Original Article

SOCIO-DEMOGRAPHIC FACTORS ASSOCIATED WITH ASYMPTOMATIC BACTERIURIA IN PRIMARY SCHOOL CHILDREN IN ENUGU, NIGERIA

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Abstract

Background: Urinary tract infection (UTI) can be symptomatic or asymptomatic (asymptomatic bacteriuria) and is a common cause of chronic kidney disease in children. It is also a common cause of morbidity and mortality among children in developing countries like Nigeria. Asymptomatic bacteriuria is said to be more common in school-age girls and children of low socio-economic class.
Objectives: To determine the relationship between asymptomatic bacteriuria and age, sex and socio-economic status of primary school children in Enugu, Nigeria.
Materials and Methods: This was a cross-sectional descriptive survey involving four hundred apparently healthy primary school children aged 6 to 12 years. A pre-tested, care-giver administered questionnaire was used to obtain information about the participants such as age, sex, socio-economic status, history of fever and antibiotic usage in the two weeks preceding the study. Following a clinical examination, a sample of spot mid-stream urine was collected from each participant for dipstick urinalysis and urine microscopy and culture.
Results: The age of the children ranged from 6 to12 years. Fifty seven of the 400 children were noted to have asymptomatic bacteriuria with a female preponderance of 14:1, although not statistically significant (P = 0.787). Forty-six (80.7%) of the fifty-seven children were aged nine years and above. There was a predominance of positive cases (54.4%) in the middle socioeconomic class but not statistically significant (P = 0.449).
Conclusion: Asymptomatic bacteriuria is commoner in adolescent school aged female and in children of middle socioeconomic class.
Keywords: Asymptomatic bacteriuria, Socio-demographic, Prevalence.

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INTRODUCTION

Infection of the urinary tract is one of the most common bacterial infections in humans, both in the community or in hospital settings and has been reported to occur in all age groups and in both genders.[1] Urinary tract infection (UTI) could present as either a clinical (symptomatic) or subclinical (asymptomatic) disease that involves the genito-urinary system.[1] Asymptomatic bacteriuria (ASB) is defined as significant bacterial count in the urine, usually 10^5 or more colony forming units (cfu) per millilitre in a child without symptoms referable to urinary tracts.[2] High prevalence of asymptomatic bacteriuria is largely due to poor hygiene, not knowing how to clean up after toilet use and sharing of towels and other clothing with older children and adults.[3] Poor toilet sanitation and maintenance coupled with non-availability of water to clean the toilets in primary schools are well known predisposing factors to poor hygiene in developing countries like ours.[4] Bacteria usually abound in such places and primary school children spend most of their active days in the school premises and inevitably make use of the toilets.[4] ASB could lead to symptomatic UTI especially in the presence of risk factors which may cause renal parenchymal infections. This renal parenchymal infection can lead to renal scarring which is the prelude to chronic morbidities associated with urinary infections, such as hypertension, reduced renal function and chronic kidney disease.[5,6] ASB in a child can also be an indication of an underlying structural or functional urinary abnormality which may require specific therapy to prevent reoccurrence of infection and progression to renal damage.[7] Asymptomatic bacteriuria has been reported to be associated with an increased risk of symptomatic UTI and may actually represent the beginning of symptomatic UTI in school

children.[8] This study determined the influence of age, sex and social status on the incidence of asymptomatic bacteriuria in primary school children in Enugu, Nigeria.

MATERIALS AND METHODS

Study Design and Population

This was a cross-sectional descriptive study conducted in Enugu South LGA of Enugu State, Nigeria which has an area of 67 square kilometres of land and a population of 196,723.[9] Enugu South LGA is one of the three local government areas that make up Enugu metropolis. Majority of the residents of Enugu South LGA are of the Igbo ethnic group. The inhabitants are people with different educational background and religious beliefs. The working-class population is made up of different groups of individuals in various occupations but most of them are civil servants.[10] There was a total of 110 officially registered primary schools in Enugu South LGA. The subjects were primary school children aged 6 to 12 years, who attended primary schools located in Enugu South LGA comprising 32 public and 78 private primary schools.

