ISSN Print: 2394-7500
ISSN Online: 2394-5869
Impact Factor: 5.2
IJAR 2018; 4(8): 21-25
www.allresearchjournal.com
Received: 06-06-2018
Accepted: 07-07-2018

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Prevalence of asymptomatic bacteriuria in married non-pregnant women

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Abstract
UTI is twice more common in pregnant women than age matched non-pregnants. UTI may occur with or without symptoms; the latter is known as asymptomatic bacteriuria (ASB). The prevalence of ASB in non-pregnant women rises with age at the rate of 1% for each decade of life. However, UTI in married non pregnant women have not been adequately studied in developing countries. This study is therefore aimed at finding out the incidence of ASB in married non pregnant women. This study was carried out over a period of two years in the Department of Microbiology in association with the Department of Obstetrics and Gynecology, of T. N. Medical College and B. Y. L. Nair Charitable Hospital. Total patients included were randomly selected 300 married, non-Pregnant women reporting to ANC clinic of the hospital of age group of 18 to 40 year. Collection, transportation, microscopic examination and antibiotic sensitivity testing was carried out by standard procedures. In the non-pregnant women, the majority cases of UTI were in 24 to 29 years (52.66%) followed by in age group 18 to 23 years (36.33%). While least was seen in age group 36-40 years (4.66%). The highest cases of ASB was found in age group of 30 to 35 years (15%) followed by in age group of 36 to 40 years (14.3%). The least was in age group of 18 to 23 years (0.9%). Prevalence of ASB in Married, non-pregnant women is 3.66% and only single pathogen could be detected and none (0%) of the sample showed mixed infection of two pathogens. *E. coli* was the predominant isolate (36.36%) in the study and the least was Klebsiella oxytoca (9.09%). The antibiotic sensitivity of uropathogens obtained were as follows. *E. coli* was highly sensitive to Piperacillin + Tazobactum (100%) and Cefotaxime (100%). *Klebsiella pneumoniae* showed 100% sensitivity to Cephalexin, while *Klebsiella oxytoca* showed 100% sensitivity to Cephalexin. *Staphylococcus aureus* were found 100% sensitive to Cephalexin, while *Staphylococcus epidermidis* were 100% sensitive to Ampicillin. The results of this work showed an association of ASB with age and gravidity and it appeared to be multifactorial. A screening for ASB in married non-pregnant women must be done to discover the infected cases, which would allow early treatment to avoid the complications.

Keywords: Asymptomatic bacteriuria, UTI, married women, non-pregnant women

1. Introduction

Urinary tract infection (UTI) is one of the most important causes of morbidity and mortality [1]. UTI is second only to the respiratory tract infection in acquiring microbial infection especially in females. It is more common in pregnant than in non-pregnant women [2]. UTI is twice more common in pregnant women than age matched non-pregnants [3]. UTI may occur with or without symptoms; the latter is known as Covert or Asymptomatic bacteriuria [4]. Asymptomatic bacteriuria *(*ASB) is often a dynamic process that is it may wax and wane in particular women. ASB is defined as 10 [5] bacteria per ml of one or more on two clean-catch cultures taken on separate days [5]. It is identified in 5-10% of women at their initial prenatal visits [6]. While ASB is generally considered not clinically important in women who are not pregnant, during pregnancy it can be associated with a variety of adverse obstetrics outcomes and medical conditions [7].

Many of the risk factors for bacteriuria in pregnancy contribute to UTI in non-pregnant women. The prevalence of ASB in non-pregnant women rises with age at the rate of 1% for each decade of life. The prevalence of bacteriuria not only increases with age but also with sexual activity and parity [8].

A number of conditions are associated with an increased prevalence of ASB, which includes low socioeconomic status, sickle cell trait, diabetes mellitus, grand multiparity [7]. In married, non-pregnant women, ASB may not have a significant impact and may not be persistent. The epidemiology of bacteriuria in pregnancy is similar to that seen in non-pregnant women. The risk factors for bacteriuria and the etiologic agents of bacteriuria in pregnancy mirror those in non-pregnant women. In most patients, bacteriuria in pregnancy usually reflects prior colonization rather than acquisition during the pregnancy itself [8].

