ANTIMICROBIAL SENSITIVITY PATTERN OF URINE ISOLATES FROM ASYMPTOMATIC BACTERIURIA DURING PREGNANCY

[1]AZIZ MARJAN KHATTAK, [2]HABIB-ULLAH KHAN, [3]IHSAN-ULLAH MASHUD
[4]BUSHRA ASHIQ AND [5]SYED HUMAYUN SHAH
Departments of [1,5]Pathology, [2,3]Medicine, Gomal Medical College, Dera Ismail Khan – Pakistan
[4]Departments of Microbiology, BMSI, JPMC, Karachi

Screening women for asymptomatic bacteriuria (ASB) on the first antenatal visit is a part of standard obstetric care. Treating women with ASB decreases the chances of maternal and foetal complications. This study was conducted to find out the spectrum of urine pathogens and their drug susceptibility pattern for ASB during pregnancy. The study was conducted in the Basic Medical Sciences Institute, Jinnah Postgraduate Medical Centre Karachi, from September 2001 to March 2002. Two hundred and ninety women, apparently normal with confirmed pregnancy, were registered. A voided midstream urine specimen was collected and cultured. A significant growth i.e. ≥10^5 organisms/ml was identified with Analytical Profile Index 20 tests for identification of Enterobacteraceae (API-20-E) and for Gram positive cocci by other standard methods. The prevalence of ASB was found 6.2%. Antimicrobial sensitivity was determined by disc diffusion Kirby Bauyer method after matching the turbidity with 0.5 McFarland·s standard. Most of the recommended drugs were found to have encouraging results, however, Escherichia coli showed 66.67% resistance to ampicillins and sulphonamides. Enterobacters showed 100% resistance to ampicillins, cephalosporins and nitrofurantoin. Staphylococcus saprophyticus showed 66.67% resistance to ampicillins and sulphonamides. It was concluded that detection of ASB during pregnancy and appropriate use of antimicrobials is only possible after culture of urine. Empirical anti-microbial therapy cannot be relied upon because of possible risk of resistance.

Key Words: *Asymptomatic bacteriuria, Pregnancy, Urine isolates, Antimicrobial sensitivity.*

INTRODUCTION

Despite impressive advances in understanding the pathogenesis, diagnosis and treatment of urinary tract infection (UTI), these infections still contribute a major problem to obstetrics patients.[1] Asymptomatic or symptomatic bacteriuria can result in temporary or permanent renal damage in the pregnant women,[2,3] and can also have effects on the foetus in the form of intra uterine growth retardation, prematurity, increased risk of perinatal death and congenital abnormalities.[4,5]

Symptomatic UTI is usually preceded by a period of asymptomatic bacteriuria.[6] It is suggested that by identifying and treating patients in the stage of asymptomatic infection, future complications can be prevented.[7] There is a great variation in clinical presentation of ASB. In some individuals bacteriuria causes no symptoms at all till perpurium and only screening programme can reveal its existence.[8] The Canadian task force recommends urine culture at 12-16 weeks of pregnancy.[5,9] Although urine screening by culture is expensive and time consuming, it remains the gold standard method for diagnosing bacteriuria during pregnancy.[10] Diagnosis and treatment should be aggressively pursued instead of being avoided because of the unsubstantial fear of foetal complications due to treatment.[11] Treatment decreases the occurrence of adverse outcome of pregnancy due to bacteriuria.[7,12,13].

The present study was conducted to find out the drug susceptibility pattern recommended as safe therapy for bacteriuria during pregnancy.

MATERIAL AND METHODS

This study was conducted in the Department of Microbiology, Basic Medical Sciences Institute, Jinnah Postgraduate Medical Center, Karachi, from September 2001 to March 2002.

Pregnant women apparently normal, attending their first antenatal visit at the Neonatal care and safe motherhood center, Jinnah Postgraduate Medical Center were registered. Patients with pyrexia and those taking antibiotics or having taken antibiotics in the previous two weeks were excluded from the study.

