Asymptomatic Bacteriuria among Students of Bellstech, Ota, Nigeria.

Felix Oluwasegun Alao, M.Sc.* and Fayokemi Ayomide Akintunde, B.Sc.

Department of Biological Sciences, College of Natural and Applied Sciences,
Bells University of Technology, PMB 1015, Ota, Ogun State, Nigeria.

E-mail: sunfelixnaija@gmail.com*
smily_bunny@yahoo.com

ABSTRACT

Asymptomatic bacteriuria (ASB) is bacteriuria without apparent symptoms of urinary tract infections (UTIs). One hundred midstream urine samples were collected from apparently healthy students made up of 50 males and 50 females. Urinalysis of the samples was carried out. Growth was observed in 95% of the samples while there was no growth in 5% of the samples. Prevalence of significant asymptomatic bacteriuria was higher in females (23%) than males (20%).The antibiotic sensitivity of the isolates was also determined.

(Keywords: asymptomatic bacteriuria (ASB), urinary tract infections (UTIs), students, urinalysis, isolates, antibiotic sensitivity)

INTRODUCTION

Asymptomatic bacteriuria (ASB) is bacteriuria without apparent symptoms of urinary tract infections (UTIs) (Barr *et al.*, 1985). Urine in bladder is normally sterile (Kass, 1962) .The presence of bacteria in urine is called bacteriuria (Whalley, 1962). The importance of asymptomatic bacteriuria lies in the insight it provides into symptomatic infections (Nicolle, 1997).

Asymptomatic bacteriuria (ABU) is defined as significant bacterial count in the urine, usually 10^5 cfu per ml in an individual without symptoms of urinary tract infections (UTI) (Smith, 1994). This has changed from $\geq 10^5$ bacteria/ml urine before 1992 to $\geq 10^4$ bacteria/ml urine. This change will increase the representation of slow-growing bacteria like enterococci and coagulase-negative staphylococci (Grude *et al*., 2001).

Stamm and Hooton, 1993 referred to UTI as a clinical (symptomatic) or subclinical (asymptomatic) disease that may involve just the lower tract or both the lower and upper tracts – infection may involve single sites. Such as urethra – urethritis, prostrate-prostitis, bladder, cystitis, kidney – pyelonephritis but the whole system is always at a risk of invasion by bacteria once any part is infected (Atlas, 1986).

Urinalysis is an important part of the initial examination of a patient and the results provide a valuable picture of the patient's general health pattern. Generally, urinalysis will indicate: The state of kidneys and the urinary tract - chemical tests for the presence of protein or blood; together with physical properties and microscopic examination for casts, cells and certain crystals are very helpful in assessing and treating renal and urinary tract disease. Information about metabolic and systemic (nonrenal) abnormalities - tests for glucose, ketone bodies, bilirubin and urobilinogen are useful parameters in the diagnosis of metabolic and systemic disorders such as diabetes and jaundice (Bulakh, 2003).

The organisms most frequently isolated in asymptomatic bacteriuria and urinary tract infection includes species of Enterobacteriaceae especially *Escherichia coli* which is predominant, followed by *Klebsiella pneumoniae, Staphylococcus saprophyticus, Pseudomonas aeruginosa, Enterococcus faecalis, and Proteus species.*

Antibiotics such as: Ceftazidime (30µg), Cefuroxime (30µg), Gentamicin (10µg), Ciprofloxacin (5µg), Cefixime (5µg), Ofloxacin (5µg), Augmentin (30µg), Nitrofurantoin (300µg) are highly effective to the organisms associated with asymptomatic bacteriuria with one or two being resistant to the organisms (Geerlings and Brouwer, 2000).

Asymptomatic bacteriuria occurred reliably more frequently in females as compared with males and it is a major criterion of urinary tract infection

(Nurullaev, 2004). Therefore, it is necessary to investigate the incidence of asymptomatic bacteriuria in both apparently healthy male and female undergraduate students of Bellstech, Ota and also to study the antibiotic sensitivity pattern of the bacteria isolated.

