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Original Article

Evaluation of Antimicrobial Susceptibility of *Streptococcus agalactiae* Isolates from Patients with Urinary Tract Infection (UTI) Symptoms

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Background: *Streptococcus agalactiae*, also known as Group B *Streptococcus* (GBS) is a commensal organism in the urogenital tract and rectum in approximately 25% of the healthy adult female population. The bacterium is the leading cause of bacterial meningitis, pneumonia, and sepsis in human infants.
Materials and Methods: Our study was performed over a three - month period from April to June 2014. Midstream specimens of urine were collected from outpatients suspected of having a bacterial urinary tract infection, which had not received any antibiotics. Group B *Streptococci* isolates were confirmed by typical colony morphology and identified by differential tests. Antibiotic susceptibility testing was carried out by disk diffusion method on Mueller Hinton agar (Merck, Germany) based on (CLSI) Guidelines 2012.
Results: GBS strains were isolated from 264 (21.1%) cases (out of 1249 positive bacterial urine cultures). The higher prevalence was recorded in the 15-44 and 45-64 age groups. Antibiotic susceptibility tests revealed that vancomycin, penicillin, and linezolid had the lowest, and tetracycline had the highest resistance rate.
Conclusion: In conclusion, the results of the present study confirm the universal susceptibility of GBS strains to the penicillin family and assert the use of penicillin or ampicillin as the first drug of choice for treatment and prophylaxis against GBS infections. However, it is important to perform antibiotic susceptibility testing whenever penicillin could not be prescribed.

Keywords: Streptococcus Group B, Urinary tract infection, Sensitivity test

1. Background

Streptococcus agalactiae, also known as Group B *Streptococcus* (GBS) is a commensal organism in the urogenital tract and rectum in approximately 25% of the healthy adult female population (1). The bacterium is the leading cause of bacterial meningitis, pneumonia, and sepsis in human infants. In addition, GBS has been reported increasingly associated with invasive infections in pregnant women and non-pregnant adults, especially those under conditions such as diabetic mellitus, malignancy, liver disease, and the elderly population (2, 3). GBS, even when it is asymptomatic, has been associated with adverse pregnancy outcomes, for example, premature rupture of the membranes, pre-term delivery, and low birth weight (4, 5). GBS has also emerged as a significant cause of acute urinary tract infection (UTI) in adults, especially in the females in the 1990's, and numerous forms of GBS infection of the urinary tract have been reported in subsequent studies(6). Urinary tract infection (UTI) is the most common bacterial infection in humans throughout the world, and one third of the women will suffer from UTI in their lifetime (7, 8). GBS is one of the most important uropathogens that causes acute bacterial urinary tract infection in older individuals and persons with chronic medical complications. The range of GBS UTI involves asymptomatic bacteriuria, pyelonephritis, cystitis, and urosepsis. GBS can be cultured from the urine of about two percent of all cases clinically suspected to UTI (9, 10).Yet, Penicillin G and ampicillin are the antibiotics of choice for prevention of perinatal GBS infections, and clindamycin and erythromycin are also prescribed as treatment for women with penicillin allergy (11). In 1992, penicillin-intermediate GBS strains were isolated from clinical practices (12). However, with the widespread use of antibiotics (mainly ampicillin and amoxicillin in Iran) for prophylaxis against some infections, there is a potential for the emergence of penicillin-tolerant GBS strains. Throughout the last decade, resistance of GBS strains to clindamycin and erythromycin has also been increased in numerous countries with some geographical variations. The increasing tendency in the rate of resistance to clindamycin and erythromycin among GBS strains has enhanced worry about the use of the antimicrobial agents as alternative agents for the prophylaxis or treatment of GBS infections. Plasmid-mediated resistance to antimicrobial agents has also been expected to occur in GBS and can facilitate the development of multidrug resistance (13-15).

The objective of this study was to determine antibiotic susceptibility pattern of GBS strains isolated from midstream urine specimens of patients with the characteristics of UTI, admitted to the Pars Hospital, Tehran, for help in the selection of proper antibiotics in the treatment of patients.