Urine specimen of at least 4 hours stay in bladder was collected in a wide mouth sterile container of 100 ml capacity.[14] The specimen was promptly transported and processed. In case of delay it was refrigerated at 4°C for not more than 1 hour. Aerobic culture was performed on Cystein Lactose Electrolyte Deficient (CLED) agar with the help of standard platinum closed loop. After 18-24 hours incubation at 37°C, the growth of $\geq 10^5$ organisms/ml was considered as significant. The count of 10^4-$<10^5$/ml was taken as doubtful and repeated. Gram-negative organisms were identified by Analytical Profile Index 20 tests for identification of Enterobacteraceae (API-20-E). The inoculums from 3-4 pure colonies of bacteria were taken to log phase in tryptic soya broth by incubation for 2-4 hours. The turbidity so obtained was compared with and adjusted to 0.5 Mac-Farmland turbidity. Mueller Hinton agar was flooded with broth. The excess of broth was drained and sides of petri dishes were cleaned with sterile gauze. Antimicrobial discs were applied within 10 minutes of charging the media. Mueller Hinton agar with 4% lysed sheep blood was used to test for Streptococcus agalactiae and sulphonamide discs. Quality control strain Escherichia coli, American Type Culture Collection (ATCC 25922) was used for gram-negative organisms and Staph. aureus (ATCC 25923) for gram-positive organisms. Zones of inhibition around discs on Mueller Hinton agar were recorded after 18-24 hours incubation according to sensitivity and resistant standards specified by the National Committee for Clinical Laboratory Standards (NCCLS) recommended by WHO.[15,16]

RESULTS

The spectrum of organisms isolated is given in Table 1. Esch. Coli was the most common pathogen 12/18 (38.89%). Enterobacter species and Staph. saprophyticus were found in 3/18 each (16.68%). The other pathogens were 1/18 each (5.55%). In terms of Gram-staining, infected urine from pregnant women had Gram-negative bacilli in 12/18 (66.67%), Gram-positive cocci in 5/18 (27.78%) and yeast cells 1/18 (5.55%) Table 2.

Antimicrobial susceptibility pattern is shown in Table-3. Esch. coli showed 88.89% sensitivity to fosfomycin and nitrofurantoin each and 77.78% to cephalothin. While resistance was shown as 77.78% to sulphamethoxazole, 66.67% to each ampicillins, trimethoprim and cotrimoxazole. Enterobacter species showed 100% sensitivity to cephaloithin, 77.78% to fosfomycin, 66.67% to both sulphamethoxazole and cotrimoxazole. While 100% resistance to ampicillins and nitrofurantoin and 66.67% to trimethoprim. Staph.saprophyticus showed 100% sensitivity to nitrofurantoin and 66.67% to each cephalothin, fosfomycin, erythromycin and sulphamethoxazole. While 66.67% resistance to ampicillins, trimethoprim and cotrimoxazole. The percentage sensitivity of other pathogens could not be given, as these were isolate only in single number.

Table 1: *Spectrum of urinary pathogens isolated from urine samples of pregnant women.*

Pathogens Isolated	No. of Positive Cultures	Percentage
Esch. coli	7	38.89%
Staph. saprophyticus	3	16.68%
Enterobacter species	3	16.68%
Staph. aureus	1	05.55%
Streptococcus agalactiae	1	05.55%
Proteus mirabilis	1	05.55%
Serratia marcescens	1	05.55%
Candida albicans	1	05.55%
Total	18	100%

Table 2: *Percentage of isolated pathogens in culture positive patients.*

S. No	Pathogen	Percentage
1	Gram negative bacilli	66.67%
2	Gram positive cocci	27.78%
3	Yeast cells	05.55%

DISCUSSION

The subjects were from indigent group of population, women attending first antenatal clinic. Midstream voided urine was taken. The samples were processed for aerobic culture and sensitivity. The causative organisms of ASB in our study were the same causing symptomatic bacteriuria during fertile life of women as reported by Nathaniel (1985).[17]

