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Prevalence of group B streptococcus among pregnant and non-pregnant women in Ismailia, Egypt

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Abstract

Streptococcus agalactiae or group B streptococcus (GBS) is among the leading life-threating infectious agents in neonates. It is also known to cause disease in pregnant women and immunocompromised patients. The aim of the current study was to determine GBS prevalence among pregnant and non-pregnant women in Ismailia, Egypt. Four-hundred sixty-two vaginal swabs were collected from pregnant and non-pregnant women. A total of 27.9% women were found as positive GBS carriers. In respect to pregnant participants, 26% of pregnant women were defined as asymptomatic GBS carriers versus 35.5% among non-pregnant participants. Age, parity, history of previous abortion or preterm labor were not significantly associated as potential risk factors for GBS carriage.

1. Introduction

Streptococcus agalactiae or group B *streptococcus* (GBS) is a commensal of the vagina and/or rectum of 10-30% of pregnant women and up to 40% in non-pregnant adults (Schuchat, 1998; Verani et al., 2010). GBS is a principle etiological agent of sever neonatal infections such as sepsis, pneumonia, and meningitis (Schuchat, 1998; Verani et al., 2010).

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GBS infections manifest as early-onset disease during the first week of life and late-onset disease during the first three months after birth (Schuchat, 1998; Lindahl et al., 2005). GBS is also an important cause of disease in pregnant women, elderly, and immunocompromised patients with underlying conditions (Schuchat, 1998; Lindahl et al., 2005).

The most important risk factor of GBS transmission is maternal carriage (Verani et al., 2010). Vertical transmission during or after labor is the most predominant route of transmission (Verani et al., 2010). In 2010, the Centers for Disease Control and Prevention (CDC) (Verani et al., 2010) released revised guidelines which recommended culture-based GBS screening at 35 to 37 weeks of gestation followed by intrapartum antibiotic prophylaxis for positive carriers. Implementation of CDC guidelines resulted in significant decline in neonatal GBS disease in the US (Schrag and Verani, 2013).

Except for very few studies (Elbaradie et al., 2009; Shabayek et al., 2009; Shabayek et al., 2010; Shabayek and Abdalla, 2014; Shabayek et al., 2014), epidemiological data on GBS carriage and neonatal disease in Egypt still sparse. The aim of the current study was to determine GBS prevalence among pregnant and non-pregnant women and evaluate potential risk factors associated with GBS carriage.

2. Materials and Methods

2.1 *Ethical approval*

All procedures involving human subjects were approved by the Research Ethics Committee at the Faculty of Pharmacy, Suez Canal University, Egypt (Reference number 201611RH1). An informed consent was obtained from all study participants.

2.2 *Study participants*

This is a cross-sectional study where a total of 462 consecutive women (372 pregnant women, during their third trimester and 90 non-pregnant women) attending the Gynecological and Obstetric department at the University Hospital of Suez Canal University in the period from November 2016 to January 2017 were enrolled in the study.

2.3 *Sample collection and transportation*

One vaginal swab was obtained from each participant as described by CDC (Verani et al., 2010). Afterwards, each swab was dipped into sterile tubes containing Amines Transport medium (Lab M) and stored at 4 °C until being processed.

2.4 Demographic data collection

Demographic data including participants` age, parity, history of previous abortion and previous preterm labor were collected.

