Antimicrobial sensitivity pattern of organisms causing urinary tract infection in children with sickle cell anaemia in Ibadan, Nigeria.

***B. J. Brown[1], A. O. Asinobi[1], O. J. Fatunde[1], K. Osinusi[1] and N. A. Fasina[2]**

Departments of Paediatrics[1] and Medical Microbiology[2],
University College Hospital, Ibadan, Nigeria.

Summary

As part of a larger project on childhood urinary tract infection, antimicrobial sensitivity tests were carried out on the bacterial isolates from the urine of febrile children seen at the University College Hospital, Ibadan, Nigeria.

***Methodology*: Midstream urine specimens were collected from 171 sickle cell anaemia children and from an equal number of haemoglobin-A controls and cultured by standard methods. Sensitivity to eleven antimicrobials was tested using the disc-diffusion technique of Stokes.**

***Results*: Significant bacteriuria was obtained from 37 children with sickle cell anemia and 27 controls. The isolates were Escherichia coli, Klebsiella species, Non-haemolytic streptococcus. β-haemolytic Streptococcus, Salmonella, Proteus and Pseudomonas species. Sensitivity was highest to Pefloxacin to which over 94% of the organisms were sensitive followed by Ceftriaxone (over 85%) and ceftazidime (over 85%). Sensitivities to nalidixic acid and cefuroxime were between 67.6% and 74.1%. Most of the isolates were resistant to gentamicin, amoxycillin, cotrimoxazole and ampicillin. In general the sensitivity pattern in the sickle cell anaemia group was similar to the pattern in the control group.**

***Conclusion*: Aetiological agents of childhood UTI in this environment are resistant to most of the drugs commonly recommended for its treatment. Nalixidic acid and cefuroxime are recommended as first line drugs while awaiting results of sensitivity testing. Ceftriazone and ceftazidime should be reserved for cases of non-response to first line drugs and in severe cases. Pefloxacin should be considered potential drug of treatment particularly in multi-drug resistant infections.**

Keywords: ***Urinary tract infection, Antimicrobial sensitivity, Sickle cell anaemia.***

Résumé

Comme une partie d'un très grand projet sur l'infection urinaire d'enfance, des épreuve de la sensibilité antimicrobienne ont été effectuées sur des isolâtes bacteriens de l'urine des enfants fébriles soignés au College hospitalo-universitaire d'Ibadan, Nigeria.

Méthodologie: Echantillons d'urine en plein milieu ont été collectionnées chez 171 enfants atteints de la drépanocytose et à travers la méthode normale d'un nombre égal de contrôle et culture hémoglobine -A. La sensibilité de onze antimicrobiens ont été examinées tout en utilisant la technique de la diffusion disque d'attaques.

Resultats: La bactériurie important a été trouvée chez 37 enfants atteints de la drépanocytose, et 27 contrôles. Les isolâtes étaient escherichia coli, l'espèce Klebsiella, streptococcie non hémolyse, streptococcie β hémolyse, salmonellose, proteus et l'espèce pseudomonas. La sensibilité était très élevée en ce qui concerne Pefloxacine dans laquelle plus de 94% organisme étaient plein de sensibilité suivi par ceftriaxone (plus de 85%) et ceftazidime plus de 85%) les sensibilités à l'égard d'acide nalidixique et cefuroxime étaient entre 67,6% et 74,1%. La majorité des isolâtes étaient rebelle à gentamicine, amoxycilline, cotrimoxazole et ampicilline. En général. la tendance de la sensibilité dans le groupe de la drépanocytose était semblable à la tendance dans le groupe de contrôle.

Conclusion: Agents étiologique d'UTI d'enfance dans ce milieu sont rebelle à la majorité des drogues fréquemment prescrit pour soigner cette maladie. Acide nalidixique et cefuroxime sont prescrit comme des drogues de première main dans l'attente du résultat d'épreuve de la sensibilité. On doit réserver l'administration de la ceftriaxone et ceftazidime pour des cas non réponse aux drogues de première main et en cas d'urgence. Pefloxacine devrait être considéré comme une droque potentielle pour le traitement des infections multi-drogues résistante en particulier.