Sample Size Estimation

The sample size of 400 was calculated using the formular[11] for cross-sectional survey:

$N = Z^2P (1-P)/d^2$.

Where n = the minimum sample size,

Z = 1.96 (a standard normal deviate, usually set at 1.96, which corresponds to the 95% confidence level),

P = Prevalence of ASB from a previous study in South-East Nigeria = 30%,[12]

d = 0.05 (degree of accuracy desired).

Sampling Method

Multi-stage sampling method was used in this study. Enugu South LGA has 110 officially registered primary schools: 32 public and 78 private schools and a total population of 22,375 primary school children comprising 11,053 males and 11,322 females[13] with male to female ratio of approximately 1:1.

The total number of primary schools selected was achieved by using a proportionate sampling outcome of 10%.[14] Since there were 110 primary schools in Enugu South LGA, 10% of 110 gave 11 primary schools. The primary schools were then stratified proportionately into public and private primary schools.

Since there were 32 public and 78 private schools in the LGA, the ratio of public to private primary school was calculated to be 1:2.5. From this ratio, it meant that out of 11 primary schools selected, 3 primary schools were chosen from public schools and 8 from private schools.

These 3 public schools and 8 private schools were then selected using simple random sampling method.

The sample size of 400 was allocated proportionately to the schools selected using the Neymann proportional allocation formula.[15]

Preliminary Activities

Three visits were made to each of the selected schools; the first was to get approval from the School authorities and introduce the researcher, the study objectives and design. The second was to give brief lectures on asymptomatic bacteriuria and select the subjects. The third was to obtain parental consent from each of the selected subject.

A female nurse was recruited and repeatedly trained by the researcher until she understood all the procedures of urine sample collection in females.

The selected subjects were then given a detailed explanation on the procedures involved in urine sample collection and the location within the school premises the urine collection will take place.

Physical examination was carried out on each of the selected subjects. The perineum of each male subject was also examined to rule out abnormal urethral orifice and to note circumcision status and urinary stream.

Data Collection

A research proforma designed for the study was used to record the information obtained. The subjects who met the study inclusion criteria were given consent forms and letters to give their parents or guardians, explaining clearly the nature and purpose of the study. The subjects were allowed to go home with the research proforma where the parents or guardians helped with filling of the proforma to get relevant demographic information and past medical history of the subjects. The parents or guardians who could not fill the proforma properly were assisted by the researcher through telephone discussions and home visits.

Specimen Collection

Inside the school compound, two separate screened enclosures were set up; these were the enclosures where the urine collection took place, one for males and the other for females. The researcher and the female nurse (who was also the assistant to the researcher) supervised the proper collection of urine samples in males and females respectively. Aseptic procedures were observed. The researcher and the nurse wore

sterile disposable hand gloves, each for every subject. The researcher put on sterile disposable gloves and cleaned the external urethral orifice of each male subject with clean, sterile swab.[8]The nurse also cleansed between the labia of each female subject from front to back with a clean and sterile swab,[8] following which urine was passed with the labia separated to avoid contamination.

The midstream urine samples were collected in pre-labelled, sterile, boric acid containing bottles without allowing the bottles to come in contact with external genitalia, perineum or adjacent skin to avoid contamination. The urine samples collected were stored in a clean container filled with ice packs and then transported to Microbiology laboratory at UNTH for storage in the refrigerator at 4^0C and were subsequently processed within 4 hours of collection.[16]

Laboratory Procedures

Each urine sample was divided into two parts: One for dipstick analysis and the other for microscopy and culture. Dipstick analysis was done using SD UroColor^{TM}10 (Standard Diagnostics, INC Korea) for nitrite, leucocyte esterase and protein. The test strip was immersed in the urine for 2 seconds,[17] such that all reagent pads were covered by urine. Thereafter, excess urine was removed from the strip by tapping the edge of the strip on the rim of the urine container while holding the strip in a horizontal position to prevent interaction from adjacent reagent pads. The colour change after one minute was compared with the colour chart on the container label under good light with the strip horizontally placed to prevent mixing of chemicals if excess urine was still present. The other urine part was used for microscopy, culture and sensitivity. It was transferred to the Microbiology Laboratory of UNTH for analysis.