MATERIALS AND METHODS

Study Site

One hundred students of Bells University of Technology, Ota comprised of 50 males and 50 females within the age of 16 to 26 years were sampled for asymptomatic bacteriuria.

The following information was obtained from participating student before sample collection e.g. name, sex, age, as well as history of UTI.

Exclusion Criteria

Students who had taken antibiotics two weeks before or were currently on antibiotics, or with any symptom suggestive of urinary tract infections (UTIs), or pregnant were excluded from the study.

Sample Collection

The participated students were instructed to wash their external genital with mild toilet soap and rinse thoroughly with clean water. Then, the early morning midstream urines were collected into the sterile universal bottles. The samples were labeled against the names of students.

Processing of Sample

Urinalyses of the samples were carried out with the aid of urine dipstick. Prior to culturing, samples were preserved in the refrigerator at 4^0C and processed within 2 hours of collection. The samples were examined macroscopically. The samples were cultured on MacConkey and CLED agar (Oxoid) and isolates were characterized biochemically as described by Cheesbrough (2000).

Antibiotic Sensitivity Test

This test was carried out to determine the antibiotic susceptibility pattern of the different isolates. Nutrient agar plates were inoculated with isolates from stock cultures. The Kirby-Bauer disc-diffusion test which conforms to the recommended standard of the clinical and laboratory standards institute (CLSI), formerly National Committee for Clinical Laboratory Standards (NCCLS) were used (Cheesbrough, 2000).

Turbidity of the inoculum of various bacterial isolates was compared with 0.5 McFarland standard and each of the isolates were inoculated onto the surface of a sterile nutrient agar plates using a sterile swab in order to ensure even distribution of the inoculum, the plates were allowed to dry and antibiotic discs with different concentrations were placed on the surface of the agar plates.

The antimicrobial discs include the following: Ceftazidime (30µg), Cefuroxime (30µg), Gentamicin (10µg), Ciprofloxacin (5µg), Cefixime (5µg),Ofloxacin (5µg), Augmentin (30µg), Nitrofurantoin (300µg). After 30 mins of applying the disc, the plates were inverted and incubated for 24 hours at 37^0C. The clear zone that developed around each disc was measured as the zones of inhibition from underneath each plate with the aid of a ruler in centimetre and then converted to millimetre (mm).

Statistical Analysis

Statistical analysis was carried out using chi-square test to compare the prevalence of asymptomatic bacteriuria among male and female students. The difference were considered significant at $p<0.05$.

RESULTS

Urine samples from one hundred (100) students of Bells University of Technology, Ota within the age range of 16 to 26 years were collected and examined for the presence of bacteria using a semi quantitative method. The color of the urine samples ranged from pale yellow, light yellow, yellow to clear or turbid in transparency. From the urinalysis, 3 samples (2 males and 1 female) were positive to bilirubin which is an indicator of liver abnormalities; 3 samples (2 males and 1 female) were positive to urobilinogen; 6 samples

were positive to protein; 3 males were positive to protein which indicate the condition known as proteinuria while 15 had traces of protein; 3 females were positive to protein while 11 females had traces of protein; 2 samples were positive to nitrite while 4 samples had traces of nitrite, in which 3 males had traces of nitrite while 2 was positive to nitrite and 1 female with traces of nitrite which is a rapid method for detecting asymptomatic urinary tract infection; 4 female samples were positive to leucocyte which indicates bacterial infection; detection of nitrite coupled with leucocyte is a rapid method for detecting asymptomatic bacteruria. The students were in the normal pH range (4.0 – 8.0).

In this study, 100 students were sampled out of which 43 (43%) showed significant bacteriuria; 23 (23%) were female while 20 (20%) were male (Table 1).

Bacterial isolates were recovered from the samples of which *Escherichia coli* 60 (60%), *Klebsiella* species 18 (18%), and *Staphylococcus saprophyticus* 8 (8%) were the most frequent isolates (Table 2).