Introduction

Urinary tract infection (UTI) is a significant cause of morbidity in childhood[1,2] and individuals with sickle cell disease have been observed to be at increased risk[3-7]. Since delays in instituting appropriate treatment may result in formation of scars and subsequent renal impairment[8,9], knowledge of the antimicrobial sensitivity pattern of the common aetiological agents is important. This will serve as a guide to first line treatment while the results of urine culture and sensitivity are being awaited. Prompt treatment will in addition to reducing the risk of renal scars also reduce that of acute sequelae of UTI in sickle cell anemia (homozygous haemoglobin S, Hb SS) individuals such as potentially fatal septiceamia and the precipitation of crises[10,11].

This study is particularly relevant since there is no previous report to guide antibiotic therapy for UTI in Sickle cell anaemia (SCA) children in this environment. The study was therefore carried out to determine the antimicrobial sensitivity pattern of bacterial agents isolated from the urine of febrile SCA children seen at the University College Hospital as part of a larger project on childhood UTI. Ethical clearance was obtained from the Joint University of Ibadan and University College Hospital Ethical Committee. Informed verbal consent was obtained from the parents or guardians.

Methods

The study population consisted of 171 SCA (Hb SS) patients aged between 1 and 15 years presenting with fever

**Correspondence*

Table 1 Antimicrobial sensitivity pattern of urinary isolates from 37 SCA children

Isolates	% Sensitivity										
	AMP	GEN	PEF	CRO	CAZ	CXM	NIT	NAL	AUG	AML	COT
E.coli (n = 24)	8.3	58.3	95.8	87.5	91.7	70.8	62.5	75.0	62.5	25.0	0.0
Klebsiella (n = 7)	0.0	28.6	85.7	85.7	85.7	57.1	57.1	85.7	28.6	0.0	0.0
β-hemolytic Streptococcus (n=1)	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	0.0
Salmonella (n = 1)	0.0	0.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	0.0
Staphylococcus aureus (n = 1)	0.0	0.0	100.0	100.0	0.0	100.0	0.0	0.0	100.0	0.0	0.0
Pseudomonas (n = 2)	0.0	50.0	100.0	50.0	50.0	0.0	50.0	50.0	0.0	0.0	0.0
Overall Sensitivity	8.1	48.6	94.6	86.5	86.5	67.6	62.2	70.3	56.6	24.3	0.0

AMP *Ampicillin* *NIT* *Nitrofurantoin* *CAZ* *Ceftazidime*
AML *Amoxycillin* *NAL* *Nalidixic acid* *PEF* *Pefloxacin*
AUG *Amoxycillin/clavulanate* *CXM* *Cefuroxime* *GEN* *Gentamicin*
COT *Cotrimoxazole* *CRO* *Ceftriaxone*

(temperature ≥37.5°C) to the children's outpatient department or children's emegency ward of the University College Hospital Ibadan during the period March 1999 to February 2000. An equal number (171) of age and sex matched haemoglobin-A cases were enrolled as controls. Children clinically suspected to have congenital abnormalities of the genito-urinary tract and confirmed radiologically were excluded from the study. Mid-stream urine specimen was collected from each child into sterile universal bottles containing boric acid crystals. The specimens were cultured immediately by inoculation into blood agar and MacConkey agar and incubated at 37°C for 24 hours[12]. Samples showing at least 10^5 colony-forming units of bacteria per ml were considered to indicate significant bacteriuria[13]. Identification of the organisms to species level was done by standard methods[14].