Asymptomatic bacteriuria is a strong predictor of symptomatic urinary tract infection later in pregnancy (MacClean 2001).[18] Detection and treatment of asymptomatic bacteriuria is important, as 25 to 30% of pregnant women will develop symptomatic urinary tract infections.[3,19] Untreated asymptomatic bacteriuria during pregnancy leads to the development of symptomatic cystitis in approximately 30% and pyelonephritis in 50% of patients.[5,20] Significant bacteriuria ($\geq 10^5$ orga-

nisms/ml) during pregnancy is an established risk factor for other serious complications e.g. pre-term delivery, foetal pre-maturity and low birth babies, reported in various studies as 20-40%[11] and 66%.[4,21]

The results of our study, showing Gram-negative bacilli in 66.67%, Gram-positive cocci in 27.78% and yeast cells in 5.55%, were similar to the findings of Van Norstrand et al (2000),[22] who also found Gram-negative Bacilli in 67%, Gram-positive cocci in 25% and yeast cells in 8%.

The discs of antimicrobials applied were based on safe oral preventive therapy during pregnancy, recommended in the literature.[5,13] The antimicrobial susceptibility tests for Escherichia coli, the primary pathogen, showed good sensitivity to fosfomycin, nitrofurantoin each 88.89%, and high resistance to ampicillins and sulphonamides (66.67%), and cephalosporins (77.78%). Our resistance results were higher than the reports of Stamm and Hooton (1993)[23] who described that 20-30% of Escherichia coli were resistant to amplicillins, cephalosporins and sulphonamides by in-vitro testing. Abbott (1994)[11] also found 30% rate of resistance of Escherichia coli urine isolates to ampicillins. In this study, Enterobacter species were found to be resistant to ampicillins, cephalosporins and nitrofurantoin as 100% and Staphylococcus saprophyticus resistance to ampicillins, trimethoprim and cotrimoxazole each 66.67%. The results are not according to the reports of Anriole and Patterson (1991),[24] who described that most organisms isolated during screening asymptomatic bacteriuria are sensitive to antimicrobials. This may be probably due to indiscriminate use of antibiotics in our society.

Although this study on anti-microbial pattern is based on limited number of urine isolates, further studies with large number of patients will broaden the base of this study.

CONCLUSION

Sensitivity pattern of urine isolates in patients with asymptomatic bacteriuria shows resistance to some drugs commonly recommended for safe use during pregnancy. Appropriate anti-microbial use is only possible after culture and sensitivity report to improve the health services during pregnancy.

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Table 3: *Antimicrobial sensitivity of urine isolates of various species.*

Micro-organism	AMP 10µg		SXT 300µg		W 5µg		COT 300µg		CEF 30µg		F 300µg		FOS 50µg		ER 15µg	
	Sensitive (%)	Resistant (%)	Senitive (%)	Resistant (%)	Sensitive (%)	Resistant (%)	Sensitive (%)	Resistant (%)	Sensitive (%)	Resistant (%)	Sensitive (%)	Resistant (%)	Sensitive (%)	Resistant (%)	Sensitive (%)	Resistant (%)
Esch. coli	33.33	66.67	22.22	77.78	33.33	66.67	33.33	66.67	77.78	22.22	88.89	11.11	88.89	11.11	-	-
Enterobacter species	0	100	66.67	33.33	33.33	66.67	66.67	33.33	100	0	0	100	77.78	33.32	-	-
Staph. saprophyticus	33.33	66.67	66.67	33.33	33.33	66.67	33.33	66.67	66.67	33.33	100	0	66.67	33.33	66.67	33.33

Key: AMP (Ampicillin), F (Nitrofurantoin), SXT (Sulphamethoxazole), FOS (Fosfomycin), W (Trimethoprim), ER (Erythromycin), COT (Cotrimoxazole), CEF (Cephalothin).