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PREVALENCE AND ANTIBIOTIC RESISTANCE PATTERNS OF *ESCHERICHIA COLI* AMONG HOSPITALISED PATIENTS AT THIKA DISTRICT HOSPITAL

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PREVALENCE AND ANTIBIOTIC RESISTANCE PATTERNS OF *ESCHERICHIA COLI* AMONG HOSPITALISED PATIENTS AT THIKA DISTRICT HOSPITAL

C. NDUNG'U, A. W. T. MUIGAI and S. KARIUKI

ABSTRACT

Background: Emerging resistance to antimicrobial drugs increases morbidity and mortality by hampering the provision of effective chemotherapy, and makes treatment more costly. The emergence of resistance to antimicrobial agents is a global public health problem, especially in pathogens causing nosocomial infections.

Objectives: To determine the carriage of *E. coli* from wounds and urine in catheterised inpatients at Thika District Hospital (TDH) and to determine antimicrobial resistance patterns to β -lactams, aminoglycosides and (fluoro) quinolones.

Design: A cross-sectional study.

Setting: Thika District Hospital among hospitalised patients.

Subjects: A total of 450 specimens were collected and forty two (42) *Escherichia coli* isolated. Pus swabs were collected from wounds and urine was collected aseptically from the inpatients with catheters. *Escherichia coli* were identified by culture methods and biochemical tests. Antimicrobial susceptibility testing was performed by Kirby-Bauer disc diffusion method and interpreted according to Clinical Laboratory Standards Institute recommendations.

Results: Susceptibility results in aminoglycosides were, resistance for amikacin, gentamicin and kanamycin was 20%, 39% and 51% respectively. Resistance in penicillin was ampicillin 85% and piperacillin 83%. Resistance for sulfamethoxazole was 83%, tetracycline 66%, nalidixic acid 44% and chloramphenicol 39%. In amoxicillin/clavulanic acid, resistance was 68%. Cephalosporins' resistance was ceftazidime 22%, cefotaxime 56%. Resistance for imipenem and tazobactam was 7% and 12% respectively.

Conclusion: Due to observations on resistance to antimicrobial agents commonly used in Thika District Hospital, this shows that there is need to revise antimicrobial policy in this region in the treatment of *E. coli* infections.

INTRODUCTION

Escherichia coli (*E. coli*) are facultative anaerobe Gram-negative rods, are motile, and some possess capsules while others are non-capsulated (1). It is normally found in the gut of both man and animals. However it also colonises the lower end of urethra and vagina. *Escherichia coli* cause urinary tract infections, neonatal meningitis, diarrhoea and septicaemia (1). It is the leading pathogen causing urinary tract infections (2). *Escherichia coli* is also among the most common pathogens that cause blood stream infections (3), wounds, otitis media and other complications in humans (4).

Resistance to antimicrobials in *E. coli* has been reported worldwide (5). There is increasing rates of resistance among *E. coli* which is a growing concern in both developed and developing countries (6). Treatment options for *E. coli* strains include β -lactam antimicrobials, aminoglycosides and (fluoro)-quinolones. In most cases combined therapy comprising, a β -lactam antimicrobials and a (fluoro) quinolone or an aminoglycoside is prescribed for serious infections. Cephalosporins alone as a monotherapy or as a β -lactam/ β -lactamase inhibitor such as amoxicillin-clavulanic acid combinations may be effective against *E. coli* infections.

MATERIALS AND METHODS

Study site: The study was conducted at Thika District Hospital, Kiambu County, Kenya.

Study population: The study population included both adults and children who gave consent to participate in this study. For children, consent was given by their parents or guardians. All study participants had been admitted in the ward for at least 48 hours. They were all in-patients with wounds, burns wounds and those with indwelling urine catheters. Information sheets explaining the purpose of the study and the procedures involved were given to the patients/guardians. Patients/guardians who agreed to participate in the study were given the consent form to sign.

Design of the study: A cross-sectional study design was used.

Sampling: Sterile cotton swabs were used to aseptically collect specimens from fresh burn sites and surgical wounds. The swab was rolled gently on the surface area of the wound for about five seconds. The cotton swab was inserted directly into sterile bottle containing Stuart transport media. The handle of the swab was snapped off, with cotton tip remaining in the transport media. The bottle was sealed and then labeled with the patient's study number, ward and date. Urine specimens were collected in sterile universal bottles from patients with catheters. The specimens were labeled with the study number, ward and date. These specimens were delivered immediately to the microbiology department in the laboratory for processing.