Antimicrobial sensitivity tests were performed using the disc-diffusion technique of Stokes[15] using Oxoid's multodiscs (Oxoid Ltd, Basing Stoke, Hampshire, England) with the following antimicrobials: ampicillin (25mcg), nirofurantoin (200mcg), cotrimoxazole (25mcg), nalidixic acid (30mcg), gentamicin (10mcg), ceftazidime (30mcg), Cefuroxime (30mcg) and Ceftriaxone (30mcg).Other antibiotic discs used were amoxycillin (20mcg), amoxycillin - clavulanate (Augmentin® 30mcg) and pefloxacin 10mcg (Pathoteq Biological lab 1, India). Sensitivity testing was done using Sensitivity Testing Agar (BIOTECH Lab, Ipswich, Suffolk, IP57RG, United Kingdom) at a pH of 7.2 – 7.6.

Results

The mean age (± standard deviation) of the SCA children was 7.2 ± 3.7 years while that for the controls was 6.9 ± 3.4 years. There were 92 males and 79 females in each category of patients giving a male: female ratio 1.2:1. Out of the 92 boys with SCA, bacteriuria was present in 16 (17.4%) of them whereas 21 (26.6%) of the 79 girls had bacteriuria. Although bacteriuria occurred in a higher proportion of girls than boys with SCA, there was no statistically significant association of bacteriuria with sex ($p=0.146$, $X^2 = 2.12$). Similarly, among the controls

Table 2 Antimicrobial sensitivity pattern of urinary isolates from 27 children with hemoglobin AA

Isolates	% Sensitivity										
	AMP	GEN	PEF	CRO	CAZ	CXM	NIT	NAL	AUG	AML	COT
E.coli (n = 17)	0.0	23.5	94.1	88.2	94.1	76.5	35.3	76.5	23.5	5.9	0.0
Klebsiella (n = 6)	0.0	33.3	100.0	100.0	100.0	66.7	66.7	66.7	16.7	16.7	0.0
Proteus (n=1)	0.0	0.0	100.0	100.0	100.0	100.0	0.0	100.0	0.0	0.0	0.0
Pseudomonas (n = 1)	0.0	100.0	100.0	100.0	100.0	0.0	0.0	0.0	0.0	0.0	0.0
Non-hemolytic Streptococcus (n = 1)	0.0	100.0	100.0	100.0	100.0	100.0	0.0	100.0	100.0	100.0	100.0
Staphylococcus aureus (n = 1)	0.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	0.0	0.0	0.0
Overall Sensitivity	0.0	33.3	96.3	92.6	96.3	74.1	40.7	74.1	22.1	11.1	3.7

AMP *Ampicillin* *NIT* *Nitrofurantoin* *CAZ* *Ceftazidime*
AML *Amoxycillin* *NAL* *Nalidixic acid* *PEF* *Pefloxacin*
AUG *Amoxycillin/clavulanate* *CXM* *Cefuroxime* *GEN* *Gentamicin*
COT *Cotrimoxazole* *CRO* *Ceftriaxone*

bacteriuria was more common among the girls than boys being found in 14(17.7%) of the 79 girls and in 13(4.1%) of the 92 boys. There was however no significant association of bacteriuria with sex ($p=0.521$, $X^2=0.41$).

Out of the 171 children in each group there was significant bacteriuria in 37 of the SCA group and in 27 of the control group. The organisms in the SCA group were Escherichia coli (64.9%), Klebsiella species (18.9%), Non-hemolytic Streptococcus (2.7%), Pseudomonas (5.4%), β-hemolytic Streptococcus (2.7%), Salmonella (2.7%) and Staphylococcus aureus (2.7%). The isolates from the controls were Escherichia coli (63.0%), Klebsiella species (22.2%), Non-haemolytic streptococcus (3.7%), Pseudomonas (3.7%), Staphylococcus aureus (3.7%) and Proteus sp (3.7%).

