Uropathogens and diagnostic potential of pH and specific gravity in a rural community of Nigeria

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ABSTRACT

Objective: To assess the association between urine pH and specific gravity (SG) and occurrence of urinary tract pathogens involved in urinary tract infection (UTI) in school pupils.
Methods: Laboratory culture techniques and biochemical tests were carried out to identify the UTI-associated bacteria in school pupils' urine of United Nursery and Primary School, Ilara-Remo, in Ikenne Local Government area, Ogun State, Nigeria. Urine dip-stick tests were carried out on the samples to take pH and SG measurements.
Results: The prevalence of urinary tract pathogens in the study population was 82.4%. This prevalence was not gender and age dependent ($P > 0.05$). *Escherichia coli* (*E. coli*) (37.0%) and *Klebsiella* (0.9%) were the highest and least prevalent uropathogens. There was significant increase in proportion of subjects diagnosed with *E. coli* (37.5%), *Staphylococcus aureus* (32.0%), *Klebsiella* (57.1%) in pH 9, pH 6 and pH 8, respectively ($P < 0.05$). However, an increase in proportion of individuals diagnosed with *Proteus* occurred in pH 6 and 9 (31.3% each); these proportions were not significant compared with results in other pH categories ($P > 0.05$). There were significant increase in proportion of subjects diagnosed with *E. coli* (50.0%), *Proteus* (56.3%), *Staphylococcus aureus* (44.0%), *Klebsiella* (71.4%) in SG 1.010, 1.015, 1.015 and 1.010, respectively ($P < 0.05$).
Conclusion: Therefore, the urine pH 9 and SG 1.010–1.015 are the best diagnostic indicators of UTI-associated uropathogens in school children.

1. Introduction

A urinary tract infection (UTI) is often caused by bacteria and affects any part of the urinary tract. It is one of the most common infections in children. A previous report has shown that childhood UTI will occur in at least 8% of girls and 2% of boys, and between 30% and 40% children will have another episode within two years[1]. The higher occurrence of UTI in females is believed to be associated with the closeness of the opening of urethral to the anus and the shorter length of urethra opening into the bladder[2].

UTIs are highly challenging, not only because of the yearly endemicity but also because the diagnosis of UTI is not always straightforward[3]. Presence of associated symptoms and laboratory investigations are necessary to confirm the diagnosis of UTIs[4]. The urine nitrites test has been widely reported as a rapid screening test for significant bacteriuria[5,6]. Another useful analyte for UTI diagnosis is leukocyte esterase which is an enzyme found in neutrophil granules that reacts with agents on the dipstick to produce a blue colour in 1–2 min[7]. However, the low sensitivity of nitrites and leukocyte esterase has greatly limited their diagnostic values[8,9].

Urease-producing bacteria *e.g. Proteus mirabilis* has been shown to be associated with pH > 7–7.5, while a decrease in specific gravity (SG) (measured by increasing hydration status) has been reported to be associated with decrease in bacteriuria in women[10,11]. Although pH and SG are among the analytes of urine dipstick often used in urinalysis, they are not usually employed for rapid UTI diagnosis. So this study seeks to assess the age and gender associated uropathogens diagnostic potential of urine dipstick pH and SG in school children.

2. Materials and methods

2.1. Subjects and ethical consideration

The study was conducted among pupils of United Nursery and Primary School, Ilara-Remo, in Ikenne Local Government Area, Ogun State, Nigeria. Approvals to conduct the study were obtained from the Local Government Education Authority, Ikenne and the State Ministry of Health, Abeokuta Ogun State. All pupils whose parents gave oral consent and who had stayed for a period of at least one year in the locality were recruited to participate in the study. Those whose parents did not give consent were excluded from the study.

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2.2. Sample collection and urine pH/SG determination

Clean catch mid stream urine samples were collected from 108 pupils in sterile labelled universal containers. The pupils were properly instructed and carefully monitored during sample collection to avoid contamination. Each label contained the pupil's bio-data (name, age and gender) and was recorded against the tagged registration number. A total of 89 subjects also provided urine samples for pH and SG screening by using chemical reagent strips (San Diego, CA 92121, USA). The screening was done according to the manufacturer's instructions.

