Original Article

Asymptomatic Bacteriuria in a University Teaching Hospital in Southern Nigeria: Prevalence, Uropathogens, and Antibiotic Susceptibility

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Abstract

Background: Asymptomatic bacteriuria (ASB) has been documented as the main risk factor for the development of symptomatic urinary tract infection in pregnancy and is associated with maternal and fetal complications. **Objective:** To determine the ASB prevalence, the causative microorganisms, their drug sensitivity patterns, and the factors associated with its occurrence in pregnant women in the Uyo, Nigeria. **Methodology:** Three hundred and twenty women were recruited during their first antenatal visit over a period of 13 weeks. A midstream urine specimen was obtained from each patient, cultured, isolates identified and antimicrobial sensitivity done. Data were analyzed using the Statistical Package for the Social Sciences version 20. **Results:** The ASB prevalence was 9.1% with the two commonest identified isolates being *Escherichia coli* (41.4%) and *Klebsiella pneumoniae* (24.1%). Imipenem (100.0%) and gentamycin (37.9%) were the two most sensitive drugs. The association between respondents' educational level and the occurrence of ASB was significant statistically. **Conclusion:** The prevalence of ASB was relatively high among the respondents. This, therefore, emphasizes the need for routine screening of our antenatal female population for ASB in all our health facilities.

Keywords: Asymptomatic bacteriuria, infection of the urinary tract, Southern Nigeria, Uyo

Introduction

Asymptomatic bacteriuria (ASB), has been documented to be a major factor associated with the development of symptomatic urinary tract infection (UTI) in pregnancy with its higher risk of maternal and fetal complications.[1]

Although ASB in women who are not pregnant may have no significant adverse effects, in pregnancy, however, relative physiologic obstruction to urine flow results in urinary stasis and increases the possibility of pyelonephritis developing[2] with its attendant complications including preterm labor, low birth weight, intrauterine fetal death, and increased perinatal mortality.[3] Hence if ASB is not identified and well managed in pregnancy, almost half of affected patients will develop symptomatic UTI and acute pyelonephritis will complicate about a third of them.[4]

Globally, ASB prevalence among pregnant women varies from 2% to 10%.[5-7] In Nigeria, the prevalence ranges from 4.0% to 14.1%[8] although a study in Benin city reported a prevalence of 86.6%,[9] which remains the highest documented rate in the world. The frequency of occurrence of bacteriuria in pregnancy increases with maternal age, coital activity, parity and has been found to be high among individuals in the lower socioeconomic class.[10,11] ASB has also been reported to occur more frequently in patients with coexisting medical conditions such as anatomic anomalies of the urinary tract, preexisting medical conditions such as diabetes mellitus, sickle cell trait, recurrent UTIs, and HIV/AIDS.[8,12]

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The causative agents implicated in patients with ASB are similar in women whether pregnant or not.[5] *Escherichia coli* is its most commonly associated pathogen, responsible for at least 80% of cases, with other Gram-negative and Gram-positive organisms including *Staphylococcus saprophyticus* and enterococci occasionally being isolated.[13] Other isolated organisms include *Staphylococcus aureus*, *Proteus* species, *Klebsiella* species, *Citrobacter* species. *Pseudomonas aeruginosa* and group B *Streptococcus*. These microorganisms are mainly of gastrointestinal origin but usually inhabit the vagina and periurethral region and so easily ascend the short and relatively wide female urethra close to the vagina and anus. Sexual intercourse is another factor that facilitates the migration of these intestinal organisms to the urinary tract through the urethral meatus.[14]

Screening patients during pregnancy for ASB as well as its treatment has become standard obstetric care and is included as one of the most cost-effective measures for ensuring the improvement of maternal and perinatal health in low-income nations.[15] The majority of prenatal guidelines emphasize routine screening for ASB.[16] This is to prevent adverse maternal and fetal outcomes associated with the condition.[12] There are several methods of diagnosis of ASB but urine culture remains the gold standard because of its high sensitivity and specificity.[16] In populations with over 2% prevalence rate for ASB, it has been shown that urine culture is a cost-effective screening tool.[17] Furthermore, urine culture utilizes the same specimen for the isolate's antimicrobial sensitivity testing.