2.5 GBS detection

GBS detection was done as per CDC recommendations (Verani et al., 2010) through a selective enrichment procedure where swabs were inoculated in Lim broth (Todd-Hewitt broth (Oxoid) supplemented with nalidixic acid 15 µg/ml and colistin 10 µg/ml) and incubated overnight at 37 °C in 5% CO_2. Afterwards, samples were subcultured onto 5% sheep blood agar (Oxoid), supplemented with nalidixic acid 15 µg/ml and colistin 10 µg/ml, and CHROMagar^{TM} StrepB agar (CHROMagar microbiology, France). Blood agar plates were incubated overnight at 37 °C in 5% CO_2 whereas CHROMagar^{TM} StrepB plates were incubated overnight at 37 °C under anaerobic conditions. Colonies on blood agar plates were examined for hemolysis, CAMP test (named after Christie, Atkins, and Munch-Petersen), catalase, Hippurate hydrolysis, Bile esculin hydrolysis and Gram stain. Colonies on CHROMagar^{TM} StrepB plates were examined for intense mauve coloration. Further confirmation was done using latex agglutination testing (Streptococcal grouping kit, AVIPATH STREP, Omega Diagnostics Ltd.). This was done on both pure colonies and primary Lim broth overnight cultures (Park et al., 2001; Guerrero et al., 2004).

2.6 GBS identification with a species-specific 23S rRNA Primer

DNA was extracted as described previously (Shabayek et al., 2010). Polymerase chain reaction (PCR) was done using the species-specific 23S rRNA primer sequences described by (Evans et al., 2008): *Forward* 5`AACAGCCTCGTATTTAAAATGATAG ATTAAC -3` and *Reverse* 5`-TCCTACCATGACACTAATGTGTC -3`. PCR was done according to (Kawata et al., 2004; Evans et al., 2008).

2.7 Statistical analysis

Continuous variables were analyzed with two-tailed unpaired t-test while categorical data were analyzed with Chi-square and Fischer`s exact test. A *p* value less than 0.05 was considered statistically significant.

3. Results

3.1 GBS identification

All isolates were beta hemolytic, Lancefield Group B, catalase negative, Gram-positive cocci arranged in pairs or chains. All isolates showed deep intense mauve coloration on

CHROMagar™ StrepB plates and were positive for CAMP and Hippurate hydrolysis and negative for Bile esculin hydrolysis. The 23S rRNA fragment was amplified for all strains.

3.2 Prevalence

Among the 462 participants, 129 (27.9%) women were found as GBS- positive carriers. In respect to pregnant participants, 26% pregnant women (n= 97/372) were defined as asymptomatic GBS carriers versus 35.5% among non-pregnant participants (n= 32/90).

3.3 Demographics

The mean age of participants was 29.5 ± 6.9. The mean age of GBS-positive carriers was 29.2 ± 6.3. The mean age of GBS-negative carriers was 29.7 ± 7.1. GBS colonization rate was the highest in the age range from 21 – 30. However, GBS carriage was not significantly associated with older age (p = 0.74). A similar scenario was observed for parity (p = 0.65) as higher GBS colonization was detected for multigravida compared to primigravida or participants with no previous pregnancies. In addition, there was no statistically significant difference between GBS-positive and GBS-negative participants in respect to age (p = 0.45) and parity (p = 0.48). The same was true for history of previous abortion (p = 0.38) or preterm birth (p = 0.32). These were not significantly associated with GBS carriage. Percentage of GBS carriage among study participants in respect to age and parity is shown in Figures 1 and 2. Demographic data is shown in Tables 1 and 2.

4. Discussion

The prevalence of GBS carriage in Ismailia, Egypt was 27.9% among all study participants including pregnant and non-pregnant women. This rate was comparable to previous work conducted in Ismailia, Egypt among pregnant and non-pregnant women where the observed colonization rates were found to be 25.3% and 27.3% (Shabayek et al., 2009; Shabayek et al., 2014). However, it was higher than that recorded in AlFayom, Egypt (Elbaradie et al., 2009) which was 17.89%.

