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Full Length Research Paper

Comparison of susceptibility patterns of *Escherichia coli* isolated from urinary tract infections in two health institutions in South-South Nigeria to commonly used antimicrobials

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Urinary tract infection is one of the most common bacterial infections with increasing resistance to antibiotics. *Escherichia coli (E. coli)* have been identified as the most common pathogen of urinary tract infections. Studies on prevalence and antimicrobial susceptibility patterns of clinical isolates from urinary tract infections in Nigeria are few. This study was carried out to assess the prevalence and antimicrobial susceptibility patterns of *E. coli* isolates from urinary tract infections in two health institutions - a general hospital and a private clinic in South South, Nigeria. A retrospective study was carried out between January 2005 and December 2009. Statistical analysis was carried out using SPSS version 14, Chicago IL. *E. coli* accounted for 1797 out of 3655 urine isolates (49.16%). Lowest susceptibility was for cotrimoxazole (13.9%), chloramphenicol (16.3%) and amoxicillin (16.8%) while highest susceptibility was for ofloxacin (65.1%) and ciprofloxacin (50%). There was a significant difference, p<0.05, in prevalence and susceptibility patterns between the two institutions with higher susceptibility levels in the general hospital. There is a need for proper surveillance and development of hospital specific antibiograms to inform appropriate empiric therapy of urinary tract infections.

Key words: *Escherichia coli,* urinary tract infections, antimicrobial susceptibility, Nigeria.

INTRODUCTION

Community acquired infections continue to be the leading cause of death in developing countries (Wenzel and Edmond, 2000). Urinary tract infection is one of the commonest diseases diagnosed in outpatients (Gales et al., 2000) and therefore accounts for a large proportion of antibacterial drug consumption (Grude et al., 2001). Empiricial treatment is usually initiated before laboratory diagnosis which results in increasing resistance of uropathogens to antimicrobials due to frequent misuse of antibiotics (Tambekar and Dhanorkar, 2005). *Escherichia coli* has been indicated as the major causative organism for uncomplicated community acquired urinary tract infections (Raka et al., 2004; Stratchounski and Rafalski, 2006; Matsumoto, 2011).

Bacterial resistance has been increasing over the past years. Knowledge about the trends in emergence of resistance and their importance in different settings is essential prerequisite to the control of antibacterial

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Table 1. Distribution of E. coli isolates by gender.

Source	Gender	
Hospital	Male	Female
General hospital	515 (31.4%)	1126 (68.6%)
Private clinic	50 (32.1%)	106 (67.9%)
Total	565 (100%)	1232 (100%)

resistance (Naaber et al., 2000). In many instances, the data available are not amenable to accurate quantitative assessment, particularly in countries like Nigeria whose systematic surveillance system is absent or rudimentary (Okeke et al., 2005). There is therefore, the need to conduct surveillance studies that details the trends in antibiotic use and the resistance patterns of various bacterial isolates with the objective of making evidence based recommendations to reduce the emergence and spread of antibiotic resistant pathogens. This knowledge will help in implementing measures for the control of the spread of antimicrobial resistant organisms.

In Nigeria, bacterial resistance to antibiotics is rampant as a result of lax or no policy that guides the usage and administration of antibiotics. In Nigeria, there is inadequate data on trends in antibiotic use and resistance patterns. Few studies have been carried out in the past but have included only a small number of clinical isolates. This study seeks to analyse the difference in antibiotic susceptibility of *E. coli* isolates from urine samples in humans over a time frame of five years from two health institutions in South South, Nigeria. The observations from this study should be useful in other resource poor settings.

MATERIALS AND METHODS

Study setting

The clinical microbiology laboratories of two health institutions in Rivers State, South-South, Nigeria were involved in the study. The first institution is a government owned general hospital. The second is a private clinic. All urinary specimens, collected from patients with clinical diagnosis of urinary tract infection, from freshly voided clean catch midstream urine with significant bacteruria ≥ 10^5 cfu/ml processed in the two laboratories between January 2005 and December 2009 from outpatients were included in the study. Information was retrieved from the laboratory records. Demographic data collected were age and gender. Age was classified as child (less than18 years) and adults (18 years and above). Only one positive urine specimen per patient was included in the study. These were inoculated unto CLED and MacConckey agar and incubated at 37°C for 24 h. Identification of *E. coli* was based on standard biochemical tests (McNulty et al., 2004).