2.3. Uropathogen detection in urine

The samples were preserved on ice and were transported to the Microbiology Laboratory, Babcock University, Ilishan-Remo for immediate bacterial culture. Analysis was conducted within 1 h of sample collection. Samples were properly agitated before inoculation to ensure even distribution of the organisms. The samples were cultured on blood and cystine lactose electrolyte deficient agar media and were incubated at 37 ℃ for 18–24 h. The characteristic bacterial isolates observed on the selective media were aseptically isolated and subjected to microscopical and appropriate biochemical tests for proper identification[12].

2.4. Statistical analysis

Data were entered in Excel and cross checked for error. Analysis was carried out by SPSS for Windows (version 17.0, Inc, Chicago USA). *Chi*-square analysis was used to determine association between categorical data like age and sex and urinary indicators of uropathogens (*i.e.* pH and SG). *P*-values less than 0.05 were considered significant.

3. Results

The overall prevalence of urinary tract pathogens in the study population was 82.4%. The prevalence of uropathogens was higher in female subjects (83.6%) than in the male subjects (80.9%), however, prevalence was not gender dependent (*P* > 0.05) (Table 1).

The occurrence of uropathogens was highest in children's age ⩾ 11 years (86.0%) compared with those in age 5–7 and 8–10 years with 57.1% and 82.8%, respectively (Table 1).

Table 1
Gender and age-related uropathogens occurrence pattern.

Categories		Total examined	No. infected	Prevalence (%)	*P*-value
Gender	Male	47	38	80.9	0.801
	Female	61	51	83.6	
Age (years)	5–7	7	4	57.1	0.176
	8–10	58	48	82.8	
	⩾ 11	43	37	86.0	
Total		108	89	82.4	

Five uropathogens including *Escherichia coli* (*E. coli*), *Proteus* sp, *Staphylococcus aureus* (*S. aureus*), *Klebsiella* spp and *Pseudomonas* were encountered with 37.0%, 14.8%, 23.1%, 6.5% and 0.9% prevalence level, respectively (*P* < 0.000 1). There was significant increase in proportion of subjects diagnosed with *E. coli* (37.5%), *S. aureus* (32.0%), *Klebsiella* (57.1) in pH 9, pH 6 and pH 8, respectively (*P* < 0.05) (Table 2). However, an increase in proportion of individuals diagnosed with *Proteus* occurred in pH 6 and 9 (both 31.3%), these proportions were not significant compared with results in other pH categories (*P* > 0.05). Overall, pH 5 and 9 showed the least (3.4%) and the highest (32.6%) uropathogens diagnostic potential (*P* < 0.000 1) (Table 2).

There were significant increase in proportion of subjects diagnosed with *E. coli* (50.0%), *Proteus* spp (56.3%), *S. aureus* (44.0%), *Klebsiella* spp (71.4%) in SG 1.010, 1.015, 1.015 and 1.010, respectively (*P* < 0.05) (Table 3). Overall, SG 1.030 and 1.010 showed the least (3.4%) and the highest (43.8%) uropathogens diagnostic potential (*P* < 0.0001) (Table 3).

4. Discussion

The prevalence of urinary tract pathogens in present study was higher than other previous reports on UTI in Nigeria and other parts of the world ranging from 48% to 72%[2,13,14]. Besides the low socio-economic status of the people in our study area, the inclusion of all uropathogens positive subjects without necessarily considering their UTI statuses could have been responsible for the higher

Table 2
Uropathogen prevalence by pH stratified. *n* (%).

Bacteria species	Total examined	Uropathogens profile in different pH values					*P*-value
		pH 5	pH 6	pH 7	pH 8	pH 9	
E. coli	40	1 (2.5)	11 (27.5)	3 (7.5)	10 (25.0)	15 (37.5)	0.000 3
Proteus	16	1 (6.3)	5 (31.3)	4 (25.0)	1 (6.3)	5 (31.3)	0.161 0
S. aureus	25	0 (0.0)	8 (32.0)	4 (16.0)	6 (24.0)	7 (28.0)	0.040 0
Klebsiella	7	1 (14.3)	1 (14.3)	0 (0.0)	4 (57.1)	1 (14.3)	0.084 0
Pseudomonas	1	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (100.0)	0.287 0
Overall	89	3 (3.4)	25 (28.1)	11 (12.4)	21 (23.6)	29 (32.6)	< 0.000 1

Table 3
Uropathogen prevalence by SG stratified. *n* (%).