The sensitivity of the isolates to the antimicrobials is shown in table 1 and 2 for the SCA children and controls respectively. Less than 10% of the isolates from the SCA children were sensitive to cotrimoxazole and ampicillin that are antimicrobials commonly prescribed for the treatment of urinary tract infection. Sensitivity to amoxycillin was only 25% among them but when augmented with clavulanate, the sensitivity was enhanced to 56.6%. Also among the sickle cell anemia children only 48 .6% of the isolates were sensitive to gentamicin which is another drug commonly recommended for the treatment of UTI. Sensitivity was higher to the quinolone: pefloxacin (94.6%) across the entire spectrum of organism from the SCA patients and it was the only antibiotic to which the isolates of pseudomonas were sensitive. Next in sensitivity were ceftriaxone and ceftazidime to each of which 86.5% of the organisms were sensitive. Sensitivity nalidixic acid, nitrofurantoin and cefuroxime were 70.3%, 62.2% and 67.6% respectively.

Similar to the SCA children, sensitivity amongst the control patients was highest (>85%) to pefloxacin, ceftriaxone and ceftazidime and lowest (<10%) to ampicillin and cotrimoxazole. In addition most isolates were resistant to amoxycillin and gentamicin. However, unlike in the SCA children where over 50% of the isolates were sensitive to amoxycillin/clavulanate and nitrofurantoin, most isolates from the controls were resistant to these antibiotics.

Discussion

There is paucity of literature on the antimicrobial sensitivity pattern of urinary isolates in individuals with sickle cell disease. Since several workers have reported increased risk of UTI in this group of individuals[3-7] it is necessary to have a guide to appropriate first line drugs to institute early treatment.

This study reports the sensitivity of urinary isolates from febrile SCA children and controls to 11 antimicrobials. The spectrum of isolates from the SCA children was similar to that from the controls: *Escherichia coli* and *Klebisiella species* accounting for over 80% of the organisms in each group. This is surprising since previous studies have shown some differences in the aetiologic agents of steomyelitis and septiceamia in individuals with SCA compared to the general population as a result of immunological deficiencies and splenic dysfunction[16]. This has not proved to be the case with UTI as shown in this study. The explanation may be that similar host and bacterial virulence factors are acting in both groups of patients predisposing them to ascending infection (rather than haematogenous infection). In addition, the immunological deficiencies and splenic dysfunction probably do not contribute appreciably to UTI in these children.

In the SCA group, most isolates were resistant to ampicillin, which is similar to findings by Elbashier and Badu[6] on sickle cell disease patients in Saudi Arabia. There was also a very high level of resistance to cotrimoxazole in the present study unlike that in the aforementioned study. However workers in Nigeia[17,18] have reported a similarly high level of resistance of UTI pathogens to cotrimoxazole. In a study involving 40 isolates of *E coli* and 35 isolates of *Klebsiella species*, Obaseiki-Ebor[17] in Benin found that 80% of the *E.coli* isolates and 74% of the *Klebsiella* isolates were resistant to cotrimoxazole. Also Adeyemo et al[18] in Ibadan observed that 100% of *E.coli* isolates and over 90% of *Klebsiella* isolates were resistant to cotrimoxazole and ampicillin. In the present study, there was also a high level of resistance to amoxycillin but sensitivity improved when it was potentiated with clavulanate in keeping with findings by Roomi et al[19]. With the level of resistance to antimicrobials observed in this study, it would seem unsafe to use cotrimoxazole, ampicillin and amoxycillin as first line drugs in the treatment of UTI in this environment.

At least 67% of the isolates from the SCA children were sensitive to cefuroxime, and nalidixic acid and nitrofurantoin were reported by Elbashier and Badu[6] in Saudi Arabia and by Adeyemo et al[18] in Ibadan. In particular there was a consistent superior sensitivity of nalidixic acid over nitrofurantoin with respect to *Klebsiella* isolates in the latter two studies as well as this study. This suggests that nalidixic acid may be a better option than nitrofurantoin in cases of UTI suspected to be caused by *Klebsiella* in instances where oral antibiotics are permissible.