Urine microscopy was carried out by centrifuging 10 ml of urine sample at 2,000 revolutions per minute (rpm) for 5 minutes. The supernatant was discarded and a drop of the urine deposit was examined under the microscope at high magnification for pus cells, red blood cells, bacteria and casts.

Culture was done employing the quantitative method as described by Guttman and Stokes.[18] Each uncentrifuged urine sample was well mixed and subsequently inoculated unto well dried plates of blood agar and MacConkey agar as described by Uqurhart and Gould,[19] using a calibrated standard wire loop of 2 mm internal diameter which delivers 0.001 ml of urine per loopful. The wire loop was sterilized over a Bunsen burner flame before and after use. The culture plates were incubated prior to inoculation to avoid contamination. Using the standard wire-loop, urine sample was collected from the well mixed specimen and streaked well on to the well dried freshly prepared blood agar and MacConkey plates. The plates were incubated aerobically at 37^{o}C for 24 hours after which the colonies were counted with a colony counter. The number of colony forming units (CFUs) was multiplied by 1,000 to determine the number of microorganisms per ml in the original specimen.

Significant bacteriuria was defined as pure growth of $\geq 10^5$ colony forming units per ml from midstream urine sample. Growth less than 10^5 CFU/ml from the midstream urine sample were regarded as insignificant.[19] The subjects with asymptomatic bacteriuria were subsequently referred to the Paediatric clinic of the UNTH for follow up.

Data Handling and Statistical Analysis

The data obtained was recorded on the study proforma and entered into the computer and analysed using the Statistical Package for Social Science (SPSS) version 18 for Windows. Data collated were summarized using frequency, percentages, means and standard deviations.

Association between categorical variables was analysed using chi-square and logistic regression and statistically significant result was attained wherever a p-value was less than the significance level of 0.05.

RESULTS

Demographic Characteristics of the Study Population

A total of 400 apparently healthy children were enrolled into the study which was carried out from 29th April 2018 to 18th July 2018. There were one hundred and seventy-five (44%) males and 225 (56%) females. Their ages ranged from 6 to 12 years with a mean age of 10.13 ± 1.81 years. Generally, there was female gender predominance across all age groups except for the 6-year olds but this was not statistically significant. (χ^2 = 2.551, p = 0.863). All the male subjects were circumcised, had normal urethral orifice with normal urinary stream. Table I shows the demographic characteristics of the study population.

Table I: Demographic characteristics of the study population.

Age (years)	Male (%)	Female (%)	Total (%)
6	9 (5.2)	8 (3.5)	17 (4.3)
7	9 (5.2)	16 (7.1)	25 (6.3)
8	15 (8.5)	24 (10.7)	39 (9.8)
9	25 (14.3)	29 (12.9)	54 (13.5)
10	34 (19.4)	42 (18.7)	76 (19.0)
11	25 (14.3)	26 (11.5)	51 (12.6)
12	58 (33.1)	80 (35.6)	138 (34.5)
Total	**175 (100.0)**	**225 (100.0)**	**400 (100.0)**

Prevalence of Asymptomatic Bacteriuria (ASB) and Colony Counts.

Seventy (17%) of the 400 urine samples had bacterial growth but only 57 (14.25%) had colony counts $\geq 10^5$ colony forming units (CFU)/ml and thus qualified as cases of asymptomatic bacteriuria (ASB). (Table II).

Table II: Urine colony counts in the bacteriuric subjects.

Colony count (CFU/ml)	Frequency	Percent
$\geq 10^5$ (significant bacteriuria)	57	14.25
10^4 - 10^5	4	1.00
$<10^4$	9	2.25
Total	**70**	**17.00**

The Gender Distribution of Children with ASB

Table III shows the gender distribution of children with ASB. Out of the 57 children with ASB, 33 (57.9%) were females and 24 (42.1%) males with a male: female ratio of 1:1.4. The gender specific prevalence of ASB in females was 14.7% (33/225) while that of males was 13.7% (24/175). The prevalence of ASB did not differ significantly between females and males. ($\chi^2 = 0.073$, p = 0.787).

