Vaginal carriage and antibiotic susceptibility profile of group B Streptococcus during late pregnancy in Ismailia, Egypt

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Summary Group B Streptococcus (GBS) infection has long been recognized as a frequent cause of morbidity and mortality in newborn infants. The purpose of this study was to determine the colonization rate with GBS and the antibiotic susceptibility profile in pregnant women attending Gynecological clinics in Egypt. One-hundred and fifty vaginal swabs were collected from pregnant women at 35—40 weeks of gestation. In comparison to culture, direct latex agglutination testing revealed 100% sensitivity and 93.75% specificity. Thirty-eight specimens (25.3%) were found to be positive for GBS. Each isolate was tested for susceptibility to penicillin G, ampicillin, cefotaxime, erythromycin, clindamycin and vancomycin. Erythromycin-resistant isolates were further classified by double-disk method. All isolates were susceptible to penicillin G, ampicillin and vancomycin. Resistance to cefotaxime was detected in three isolates (7.89%). Five isolates (13.15%) were resistant to erythromycin and nine isolates (23.68%) were resistant to clindamycin. Four (80%) isolates had constitutive macrolide-lincosamide-Streptogramin B resistance (cMLSBB) resistance and one (20%) isolate had inducible resistance (iMLS) resistance. GBS colonization was found to be high in our region. Latex agglutination testing and Islam medium are reliable methods to detect GBS in late pregnancy; however, latex agglutination test is rapid and simpler. Penicillin G remains the first choice antibiotic for treatment of GBS infections.

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Introduction

Streptococcus agalactiae or group B Streptococcus (GBS) infection has long been recognized as a frequent cause of morbidity and mortality in newborn infants [1]. Life-threatening complications of GBS bacteremia, such as endocarditis, meningitis and fatal septicemia with multiorgan failure, have been described over the last decades [2].

GBS is found in the vagina and/or rectum of 10—30% of pregnant women as normal flora, and infants born to these women may develop disease due to exposure to bacteria before birth or during the neonatal period [3]. These neonatal GBS infections are divided into early-onset infection and late-onset infection. Early-onset infection, which is the most common type of neonatal GBS disease, occurs within the first week of life, while late-onset infection occurs in infants between 1 week and 3 months of age [4].

Maternal colonization with GBS is the most predominant risk factor for the development of invasive neonatal GBS disease [5], since vertical transmission of GBS before or during delivery can be found in the vast majority of all cases [5]. The revised Centers for Disease Control and Prevention (CDC) guidelines issued in 2002 recommended a culture-based screening for vaginal rectal colonization with GBS for all pregnant women at 35—37 weeks of gestation for prevention of early-onset GBS disease. If colonization is detected, intrapartum antibiotic prophylaxis is recommended [6]. Implementation of these guidelines has been associated with a falling incidence of neonatal GBS disease in USA [7—9]. No epidemiological data has been published on the colonization rate with GBS of pregnant women in Egypt. The purpose of this study was to determine the current colonization rate with GBS and the antibiotic susceptibility profile in our region.

Materials and methods

Culture and identification procedures

One-hundred and fifty pregnant women (17—43 years) at 35—40 weeks of gestation attending the Gynecological clinic at the Ismailia General Hospital (400 beds) and the Gynecological Department at the University Hospital of Suez Canal University (415 beds) from September 2007 to April 2008 were enrolled in this study. One vaginal swab was collected from each patient with an informed consent, placed in Amies transport medium (Lab M) and kept at 4°C. Swabs were inoculated in selective enrichment broth medium (Todd-Hewitt broth (oxoid) supplemented with nalidixic acid 15 μg/ml and colistin 10 μg/ml). After 18—24 h incubation at 37°C in 5% CO2, broths were subcultured onto Islam medium (Oxoid). Islam media plates were examined for orange pigmented colonies after 24—48 h of anaerobic incubation. In addition, latex agglutination testing (Streptococcal grouping kit, AVIPATH STREP, Omega Diagnostics Ltd.) was performed directly on primary selective broth cultures. Positive selective broth cultures for GBS (either by latex agglutination testing or Islam medium) were further confirmed by the traditional culture method through subculturing onto 5% columbia blood agar (oxoid) supplemented with nalidixic acid 15 μg/ml and colistin 10 μg/ml (CNA medium), incubated for 18—24 h at 37°C in 5% CO2. Colonies on CNA plates were identified by Gram stain, hemolysis, catalase, CAMP test (named after Christie, Atkins and Munch-Petersen), hippurate hydrolysis test, and bile esculin hydrolysis test [10,11].

Antibiotic susceptibility testing and macrolide resistance phenotypes

Each isolate was tested for susceptibility to penicillin G, ampicillin, cefotaxime, erythromycin, clindamycin and vancomycin using disk diffusion method according to CLSI (formerly NCCLS) guidelines [12]. Erythromycin-resistant isolates were further classified as having cMLSBo (constitutive macrolide—lincosamide-Streptogramin B resistance), iMLSBo (inducible resistance), or M phenotype (macrolide—Streptogramin B resistance and Lincosamide susceptibility) by double-disk method [12,13].