Susceptibility tests

In vitro susceptibility of *E. coli* isolates to nine commonly used antibiotics was performed using antibiotic discs on Mueller Hinton agar, modified by Kirby-Bauer (Baron et al., 1994). Antibiotic discs used were, amoxicillin (30 mcg), chloramphenicol (20 mcg), gentamycin (10 mcg), cotrimoxazole (25 mcg), ciprofloxacin (10 mcg), ofloxacin (10 mcg), amoxicillin-clavulanic acid (30 mcg), streptomycin (30 mcg) and nalidixic acid (30 mcg). Inhibition zone diameter was interpreted based on CLSI (2011) guidelines. Intermediate and resistant strains were grouped together. For quality control of susceptibility tests, *E. coli* ATCC 27922 was used.

Statistical analysis

Statistical analysis was carried out using Statistical Pacvkage for the Social Sciences (SPSS) version 14, Chicago IL. Susceptibility was expressed as percentages. Chi square test with Fisher's exact two-tailed test was used to assess the association between institution, gender and age with the prevalence of resistant isolates. A p-value of <0.05 was considered as statistically significant.

RESULTS

E. coli was isolated from 1797 out of 3655 of urine isolates accounting for 49.16% of urinary tract infections. Out of the 1797 *E. coli* isolates, 1641 were from the general hospital and 156 were from the private clinic. This was isolated from 1232 females (68.56%) and 565 men (31.44%) as shown in Table 1. There was no significant difference in the distribution of *E. coli* between both genders in the two health institutions. There was a significant difference, $p < 0.05$, in occurrence of *E. coli* in the two institutions. *E. coli* accounted for 52.24% of isolates from the general hospital and 37.7% of urine isolates in the private clinic.

Table 2 shows results of the resistance rates of *E. coli* to antimicrobials. Antimicrobials with the lowest susceptibility levels include cotrimoxazole (13.9%), chloramphenicol (16.3%) and amoxicillin (16.8%). Highest susceptibility levels were for the fluoroquinolones, 65.1, 50 and 42.9% for ofloxacin, ciprofloxacin and nalidixic acid, respectively. A significant difference was observed in the levels of susceptibility in the two clinics for amoxicillin (4.16 and 1.3%), chloramphenicol (28.7 and 12%), nalidixic acid (46.16 and 24.4%), ofloxacin (68.47 and 47.4%) and amoxicillin/clavulanic acid (30 and 21.8%) at $p< 0.05$ with lower susceptibility levels observed in the private clinic. However, for the aminoglycosides, gentamycin (23.53 and 48%) and streptomycin (29.71 and 46.2%), there was a significant difference ($p< 0.05$) in susceptibility levels between the two clinics with lower susceptibility levels in the general hospital. The susceptibility levels were higher in the males than in the females except for chloramphenicol and gentamycin. Susceptibility levels among males for amoxicillin, chloramphenicol, gentamycin, nalidixic acid, ciprofloxacin, ofloxacin, streptomycin, cotrimoxazole and amoxicillin clavulanic acid were 17.4, 13.4, 27.1, 43.9, 54.4, 67.4, 37.6, 17.4 and 31.5%, respectively. For the same antibiotics among females, susceptibility levels were 14.0, 13.8, 28.5, 41.0, 51.9, 62.6, 34.0, 15.2 and 30.2%, respectively. However the difference was not significant. There was no significant difference also

Table 2. Susceptibility rates of *E. coli* to antimicrobials.

Antimicrobial agent	General Hospital		Private Clinic		Total	
	Percentage (%)	Number sensitive	Percentage (%)	Number sensitive	Percentage (%)	Number sensitive
Amoxicillin	24.16	397/1641	1.30	2/156	16.80	302/1797
Chloramphenicol	28.70	471/1641	12.00	19/156	16.30	293/1797
Gentamycin	23.53	386/1641	48.00	75/156	26.70	480/1797
Nalidixic Acid	46.16	757/1641	24.40	38/156	42.90	771/1797
Ofloxacin	68.49	1124/1641	47.44	74/156	65.10	1170/1797
Ciprofloxacin	59.54	977/1641	47.44	74/156	50.00	899/1797
Streptomycin	29.71	488/1641	46.20	72/156	34.22	615/1797
Cotrimoxazole	12.07	198/1641	17.95	28/156	13.90	250/1797
Amoxicillin/clavulanic acid	30.00	492/1641	21.80	34/156	28.50	512/1797

in the levels of susceptibility between the two age groups.