Table III: Distribution of ASB by Gender

	ASB				
	No (%)	**Yes (%)**	**Total**	χ^2	**p-value**
Gender					
Male	151 (86.1)	24 (13.7)	175	0.073	0.787
Female	192 (85.3)	33 (14.7)	225		
Total	**343**	**57**	**400**		

The Age Distribution of Children with ASB

Table IV shows that the prevalence of ASB in the early adolescent group (9 to 12 years) was 14.4% (46/319) while a prevalence of 13.6% (11/81) occurred in the preadolescent group (6 to 8 years) but the prevalence of ASB between these two age groups was not statistically significant. ($\chi^2 = 1.514$, p = 0.218).

Table IV: Distribution of ASB by Age

	ASB				
	No (%)	**Yes (%)**	**Total**	χ^2	**p-value**
Age Group (years)					
6-8	70 (86.4)	11 (13.6)	81	1.514	0.218
9-12	273 (85.6)	46 (14.4)	319		
Total	**343**	**57**	**400**		

Socioeconomic Class Distribution of Subjects with ASB.

Table V shows the family socioeconomic distribution of subjects with ASB. There was a predominance of ASB in the children of the middle socioeconomic class (SEC), 31 out of 57 (54.4%), followed by the upper SEC, 14 out of 57 (24.6%), then the lower SEC, 12 out of 57 (21.0%). There was no significant association between prevalence of ASB and socioeconomic classes. ($\chi^2 = 1.600$, p = 0.449).

Table V: Socioeconomic class distribution of subjects with ASB.

Socioeconomic class by Oyedeji	Significant bacteriuria		
	Yes (%)	**No (%)**	**Total**
Upper or Social class 1	14 (24.6)	105 (30.6)	119
Middle or Social class 2	31 (54.4)	156 (45.5)	187
Lower or Social class 3	12 (21.0)	82 (23.9)	94
Total	**57 (100.0)**	**343 (100.0)**	**400**

DISCUSSION

The prevalence of 14.25% for ASB obtained in this study was similar to what was observed in other studies in Nigeria.[20,21,22] Mbakwem-Aniebo *et al*[20] in Port Harcourt, Nigeria documented a prevalence of 14.5% amongst primary school children, Akor *et al*[21] studied primary school children in Jos and found a prevalence of 14% while Onifade *et al*[22] in Ibadan reported a prevalence of 15.5% which are all comparable with the present study. Just like the present study, Mbakwem-Aniebo *et al*,[20] Akor *et al*[21] and Onifade *et al*[22] equally enrolled apparently healthy primary school pupils using the midstream urine and 100,000 colony forming units were used as the diagnostic criterion.

In contradistinction, some studies have reported ASB prevalence rates much higher than that obtained in the present study. Salem *et al*[23] in 2009 documented a prevalence rate of 30% in Egyptian school children which was also higher than the prevalence rate gotten from the present study. The fact that Salem *et al*[23] studied children with type 1 diabetes could have also contributed to the high values they found. Children with diabetes mellitus tend to have higher rates of ASB because of reduced immunity and their urines which are laden with sugar could be a culture medium.[24] While this present study was carried out in a community, the one by Salem *et al*[23] was a hospital-based study. Such children are considered to have a higher risk of ASB.[25] Sample contamination may also be more in them.[25]

In Nigeria, reports of higher ASB rates include 30% by Azubuike *et al*[26] among primary school children in Awka in 1994 and 48% by Alo *et al*[5] in rural primary school children in Ebonyi State in 2012. Azubuike *et al*[26] enrolled a relatively lower sample size of 200, compared to the 400 in the current study. The sample size of 200 in the study by Azubuike *et al*[26] did not represent the appropriate percentage of the total population of the primary school children in Awka[14] and one primary school was used in their study which could lead to poor distribution of subjects and may perhaps be responsible for their high ASB prevalence.