Results

Colonization rate

Among the 150 patients that were enrolled in the study a total of 38 specimens (25.3%) were found to be positive for GBS by Islam medium. The direct latex agglutination testing on primary selective broth cultures detected 45 (30%) positive specimens.

All specimens found to be positive by culture on Islam medium were also positive by latex agglutination testing. Among the 112 Islam medium negative specimens, 7 were positive by latex agglutination testing. It was noticed that all these seven samples showed positive agglutination testing with
Streptococcus group B and group D. Presumptive identification of Enterococcus spp. was achieved using bile esculin test. When these samples were analysed by traditional culturing method, GBS failed to be recovered from primary selective broth cultures and were considered false positive by latex agglutination testing. Thus latex agglutination testing directly on primary selective broth cultures revealed 100% sensitivity and 93.75% specificity.

Antibiotic susceptibility testing and macrolide resistance phenotypes

All the 38 isolates were uniformly susceptible to penicillin G, ampicillin and vancomycin. Resistance to cefotaxime was detected in three isolates (7.89%) while 1 isolate (2.63%) was intermediate. Five isolates (13.15%) were resistant to erythromycin and three (7.89%) were intermediate. Nine isolates (23.68%) were resistant to clindamycin.

Among the erythromycin resistance isolates, four (80%) isolates had constitutive cMLSB\textsubscript{B} resistance and one (20%) isolate had inducible iMLS\textsubscript{B} resistance.

Discussion

In the present study, the prevalence of GBS in Ismailia, Egypt was found to be 25.3%. Such finding was in accordance with studies from the United States and Europe that reported colonization rates between 6.5% and 36% [14–16,4]. Recent Arabian studies showed similar colonization rates ranging from 10.1% in United Arab Emirates [17], 16.4% in Kuwait [18], 17% in Tunisia [19], and 27.6% in Saudi Arabia [20]. This was consistent with a recent survey by Stoll and Schuchat [21] who demonstrated that heavy growth of GBS usually lacked concomitant growth or had only very light growth of Enterococcus fecalis. Conversely, very light growth of GBS revealed heavy growth of E. fecalis. Bourbeau et al. [28] had a similar experience when five of nine GBS specimens failed to multiply in selective enrichment broth. A possible explanation was suggested by Dunne and Holland-Staley [29], who noticed that certain strains of GBS appear to be suppressed by a moderate to heavy growth of E. fecalis. Other explanations may include presence of nonviable GBS that could be detected by antigen-detection method but not by culture, as well as inability of culture to detect low bacterial numbers. Antibiotics and feminine hygiene products have also shown to inhibit the detection of GBS by culture but have no detrimental effect on antigenic detection [30]. Inadequate specimen collection and transport from obstetric clinics to the laboratory may have some effect especially in case of light colonization [31,32].

Susceptibility testing revealed that GBS is uniformly susceptible to penicillin G, ampicillin and vancomycin. Most other studies have found the same [33–36]. Penicillin G is the drug of choice when diagnosis is established. However, an increased resistance to erythromycin and clindamycin, the drugs of choice for women with serious penicillin allergy, has been observed. In this regard, 13.15% of the tested GBS isolates were resistant to erythromycin and 23.68% to clindamycin. Compared to recent Arabian studies,
the present study showed much lower resistance rates to erythromycin and clindamycin than those reported by Dunia et al. in Syria [37]. However, it revealed higher resistance rates to these antibiotics than those reported by Al-Seweih et al. in Kuwait [38]. In regard to recent studies from America and Europe, the present findings were consistent with those reported by Barbaros et al. in Turkey [39], much lower than those reported by DiPersio and DiPersio in USA [40] and higher than those reported by Schoening et al. in Germany [41] and Figueiracoelho et al. in Portugal [42]. The majority of the erythromycin-resistant isolates showed cMLS\(_B\) resistance and one had iMLS\(_B\) resistance. Quite similar distribution resistance phenotypes were described in studies from Portugal, France and Spain [42–45].

In conclusion, the vaginal GBS colonization rate among Egyptian women was found to be 25.3% and thereby constitutes a group of women whose infants are at great risk of GBS invasive infections. Latex agglutination testing and Islam medium are reliable methods to detect GBS in late pregnancy; however, latex agglutination test is rapid and simpler. Finally, Penicillin G remains the first choice antibiotic for the treatment of GBS infections. The majority of the erythromycin-resistant isolates showed cMLS\(_B\) resistance. Continuous monitoring of the occurrence of resistance to antibiotics is essential, as the emergence of resistant strains has become a growing concern on the therapy of GBS. The increasing rates and continued spread of macrolide and lincosamide resistance in GBS have broad implications for both prophylactic and empiric drug treatment strategies.

**Conflict of interest statement**

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**References**