Table 3 shows antibiotic sensitivity pattern of the isolates. Majority of the isolates were sensitive to nitrofurantoin, ofloxacin, and ciprofloxacin.

Table 1: Prevalence of Significant Bacteriuria (≥ 10^4 CFU/ml)

Total	Subjects with significant bacteriuria	%	Subjects with non-significant bacteriuria	%	Subjects with mixed culture	%	Subjects with no growth	%
Females (50)	23	46	24	48	1	2	2	4
Males (50)	20	40	25	50	2	4	3	6
Total (100)	43	43	49	49	3	3	5	5

Table 2: Prevalence of Bacterial Isolates among Students.

Organism	Male Isolates	Female Isolates	Total number of isolates (%)
Escherichia coli	35	25	60 (60)
Klebsiella species	5	13	18 (18)
Staphylococcus saprophyticus	4	4	8 (8)
Staphylococcus aureus	3	2	5 (5)
Enterococcus faecalis	2	3	5 (5)
Proteus Species	1	2	3 (3)
Pseudomonas aeruginosa	-	1	1 (1)
Total	50	50	100 (100%)

Table 3: Antibiotic Susceptibility Patterns of the Isolates with ≥ 10^4 CFU/ml.

Organism	Isolates	Antibiotics							
		CAZ (30µg)	CRX (30µg)	GEN (10µg)	CXM (5µg)	OFL (5µg)	AUG (30µg)	NIT (300µg)	CPR (5µg)
Escherichia coli	20	0(0%)	2(10%)	10(50%)	2(10%)	18(90%)	0(0%)	16(80%)	17(85%)
Klebsiella species	13	0(0%)	0(0%)	6(46.2%)	0(0%)	11(84.6%)	0(0%)	9(69.2%)	11(84.6%)
Proteus species	1	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)	1(100%)	1(100%)
Pseudomonas aeruginosa	1	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)	1(100%)	0(0%)
Enterococcus faecalis	2	0(0%)	0(0%)	1(50%)	0(0%)	1(50%)	0(0%)	2(100%)	1(50%)
Staphylococcus saprophyticus	5	0(0%)	0(0%)	2(40%)	0(0%)	4(80%)	0(0%)	3(60%)	3(60%)
Staphylococcus aureus	1	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)	1(100%)	0(0%)

KEYS: CAZ-CEFTAZIDIME; CRX-CEFUROXIME; GEN- GENTAMICIN; CXM- CEFIXIME; OFL- OFLOXACIN; AUG- AUGMENTIN; NIT- NITROFURANTOIN; CPR- CIPROFLOXACIN

DISCUSSION

For this study, the significant asymptomatic bacteriuria is 10^4cfu/ml, although according to Cheesbrough (2000), 10^4cfu/ml is doubtful significant. However, based on the result of urinalysis, 10^4cfu/ml used as significant asymptomatic bacteriuria in this study was in accordance with the earlier submission by Brooks and co-workers (1998) that the presence of 10^4cfu/ml of a single type of enteric Gram negative rod is strongly suggestive of urinary tract infections. In addition, the American Society of Microbiology's Manual of Clinical Microbiology (Thompson and Miller, 2003) recommends a cut-off value of between 10^2 and 10^5 cfu/ml for the diagnosis of significant bacteriuria, but there is not a worldwide consensus for these values (Raz *et al*., 2005).

The urinalysis in this study showed that 6 samples were positive to nitrite which is a rapid method for detecting asymptomatic urinary tract infections (UTIs), 4 samples were positive to leucocyte which indicates bacterial infection and detection of nitrite with leucocyte is a rapid method for detecting asymptomatic bacteriuria. This report is in line with (Bent, 2002) that dipstick urinalysis in practice may provide useful information when the diagnosis of UTI is in doubt. The presence of nitrite has a predictive value for UTI, since nitrite are formed as a metabolic product from bacteria that breakdown nitrate to nitrite (e.g. *Escherichia coli, Proteus species, Klebsiella species,* etc.). If the dipstick is positive to either nitrite or leucocyte this increases the probability of UTI to about 80% while dipstick negative in both nitrite and leucocyte reduces the probability of UTI to about 20% (Bent, 2002).