Laboratory procedures: Processing of urine and pus swabs specimens. Swabs collected from wounds and urine from catheterised patients was processed according to standard operating procedures. Swab specimens were first inoculated on blood agar media and then in MacConkey media for lactose fermentation determination. Smears were then prepared and a Gram stain performed. Culture

plates were incubated aerobically at 37 °C for 18-24 hours. The identity of the isolates was confirmed by standard laboratory methods which included colony morphology, lactose fermentation, gram staining, oxidase, triple sugar iron, motility, and indole and citrate utilisation.

Antibiotic Susceptibility Testing: Antibiotic susceptibility test was done using Kirby-Bauer Disc diffusion method. At least four to five isolated colonies of the same morphologic type was selected from an overnight agar plate of a pure culture and emulsified in 10 ml sterile normal saline (0.85% sodium chloride) in sterile glass tubes. Susceptibilities were tested to antimicrobials commonly used for treatment of infections caused by *E. coli* including aminoglycosides thus gentamycin (10µg), amikacin (30µg), and kanamycin (30µg). Beta-lactams included piperacillin (100µg), amoxicillin/clavulanic acid (20/10 µg), piperacillin/tazobactam (100/10 µg), cefotaxime (30µg), ceftazidime (30µg) and imipenem (10µg). Fluoro-quinolones included nalidixic acid (30µg), ciprofloxacin (5µg) and ofloxacin (5 µg). Other agents were trimethoprim/ sulfamethoxazole (25µg) and tetracycline (30µg) (all from Oxoid, Basingstoke, UK).

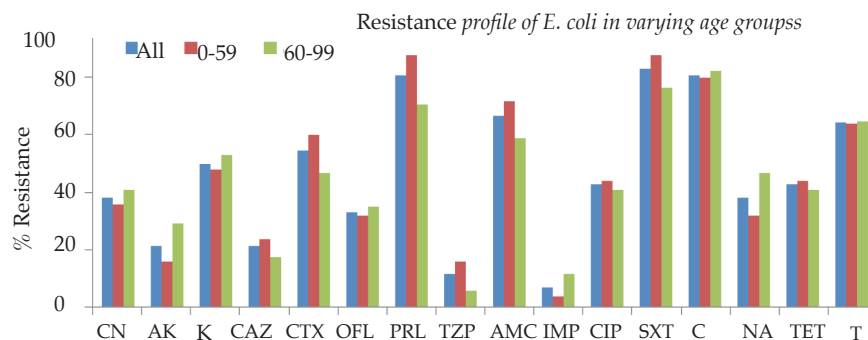
Quality control organisms used to test for disc potency and media quality were ATCC *E. coli* 25922. The plates were incubated at 37°C for 18-24 hours. Susceptibility results were interpreted according to the Clinical Laboratory Standards Institute (CLSI) 2011.

Data entry and analysis were performed using Ms excel and STATA tool respectively. Categorical data were tested using chi-square test. P-values of < 0.05 were considered statistically significant. Ethical clearance for the study was obtained from Kenya Medical Research Institute (KEMRI) Scientific Steering Committee and Ethical Review Committee (ERC No. 2081). Approval was also obtained from Medical superintendent, Thika District Hospital and informed consent from the patients or their guardians.