In pregnancy, it is recommended that screening for ASB be done by culture of urine between the 12th and 16th gestational week or during the booking visit if after this period.[16]

Prospective, comparative, and randomized clinical trials, as well as meta– analyses, have consistently revealed that effective management of ASB with antimicrobials during pregnancy decreases the risk of pyelonephritis developing from 20%–35% to 1%–4%.[6] and also reduces the rate of development of low-birth-weight babies as well as preterm delivery.[13] The consistency and robustness of observations from several researches led to screening for ASB and its treatment during pregnancy becoming a standard of management in the developed world, a practice yet to be full adopted in the developing countries.[1]

In the University of Uyo Teaching hospital (UUTH), Uyo in Akwa Ibom State, Nigeria, the urine of women receiving antenatal care is not routinely tested by culture. This may probably be due to a lack of documented information regarding the ASB prevalence among antenatal attendees in the state. Hence, the proportion of pregnant women who have ASB and the associated adverse effects in our environment may not be known. This study seeks to contribute in this regard by assessing the prevalence and pattern of ASB among antenatal attendees at UUTH, Uyo, Akwa Ibom State. Our studies findings may lean support to the need to routinely offer women who receive antenatal care in our facility, screening for ASB. This would also provide an objective platform upon which decisions about the choices of antibiotics are made for those patients that need to be treated.

Methodoloy Study Location

This is a descriptive cross-sectional study that was conducted in the antenatal clinic of UUTH, Uyo, the capital city of Akwa Ibom State in Nigeria. The Hospital is a 500-bed health facility and the only one that offers tertiary health care services in the state. The institution receives referrals from peripheral health centers in the state (and surrounding) covering a population of over 3.9 million people[18] in southern Nigeria.

The sample size's estimation

The ASB prevalence rate of 27.3% from a study in Calabar, South-south Nigeria[19] was used to calculate the minimum sample size for this study using the statistical formula of Fische $n = \frac{Z^2pq}{d2}$.

From this, a minimum sample size of 305 was calculated for the study. However, this number was increased to 320 by adding 5% of the calculated minimum sample size to provide for nonresponse rate. Thus, the minimum sample size for the study was 320.

Patients selection

The study's details were explained to all the patients during the health talk session at the beginning of the booking clinic. Systematic random sampling technique was used with a sampling interval of 3. The first woman was chosen by random sampling. The sampling frame used was the attendance list of the patients at each booking clinic. Separate pieces of paper were then used to write the attendance number of each woman. A container was used to mix these pieces of papers from which through balloting, the first participant was picked randomly. Where the inclusion criteria were not met by the patients, a new number was drawn until the criteria were met. Systematic sampling was then used to select the remaining number of participants.

Inclusion criteria

Consenting patients who attended the booking clinic during the study period.

Exclusion criteria

Those who had symptoms that were suggestive of UTI, those who took antibiotics in the preceding two weeks and women who declined consent.

Collection of data

Data were collected over a 13 weeks period (04/06/2014–27/08/2014) during which time 320 pregnant women were recruited during their antenatal visit. A pretested structured questionnaire was used to interview the patients by the investigator and some resident doctors in the Department of Obstetrics and Gynaecology after they were trained on

the subject of study. Information sought in the questionnaire included; age of the patient, marital status, parity, gestational age, educational level, occupation and that of their spouse, history suggestive of infection of the urinary tract, and ingestion of antibiotic in the preceding two weeks.

Socioeconomic status was determined using the levels of education and occupation of the patient as well as their spouses. Each patient was apportioned to one of five social classes based on her level of education level and spouses' occupation in accordance with Olusanya *et al*.'s scoring system.[20] The social class for the unmarried women or those separated was based on the patient's occupation and educational status.

Specimen collection

After seeking and obtaining consent, administering the questionnaire and instructing each client on the right way to obtain a clean catch mid-stream urine as described by Chessbrough,[21] about 5–10 ml of freshly voided clean catch mid-stream specimen of urine was collected from each participant in a sterile disposable plastic universal bottle. These were taken immediately to the Medical Microbiology laboratory of UUTH, Uyo for culture.