In general, sensitivity pattern of the isolates in the control group was similar to that in the SCA group but for those of nitrofurantoin and amoxycillin/clavulanate, which were less sensitive in the former group. The 40.7% sensitivity to nitrofurantoin in the control group in the present study is in contrast to the high sensitivity reported earlier by Adeyemo et al in the same environment. This suggests increasing resistance and may be due to the effect of drug pressure from increased awareness and use of the drug in the treatment of UTI. The finding of resistance to cotrimoxazole and ampicillin in the isolates from the controls is in keeping with findings by other workers in Nigeria[17,18]. These are oral drugs that are widely prescribed for uninvestigated fevers even without prescription.

The finding of greatest sensitivity to pefloxacin in this study is similar to findings by Adeyemo et al [18] of 100% sensitivity to ofloxacin a quinolone in childhood UTI in Ibadan. Although the quinolones are generally not recommended in young children, they may be of use in multi-drug resistant infections[20] as was the case of the Pseudomonas isolate in this study. While the risk of joint damage has caused clinicians to exercise caution in prescribing these agents in children, close monitoring of paediatric patients receiving ciprofloxacin, a quinolone has failed to reveal cartilage toxicity[21-23]. Studies evaluating joint changes using magnetic resonance imaging, skeletal function tests, height velocity, laboratory testing and physical examinations have revealed no abnormal development[21-23]. In the case of pefloxacin whose use has been associated with arthropathy, it is reversible and subsides after the drug is discontiued[24]. In addition, the incidence of arthropathy following its use seems to be age-related being greater when the drug is first used between the ages 15 and 20 years[24]. It therefore seems relatively safe to use it in multi-drug resistant infections below the age of 15 years. Moreover, since a review of available data on the pharmacokinetics of the quinolones in children suggests no risk of nephrotoxicity, it may be considered a potentially useful drug in the treatment of

childhood UTI[25].

Conclusion

The pattern of antimicrobial sensitivity and resistance observed in this study calls for a review of the drugs routinely used as first line in the treatment of UTI in this environment. Nalidixic acid and cefuroxime are recommended as first line drugs while awaiting the results of urine culture and sensitivity. Because of cost and non-availability of oral formulations, ceftriaxone and ceftazidime should be reserved for cases of non-response to first line drugs or in severe cases. There is need to review the indications for use of quinolones particularly in infections caused by multi-drug resistant organisms; consideration should be given to pefloxacin as a potential drug for the treatment in such cases.

Acknowledgement

This study was partly funded by the Mobolaji-Bank Anthony grant for Fellowship in Nephrology. Our appreciation also goes to Glaxo Wellcome (Nigeria), Swiss pharma Nigeria and Fidson drugs that provided some antibiotic discs for sensitivity testing.

References

1. Kunin CM. Epidemiology and natural history of urinary tract infection in children. Pediatr Clin North Am 1971; 18: 509 – 527.

2. Neumann CG, Pryles CV. Pyelonephritis in infants and children: autopsy experience at Boston City Hospital, 1933 – 1960. Am J Dis child 1962; 104: 215 – 229.

3. Robinson MG, Halpern C. Infections, *Escherichia coli* and sickle cell anaemia JAMA 1974; 230:1145 – 1148.

4. Karayalcin G, Rosner F, Kim KY, Chandra P, Aballi AJ. Sickle cell anaemia clinical manifestations in 100 patients and review of the literature. Am J Med Sci 1975; 269: 51 – 68.

5. Tarry WF, Duckett JW Jr, Snyder HM. Urological complications of sickle cell disease in a paediatric population J Urol 1987; 138: 592 – 594.

6. Elbashier AM, Badu GA. Pattern of bacteriuria in patients with sickle cell disease in Qatif Central Hospital Saud Med J 1991; 12: 121 – 124.

7. Ajasin MA, Adegbola RA. symptomatic bacteriuria in children with sickle cell anaemia. Nig. J Paediatr 1997; 24: 40 –44.