DISCUSSION

E. coli has been identified as the most common uropathogen. This is in line with other studies. In a study of the prevalence and antimicrobial susceptibility patterns of uropathogens in Ireland, *E. coli* had the highest prevalence of 56.7% (Cullen et al., 2013). However, in another study carried out by Okesola and Aroundegbe (2011) in South West Nigeria, the most prevalent uropathogen was *Klebsiella spp* with a prevalence of 40 %, while *E. coli* had the second highest prevalence at 25 %. This shows that the aetiology of urinary tract infections could vary geographically. With the high prevalence of *E. coli* as the causative agent of urinary tract infections, the study of its susceptibility pattern therefore is very important in the development of empiric treatment guidelines for urinary tract infections. Results from this study portray *E. coli* as having low susceptibility levels to cotrimoxazole (13.9%), chloramphenicol (16.3%) and amoxicillin (16.8%). These results are comparable with results reported in another study carried out in South South Nigeria by Wariso et al. 2010), in which susceptibility to cotrimoxazole by all uropathogens was 7.1%. Similar studies carried out in South-West Nigeria (Odusanya, 2002; Dada-Adegbola and Muili, 2010) reported cotrimoxazole had lower sensitivity 2.0 and 5.1% than the results here reported. In Ghana, however, cotrimoxazole and chloramphenicol susceptibilities, 27 and 25%, respectively (Nambodiri et al., 2011) though low, were higher than reports from studies in Nigeria. In an 11 year study carried out in Dublin, susceptibility levels were lowest for ampicillin and trimethoprim at 39.2 and 68.5%, respectively (Cullen et al., 2013). The high level of resistance to these medications could probably be because they have been in the market for a long time, thus allowing micro organisms time to develop resistance mechanisms towards the antibiotics above mentioned. Highest susceptibility levels were seen in the fluoroquinolones, ofloxacin (65.1%) and ciprofloxacin (50%).

In the study carried out by Wariso et al (2010) in South South Nigeria, susceptibility levels to ciprofloxacin and ofloxacin were 75.8 and 76.1%, respectively, while the level of susceptibility of ciprofloxacin in the study in Ghana by Namboodiri et al. (2011) was 89%. The levels of fluoroquinolone susceptibility were lower in this study. Significance test shows however, that there was a lower level of susceptibility for the fluoroquinolones, ciprofloxacin and ofloxacin, in isolates from the private clinic when compared to isolates from the general hospital ($p<0.05$). The susceptibility levels for isolates from the general hospitals for ofloxacin, ciprofloxacin, nalidixic acid at 68.47, 59.52 and 46.16% were a bit more comparable with other results from Nigeria and Ghana. Fluoroquinolones have been in the market for a shorter duration which might account for a higher level of susceptibility when compared to older antibiotics such as cotrimoxazole, amoxicillin and others.

Also, the higher costs of these antibiotics contribute to less usage. The higher level of resistance in the private clinics might be as a result of greater use in that setting as patients that use these private clinics are generally of a higher socioeconomic status and can afford the more expensive medications, whereas cheaper medicines are used in general hospitals. However, more research has to be done on antimicrobial use in these two institutions to ascertain if there is an association between the level of utilisation and levels of resistance. Susceptibility levels in this study was far lower than results in developed countries including Europe, USA, Canada, Japan (Grude et al., 2001; Stratchounski and Rafalski, 2006; Matsumoto, 2011; Alos et al., 2005; Sahm et al., 2001; Zhanel et al., 2000; Karlowsky et al., 2002).

Cullen et al. (2013) reported susceptibility levels of 89.4% for ciprofloxacin in Dublin. A recent study carried out in America showed an increase in resistance to ciprofloxacin from 3 to 17.1 % between 2000 and 2010, which was attributed to the higher provider use of fluoroquinolones (Sanchez et al., 2012). This high level of resistance in Nigeria could be explained by the easy access of antibiotics across the counter in developing countries and the running of pharmacies by unlicensed personnel. The need for the development and enforcement of antibiotic policies and proper antibiotic stewardship in developing countries cannot be overemphasized. Varying levels of resistance in the two institutions under study, shows that there is a need to develop hospital specific antibiograms to improve on the outcome of empiric therapy.

It is important to note that the method of susceptibility testing used in this study was the disk diffusion method. Bond et al. (2012) reported that disc diffusion assays could show resistance levels for antimicrobial agents whereas the minimum inhibitory concentration (MIC) tests show sensitivity to the same antimicrobial agents. MIC tests were not carried out to confirm the sensitivity of *E. coli* to these antimicrobials in this study. This could be a limitation of this study. It will be recommended that in subsequent research carried out in Nigeria, disc diffusion tests should be compared with MIC tests such as Etests and Vitek tests to confirm results.

