BACTERIOLOGY OF ASYMPTOMATIC BACTERIURIA IN PRE-SCHOOL CHILDREN IN ENUGU

BY

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SUMMARY

Objective: This study was undertaken to ascertain the predominant bacterial isolates in the urine of pre-school children with asymptomatic bacteriuria and determine their sensitivity pattern.

Methods: This was a cross-sectional study in which analysis of urine cultures obtained from 800 nursery school children aged 2- 5 years in Enugu, South East Nigeria, between January and November 1989 was done.

Results: Three hundred and ninety-one (48.8%) of the children were females while 409 were males, with a male female ratio of 1.05:1.The mean age was 3.79 ±1.3yrs and seventeen out of 800 (2.1%) cultures were positive, 2 (12%) from male children and 15 (88%) from female children. Nine out of the 17 (53%) positive cultures yielded ***Escherichia Coli***.(E. Coli), 5 specimens (29%) yielded ***streptococcus faecalis*** *(strept. faecalis)* while the remaining 3 specimens yielded ***Proteus*** species (6%), ***staphylococcus pyogenes*** (6%) and Micrococcus (6%) respectively. There was 100% resistance of the ***E. Coli, staphylococcus pyogenes*** *(staph. pyogenes)* ***and proteus*** *species* to Ampicillin, while there was 100% sensitivity of all the organisms to nitrofurantoin. Seventy-eight percent of the ***E. Coli*** were sensitive to gentamycin while 100% sensitivity to this drug was obtained for ***staphylococcus. pyogenes***, ***micrococcus*** and ***proteus spp***. However only 40% of the ***Strept. faecalis*** was sensitive to gentamycin. Most of the organisms (88%) were resistant to co-trimoxazole.

Conclusion: It is concluded that nitrofurantoin or gentamycin should be used as first line drugs in urinary infections rather than co-trimoxazole which is the practice at present.

Keywords: *Asymptomatic, Bacteriuria, Pathogens, Pre-School Children.*

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INTRODUCTION

In the paediatric age group, urinary tract infections have been shown to be significant causes of morbidity [1, 2, 3] and mortality [4] .Studies [5, 6, 7] have also shown that they are of greater long term significance in childhood than later in life. These infections may be symptomatic or asymptomatic. Earlier studies revealed a higher prevalence of symptomatic bacteriuria in Nigeria compared to the Western world [8, 9]. The commonest organisms isolated in both symptomatic and asymptomatic infections are Escherichia coli [10] reflecting the origin of the infecting species which is usually from the microflora of the intestine and perineum. Recent studies from some parts of the country have shown a changing trend in the bacteriology of urinary tract infections[11]. It is thus most desirable to determine the prevalent organisms causing urinary infections as well as their sensitivity pattern in our environment. The information obtained therefore will guide management of this condition.

MATERIALS AND METHODS

The study was done in Enugu, the capital of Enugu State, South East Nigeria. It is a metropolitan city with a total of 31 registered nursery schools distributed within high and low population density areas of the city.

We did a cross-sectional study whereby 800 children aged 2-5 years were randomly selected from 14 of the registered nursery schools within Enugu metropolis. The nursery schools were selected by systematic random sampling from a constructed frame of 31 registered nursery schools in Enugu, whereby every second nursery school was selected. Verbal and written consent of the parents and school proprietors were obtained at commencement of the study The selected schools had a total pupil population of 4,560. The number of children screened in each school was proportional to the school population and there was no sex stratification of the children.

In each school, the pupils were assigned serial numbers, and alternate numbers enrolled into the study until the predetermined target number for the school was attained. Drop outs were replaced with the next alternate number. A brief history of each child was obtained through a self-administered questionnaire distributed to the mothers and any child on antimicrobial treatment a week before enrolment was excluded from the study.

All the children were toilet-trained since this was a pre-requisite for admission into the selected schools. Midstream urine specimens were collected from these children into sterile bottles containing 1.8% boric acid, (no special preparation of the perineum was made[1, 12] and urine collection was supervised by one of the authors (H.U.O)). After collection, the urine specimens were immediately stored in an ice box at 4^0C and transported to the microbiology laboratory of the University of Nigeria Teaching Hospital , where they were cultured within 2 –3 hours. Second urine specimens were obtained from patients with positive cultures of the first specimens.