Specimen processing urine culture

A loopful of well-mixed uncentrifuged urine specimen of each patient was inoculated aseptically on dried plates of cysteine lactose electrolyte deficient agar and blood agar using a standard inoculating wire loop that delivered 0.002 ml of urine. The inoculated agar plates were incubated aerobically at 37°C for 24 h.

Significant bacteriuria determination

After 24 h of incubation at 37°C, the plates were read and colony counts yielding bacterial growth of 100,000 pure isolates or more per milliliter ($\geq 10^5$/ml) of urine were regarded as significant. This was determined by counting the number of colonies on the plates with the help of a magnifying lens and dividing the number gotten by 0.002 ml which was the urine volume the loop delivered.

Subculture of isolate

The isolates were sub-cultured on blood agar and incubated at 37°C for 24 h to obtain colonies for identification of the organism and antibiotic susceptibility testing.

Identification of isolates

Identification of bacterial isolates was based on morphological characteristics of isolates on the culture medium. Gram staining and biochemical testing were done according to standard procedure. Gram Staining was used to distinguish isolates into Gram-negative or Gram-positive bacteria**.** The identification of bacterial isolates to species level was done biochemically using the Microbact 24E (MB24E). Microbact 24E (MB24E) is a commercial identification system for enterobactericae and other Gram-negative bacteria.

Antibiotic susceptibility testing

The modified Kirby Bauer disk diffusion method[22] was used to determine the antibiotic susceptibility pattern of the isolates as recommended by the Clinical Laboratory Standard Institute with the commercially available disc (Oxoid Ltd). The antibiotics tested were selected from each class based on their safety profile in pregnancy. They included: Augmentin (30 µg), nitrofurantoin (300 µg), gentamicin (10 µg), erythromycin (5 µg), ceftriazone (30 µg), and imipenem (10 µg).

Data analysis and presentation

All generated data in the study including results from laboratory analysis of specimens were coded and fed into a computer using the Statistical package for social sciences (SPSS) version 20, IBM, Chicago, Illinois, USA. Mean and standard deviation were calculated for quantitative variables. Qualitative variables were summarized in frequencies and percentages. Chi-square test was used to test for association between ASB and maternal age, parity, trimester of pregnancy, and socioeconomic status. A $P < 0.05$ was considered statistically significant.

Ethical considerations

Ethical approval was obtained from the Intuitional review committee of UUTH. All women registering for ANC had the details of the study explained to them at the commencement of the clinic and written informed consent was collected before recruiting each patient into the study. The patients reserved the right and freedom to refuse to take part as well as leave the study at any time without any consequence. Initials and serial numbers on the patient's laboratory forms, questionnaires, and specimen containers were used to identify the patients and their samples and the study was carried out with the assurance of confidentiality of information.

Results

Out of the 320 women studied, 29 had significant bacteriuria giving a prevalence of 9.1%. The respondents had a mean age of 27.84 ± 5.05 years with a range of 15–43 years. Most (65.6%) of the respondents were in the 21–30 years age bracket, were married (94.1%) and 34.1% belonged to social classes 1. About half (51.9%) of the respondents and 50.8% of spouses completed secondary education and postsecondary education respectively. Many respondents (58.1%) booked for ANC in their second trimester and 40.9% were nulliparous [Table 1].

The common bacterial isolates responsible for ASB among the women were; *E. coli* 12 (41.1%), *Klebsiella pneumoniae* 7 (24.1%), and *Enterobacter cloacae* 5 (17.2%) with *E. coli* responsible for the majority of cases. Others were *Citrobacter amalonaticus* 3 (10.3%), *Pseudomonas luminescens* 1 (3.5%), and *Acinebacter haemolyticus* 1 (3.5%).

The bacterial isolates' sensitivity pattern is shown in Table 2. Imipenem was the only antibiotic that was sensitive to all bacterial isolates 29 (100.0%). Others were gentamycin 11 (37.9%), ceftriazone 8 (27.6%), nitrofurantion 8 (27.6%), augmentin 2 (6.9%), and erythromycin 1 (3.4%) which had the least sensitivity.