Determination of the number and type of bacteria in the urine is an extremely important diagnostic procedure [9]. The quantitative confirmation of bacteriuria following an initial positive non specific screen is useful in establishing the etiologic agent and susceptibility [8]. For a women with ASB two consecutive positive specimens are necessary for diagnosis [10]. In India, work done on this topic is scanty.Studies conducted in India suggest a prevalence of bacteriuria as 2 to 12% among pregnant women that is slightly higher than that from the west [3]. However, UTI in married non pregnant women have not been adequately studied in developing countries. This study is therefore aimed at finding out the incidence of bacteriuria in married non pregnant women.

2. Material and methods

This prospective longitudinal study was carried out over a period of two years, from January 2003 to December 2004 after taking the permission from Institutional Ethics committee of T. N. Medical College and B. Y. L. Nair Charitable Hospital, Mumbai. It was conducted in the Department of Microbiology in association with the Department of Obstetrics and Gynecology, of T. N. Medical College and B. Y. L. Nair Charitable Hospital.

Participants

The study included the patients from Out-door and Indoor patients of Gynecology department which were recruited for bacteriologic evidence of ASB by microscopy, culture and chemical examination. Total patients included were randomly selected 300 married, non-Pregnant women reporting to ANC clinic of the hospital of age group of 18 to 40 years. Those subject who were showing sign and symptoms of urinary tract infection, suffering from diabetes, under antibiotic treatment in the past or treatment with steroids for any infection/ indication was excluded from our study. Counseling for enrollment procedure in the study was done. Detailed data from the patients were recorded in a specially formulated structured proforma.

Collection and microbiological analysis

Collection and transportation of urine was carried out by standard procedures [11]. Further microscopic examination of urines was done by Wet film examination [12] and Gram staining using Hucker's modification [13]. Culturing of uncentrifuged urine specimen was done by standard methods using growth promoting mediums for presence or absence of haemolysis and lactose fermentation. Identification of the isolate was done on the basis of morphological, cultural characteristics and Rapid Biochemical Identification Kit [14].

Antibiotic sensitivity testing

Antibiotic sensitivity testing of the isolate obtained from clinical samples was carried out by Kirby - Bauer disk diffusion method according to HiMedia Manual 1998 [14, 15]. Quality control checks of all the medium was carried out by inoculating standard strains of bacteria.

3. Results

Table 1: Age distribution of Non-pregnant married women population studied for ASB. n_1=300 and n_2=11

Sr. No.	Age in years	Total number of cases screened (n_1)	Cases with asymptomatic bacteriuria (n_2)
1	18-23	36.33%	0.9%
2	24-29	52.66%	3.16%
3	30-35	6.33%	15.0%
4	36-40	4.66%	14.3%

Table 1 shows the age distribution of the non-pregnant women group having ASB.In the non-pregnant women, the majority cases were in 24 to 29 years (52.66%) followed by in age group 18 to 23 years (36.33%). While least was seen in age group 36-40 years (4.66%).

The highest cases of ASB was found in age group of 30 to 35 years (15%) followed by in age group of 36 to 40 years (14.3%). The least was in age group of 18 to 23 years (0.9%).

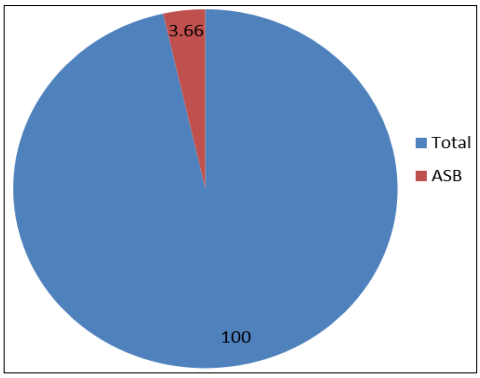

Fig 1: Positive cases of ASB in married, non pregnant women population n=300

From the above Chart 1, it is seen that the prevalence of ASB in Married, non-pregnant women is 3.66%.

Table 2: Culture findings in ASB in Married, non-pregnant group.

