INTRODUCTION

Urinary Tract Infection (UTI) refers to both microbial colonization of the urine and tissue invasion of any structure of the urinary tract. Pregnancy enhances the progression from asymptomatic to symptomatic bacteriuria which could lead to pyelonephritis and adverse obstetric outcomes such as preterm delivery, low-birth weight and higher fetal mortality rates. This study was undertaken to determine the prevalence of asymptomatic bacteriuria in pregnancy, its causative agents and their antimicrobial susceptibility pattern, and also to determine the relationship between asymptomatic bacteriuria and pyuria. The total number of participants who finished the study was 392. The mean age of the participants was 29.76 ± 6.71 (range, 21-37 years). Of the 392 urine specimens processed, 102 (26.0%) showed significant bacteriuria. The most common organism causing bacteriuria was Escherichia coli. The sensitivity pattern of the isolated organisms revealed that all were sensitive to ciprofloxacin and gentamicin at very high percentages. 200 (51.5%) women had more than 5 pus cells in urine specimens from which 50 (12.75%) had positive cultures. Women with higher number of pus cells in urine specimen had significantly higher asymptomatic bacteriuria (p < 0.0001). In conclusion, screening of bacteriuria in pregnancy and proper treatment must be considered as an essential part of antenatal care in Nepalese community. To prevent asymptomatic bacteriuria complications, all pregnant women should be screened at the first antenatal visit. A negative test for pyuria is not a reliable indicator of the absence of asymptomatic bacteriuria in pregnant women. Keywords: asymptomatic bacteriuria, pyuria, antenatal care, urinary tract infection, E.coli

MATERIALS AND METHODS

Sample collection and setting

The study was conducted in outpatient clinic of gynecology department of Chitwan Medical College Teaching Hospital and has been approved by Institutional Review Committee of Chitwan Medical College (CMC-IRC) on August, 2012. A

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ABSTRACT

Urinary Tract Infection (UTI) refers to both microbial colonization of the urine and tissue invasion of any structure of the urinary tract. 1 Bacteria are most commonly responsible although yeast and viruses may also be involved. Asymptomatic bacteriuria, in which urine culture reveals a significant growth of microorganisms, accounts for approximately 10 percent of OPD visits by pregnant women. 2 Approximately 10 percent during pregnancy are the same as those found in non-pregnant patients. Escherichia coli accounts for 80 to 90% of infections. 3 Other gram-negative rods such as Proteus mirabilis and Klebsiella pneumoniae are also common. In the 1960s, Kass et al. noted the subsequent increased risk of developing pyelonephritis in patients with asymptomatic bacteriuria. 4-6

Asymptomatic bacteriuria is common, with a prevalence of 10 percent during pregnancy. 1,4 Thus, routine screening for bacteriuria is advocated. Untreated asymptomatic bacteriuria leads to the development of symptomatic cystitis in approximately 30 percent of patients and can lead to the development of pyelonephritis in up to 50 percent. 5 Asymptomatic bacteriuria is associated with an increased risk of intrauterine growth retardation and low-birth-weight infants. 6,7 There are a number of conditions associated with an increased prevalence of asymptomatic bacteriuria in pregnancy. Low socioeconomic status, sickle cell traits, diabetes mellitus and grand multiparity have been reported; each is associated with two-fold increase in the rate of bacteriuria. 8 A major study comparing normal and high-risk pregnant women reported 6.0% prevalence in healthy women; 12.2% rate in diabetic women and 18.7% in women with a previous history of urinary tract infection. 9 Maternal anemia has been reported to be associated with both asymptomatic bacteriuria and pyelonephritis, but an association with covert bacteriuria has not yet been confirmed. Also there is evidence that when there is no symptom, untreated bacteriuria in pregnancy may lead to less favorable pregnancy outcomes and complications like preterm delivery, low birth weight, pre-eclamptic toxemia and anemia of pregnancy. 10-12 Some studies have shown that 18 weeks of gestation is the optimal time for performing screening culture for detecting bacteriuria. 13 The present study was undertaken to determine the prevalence of asymptomatic bacteriuria in pregnancy; its causative agents and their antimicrobial susceptibility pattern, and also to determine the relationship between asymptomatic bacteriuria and pyuria.
Data Analysis

Antibiotic susceptibility testing was conducted on 392 urine specimens collected from asymptomatic bacteriuric women during April, 2012 to September, 2012. All participants were examined at the first antenatal visit and the maximum gestational age was 18 weeks. None of the patients had any signs or symptoms of classical UTI on examination.