This study showed that prevalence of asymptomatic bacteriuria was observed in 43% of the population. The females had a higher prevalence (53.5%) than the males (46.5%).This observation is in agreement with Prescott and co-workers (1999) who reported that bacteriuria is more common in females and is often asymptomatic with frequent reoccurrence. Higher prevalence of asymptomatic UTI in females than males has also been observed by other workers (Abbey, 1987; Frank-Peterside and Oguike, 2006; Frank-Peterside and Eton, 2007). While 57% of the population studied showed insignificant growth, it will not be ruled out that the mixed bacterial growth obtained could be common contaminants or from careless sample collection. It can also be due to the presence of intestinal bacteria or contaminants from the vagina, feces or perineal skin especially in females. The bacterial species isolated in this study were seven as shown in table 2. These species have also been isolated in similar studies by different investigators of asymptomatic bacteriuria (Abbey, 1987; Olusanya *et al.,* 1993; Roos *et al*., 2006).

The most prevalent organism isolated in this study was *Escherichia coli.* The predominance of *E. coli* as the commonest aetiology agent of UTI has also been reported in previous studies (De Francesco *et al.*, 2001; Al Sweith *et al.*, 2005). It is not surprised that out of 60 isolates of this organism identified in this study; 35 were from male students while 25 from female. From the urinalysis test, 2 males that tested positive to nitrite harbored *E. coli* while out of 3 males with traces of nitrite, one harbored *Klebsiella* species and 2 with *Staphylococcus saprophyticus*. The second most common bacterial agent obtained in this study was *Klebsiella* species with 13 females and only 5 males infected with this organism.

This is followed by *Staphylococcus saprophyticus* which is usually found in infection among sexually active young women (Cheesbrough, 2004) and can also cause UTI in males of all ages; the organism has been isolated in young boys (Tolaymat and Al-Jayousi, 1991), male homosexuals (Hovelius *et al*., 1984); 4 females and 4 males were infected with this organism. It has often been postulated that men and women in the age range of the sample collection (16-26) are sexually active and this may predispose them to UTI (Cheesbrough, 2004). *Staphylococcus aureus* which has often been said as a leading cause of urinary tract infections and sexually transmitted diseases in Nigeria (Frank-Peterside and Oguike, 2006) accounted for only 5% of the isolates. *Enterococcus faecalis* accounted for 5% of the isolates and it has been reported as an agent of UTI. *Proteus species* and *Pseudomonas aeruginosa* accounted for 3% and 1% of the isolates respectively and their presence in the urine sample should however not be overlooked because they have been reported as agents of UTIs (Chessbrough, 2002).

This study has showed that some bacterial isolates are more susceptible to treatment with certain antibiotics than others. This should be the consideration for the choice of an antibiotic. Three antibiotics, Ciprofloxacin, Nitrofurantoin and Ofloxacin were found to be most effective for

Gram negative and Gram positive organisms. Ofloxacin and ciprofloxacin are members of the quinolones which are effective against a wide range of organisms (Al Sweith *et al*, 2005).

At the present time, the cost of obtaining this antibiotic is quite high and because of this, fewer numbers of people have access to it, thus diminishing the chances of misuse and of organisms developing resistance to it. However, a large proportion of the isolates were sensitive to nitrofurantoin and this should be considered among the first line of drugs for treating cases of urinary tract infection. The result is comparable with Onifade *et al.*, 2005 and Aiyegoro *et al*., 2007 in a similar study on UTIs. Some of the organisms were also sensitive to Gentamicin; while *Staphylococcus aureus*, *Proteus species*, and *Pseudomonas aeruginosa* were resistant to it. The isolates however showed high resistance to ceftazidime (0%), cefuroxime (10%), cefixime (10%) and augmentin (0%). These drugs cannot be used in treating urinary tract infections.