Considering other Arabian-African countries in the neighborhood, a lower colonization rate was found in Tunisia (17%) (Ferjani et al., 2006) which is similar to that reported in AlFayom, Egypt. Considering other Arabian-Asian countries in the vicinity, similar rates were reported in Saudi Arabia (27.6%)

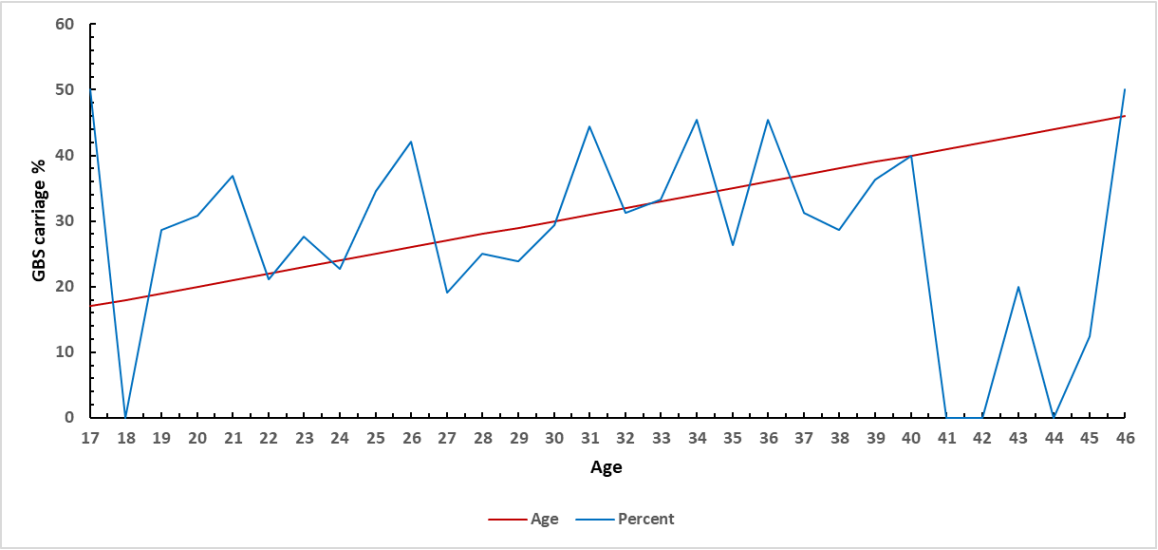

Figure 1: Percentage of GBS-positive participants in respect to age.

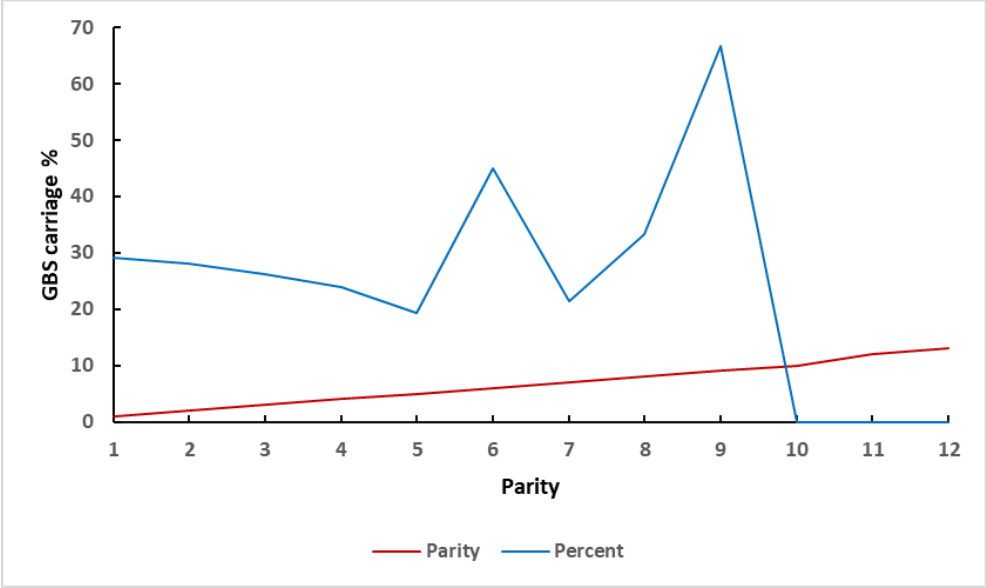

Figure 2: Percentage of GBS-positive participants in respect to parity.