Guidelines for the treatment of uncomplicated acute urinary tract infections recommend the use of cotrimoxazole in the treatment in urinary tract infections when resistance levels are less than 70% (Warren et al., 1999). Fosfmycin has been recommended in the management of urinary tract infections in areas of high resistance to fluoroquinolones (Warren et al., 1999). It could be recommended that fosfomycin be included in the essential drug lists for health institutions, as it is not currently in use in Nigeria, since there is a widespread resistance to the first line treatment of urinary tract infections (Okesola and Aroundegbe, 2011, Adedeji and Abdulkadir, 2009). Despite many years of use, fosfomycin continues to have low incidence of resistance (Alos et al., 2005, Schito et al., 2009). Fosfomycin has retained its activity against quinolone resistant strains of *E. coli* and cross resistance with other classes of antibiotics is not a problem (Ungheri et al., 2002).

In conclusion, results of this study show that there is need to evaluate and develop empiric treatment strategy for urinary tract infections. Surveillance of antimicrobial susceptibility patterns should be ongoing; hospital specific antibiograms should be developed and most importantly, health professionals should be trained on antibiotic stewardship. There might be a possibility of bias resulting from over reporting of resistance because treatment is generally empirical in the region and people that eventually get to the laboratories for culture and sensitivity testing are those in whom previous antibiotic treatment has failed.

REFERENCES

Adedeji BAM, Abdulkadir OA (2009). Etiology and antimicrobial resistance pattern of bacterial agents of urinary tract infections in students of tertiary institutions in Yola Metropolis. Acad. J. Plant Sci. 2(1): 25-28.

Alos JI, Serrano MJ, Gomez-Garces JL, Perianes J (2005). Antibiotic resistance of *Escherichia coli* from community acquired urinary tract infections in relation to demographic and clinical data, Clin Microbiol and Infect 11(3): 199 - 203.

Baron E, Peterson LR, Finegold SM (1994). Methods of testing antimicrobial effectiveness (In: Bailey and Scott's Diagnostic Microbiology). 9th edn.; Musby Publishers, St Louis, pp 168-188.

Bond A, Plumb H, Turner PJ (2012). Susceptibility testing for *Escherichia coli* isolates from urines: are we at risk of reporting false antibiotic resistance to co-amoxiclav? J. Antimicrob. Chemother. 67(6): 1557 - 1558.

Clinical and Laboratory Standards Institute (CLS) (2011). Performance standards for antimicrobial susceptibility tests, 21st informational supplement. 31(1).

Cullen IM, Manecksha RP, McCullagh E, Ahmad S, O'Kelly F, Flynn R, McDermott TE, Murphy P, Grainger R, Fennell JP, Thornhill JA (2013). An 11 year analysis of the prevalent uropathogens and the changing pattern of Escherichia coli antibiotic resistance in 38,530 community urinary tract infections, Dublin 1999–2009. Ir. J. Med. Sci. 182:81–89.

Dada-Adegbola H, Muili KA (2010). Antibiotic susceptibility pattern of urinary tract pathogens in Ibadan, Nigeria. Afr. J. Med. Sci. 39(3): 173 - 179.

Gales AC, Jones RN, Gordon KA, Sader HS, Wike WW, Beach ML, Pfaller MA, Doem GV (2000). Activity and spectrum of 22 antimicrobial agents tested against urinary tract infection pathogens in hospitalized patients in Latin America: report from the second year of SENTRY Antimicrobial Surveillance Program(1998). J. Antimicrob. Chemother. 45:295-303.

Grude N, Tveten K, Kristiansen B (2001). Urinary tract infections in Norway: bacterial etiology and susceptibility. A retrospective study of clinical isolates. Clin. MicrobiolInfect. 7: 543 - 547.

Karlowsky JA, Kelly LJ, Thornsberry C, Jones ME, Sahm DF (2002). Trends in antimicrobial resistance among urinary tract infection isolates of Escherichia coli from female outpatients in the United States. Antimicrob. Agents Chemother. 46: 2540-2545.