The association between some socio-demographic and clinical factors of the patients and the presence of ASB is shown in

Table 1: Socio demographic profile of the women at booking

Characteristic	Frequency, *n* (%)
Age group (years)	
<20	24 (7.5)
21-30	210 (65.6)
31-40	85 (26.6)
>40	1 (0.3)
Marital Status	
Single	15 (4.7)
Married	301 (94.1)
Divorced	4 (1.3)
Social class of respondents	
1	109 (34.1)
2	47 (14.7)
3	104 (32.5)
4	46 (14.4)
5	14 (4.4)
Respondent's educational status	
No formal education	1 (0.3)
Primary education	25 (7.8)
Secondary education	166 (51.9)
Postsecondary education	128 (40.0)
Spouse educational status (*n*=305)	
No formal education	1 (0.3)
Primary education	13 (4.3)
Secondary education	136 (44.6)
Postsecondary education	155 (50.8)
Trimesters of pregnancy (months)	
1-3	24 (7.5)
4-6	186 (58.1)
7-9	110 (34.4)
Parity	
Nullipara	131 (40.9)
Primipara	91 (28.4)
Multipara	92 (28.8)
Grandmultipara	6 (1.9)

Table 3. The association between the educational status of the respondents and the occurrence of ASB was significant statistically. However, the association between age groups, social classes of respondents, educational level of spouses, trimesters of pregnancy, parity, and occurrence of ASB was not statistically significant.

Discussion

Asymptomatic bacteuria's diagnosis during booking is essential because early treatment can be instituted, thereby reducing the risk of adverse effects on maternal and fetal health.[12] This is especially important because ASB in pregnancy is a subclinical infection and is associated with adverse perinatal outcome.[23]

This study revealed that the respondents were mostly in their twenties, this being about the height of the reproductive period in our society and indeed in most other parts of the country. Virtually all the women were married and this could result in a higher chance of their pregnancies been planned, hence their presentation for orthodox antenatal care compared to single women. The women were also well educated with almost half of the women having attained tertiary level education. A significant proportion of the study population was of low parity as 40.9% were nulliparous. These are in agreement with findings from other researchers whose studies revealed antenatal booking to be high in well-educated women and those of low parity.[24] As it is common practice for pregnant women in this environment to book late for ANC,[25] it is not surprising that only 7.5% of the women were in their first trimester.

The ASB prevalence in our study was 9.1% and this is in concordance with global rates of 2%–10% reported in other studies.[6,12] It is however slightly higher than the rates reported from other African countries[26] and the United States[16] but far below the 27.3% and 86.6% reported in Calabar,[19] and Benin City, Nigeria,[9] respectively.

These variations may be due to the differences in characteristics of the different populations such as maternal age, parity, socioeconomic status, setting of study (primary care, community based or hospital based), methods of screening (dipstick, microscopy, or culture), and interpretation of significant bacteriuria.[12]

The most common microorganism responsible for the occurrence of ASB was *E. coli* in our study population. This is similar to findings from other studies where *E. coli* accounted for over 80% of isolates.[12,14,26] This may be due to its virulent characteristics which include; resistance to vaginal acidity, rapid multiplication in urine, possession of adhesions (characterized as fimbriae) which allows adherence to the uroepithelial cells, and production of toxins that decrease ureteric peristalsis and inhibit phagocytosis.[14] These finding, however, is at variance with reports from some Nigerian studies[1,9,22,24] where *S. aureus* was the commonest isolated organism among pregnant women with ASB. *S. aureus* is not considered a typical urinary tract pathogen as its colonization of the urinary tract is mostly by hematogenous seeding from bacteremia.[27] However, the role of *S. aureus* in ascending UTI remains unclear and the interpretation of its isolation in urine in those studies really does require further evaluation as indicated in the studies.[14]

Imipenem and gentamicin were the most sensitive antibiotics to the bacterial isolates in this study with all of them sensitive to imipenem. This is in concordance with findings of similar studies from Minna, Nigeria[28] and Ethiopia[29] This sensitivity pattern may be as a result of their potency against Gram-negative rods. In addition, the fact that they are administered parenterally makes them less subject to abuse. Nitrofurantoin and ceftriazone were moderately effective against the urinary isolates with the most resistance being demonstrated to augmentin and erythromycin.