8 Newcastle Covert Bacteriuria Research Group. Covert bacteriuria in schoolgirls in Newcastle upon Tyne: a 5-year follow-up. Arch Dis Child 1981; 56: 585 – 592.

9. Jacobson SH, Eklof O, Eriksson CG, Lins L, Tidgren B, Winberg J. Development of hypertension and uraemia after pyelonephritis in childhood: 27-year follow-up. Br Med J 1989; 299: 703 – 706.

10. Zarkosky HS, Gallagher D, Gill FM, Wang WC, Falletta JM, Lande WM et al. Bacteriuria in sickle hemoglobinopathies. J Pediatr 1986; 109: 579 – 585.

11. Konotey-Ahulu FID. The sickle cell disease; clinical manifestations including the 'sickle crises'. Arch Intern Med 1974; 133: 611 – 619.

12. Duguid JP, Collee JG, Fraser AG. Laboratory strategy in the diagnosis of infective syndromes. In: Collee JG, Duguid JP, Fraser AG, Marmion BP, eds. Mackie and McCartney Practical Medical Microbiology. New York: Churchill Livingstone, 1989: 600 – 649.

13. Kass EH. Bacteriuria and the diagnosis of the urinary tract. Arch Intern Med 1957; 100: 709 – 714.

14. Cheesbrough M. Medical Laboratory Manual for Tropical countries. Volume II: Microbiology. Cambridge: Cambridge University Press, 1984: 225 – 273.

15. Cheesbrough M. Medical Laboratory Manual for Tropical Countries. Volume II Microbiology. Cambridge University Press 1984: 146 – 205.

16. Serjeant GR. The immune system. In: Serjeant GR. Sickle cell disease. Oxford: Oxford University Press, 1988: 124 – 134.

17. Obaseiki-Ebor EE. Trimethoprim/Sulphamethoxazole resistance in *Escherichia coli* and Klebsiella spp urinary isolates. Afr J Med Sci 1988; 17: 133 – 140.

18. Adeyemo AA, Gbadegesin RA, Oyemenem TN, Ekweozor CC. Urinary tract pathogens and antimicrobial sensitivity patterns in children in Ibadan, Nigeria Ann Trop Paediatr 1994; 4: 271 – 274.

19. Roomi LGA, Sutton AM, Cockburn F, McAllister. Amoxycillin and Clavulanic acid in the treatment of urinary infection. Arch Dis Child 1984; 59: 256 – 259.

20. Khan DM, Bhutta ZA. Ciprofloxacin in multi-resistant infections in childhood: an audit. J Pak Med Assoc 1995: 45: 147 – 150.

21. Schaad UB, Stoupis C, Wedgewood J, Tschaeppeler H, Vock P. Clinical, radiologic and magnetic resonance monitoring for skeletal toxicity in pediatric patients with cystic fibrosis receiving a three-month course of ciprofloxacin. Pediatr Infec Dis J 1991; 10: 723 – 729.

22. Danisovicova A, Kremeryova T, Belan S, et al. Magnetic resonance imaging in diagnosis of potential arthropathogencity in children receiving quinilones: No evidence for quinolone-induced arthropathy. Drugs 1995; 49 (Suppl 2): 492 – 4.

23. Bethell DB, Hien TT, Phil LT, et al. Effects on growth of single short courses of fluoroquinolones. Arch Dis Child 1996; 74: 44 – 6.

24. Pertuiset E, Lenoir G, Jehanne M, Douchain F, Guillot M, Menkes CJ. Joint tolerance of pefloxacin and ofloxacin in children and adolescents with cystic fibrosis. Rev Rhum Mal Osteoartic 1989; 56: 735 – 40.

25. Fanos V, Cuzzolin L. Fluoroquinolones in pediatrics and their nephrotoxicity in adults: minireview. J Chem 2000; 12: 228 – 31.