Table 1: Study participants.

GBS status (number)	Age (mean ± SD)	*p value*	Parity	*p value*
GBS-positive (129)	29.17 ± 6.34	0.45	2.86 ± 1.89	0.48
GBS-negative (333)	29.70 ± 7.10		2.99 ± 1.84	

Two-tailed unpaired t-test was calculated to evaluate the statistical differences between GBS -positive and GBS -negative participants for age and parity. P value < 0.05 is considered statistically significant.

Table 2: Detailed demographic data.

Variable	GBS-positive	GBS-negative	*p* value
Age			0.74
≤ 20	9	24	
21 - 30	71	188	
>30	49	121	
Parity			
No previous pregnancies	6	5	0.65
Primigravida	28	68	
Multigravida	95	260	
Previous abortion			0.38
Yes	41	122	
No	88	211	
Previous preterm labor			0.32
Yes	11	41	
No	118	292	

Two-tailed Chi-square and Fischer`s exact tests were calculated to evaluate the statistical differences between GBS -positive and GBS -negative participants for age, parity, previous abortion, and history of preterm labor. P value < 0.05 is considered statistically significant.

(El-Kersh et al., 2002). However, lower rates were demonstrated in Lebanon (17.7%) (Seoud et al., 2010), Kuwait (16.4%) (Al-Sweih et al., 2004), United Arab Emirates (10%) (Amin et al., 2002), and Iran (9.1%) (Namavar Jahromi et al., 2008). These later were also like those reported in AlFayom, Egypt (Elbaradie et al., 2009).

Nevertheless, all detected colonization rates were within the expected ranges. This was clear when considering the GBS carriage rates reported in the US and Europe showing a rage between 6.5% and up to 36% (Campbell et al., 2000; Barcaite et al., 2008). According to (Stoll and Schuchat, 1998) comparable colonization rates can still be observed in developing and developed countries despite the geographical differences. According to (Benitz et al., 1999), differences in colonization rates are expectable across various geographical locations.

Moreover, (Benitz et al., 1999) demonstrated that variations in GBS colonization rates can also be attributed to ethnicity, parity, age, marital status, multiple sex partners, smoking and even to education level. In the present study, non-statistically significant associations were demonstrated between GBS carriage, age, and parity although higher percentage of GBS carriage was observed with older age and multigravida.

Furthermore, (Philipson et al., 1995) reported that the true burden of GBS colonization might be underestimated by sampling technique. They demonstrated enhanced antenatal GBS detection when vaginal swabs were combined with rectal swabs. In the current study, only one vaginal swab was obtained from each participant.

It is noteworthy that the genera *Enterococcus, Staphylococcus* and *Candida* were the most predominant co-colonizers easily detected during GBS screening. Such observation was in concordance with (Kubota et al., 2002; Altoparlak et al., 2004). The possibility of GBS masking due to other predominant vaginal colonizers has been reported before (Ostroff and Steaffens, 1995; Bourbeau et al., 1997; Dunne and Holland-Staley, 1998). This highlights the importance of selective enrichment techniques for reliable detection of GBS in the vaginal swabs through inhibiting the overgrowth of other co-colonizers.

5. Conclusion

In the current study, GBS prevalence among pregnant and non-pregnant women in Ismailia, Egypt was 27.9% which is comparable to other regions in the world.

GBS is among the leading causes of neonatal mortality and morbidity worldwide. However, till now, no national guidelines or recommendations are available for GBS monitoring among Egyptian pregnant women. Further GBS investigations should be done to reveal the true burden and etiological agents of neonatal diseases and deaths in Egypt. This should also include deeper epidemiological characterization of the Egyptian GBS isolates in order to be considered in the ongoing vaccine clinical trials to combat neonatal GBS disease.

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