Bacteria species	Total examined	Uropathogens profile in different SG						*P*-value
		SG 1.005	SG 1.010	SG 1.015	SG 1.020	SG 1.025	SG 1.030	
E. coli	40	6 (15.0)	20 (50.0)	6 (15.0)	5 (12.5)	2 (5.0)	1 (2.5)	< 0.000 1
Proteus	16	1 (6.3)	4 (25.0)	9 (56.3)	1 (6.3)	1 (6.3)	0 (0.0)	< 0.000 1
S. aureus	25	1 (4.0)	9 (36.0)	11 (44.0)	0 (0.0)	2 (8.0)	2 (8.0)	< 0.000 1
Klebsiella	7	0 (0.0)	5 (71.4)	1 (14.3)	1 (14.3)	0 (0.0)	0 (0.0)	0.002 0
Pseudomonas	1	0 (0.0)	1 (100.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0.306 0
Overall	89	8 (9.0)	39 (43.8)	27 (30.3)	7 (7.9)	5 (5.6)	3 (3.4)	< 0.000 1

occurrence. The lack of association between bacterial occurrence and gender showed that all groups are equally predisposed to the risk factors of UTI. Notable risk factors include foreskin, fecal and perineal colonization, urinary tract abnormalies, functional abnormalities and immunocompromised states[15].

The higher prevalence in female subjects which was similar to other observations in Nigeria and other parts of the world[16,17], was however probably due to the anatomical design of female genitalia evidenced in the close proximity of the urethral meatus to the anus and shorter urethra[12]. Our result that showed *E. coli* as the most frequently encountered pathogen was in consonance with other reports[18,19]. *E. coli*, a member of Gram negative bacteria is known to colonize the urogenital mucosa with adhesins, pili, fimbriae, and P-1 blood group phenotype receptor[20].

Urinary leukocytes esterase and nitrite have been the usual indicators of UTI, while little is known about pH and SG. Unlike a previous study that showed dominance of pH 5 and pH 6 in UTI asymptomatic and symptomatic subjects[21], the most frequently observed urinary pH in our study was 9. The low and high prevalence of uropathogens in urine pH 5 and pH 9 indicates a bacteriostatic and pathogens supporting media, respectively. The high occurrence of urease producing bacteria (*i.e. Proteus* sp, *S. aureus*, and *Klebsiella* spp) in pH 8 and pH 9 suggests the bacteria ability to split urinary urea into CO_2 and ammonia and this has been implicated in hyperammonemic encephalopathy[22], however, increase of these bacteria in pH 6 requires further studies. The bacterium-specific diagnostic potential of some pH values *e.g.* pH 9/*E. coli*, pH 6, 9/*Proteus*, pH 6/*S. aureus*, and pH 8/*Klebsiella* requires further studies to ascertain the actual relationship between certain bacterium and hydrogen ion gradient in patient urine.

The SG is a measure of urine concentration. As earlier discussed, it is not usually employed by clinicians as a diagnostic indicator of UTI in human. This study suggested that the SG of the range 1.010–1.015 was best for diagnosing uropathogens; an observation that was similar to an elsewhere study on cat[23]. Concentrated urine (SG $\geq$ 1.020) seems to cause inhibition in bacterial growth. Similar observation had been earlier reported[24].

This study showed high occurrence of bacteriuria in the school children and also showed that pH 9 and SG 1.010–1.015 were good diagnostic indicators of uropathogens. This approach will compliment the routine microscopic and urine culture method often adopted in tropical countries laboratories. It will further improve the sensitivity of other commonly utilized urinalysis diagnostic approach.

Conflict of interest statement

We declare that we have no conflict of interest.

Acknowledgments

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