Alo *et al*[5] used a diagnostic cut-off of $>10^4$ CFU/ml instead of $>10^5$ CFU/ml as their definition of significant bacteriuria using midstream urine and this could also be one of the reasons for the high ASB rate. Moreover, this high ASB rate of 30% and 48% documented by Azubuike *et al*[26] and Alo *et al*[5] respectively could be attributed to the fact that the studies were carried out among children in rural areas who are known to have low levels of hygiene and poor health consciousness and Awka was considered rural two decades ago.[26]
On the other hand, some other studies documented ASB prevalence rates much lower than that observed in the present study. Elegbe *et al*[34] in Ile-Ife in 1987 and Akinkugbe *et al*[27] in Ibadan in 1988 documented prevalence rates of 5% and 4.7% respectively among school children which were quite lower than the present study. These low rates were attributed to the greater attention paid to health education in their school curriculum where all the rudiments of environmental and personal hygiene were taught.

There was no statistically significant difference in the prevalence of ASB across the two age groups (pre-adolescent and early adolescent) in this study. Nonetheless, the higher rate of ASB was found in the 9-12 years group (the early adolescent age group). This observation is similar to the findings of Mbakwem-Aniebo *et al*[20] who recorded higher ASB prevalence in the 10-12 years age group. The higher prevalence of ASB in this age group may not be unconnected with the fact that this age group is the early adolescent age and some of the subjects may have been sexually exposed.[15] Moreover, education on basic perineal hygiene may not have been properly inculcated in the early adolescent age groups and they, more often than not, will want to do the perineal cleaning themselves unlike the pre-adolescents, where the perineal cleaning are usually done by the parents or older relatives.

In the present study, ASB was commoner in females than in males. The observed trend is in tandem with findings contained in the studies done by Alo *et al*,[5] Sawalha *et al*,[28] Saleh *et al*[29] and Mbakwem-Aniebo *et al*.[20] The female predominance could be attributed to the short female urethra, which is in close proximity to the anus from where it can be easily contaminated by faecal matter, alterations in vaginal microflora which play a role in encouraging vaginal colonization by bacterial pathogens and incomplete voiding of urine among school girls.[30] Some predisposing factors such as non-circumcision that predisposes males to ASB[31] were not observed in this study as all male children in this study were circumcised. This may also have contributed to the lower prevalence of ASB in males as seen in this present study. This observation is however in disagreement with results obtained by Wennerstrom *et al*[32] who found higher ASB rates in males compared to females. This male predominance seen in the study by Wennerstrom *et al*[32] was attributed to high incidence of uncircumcised males in their study.

In the present study, higher prevalence of ASB was found in the subjects drawn from the middle socioeconomic families, although there was no significant association between prevalence of ASB and family socioeconomic classes. This is however different from the findings of Savage *et al*[33] who documented a higher prevalence of asymptomatic bacteriuria in school children drawn from lower socioeconomic families. A similar trend was noted by Elegba *et al*[34] in Ile-Ife, Nigeria. Observed trend in this study is in tandem with those noted in the studies done by the Newcastle Asymptomatic Bacteriuria Research group[35], Kunin *et al*[36] and Chukwu *et al*[37] that did not find any significant association between asymptomatic bacteriuria and socioeconomic class.

CONCLUSION

In conclusion, ASB remains an important problem in school children. The prevalence of ASB in Enugu South LGA was 14.25% and is commoner in females and children aged 9-12 years.

Hence, it is advocated that periodic urinary screening should be introduced in primary school system and a robust health education should be included in their school curriculum focusing on the importance of all-round cleanliness and regular environmental sanitation.

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CONFLIT OF INTEREST

The authors declare that there are no conflicts of interest.

CONSENT

An informed written consent was obtained from parents/guardians of each selected pupil.

ETHICAL APPROVAL

Ethical clearance/consent was obtained from the Health Research and Ethics Committee of the University of Nigeria Teaching Hospital (UNTH).

Permission was also obtained from the Enugu State Universal Basic Education Board (ESUBEB), Enugu State Ministry of Education as well as from various head teachers of the selected primary schools.

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