RESULTS

Figure 1

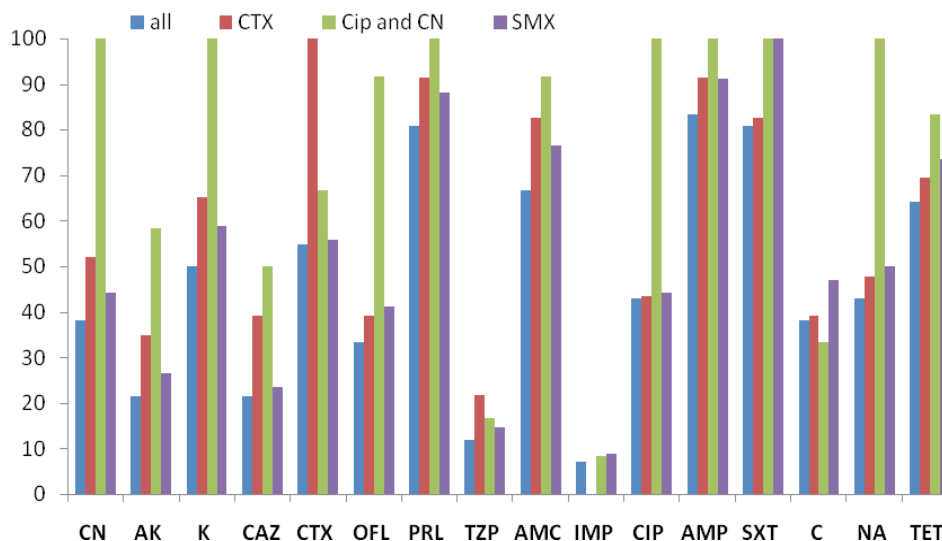
Resistance profile of *E. coli* to various classes of antimicrobials in relation to two age groups 0



A comparison of the resistance profiles of strains obtained from two age groups was done, that is, those between 0 and 59 years and 60 years and above. It was noted that generally, there were no significant differences in resistance to combinations of antimicrobial agents between the two groups (P: 0.9115).

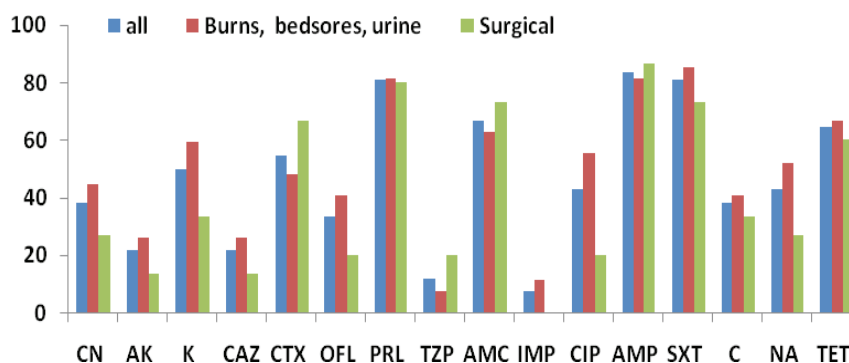
Figure 2

Comparison of resistance patterns of *E. coli* among a cephalosporin, combined resistance to gentamicin and ciprofloxacin, and sulfamethoxazole



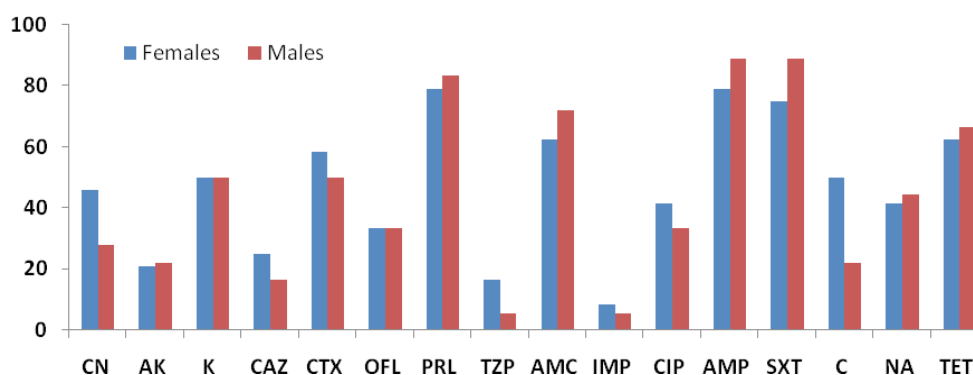
Resistance patterns were compared among strains showing resistance to at least a cephalosporin such as cefotaxime and those with combined resistance to ciprofloxacin, gentamicin and those resistant to sulfamethoxazole. Those with combined resistance to ciprofloxacin and gentamicin were resistant to more combinations of antimicrobials than those susceptible to these antimicrobials (P: 0.0066).

Figure 3
Resistance patterns of *E. coli* from different sources of specimen



Investigations were done on whether there were significant differences in resistance patterns of strains depending on the source of specimen. A comparison of resistance of strains obtained from surgical sites and those from burns, bedsores and urine was done and it was found that isolates from non-surgical sites were slightly more resistant than those from surgical specimens ($P: 0.0261$).