The increased antibiotic-resistant pattern to the oral antibiotics observed in this study could be due to self-medication as

Table 2: Antibiotic sensitivity pattern of bacterial isolates

Antimicrobials (μg)	Antibiotic sensitivity pattern by isolates											
	Escherichia coli (*n*=12)		*Klebsiella Pneumonia* (*n*=7)		*Enterobacter cloaca* (*n*=5)		*Citrobacter amalonaticus* (*n*=3)		*Pseudomonas luminescens* (*n*=1)		*Acinetobacter haemolyticus* (*n*=1)	
	S, *n* (%)	R, *n* (%)	S, *n* (%)	R, *n* (%)	S, *n* (%)	R, *n* (%)	S, *n* (%)	R, *n* (%)	S, *n* (%)	R, *n* (%)	S, *n* (%)	R, *n* (%)
Imipenem (10)	12 (100.0)	0	7 (100.0)	0	5 (100.0)	0	3 (100.0)	0	1 (100.0)	0	1 (100.0)	0
Genticin (10)	5 (41.7)	7 (58.3)	2 (28.6)	5 (71.4)	2 (40.0)	3 (60.0)	1 (33.3)	2 (66.7)	1 (100.0)	0	0	1 (100.0)
Ceftriazone (30)	5 (41.7)	7 (58.3)	0	7 (100.0)	1 (20.0)	4 (80.0)	1 (33.3)	2 (66.7)	1 (100.0)	0	0	1 (100.0)
Augmentin (30)	1 (8.3)	11 (91.7)	0	7 (100.0)	0	5 (100.0)	0	3 (100.0)	1 (100.0)	0	0	1 (100.0)
Erythromycin (5)	0	12 (100.0)	0	7	0	5 (100.0)	1 (33.3)	2 (66.7)	0	1 (100.0)	0	1 (100.0)
Nitrofurantoin (300)	4 (33.3)	8 (66.7)	0	0	1 (20.0)	4 (80.0)	2 (66.7)	1 (33.3)	1 (100.0)	0	0	1 (100.0)

All the bacterial isolates demonstrated 100% sensitivity to imipenem. S: Sensitivity, R: Resistance

Table 3: Association between some sociodemographic and clinical factors and presence of asymptomatic bacteriuria

Characteristic	Presence of ASB		χ^2	*P*
	Yes	No		
Age group				
<20	4 (16.7)	20 (83.3)	2.196	0.379*
21-30	19 (9.0)	191 (91.0)		
31-40	6 (7.1)	79 (92.9)		
40 and above	0	1 (100.0)		
Social class of respondents				
1	7 (6.4)	102 (93.6)	4.721	0.259*
2	6 (12.8)	41 (87.2)		
3	8 (7.7)	96 (92.3)		
4	5 (10.9)	41 (89.1)		
5	3 (21.4)	11 (78.6)		
Respondents educational status				
No formal education	1 (100.00)	0	12.790	0.037**
Primary education	4 (16.0)	21 (84.0)		
Secondary education	16 (9.6)	150 (90.4)		
Tertiary education	8 (6.3)	120 (93.8)		
Spouse educational status				
No formal education	0	1 (100.0)	0.103	1.000*
Primary education	1 (7.7)	12 (92.3)		
Secondary education	11 (8.1)	125 (91.9)		
Tertiary education	13 (8.4)	142 (91.6)		
Trimesters of pregnancy				
1-3	2 (8.3)	22 (91.7)	0.181	0.908*
4-6	16 (8.6)	170 (91.4)		
7-9	11 (10.0)	99 (90.0)		
Parity				
Nullipara	14 (10.7)	117 (89.3)	2.385	0.339*
Primipara	9 (9.9)	82 (90.1)		
Multipara	5 (5.4)	87 (94.6)		
Grandmultipara	1 (16.7)	5 (83.3)		

*Fisher Exact *P* value, **Statistically significant *P* value. There was a statistically significant association between the educational status of respondents and occurrence of ASB. However, the association between age groups, social classes of respondents, educational level of spouses, trimesters of pregnancy, parity and occurrence of ASB was not significant statistically. ASB: Asymptomatic bacteriuria

well as drug abuse which are not uncommonly practiced in many low-income countries including Nigeria.[30] In addition, low cost, easy availability of these drugs without a physician's prescription, and poor regulation could be other factors contributing to antibiotic resistance in our society.[30] This, therefore, necessitates that treatment of ASB be based on antibiotic susceptibility testing with considerations for maternal and fetal safety profile.