Asymptomatic bacteriuria was defined as the presence of ≥105/ml colonies of the same bacterial species in two consecutive midstream urine samples without any symptom of urinary tract infection. On each visit, two consecutive midstream clean catch urine samples, one on the day of the visit and another on the next day of the visit, were collected in a sterile wide mouth screw capped container with aseptic precautions. The patients were individually instructed about the technique for collecting clean voided midstream urine specimens. All patients were asked to wipe their labia with soapy water and rinse well, then after urinating a little in the toilet fill the container (clean catch method). All the specimens were sent to the microbiology laboratory and processed on the same day. Standard microbiological techniques were used in the culture of all urine specimens and in the identification of the isolates. One μl of uncentrifuged urine specimens were aseptically inoculated, using standard loops, onto sheep blood agar (SBA) plates containing 6% blood and cysteine lactose electrolyte deficient (CLED) agar plates. The plates were incubated aerobically for 24 to 48 hours at 37°C. The plates were read at the end of the incubation period. Colony counts equal to or more than 105/ml was considered as significant growth.

Antibiotic sensitivity test

Antibiotic susceptibility testing was carried out using the Kirby-Bauer disc diffusion technique on Muller-Hinton agar and commercial antibiotic discs (Oxoid, United Kingdom) were used for antimicrobial testing.14 The antibiotic discs used were: Ampicillin (10 μg), Nalidixic Acid (30 μg), Nitrofurantoin (300mg), Cephalexin (30 μg), Gentamicin (10 μg), Trimethoprim-Sulphamethoxazole (1.25/23.75 μg) and Ciprofloxacin (5 μg). The antibiotic disc impregnated culture plates were incubated at 37°C overnight. The diameter of the zone of inhibition was measured and recorded as resistant or susceptible. For the test of proteinuria, the end of the reagent strip was dipped in the fresh urine for approximately 1 second and shaken off by tapping the strip on the side of the container. After 30 to 60 seconds the test strip was compared with the color scale (color range from yellow for “negative” and through yellow-green and green blue for “positive”). For microscopy about 10 ml of well-mixed urine sample was centrifuged at 2000xg for 5 minutes. A drop of the deposit was examined microscopically at 40x for the presence of pus cells, red blood cells, epithelial cells, casts and crystals.

Data Analysis

The Sigma plot version 12 for windows was used for data analysis. The results are expressed as mean value ± standard deviation. Chi-square test was used for comparison between groups. A two-tailed p-value less than 0.05 was considered statistically significant.

RESULTS

The total number of participants who finished the study was 392. The mean age of the participants was 29.76 ± 6.71 (range, 21-37 years). Of the 392 urine specimens processed, 102 (26.0%) showed significant bacteriuria. Thus the prevalence of different types of causative organism of significant bacteriuria was 26.0%. The frequency of the microorganisms isolated is shown in Table 1. The commonest organism causing bacteriuria was Escherichia coli. The sensitivity pattern of the isolated organisms revealed that all were sensitive to ciprofloxacin and gentamicin at very high percentage (Table 2). The organisms showed resistance to currently preferred urinary antibiotics and chemotherapeutic agents like co-trimoxazole, norfloxacin, and cephalexin (Table 2). 200 (51.5%) women had more than 5 pus cells in urine specimens from which 50 (12.75%) had positive cultures. Women with higher number of pus cells in urine specimen had significantly higher asymptomatic bacteriuria (p=0.000, p<0.0001).