Figure 4
Comparison of resistance patterns of *E. coli* between males and females



Analysis to compare resistance patterns between *E. coli* strains obtained from males and females revealed no differences between the two populations ($P: 0.8575$).

DISCUSSION

Escherichia coli have widely been associated with various clinical hospital acquired infections (7). The results of antimicrobial resistance pattern give serious cause for concern because the predominant bacterial isolates were highly resistant to the commonly available antimicrobial agents in Kenya. Majority of strains from all specimen types were found to be multi-drug resistant (MDR). Similar results have been published before (8). These findings show that in general, isolates from burns and surgical wounds are the second most resistant strains after those from urinary tracts.

Although majority of strains were susceptible to carbapenems, some *E. coli* strains that were resistant to imipenem were identified. Carbapenems however

remain the drugs of choice against the strains investigated as reported in a related study (9).

This study reveals that a significant proportion of isolates from wounds and burns are highly multidrug resistance. The burn patients are particularly predisposed to different infections which are linked to impaired resistance from disruption of the skin's mechanical integrity and generalised immune suppression (10). In these patients, the skin barrier is replaced by a protein rich, avascular environment that provides a favorable niche for microbial colonisation and proliferation (11). It is also common for patients with wounds and burns to use topical microbial applications especially those containing heavy metals such as silver and those with neomycin and tetracycline. Such practices may promote emergence of highly resistant strains.

This study shows that *E. coli* isolates from urine were significantly more resistant than those from wounds that is burns and surgical sites. Isolates from non-surgical sites especially from urine, were slightly more resistant than those from surgical specimen ($P: 0.0261$). Similar high resistances among urine strains have been reported in related studies for instance, resistance rates among strains of *E. coli* isolated from women with urinary tract infection (UTI) averages 30% for both sulphonamides and ampicillin, varying from 17% to 54% in different countries (12). Trimethoprim resistance ranges from 11% in Scandinavian countries to 34% in Spain and Portugal. Most Scandinavian countries record low resistance to fluoroquinolones but resistance to fluoroquinolones may reach 20% in southern Europe (12).

In Spain where antimicrobials can be used without restrictions, reduced susceptibility of *E. coli* strains isolated from patients with UTI to sulfamethoxazole-trimethoprim (SXT-TRIM) combinations has been reported to be as high as 26% and 16% to fluoroquinolones (13).

This current study shows that isolates obtained from urine were fairly susceptible to common agents used for the treatment of UTIs compared to those reported in other countries. These results differ from the expectation given the fact that the sample population was hospitalised patients who are generally reported to have isolates that are highly resistant (14). Majority of isolates from urinary tracts were MDR and a significant proportion of these were resistant to sulfamethoxazole-trimethoprim (SXT-TRIM). These strains were also resistant to other classes of antimicrobials further limiting the choice of alternative antimicrobials. These results indicated similar sensitivity patterns amongst the uropathogens as those observed in Kolkata, India. The World Health Organization (WHO) guidelines recommend sulfamethoxazole-trimethoprim and ampicillin as the first choice for the UTI treatment (15) and empiric treatment is recommended for treatment of uncomplicated urinary tract infection (uUTI) (16). However, as was revealed in the present study, SXT-TRIM cannot serve as treatment of choice among majority of these isolates.

A recent report also indicated that SXT-TRIM and ampicillin were not suitable drugs of choice for treating UTIs (17). However, another study showed that amikacin and nitrofurantoin are the most effective treatments for UTIs in China (18). Since inappropriate use of antibiotics may lead to complications and treatment failure, this study suggest that there is a need to have an idea of the resistance phenotypes of isolates from urine before commencing treatment. It is therefore important to conduct culture and susceptibility testing for serious infections.

In conclusions, this study shows that isolates from urine and wounds (bedsore, burns and surgical wounds) are significantly resistant to common antimicrobials available for use in Kenya. The *E. coli* strains from urine are particularly resistant. A few isolates are resistant to carbapenems indicating that treatment of such strains would present a great challenge in clinical settings. This study shows that SMX-TRIM is no longer effective against most *E. coli* isolates and that a significant proportion of these strains are MDR. It has also been shown that women may present a significant reservoir for MDR strains. There is therefore need to check antimicrobial use patterns in the management of burns and surgical wound, UTIs and bedsore. There is also need to promote culture and susceptibility testing for isolates implicated in serious infections.

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