Matsumoto TH, Hasmasuma R, Ishikawa K, Takahashi S, Yasuda M, Havami H, Tanaka K, Kiyota H, Muratani T, Monden K, Arakawa S, Yamamoto S (2011). Nationwide survey of antibacterial activity against clinical isolates from urinary tact infections in Japan (2008). Int. J. Antimicrob. Agents, doi: 10.1016/j.ijantimicag.2010.10.032

McNulty CA, Bowen J, Clark G, Charlett A, Cartwright K, South West G, (2004). Microbiology Laboratory Use Group: How should general practitioners investigate suspected urinary tract infection? Variations in laboratory-confirmed bacteriuria in South West England. Commun. Dis. Pub. Health, 7 (3)**:**220-6.

Naaber P, Koljalg L, Maimets M (2000). Antibiotic usage and resistance - trends in Estonian University Hospitals. Int. J. Antimicrob. Agents 16: 309 - 315.

Namboodiri SS, Opintan JA, Lijek RS, Newman MJ, Okeke IN (2011). Quinolone resistance in Escherichia coli from Accra, Ghana. BMC Microbiol. 11: 44.

Odusanya MD (2002). Antibiotic susceptibility of microorganism at a general hospital in Lagos, Nigeria. J. Natl. Med. Assoc. 94: 994-998.

Okeke IN, Klugman KP, Bhutta ZA, Duse AG, Jenkins P, O'Brien TF, Pablos-Mendez A, Klugman KA (2005). Antimicrobial resistance in developing countries. Part II: strategies for containment. Lancet. Infect. Dis. 5:568–580.

Okesola AO, Aroundegbe TI (2011). Antibiotic resistance pattern of uropathogenic *Escherichia coli* in South-West Nigeria. Afr. J. Med. Med. Sci. 40(3): 235 - 238.

Raka L, Mulliqi-Osmani G, Berisha L, Begolli L, Omeragiq S, Parsons L, Saffinger M, Jake A, Kurh A, Jakupi S (2004). Etiology and susceptibility of urinary tract isolates in Kosova. Int. J. Antimicrob. Agents 23S1: S2-S5.

Sahm D, Thornsbery C, Mayfield D, Jones M, Karlowsky J (2001). Multidrug- resistant urinary tract isolates of *Escherichia coli*: prevalence and patient demographics in the United States in 2000. Antimicrob. Agents Chemother. 45: 1402 - 1406.

Sanchez GV, Master RN, Karlowsky JA, Bordon JM (2012). In vitro antimicrobial resistance of urinary *Escherichia coli* isolates among US outpaitients from 2000 to 2010, Antimicrob. Agents Chemother. 56(4): 2181 – 2183.

Schito GC, Naber KG, Botto H, Palou J, Mazzei T, Gualco L, Marchese

A (2009). The ARESC study: an international survey on the antimicrobial resistance of pathogens involved in uncomplicated urinary tract infections. Int. J. Antimicrob. Agents 34: 407 - 413.

Stratchounski L, Rafalski V (2006). Antimicrobial susceptibility of pathogens isolated from adult patients with uncomplicated community-acquired urinary tract infections in the Russian Federation: two multicentre studies, UTIAP -1 and UTIAP-2. Int. J. Antimicrob. Agents 28: S4 - S9.

Tambekar DH, Dhanorkar DV (2005). The prevalence antibiogram of potential bacterial pathogens in clinical specimens, 46th annual conference of Association of Microbiologists of India, Dept of Microbiology, Osmania University, Hyperdabad.

Ungheri D, Albini E, Bellico G (2002). In vitro susceptibility of quinolone resistant clinical isolates of *Escherichia coli* to fosfomycin trometamol. J. Antimicrob. Chemother. 14: 237 - 40.

Wariso KT, Siminialayi IM, Odigie JO (2010). Pattern and antibiogram of urinary tract infection at the University of Port Harcourt Teaching Hospital, Asian Pac. J. Trop. Med. 1-5.

Warren JW, Abrutyn E, Hebel JR, Johnson JR, Schaeffer AJ, Stamm AE (1999). Guidelines for antimicrobial treatment of uncomplicated acute bacterial cystitis and acute pyelonephritis in women. Clin. Infect. Dis. 29: 745 - 758.

Wenzel RB, Edmond MB (2000). Managing antibiotic resistance. New Engl. J. Med. 343:1961-63.

Zhanel G, Karlowsky J, Harding G, Carrie A, Mozzuli T, Low D (2000). The Canadian Urinary Isolate Study Group. A Canadian national surveillance study of urinary tract isolates from outpatients: comparison of the activities of trimethoprim-sulfamethoxazole, ampicillin, mecillianam, nitrofurantoin and ciprofloxacin. Antimicrob. Agents Chemother. 44: 1089 - 1092.