2. Objectives

Objectives: The objective of this study was to determine antibiotic susceptibility pattern of GBS strains isolated from midstream urine specimens of patients with the characteristics of UTI, admitted to the Pars hospital, Tehran, Iran.

3. Materials and Methods

Our study was performed over a three-month period from April to June 2014. Midstream specimens of urine were collected from outpatients suspected of having a microbial urinary tract infection, which had not received any antibiotics and referred to the Pars hospital, a general private hospital in Tehran, Iran. The age range of the patients was from 1 to 96 years. Primary isolation of uropathogens was performed by a surface streak plate technique on Tryptic Soy agar (TSA) with 5% sheep blood (BAP) and incubation of the petri dishes for 24 hours at 35°C in a 5% CO2 incubator (all media were provided from Merck Co., Germany). Then suspected colonies with beta-hemolytic appearance, whose colony counts were $\geq 10^4 CFU.mL^{-1}$, examined by Gram stain and catalase test. The definitive identification of GBS isolates carried out using gram stain, catalase test, CAMP reaction, bacitracin and trimethoprim sulfamethoxazole susceptibilities, bileesculin, and 6.5% NaCl tests. After identification of uropathogenic GBSs, antibiotic susceptibility test was performed according to the Clinical Laboratory Standards Institute (CLSI) Guidelines 2012, using antibiotic disks purchased from Rosco, Co.

3.1 Ethics statement

All data in this research article were analyzed anonymously. So, no consent from the patients was required and the ethics committee did not have to be approached

4. Results

Patients with urine specimens demonstrating pure cultures of ≥ 104 $CFU.mL^{-1}$ were considered cases of UTI. Gram positive cocci, catalase negative, susceptible strains to bacitracin, and positive reaction to CAMP test considered as GBS strains. A total of 2101 urine specimens were received during a three-month period from April to June 2014. GBS strains were isolated from 264 (21.1%) cases (out of 1249 positive bacterial urine cultures). One hundred and eleven strains were isolated from male patients (42.4%) and one hundred and fifty-three from female patients (57.95%). The higher prevalence was recorded in the 15-44 and 45-64 age groups. Figure 1 shows the age distribution of the studied patients.

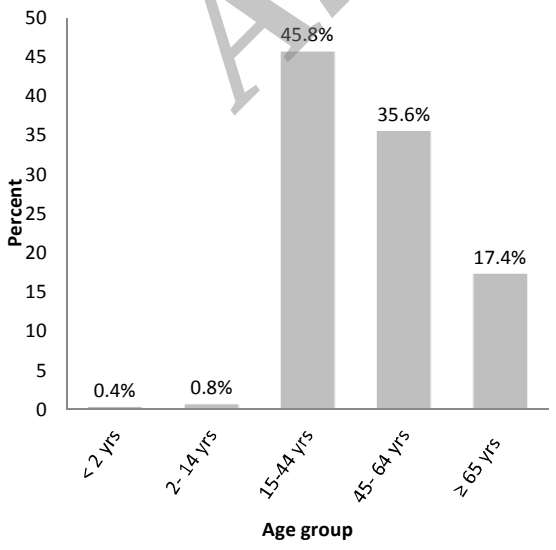

Figure. 1. The age distribution of the patients with GBS UTI in our study

Table 1 shows the antibiotic susceptibility pattern of GBS strains isolated from urine cultures. All of GBS strains showed susceptibility to penicillin, vancomycin, and linezolid. A majority of the strains (>80%) also showed susceptibility to nitrofurantoin, ampicillin, ciprofloxacin, levofloxacin, erythromycin, and clindamycin. The least effective antibiotic was tetracycline, to which only about 12% of the isolates showed susceptibility.