Prior incubation of plates before culture was done to rule out contamination. A semi-quantitative method of culture as described by Guttman and Stokes [12] was employed using a calibrated standard loop with internal diameter of 3mm and delivery of 0.003ml of urine per loopful. The loop was sterilized using a Bunsen burner before immersion in the well-mixed uncentrifuged urine specimen which was streaked unto well dried plates of CLED medium using the method of Uqurhant and Gould [13].

The plates were incubated at 37^0C for 24 hours after which the colonies were counted with a colony counter. Bacterial counts of $\leq 10^4$/ml were regarded as negative while those $\geq 10^5$/ml of pure growth on 2 consecutive cultures were taken as positive.

Mixed growths and growths giving bacterial counts 10^5/ml were repeated and only those that gave pure growth of counts $\geq 10^5$/ml were then regarded as positive. In all

positive cases organisms were identified using standard methods of Collins and Lyne [14]. A sensitivity test by the disc diffusion technique of Stokes 15 was used to determine the sensitivity pattern of the isolates. Sensitivity test agar (Oxoid) was used to produce the sensitivity lawn. Antibiotic sensitivity disc oxoid 178E containing the following; Nalidixic acid 30mg; nitrofurantoin 200mcg; Compound sulphonamide 30mcg, Streptomycin 25mcg; Tetracycline 50mcg; Co-trimoxazole 25mcg and Single discs of gentamicin were also used. The control panel consisted of Oxford Staphylococcus aureus (NCTC 6571) and Escherichia coli (NCTC 104 [18]). Sensitivity was determined by comparing the zones of inhibition of the test organisms with those of the control organisms. Inhibition zones less than those of control organisms by 2mm across were regarded resistant.

RESULTS

Three hundred and ninety-one (48.8%) of the 800 children were females while 409 were males, with a male: female ratio of 1.05:1.The mean age was 3.79 ±1.3yrs (see table 1).

Table 1: AGE AND SEX DISTRIBUTION OF THE 800 PUPILS

Age last birthday (Years)	Total No	Male(%)	Female (%)	% of Total
2	59	29(49.2)	30(50.8)	7.4
3	275	142(51.6)	133(48.4)	34.4
4	242	124(51.2)	118(48.8)	30.2
5	224	114(50.9)	110(49.1)	28
Total	800	409(51.2)	391(48.8)	100

After 24 hours of incubation 17 (88%) out of the 800 specimens (2.1%) had significant bacteriuria i.e.$\geq 10^5$ colonies /ml on 2 consecutive cultures (see table2), 776

TABLE 2
CULTURE RESULTS OF URINE SPECIMENS

No of Colonies CFU/ml	No of Specimens from males	No of Specimens from Females	Total No of Specimen
$\leq 10^5$	1	6	7(0.9%)
$\geq 10^5$	2	15	17(2.1%)
No growth	406	370	776(97%)
TOTAL	409	391	800

specimens yielded no growth while 7 specimens had non-significant growth. Of the 17 positive specimens, fifteen (88%) were from Females while 2 (12%) were from males (table 3). ***Escherichia Coli*** was isolated in 9 (53%) out of the 17 positive cultures.

TABLE 3
AGE AND SEX DISTRIBUTION OF PUPILS WITH POSITIVE CULTURES

S/NO	AGE	SEX	ORGANISM ISOLATED
7	2yrs	F	E. Coli
27	3½yrs	F	E. Col
46	4yrs	M	Strept. faecalis
52	5yrs	F	Strept faecalis
264	5yrs	F	Strept. faecalis
395	4yrs	F	E. Coli
431	4yrs	F	E. Coli
462	2yrs	F	E. Coli
472	3yrs	F	E. Coli
487	3yrs	F	E. Coli
583	5yrs	F	Micrococci
590	5yrs	F	Staph. Pyogenes
599	3yrs	M	Proteus spp
608	3yrs	F	Strept. faecalis
673	5yrs	F	E. Coli
708	3yrs	F	Strept. faecalis
763	4yrs	F	E. Coli

Escherichia Coli was 100% resistant to ampicillin and co-trimoxazole, and 100% sensitive to nitrofurantoin and compound sulphonamide (table4). It was also over 88% sensitive to colistin and kanamycin and over 77% sensitive to gentamycin and streptomycin. ***Streptococcus faecalis*** was 100% sensitive to nitrofurantoin and 80% sensitive to ampicillin and kanamycin, while

being 80% resistant to co-trimoxazole. The Staphylococcus isolated was sensitive to gentamycin and nitrofurantoin. The ***Proteus*** isolates were sensitive to nitrofurantoin, gentamycin and nalidixic acid. However ***Micrococcus*** was sensitive to all the antibiotics tested except colistin, tetracycline and compound sulphonamide.