This study showed that the younger the women, the higher the risk of ASB demonstrating an inverse relationship between maternal age and prevalence of ASB. This relationship, though not statistically significant, was clinically significant. This is because sexual activity tends to be more common in younger women and the frequency of occurrence of bacteriuria among young women has been found to be strongly related to coitus.[31,32] Other studies, however, have reported an increasing prevalence of ASB with advancing maternal age[1] because increasing age of women is naturally associated with increasing parity, the later having been documented as a risk factor for ASB.[10] Interestingly, other studies have equally found no associations between maternal age and prevalence of ASB.[33]

Alhough the percentage of respondents with ASB increased with worsening social class with the least percentage of respondents in social class 1 having ASB and those in social class 5 having the highest prevalence, this difference was statistically insignificant. This finding is in agreement with the findings of a similar study in Ibadan, Nigeria.[33] Other studies, however, have reported a statistically significant association between social class and the prevalence of ASB.[10]

The prevalence of ASB, in this study was statistically associated with respondents' level of education revealing that as the level of education of respondents improved, prevalence of ASB decreased. This finding indicates that educational level is a risk factor and is in agreement with the results of a similar study carried out in Parkistan.[34] In Iran,[33] however, no particular trend was found as the prevalence of 2.6%, 7.1%, 6.1%, 6.5%, and 2.3% were reported for illiterates, primary, secondary, high school degree and higher education, respectively. Probably, the improved personal hygiene that is associated with a higher level of education may explain the decrease in the bacteriuria prevalence noticed in this study among those with higher education.

The proportion of respondents with ASB increased with increasing trimester with the highest proportion of respondents with ASB being in the third trimester though this difference in proportion was not statistically significant. This could be because as pregnancy progresses, the combined mechanical compression of the ureters by the enlarging uterus and relaxation of smooth muscles induced by the hormone progesterone decreases ureteral peristalsis and bladder tone causing relative urinary stasis which encourages bacterial infection in the upper urinary tract.[3] This is in concordance with reports from other studies.[25] However, a Nigerian study[1] suggested no particular trend as their study revealed a high frequency of bacteriuria in the third trimester of pregnancy (21.9%) when compared with that in the first trimester (7.7%), while the level in the second trimester was 22.5%.[1]

The association between parity and ASB was not significant statistically in this study. Though a higher proportion of grandmultiparous respondents (16.7%) had ASB compared to 10.7% of nulliparous women, there was no defined trend as the prevalence fell to 9.9% and 5.4% in the primiparous and multiparous women respectively before rising to 16.7% in the grand multiparous women. This finding agrees with a similar study in Ghana.[34] Other studies that found multiparity to be significantly associated with an increased frequency of bacteriuria in pregnancy reasoned this to be secondary to urinary tract changes, like dilatation of the ureters and decrease in the tone of the bladder resulting in increased urinary stasis, occurring during each pregnancy which often do not perfectly return to normal even after delivery, perhaps making multiparous women more prone to ASB.[1]

This study's strength, however, lies in the fact that steps were taken to make sure the urine samples were properly obtained and analyzed within 2 hours of collection.

This study had some limitations. The study was hospital based hence the prevalence obtained may not be a true reflection of that of the community. The study depended on results gotten from processing the samples in the laboratory, which may not devoid of errors. Such errors could arise from the human factor or from faulty equipment used for the procedure. The specimen collection was done by the patients, thus, the findings depended on their adherence to the instructions for a clean-catch urine sample collection.

Conclusion and Recommendation

The ASB prevalence among pregnant women at booking was 9.1%. The most common bacterial isolates responsible were *E. coli* and *K. pneumoniae*. The bacterial agents were generally susceptible to antibiotics tested, especially imipenem and gentamicin and more resistant to augmentin and erythromycin. We recommend community-based research on ASB in our environment to guide general policy on diagnosis and management.

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Conflicts of interest

There are no conflicts of interest.

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