Table 1. The antibiotic susceptibility pattern of GBS strains isolated from urine of patients in Tehran, Iran

Antibiotic	Valid percentage of:		
	S	I	R
Ampicillin	99.6 (263of 264)	0	0.4 (1of264)
Clindamycin	84.7 (224of264)	0.8 (2of264)	14.6 (38of264)
Ciprofloxacin	93.5 (247of 264)	1.5 (4of 264)	5 (13of 264)
Erythromycin	82.6 (218of 264)	1.2 (3of 264)	16.2 (43of 264)
Levofloxacin	95.8 (253of 264)	0	4.2 (11of 264)
Linezolid	100 (264of 264)	0	0
Nitrofurantoin	99.6 (236of 264)	0	0.4 (1of 264)
Penicillin	100 (264of 264)	0	0
Tetracycline	12 (32of 264)	7.8 (20of 264)	80.2 (212of 264)
Vancomycin	100 (264of 264)	0	0

*I= Intermediate, *R= Resistance, *S= Susceptible

5. Discussion

GBS is still susceptible to many antibacterial agents, particularly beta-lactam antibiotics. Penicillin G and ampicillin are the antibiotics widely used against GBS. In this study, antibiotic susceptibility testing was performed for 264 GBS strains isolated from urine samples of patients with suspected UTI. The antimicrobial susceptibility testing was carried out using the antibiotics that were considered to have potential clinical utility for the prophylaxis and treatment of GBS infections and/or colonization therefore had susceptibility breakpoints recommended by the (CLSI) Guidelines 2012. The evaluated results of antibiotic susceptibility tests in this study indicated that approximately all isolated uropathogenic GBS strains were uniformly sensitive to penicillinin vitro, as different authors around the world reported it (15, 16). In the reports from Iran, many studies have also shown high susceptibility of GBS isolates to Penicillin G and ampicillin; for example, Jannati *et al.* indicated susceptibility of all GBS isolates to these antibiotics, although there is a report of high resistance (89.4%) to penicillin among uropathogenic GBS isolates by Rahbar and colleagues (17, 18). Yasini and colleagues also showed that all GBS strains isolated from the vagina were sensitive to penicillin and cefazolin. Also, of all the isolates, 97.2% were sensitive to ampicilin, 80.5% to erythromycin, and 83.4% to clindamycin (19). In our study, there was also a less resistance rate to ampicillin, erythromycin, and clindamycin incomparison with the Rahbar*'s* study (18).

In the present study, we detected ampicillin resistant GBS isolates. Comparable results had been described in other studies worldwide (20). It is indicated that decrease in

susceptibility to beta-lactam antibiotics in GBS strains is due to structural modification in penicillin binding proteins (21). It is important to monitor the possible emergence of penicillin treatment failure in vivo. The emergence of GBS strains with higher MIC levels to penicillin and ampicillin could show an emerging public health problem emphasizing the need for monitoring the susceptibility of these organisms over the time.

In the present study, some GBS isolates were shown to be resistant to clindamycin and erythromycin, and only 12% of the isolates were shown to be susceptible to tetracycline. Resistance to these antibacterial agents in GBS populations is common worldwide; however, the antibiotic resistance rate could be varied based on the kind of geographical area and the length of study conduction. Routine examinations should be carried out to determine the susceptibility of GBS strains against macrolides and lincosamides because identification of erythromycin-sensitive phenotypes can be advantageous in the choice of an appropriate alternative therapy for penicillin-allergic patients.

6. Conclusion

In conclusion, the results of the present study confirm the universal susceptibility of GBS strains to the penicillin family and assert the use of penicillin or ampicillin as the first drug of choice for treatment and prophylaxis against GBS infections. However, it is important to perform antibiotic susceptibility testing whenever penicillin could not be prescribed.

Conflict of Interests

The authors declare there is no conflict of interest regarding the publication of this paper.

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Authors' Contribution

The study idea and design was done by Zahra Tayebi and Horieh Saderi: analysis and interpretation was done by Hamidreza Houri and Mehrdad Gholami; study supervision was done by Saeed Samie and Shahram Boroumandi; drafting of the manuscript, critical revision of the articlewas done by Mehrdad Gholamiand Hamidreza HouriandZahra Tayebi.

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