Correspondence:

F. O. Alao
sunfelixnaija@gmail.com
Department of Biological Sciences, College of Natural and Applied Sciences, Bells University of Technology, P. M. B 1015, Ota, Ogun State.

REFERENCES

1. Abbey, S.D. 1987. “Asymptomatic Bacteriuria in the Port Harcourt Metropolis of Nigeria”. *Microbios*. 49(199):73-75.

2. Aiyegoro, O.A., O.O. Igbinosa, I.N. Ogunmwonyi, E.E. Odjadjare, O.E. Igbinosa, and A.I. Okoh. 2007. “Incidence of Urinary Tract Infections (UTI) among Children and Adolescents in Ile-Ife, Nigeria”. *Afri. J. Microbiol. Res.* 013-019.

3. Al Sweith, N., W.M. Jamal, and V.O. Rotimi. 2005. “Spectrum and Antibiotic Resistance of Uropathogens isolated from Hospital and Community Patients with Urinary Tract Infections in 2 Large Hospitals in Kuwait”. *Med. Princ. Practice.* 14(6):401-407

4. Atlas, R.M. 1986. *Basic and Practical Microbiology Textbook*. Macmillan Co: New York, NY. 53.

5. Barr, J.G., J.W.K. Ritchie, O. Henry, M.E.L. Sheikh, and K.E.L Deeb. 1985. “Microaerophilic/Anaerobic Bacteria as a Cause of Urinary Tract Infection in Pregnancy”. *J. Obstet. Gynecol.* 92:506-510.

6. Bents. 2002. “This Woman has an Acute Uncomplicated Urinary Tract Infection?”. *JAMA*. 287:2701-2710.

7. Brooks, G.F., J.S. Butel, and S.A. Morse. 1998. *Jawetz, Melnick and Adelberg’s Medical Microbiology*. Appleton Large: Stamford CT. 665-666.

8. Bulakh, P.M. 2003. “Renal Function Tests and Urine Analysis”. *Medical Laboratory Science Theory*. 111-151.

9. Cheesbrough, M. 2000. *District Laboratory Practice in Tropical Countries (Part 2)*. Cambridge University Press: Cambridge, UK. 99-107.

10. Cheesbrough, M. 2002. *District Laboratory Practice in Tropical Countries (Part 2)*. Cambridge University Press: Cambridge, UK. 120.

11. Cheesbrough, M. 2004. *District Laboratory Practice in Tropical Countries (Part 2)*. Cambridge University Press: Cambridge, UK. 115.

12. De Francesco, M.A., G. Ravizzola, L. Peroni, R. Negrini, and N. Manca. 2007. “Urinary Infections in Brescia, Italy: Etiology of Uropathogen and Antimicrobial Resistance of Common Uropathogens”. *Med. Sci. Monit*. 13(6):136-144.

13. Frank-Peterside, N. and N. Oguike. 2006. “Asymptomatic Significant Bacteriuria among Students of the University of Port-Harcourt”. *Nigeria. Nig. J. of Microbiology*. 20(3):1252-1257.

14. Frank-Peterside, N. and E. Eton. 2007. “Asymptomatic Bacteriuria among Pupils of the University of Port-Harcourt Demonstration Primary School, Rivers-State, Nigeria”. *Asian J. Microbiol. Biotech. and Env. Sci*. 9(3):543-546.

15. Geerlings, S.E. and E.C. Brouwer. 2000. “Is a Second Urine Specimen Necessary for the Diagnosis of Asymptomatic Bacteriuria”. *Clin. Infect. Dis*. 31(3):3-4.

16. Grude, N., Y. Tveten, and B.E. Kristiansen. 2001. “Urinary Tract Infections in Norway: Bacterial Etiology and Susceptibility”. *Clin. Microbial. Infec*. 7:543-547.

17. Hovelius, B.S., S. Collen, and P.A. Mardh. 1984. “Urinary Tract Infections in Men Caused by *Staphylococcus saprophyticus*”. *Scan. J. Infect. Dis.* 16:37 – 41.