DISCUSSION

Significant bacteriuria in this study was taken as 10^5cfu/ml and based on this, prevalence obtained was 2.1%. This result is comparable to results from developed countries [16]. This study also revealed that Escherichia Coli is the predominant causative organism especially in females which is also the case in developed countries.[16] Other studies [9,17] from other parts of this country have given higher prevalence of bacteriuria, though in an older age group. At Ibadan, Akinkugbe et al had a prevalence of 4.7% in children aged 3 –12 years, while Eliegbe et al at Ile-Ife had a prevalence of 5%. Studies within Nigeria have shown *Escherichia Coli* to be the predominant causative organism in urinary infections.[17, 18]. At Ile-Ife Eliegbe[17] recovered *Escherichia Coli (E. coli)* in 68% of his cases while Abdurrhaman at Zaria recovered it in 7 out of 16 (44%) positive cultures, the remaining 9 cultures in his series grew various other organisms. However Akinkugbe et al [8] at Ibadan recovered *E. Coli* in only 3 out of 12 (25%) positive cultures in infants from a rural area while *E. Coli* was recovered in 4 out of 6 (67%) positive cultures in infants from an urban area. There was no explanation for this variation .This may be due to higher risk of faecal contamination in urban infants who wear diapers.

In another study by Akinkugbe [9] on older children aged 3 – 12 years *Klebsiella spp.* was found to be the commonest causative organism being recovered in 21 out of 61 (34%) positive cultures, and *E. Coli* in 16 out of 61 (26%) positive cultures. In this study there was 100% resistance of the E. Coli recovered, to ampicillin and co-trimoxazole. In contrast to this Akinkugbe et al [9] in their study found that the E. Coli they recovered was sensitive to co-trimoxazole in 81% of cases and they recommended co-trimoxazole as a first line drug in the treatment of urinary tract infections in children. The 100% resistance of ampicillin and co-trimoxazole obtained in this study may be explained by the wide spread and indiscriminate use of these antibiotics in our community Ibe et al[19] had noted this high resistance to ampicillin of bacterial cultures from the Newborn special care unit of University of Nigeria Teaching Hospital, Enugu The significance of asymptomatic bacteriuria lies in the fact that same organisms can cause symptomatic infections, and the same percentage of children developing renal scars after symptomatic infections do so following asymptomatic infections[10]. Hence knowledge of the bacteriology and sensitivity pattern of organisms causing urinary infections provides a guide as to the choice of antibiotics to be administered prior to the culture results; and for empirical treatment.

In conclusion, most urinary pathogens in our environment were found to be more sensitive

to gentamycin and nitrofurantoin rather than to co- trimoxazole and ampicillin which were more commonly used.

We recommend that nitrofurantoin or gentamycin should be used as first line drugs in urinary

infections rather than co-trimoxazole. This however should be subject to modification based on the culture results in each individual case.

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ANTIBIOTICS	E. COLI (9)		S. FAECALIS (5)		MICROCOCCUS (1)		PROTEUS (1)		S. PYOGENES (1)	
	S	R	S	R	S	R	S	R	S	R
Ampicillin	-	9	4	1	-	1	1	-	-	1
Colistin	8	1	2	3	1	-	-	1	1	-
Nitrofurantoin	9	-	5	-	1	-	1	-		-
Nalidixic acid	6	3	1	4	1	-	1	-	1	-
Tetracycline	2	7	-	5	-	1	-	1	-	1
Co-trimoxazole	-	9	1	4	-	1	1	-	-	1
Kanamycin	8	1	4	1	1	-	1	-	1	-
Gentamycin	7	2	2	3	1	-	1	-	1	-
Compound Sulphonamide	9	-	3	2	-	1	-	1	-	1
Streptomycin	7	2	2	3	1	-	1	-	1	-

TABLE 4
ANTIBIOTIC SENSITITIVITY OF BACTERIA ISOLATED FROM URINE SPECIMENS WITH POSITIVIE CULTURES