Sr. No.	Isolates obtained	Married, non-pregnant group	
		n = 300	%
1	Single growth	11	3.66
2	Growth of two organisms	0	0

Table 2 elaborates the microbiological findings in the Married, non-pregnant group. In 3.66% of Married, non-pregnant women, only single pathogen could be detected and none (0%) of the sample showed mixed infection of two pathogens.

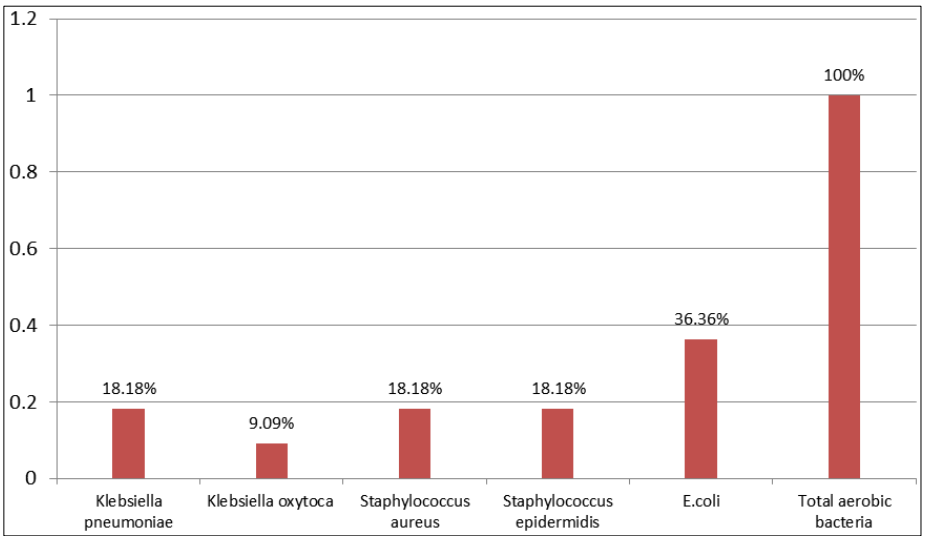

Fig 2: Incidence of aerobic bacterial growth in urine samples. n=11

Table 3 shows the microbiological findings of ASB in Married non-pregnant women. *E. coli* was the predominant isolate (36.36%) in the study and the least was Klebsiella oxytoca (9.09%).

Table 3: Antibiotic susceptibility pattern of isolated organisms.

Antibiotics (µg)		*E. coli* n = 4	Klebsiella pneumonia n = 2	Klebsiella oxytoca n = 1	Staphylococcus aureus n = 2	Staphylococcus epidermidis n = 2
Ampicillin	R	4 (100)	1 (50)	1 (100)	1 (50)	0 (0)
	S	0 (0)	1 (50)	0 (0)	1 (50)	2 (100)
Cephalexin	R	3 (75)	0 (0)	0 (0)	0 (0)	0 (0)
	S	1 (25)	2 (100)	1 (100)	2 (100)	2 (100)
Nalidixic acid	R	3 (75)	0 (0)	0 (0)	Not done	Not done
	S	1 (25)	2 (100)	1 (100)	Not done	Not done
Norfloxacin	R	2 (50)	0 (0)	1 (100)	0 (0)	1 (50)
	S	2 (50)	2 (100)	0 (0)	2 (100)	1 (50)
Nitrofurantoin	R	1 (25)	0 (0)	0 (0)	0 (0)	0 (00)
	S	3 (75)	2 (100)	1 (100)	2 (100)	2 (100)
Cotrimoxazole	R	1 (25)	1 (50)	1 (100)	0 (0)	1 (50)
	S	3 (75)	1 (50)	0 (0)	2 (100)	1 (50)
Augmentin	R	3 (75)	0 (0)	0 (0)	0 (0)	0 (0)
	S	1 (25)	2 (100)	1 (100)	2 (100)	2 (100)
Ceftriaxone	R	1 (25)	0 (0)	0 (0)	0 (0)	0 (0)
	S	3 (75)	2 (100)	1 (100)	2 (100)	2 (100)
Cefotaxime	R	0 (0)	1 (50)	0 (0)	0 (0)	0 (0)
	S	4 (100)	1 (50)	1 (100)	2 (100)	2 (100)
Piperacillin + Tazobactum	R	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
	S	4(100)	2 (100)	1 (100)	2 (100)	2 (100)
Cefuroxime	R	2 (50)	0 (0)	0 (0)	Not done	Not done
	S	2 (50)	2 (100)	1 (100)	Not done	Not done
Amikacin	R	1 (25)	0 (0)	0 (0)	0(00)	0 (0)
	S	3 (75)	2 (100)	1 (100)	2(100)	2 (100)
Piperacillin.	R	2 (50)	0 (0)	0 (0)	Not done	Not done
	S	2 (50)	2 (100)	1 (100)	Not done	Not done
Ciprofloxacin	R	Not done	Not done	Not done	1 (50)	1 (50)
	S	Not done	Not done	Not done	1 (50)	1 (50)
Erythromycin,	R	Not done	Not done	Not done	1 (50)	0 (0)
	S	Not done	Not done	Not done	1 (50)	2 (100)
Vancomycin	R	Not done	Not done	Not done	0 (00)	0 (0)
	S	Not done	Not done	Not done	2 (100)	2 (100)
Penicillin G.	R	Not done	Not done	Not done	0 (00)	0 (0)
	S	Not done	Not done	Not done	2 (100)	2 (100)