18. Kass, E.H. 1962. “Pyelonephritis and Bacteriuria a Major Problem in Preventive Medicine”. *An. Intern. Med.* 65(1): 46-53.

19. Nicolle, L.E. 1997. “Asymptomatic Bacteriuria in the Elderly”. *Infect .Dis. Clin. N. Amer*. 11(3):642-647.

20. Nurullaev, R.B. 2004. “The Role of Asymptomatic Bacteriuria in Epidemiologic Study of the Urinary Tract Infection (UTI)”. *Lik Aprava*. 7:23-25.

21. Olusanya, O. A. Oguniedun, and T.A. Fakoya. 1993. “Asymptomatic Significant Bacteriuria among Pregnant and Non-Pregnant Women in Sagamu, Nigeria.” *West. Afri. J. Med.* 12(1):27-34.

22. Onifade, A.K., F.O. Omoya, and D.V. Adegunloye. 2005. “Incidence and Control of Urinary Tract Infections among Pregnant Women attending Antenatal Clinics in Government Hospitals in Ondo State, Nigeria”. *J. Food. Agric. Environ.* 3(1):37-38.

23. Prescott, M., P. Harley, and A. Klein. 1999. *Microbiology, 4th edition*. Mc Graw-Hill Book Co.: New York, NY. 869.

24. Raz, R., R. Colodner, and C.M. Kunin. 2005. “Who are you - *Staphylococcus saprophyticus*?”. *Clin. Infect. Dis.* 40:896–898.

25. Roos, V., G.G. Ulett, M.A. Schembri, and P. Klemm. 2006. “The Asymptomatic Bacteriuria *Escherichia coli* strain 83972 Outcompetes Uropathogenic *E. coli* Strains in Human Urine”. *Infect Immun.* 74(1):615-24.

26. Smith, M.B.H. 1994. “Screening for Urinary Tract Infection in Asymptomatic Infants and Children”. *Canadian Guide to Clinical Preventive Health Care*. 220 – 300.

27. Stamm, W.E. and T.M. Hooton. 1993. “Management of UTIs in Adults”. *N. Eng. Med.* 239:1328–1334.

28. Thompson, R.B. and J.M. Miller. 2003. *Manual of Clinical Microbiology. 8th ed.* ASM Press: Washington, DC. 322.

29. Tolaymat, A. and Z. Al-Jayousi. 1991. “*Staphylococcus saprophyticus* Urinary-Tract Infection in Male Children”. *Child Nephrol. Urol.* 11: 100-102.

30. Whalley, P. 1967. “Bacteriuria of Pregnancy”. *Am. J. Obstet. Gynecol*. 97(5):723-38.

ABOUT THE AUTHORS

Felix Oluwasegun Alao, is an Assistant Lecturer at the Department of Biological Sciences, Bells University of Technology, Ota, Nigeria. He holds a B.Sc. and M.Sc. in Medical Microbiology from the University of Lagos, and is currently a doctoral student at the same institution. He is a member of the American Society for Microbiology and the Association of Industrial Microbiologists of Nigeria. His research interest is in the areas of medical microbiology /molecular epidemiology of virulence genes of *Staphylococcus saprophyticus* from women presenting urinary tract infections

Fayokemi Ayotunde Akintunde, holds a B.Sc. in Microbiology at the Bells University of Technology, Ota, Ogun State, Nigeria. He undertook Industrial training in the Laboratory of Ibeju-Lekki General Hospital Akodo, Lagos and is planning to obtain a Masters degree in Public Health and Hygiene following his mandatory National Youth Service Corps. His research interests include microbiology, public health, and hygiene research.

SUGGESTED CITATION

Alao, F.O. and F.A. Akintunde. 2012. “Asymptomatic Bacteriuria among Students of Bellstech, Ota, Nigeria”. *Pacific Journal of Science and Technology*. 13(2):342-347.

Pacific Journal of Science and Technology