<table>
<thead>
<tr>
<th>Isolates</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Escherichia coli</td>
<td>70.8</td>
</tr>
<tr>
<td>Klebsiella</td>
<td>16.7</td>
</tr>
<tr>
<td>Group B Streptococcus</td>
<td>8.3</td>
</tr>
<tr>
<td>Proteus mirabilis</td>
<td>4.2</td>
</tr>
</tbody>
</table>

Table 1: Microorganisms isolated in positive cultures

<table>
<thead>
<tr>
<th>Drugs</th>
<th>E. coli</th>
<th>Klebsiella</th>
<th>Group B Streptococcus</th>
<th>Proteus mirabilis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ciprofloxacin</td>
<td>89.2</td>
<td>87.1</td>
<td>95.4</td>
<td>95.7</td>
</tr>
<tr>
<td>Gentamicin</td>
<td>89.6</td>
<td>88.1</td>
<td>69.7</td>
<td>65.1</td>
</tr>
<tr>
<td>Ampicillin</td>
<td>50.3</td>
<td>0</td>
<td>35.1</td>
<td>10.2</td>
</tr>
<tr>
<td>Nitrofurantoin</td>
<td>65.1</td>
<td>73.1</td>
<td>82.2</td>
<td>0</td>
</tr>
<tr>
<td>Cephalexin</td>
<td>28.0</td>
<td>19.2</td>
<td>17.7</td>
<td>15.4</td>
</tr>
<tr>
<td>Nalidixic acid</td>
<td>68.2</td>
<td>42.5</td>
<td>48.6</td>
<td>31.1</td>
</tr>
<tr>
<td>Co-trimoxazole</td>
<td>12.4</td>
<td>29.4</td>
<td>0</td>
<td>2</td>
</tr>
</tbody>
</table>

Table 2: Antimicrobial susceptibility pattern of organisms causing bacteriuria in pregnant women

DISCUSSION

Urinary tract infections are remarkably common in women. Some 20% women in the age range 20-65 years suffer from at least one attack per year, 50% develop a urinary tract infection within their life time.16 Not surprisingly infections of the urinary tract are the most common bacterial infections encountered during pregnancy. These can be both symptomatic and asymptomatic. Asymptomatic bacteriuria during pregnancy is a common and important medical condition, which will result in overt renal infections such as pyelonephritis if not detected and treated. Smaill et al,18 showed that on an average treating seven pregnant women with asymptomatic bacteriuria results in prevention of one episode of pyelonephritis. Ten percent of pregnant women attended in an antenatal clinic had symptomatic urinary tract infections 4. In another study by Khatun et al. (1985),6 it was found that 30% of clinically healthy pregnant women had asymptomatic bacteriuria. Findings of the present community based study indicate that the asymptomatic bacteriuria in pregnancy is a major health problem in Srinagar city. Observed from this study that E. Coli was the commonest pathogen responsible for bacteriuria. It is consistent with the findings of Rahman et al. (1990).19, 14 and Ahmed et al. (1996).16, 17, 18 Like the other studies,16, 17, 18 the findings of our study also indicate that ciprofloxacin is highly effective. The most effective in-vitro agents were found to be gentamicin among the injectables and ciprofloxacin among the orally administered ones. Other useful oral antibiotics were nitrofurantoin and nalidixic acid. The organisms showed resistance to currently prefer urinary antibiotics and chemotherapeutic agents like co-trimoxazole and cephalexin. This fact indicates that urinary pathogens became resistant day by day to the commonly used antibiotics in Nepal. This may be due to wide spread and indiscriminate use of the drugs. There are many studies,19, 20 that link so many pregnancy complications like hypertensive disorders in pregnancy, low birth weight, premature with symptomatic bacteriuria.
Moreover, for the last two decades, asymptomatic bacteriuria has also been identified as a risk factor of similar pregnancy complications. The results of the present study also agree with these findings. The association between asymptomatic bacteriuria and prematurity is established. But the mechanism is not well defined yet. Several investigators have observed a high incidence of pyelonephritis in bacteriuric pregnant mothers.

In conclusion, screening for bacteriuria in pregnancy and proper treatment must be considered as an essential part of antenatal care in Nepalese community. A negative test for pyuria is not a reliable indicator of the absence of asymptomatic bacteriuria in pregnant women. Thus, all urine specimens, regardless of leukocyte count, should be sent for culture and sensitivity.

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REFERENCES

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