*figures in parenthesis indicate % value. Key: R =resistant strain S= Susceptible strain

4. Discussion

UTI are among common bacterial infections. Many investigations suggest that bacteria which cause urinary tract infections, possess certain virulence factors that enhance their ability to colonize and invade the urinary tract. Some of these factors include increased adherence to vaginal and uroepithelial cells by bacterial surface structure (adhesions in particular pili), α-haemolysin production and resistance to serum-killing activity. The other bacterial characteristics are also important in causing urinary tract infections. Motility may be important for bacteria to ascend to the upper urinary tract against the flow of urine and cause pyelonephritis. Some strains of bacteria demonstrate greater production of capsular antigen, which protects them from being phagocytosed [16].

In this study all urine samples were cultured on appropriate media and examined for the growth of aerobic bacterial pathogens. Anaerobic studies were not carried out, as the incidence of anaerobic is very low in UTI. Urine samples were studied to determine bacteriological flora of urinary tract in 300 married nonpregnant women.

UTI namely ASB, cystitis and pyelonephritis are frequently encountered medical complications in females. ASB is defined as the presence of bacteria in urine without having signs and symptoms. This condition affects all groups, but women, particularly pregnant women, are more susceptible than men because of a short urethra and easy contamination of the tract with fecal flora [17]. Empirical treatment of ASB without proper microbiological confirmation of diagnosis may be hazardous to the patient. Hence, the main objective of this prospective, longitudinal study was to determine the incidence, etiology and antimicrobial testing in patients suffering with ASB in order to provide prophylactic treatment [18].

In 300 married non pregnant women studied, cases of ASB were 3.66% and the incidence of ASB was higher in 30-35years (15.0%) followed by 36-40years (14.3%). In this population, *E. coli* was predominant organism isolated accounting for 36.36%. These were followed by *Klebsiella pneumoniae* (18.18%), *Staphylococcus aureus* and *Staphylococcus epidermidis* (18.18%). *Enterococcus faecalis*, *Acinetobacter baumanii*, *Streptococcus agalactiae* and *Proteus mirabilis* and *Pseudomonas aeruginosa* were not isolated.

In one study the prevalence of ASB in healthy women 18 to 40 years of age was approximately 5 percent, and it increased with age to 20 percent or more in ambulatory elderly women [19]. In another study, the prevalence of ASB adjusted for multiple observations per woman, was 5 percent in the university group and *E. coli* was the urinary tract pathogen in 77% women. In univariate analyses, factors significantly associated with asymptomatic bacteriuria were the use of a diaphragm plus spermicide, sexual intercourse, the use of spermicide alone, and the use of a cervical cap. There were no significant associations between ASB and age, marital status, race, history with respect to symptomatic urinary tract infection, or secretor status. In multivariate analyses, the recent use of a diaphragm plus spermicide and recent sexual intercourse remained strongly associated with ASB, whereas use of spermicide alone and use of a cervical cap were significantly associated in only one of the group [20].

