chemoprophylaxis intrapatum in the positive cases to prevent serious neonatal sepsis or meningitis are recommended. At present, culture of specimens from the rectum and vagina remain the gold standard for detection of GBS in pregnant women<sup>(3)</sup>. However, detection of GBS in pregnant women is probably underestimated if the culture method had low sensitivity. In the present study, the authors found that the prevalence of GBS colonization is higher than that reported in other parts of Thailand when the appropriate culture method as recommended by the CDC was used. In addition, laboratory skill of investigators picking up colonies of GBS is one of the most important factors for isolation of group B streptococci from clinical specimens. In 2002, the American Committee on Obstetric Practice supported the new CDC recommendation that obstetric providers adopt a culture-based strategy for the prevention of earlyonset GBS disease in the newborn because a riskbased approach does not rely on microbiological detection<sup>(15)</sup>.

### Acknowledgement

This project was financially supported by the Academic Promotion Research Grant, Thammasat University.

#### References

- Gil EG, Rodriguez MC, Bartolome R, Berjano B, Cabero L, Andreu A. Evaluation of the Granada agar plate for detection of vaginal and rectal group B streptococci in pregnant women. J Clin Microbiol 1999; 37: 2648-51.
- Baker CJ. Group B streptococcal infections. Clin Perinatol 1997; 24: 59-70.
- 3. 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: 1-22.
- 4. Kulkani AA, Pawar SG, Dharmadhikari CA, Kulkarni RD. Colonization of pregnant women and their newborn infants with group B streptococci. Indian J Med Microbiol 2001; 19: 1-4.
- Werawatakul Y, Wilailuckana C, Taksaphan S, Thinkumrup J, Pragarasung M, Chouwajaroen P, et al. Prevalence and risk factors of Streptococcus agalactiae (group B) colonization in mothers and neonatal contamination at Srinagarind Hospital. J Med Assoc Thai 2001; 84: 1422-9.
- Thinkhamrop J, Limpongsanurak S, Festin MR, Daly S, Schuchat A, Lumbiganon P, et al. Infections in international pregnancy study: performance of the optical immunoassay test for detection of group B streptococcus. J Clin Microbiol 2003; 41: 5288-90.
- Citernesi A, Formica G, Caruso S, Curiel P. Vaginal colonization of Streptococcus B in pregnancy. Minerva Ginecol 1996; 48: 227-33.
- 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.
- de Lourdes CM, Kretschmer RR, Becker I, Guzman A, Gallardo L, Lepe CM. Colonization of Mexican pregnant women with group B streptococcus. J Infect Dis 1981; 143: 134.
- 10. Collins TS, Calderon M, Gilman RH, Vivar A, Charache P. Group B streptococcal colonization in

a developing country: its association with sexually transmitted disease and socioeconomic factors. Am J Trop Med Hyg 1998; 59: 633-6.

- Ayata A, Guvenc H, Felek S, Aygun AD, Kocabay K, Bektas S. Maternal carriage and neonatal colonisation of group B streptococci in labour are uncommon in Turkey. Paediatr Perinat Epidemiol 1994; 8: 188-92.
- Schmidt J, Halle E, Halle H, Mohammed T, Gunther E. Colonization of pregnant women and their newborn infants with group B streptococci in the Gondar College of Medical Sciences. Ethiop Med

J 1989; 27: 115-9.

- 13. Sunna E, el Daher N, Bustami K, Na'was T. A study of group B streptococcal carrier state during late pregnancy. Trop Geogr Med 1991; 43: 161-4.
- Orrett FA, Olagundoye V. Prevalence of group B streptococcal colonization in pregnant third trimester women in Trinidad. J Hosp Infect 1994; 27: 43-8.
- ACOG Committee Opinion: number 279, December 2002. Prevention of early-onset group B streptococcal disease in newborns. Obstet Gynecol 2002; 100: 1405-12.

# อัตราชุกของการมีเชื้อสเตรปโตคอกคัสกลุ่มบีในหญิงตั้งครรภ์ที่โรงพยาบาลธรรมศาสตร์ เฉลิมพระเกียรติ

# ศิริเพ็ญ ต่ออุดม, พฤหัส ต่ออุดม, วรรณวรางค์ หิริโอตป

เชื้อสเตรปโตคอกคัสกลุ่มบีเป็นสาเหตุสำคัญที่ทำให้เกิดการติดเชื้อรุนแรงในเด็กแรกเกิดโดยเฉพาะโลหิต เป็นพิษและเยื่อหุ้มสมองอักเสบ การศึกษานี้เป็นการหาอัตราชุกของการมีเชื้อสเตรปโตคอกคัสกลุ่มบีในหญิงตั้งครรภ์ ที่มาฝากครรภที่โรงพยาบาลธรรมศาสตร์เฉลิมพระเกียรติ เพื่อกำหนดแนวทางในการดูแลผู้ป่วยที่เหมาะสม เพื่อป้องกัน การติดเชื้อสเตรปโตคอกคัสกลุ่มบีจากมารดาสู่ทารกขณะคลอด ผู้วิจัยได้ทำการเก็บสิ่งส่งตรวจจากหญิงตั้งครรภ์ 406 คน ระหว่างเดือนพฤศจิกายน พ.ศ. 2547 ถึงเดือนกุมภาพันธ์ พ.ศ. 2548 ตรวจพบเชื้อ 65 คน ไม่พบเชื้อ 341 คน มีอัตราชุกของการติดเชื้อร้อยละ 16 ผูวิจัยได้ทำการศึกษาความไวต่อยาปฏิชีวนะของเชื้อที่แยกได้จากหญิงตั้งครรภ์ พบว่า เชื้อที่แยกได้ทั้งหมดคิดเป็นร้อยละ 100 ไวต่อยาแอมพิชิลลิน เพนนิชิลลิน แวนโคมัยชิน และเซฟาโชลิน การดื้อ ต่อยาปฏิชีวนะพบว่าเชื้อดื้อต่อยาคลินดามัยชิน และ อีรีโทรมัยซินร้อยละ 3 และร้อยละ 1.5 ตามลำดับ