In another study [21], only 26 percent of 65 cultures showing asymptomatic *E. coli* bacteriuria were followed by cultures showing asymptomatic bacteriuria with the same strain of *E. coli* and persistent asymptomatic bacteriuria was rare. These numbers are considerably lower than those reported previously and suggest that the majority of episodes of asymptomatic *E. coli* bacteriuria are transient**.**

It is quite evident that *E. coli* is most predominant causative agent of urinary tract infection followed by either *Klebsiella species* and *Pseudomonas species. E. coli and Klebsiella species* being the natural flora of intestine would cause urinary tract infection in individuals with poor hygienic conditions, particularly the individuals who do not keep up the proper hygienic condition of anus and perianal region. However, *Proteus species* being the partial intestinal flora, it is mainly expected to cause urinary tract infection, as it is predominantly present in the faeces. *Pseudomonas species* appears to be the fourth predominant causative agent of urinary tract infection.

In study in Libya, the prevalence of bacteriuria in women was found to be10% (6/60) in non pregnant women. ASB in non pregnant women was 5% (3/60).

In a study performed in turkey, the prevalence of ASB was reported to be 8.1% [22]. The results of the Libayan study found significantly high relation between age and bacteriuria and revealed that the bacteriuria in women was commonest in the age group 25-30 years (62.5%), and these results agreed with study by Buzayan in Libya [23], but contrast with study in Yemen that observed the bacteriuria was more in the age group 15-24 years (53.7%) [24]. In our study we observed that bacteriuria was highest in age group 24-29, that is 52.66% and ASB highest in age group 30-35, that is 15%. This difference may be due to social factors such as early age of marriage and sexual activity.

The primary objective of using antimicrobials is to eradicate the offending microbe and thus ensure total clinical recovery of the patient without giving a chance for the microbe to develop resistance. The antimicrobial sensitivity pattern of bacteria can sometimes be sufficiently characteristic to be useful as an epidemiological marker for tracing strains.

The antibiotic sensitivity of uropathogens obtained in the study were as follows. *E. coli* was highly sensitive to Piperacillin + Tazobactum (100%) and Cefotaxime (100%). *Klebsiella pneumoniae* showed 100% sensitivity to Cephalexin, while *Klebsiella oxytoca* showed 100% sensitivity to Cephalexin. *Staphylococcus aureus* were found 100% sensitive to Cephalexin, while *Staphylococcus epidermidis* were 100% sensitive to Ampicillin. According to *Gruneber 1984* [25] Nalidixic acid resistance is becoming more important proportion of gram positive urinary pathogens. In one study. The most effective antibiotics tested on the isolated bacteria were gentamycin (87.5%), azithromycin (75%) and ciprofloxacin (68.75%), and the less effective antibiotics were cephalexine (6.25%), and ampicillin (12.5%). The susceptibility study showed that *E. coli* isolates were resistant to most of the antibiotic tested followed by *Klebsiella* species [26].

5. Conclusion

Changing pattern of antibiotic resistance among common urinary pathogens is being noticed worldwide. The general urinary bacterial pathogens *Escherichia coli*, *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* have acquired resistance to the commonly used antibiotics to varying degrees in the developed as well as developing countries. Since ASB are most often treated empirically, knowledge of

antimicrobial resistance determined on the basis of studies is essential. But it is important to emphasize that antibiotic susceptibility tests are intended to be a guide for the clinician, not a guarantee that an antimicrobial agent will be effective in therapy. The results of this study agreed with other studies and urine culture was the gold standard method of diagnosis for ASB. The results of this work showed an association of ASB with age and gravidity and it appeared to be multifactorial. A screening for ASB in married non-pregnant women must be done to discover the infected cases, which would allow early treatment to avoid the complications.

6. Acknowledgement

Authors are grateful to Nair Golden Jubilee Research Foundation for providing financial help to carry out this study and staff of the Microbiology Department of T. N. Medical College and B. Y. L. Nair Charitable Hospital for their moral support.

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