Original Article

Antibiotic resistance pattern of *Escherichia coli* isolates from outpatients with urinary tract infections in Somalia

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Abstract

Introduction: Several studies suggest increasing rates of antibiotic resistance among adult populations with Urinary tract infections (UTI). *Escherichia coli* (*E. coli*), is the predominant bacterium both in the community and in hospital environments causing uropathogenic infections. This study aimed to estimate the common uropathogen bacteria that cause UTI among outpatients as well as to determine the antibiotic resistance pattern of *E. coli* isolates among outpatients with UTI infections at Shaafi hospital, Mogadishu, Somalia.

Methodology: A cross-sectional study was conducted at Shaafi Hospital, Mogadishu, Somalia. A total of 128 samples were collected from outpatients suspected of UTI and tested through bacteriological investigations and antimicrobial susceptibility tests following the Kirby-Bauer agar disc diffusion method.

Results: *E. coli* was isolated in 34 (41%) out of the total 83 samples that showed growth followed by *Staphylococcus aureus* 22 (26.5%), *Pseudomonas aeruginosa*, 13 (15.7%), *Klebsiella pneumoniae* 8 (9.6 %) and *Proteus spp.* 6 (7.2%). *E. coli* was highly sensitive to nitrofurantoin, 29 (85.3%), and ciprofloxacin (n = 23, 67.6%), and had the highest resistance rate of ceftriaxone, 33 (97.1%). The odds of having UTI were higher in patients with a history of UTI (Odds ratio OR = 0.211, 95% confidence interval CI: 0.080, 0.553) and history of antibiotic use (OR = 0.322, 95% CI: 0.113, 0.917). Increased resistance rate of *E. coli* against cephalosporins could be due to its excessive use as empirical therapy.

Conclusion: The study indicates that outpatients with UTI could be at high risk of antibiotic resistance, suggesting regular surveillance and monitoring of antibiotics.

Key words: Antibiotic resistance; antimicrobial susceptibility test; *Escherichia coli*; urinary tract infections, Somalia.

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Introduction

Urinary tract infection (UTI) mainly results from the colonization of microorganisms in the genitourinary system [1]. It is a serious public health problem in developing countries, as encountered in both hospital and community settings, causing significant morbidity and mortality [2]. The factors associated with the high prevalence of UTI within the population include age, gender, sexual activity, contraceptive use, previous history of UTI, indwelling catheter equipment and hygiene problems [1,3,4]. Amongst the uropathogenic bacteria known to cause urinary tract infection, *Escherichia coli* is the predominant bacteria in both community and hospital environments [1,5,6]. However, the diversity of bacterial uropathogens that cause UTI varies geographically [5,7]. The emergence of antibiotic resistance is also a significant problem in sub-Saharan Africa, particularly Somalia, a country crippling with inadequate laboratory facilities for microbiological investigation, despite this being an inevitable part of decision-making in antibiotic prescription to patients, clinicians mainly prescribe antibiotics empirically. Also, requesting this test for the few hospitals with the laboratory facilities is rare, as it incurs extra cost to the patient of which the majority cannot afford as they hail from a low-income family. Therefore, this alarming situation is growing at a high rate and aggravated by the inappropriate and excessive use of antibiotics [8]. As over-use of antibiotics at the study setting is mainly due to self-prescription of antibiotics by the patients in case when they suspect of UTI infections one drug after the other without doctor’s consultation. This could be due to lack of awareness by the patients or non-existence of regulating policies and guidelines for antibiotic use. The ease of access to antibiotics in the communities by buying from the over-the-counter without doctor’s prescription at a low price in addition to the poor quality adds to escalating antibiotic resistance [9]. However, knowledge of the antimicrobial resistance patterns of common
In uropathogens is essential and necessary to guide the clinicians on appropriate and cost-effective antibiotic therapy. It also encourages clinicians to consider local susceptibility patterns of urinary pathogens prescribing antibiotics before laboratory results of urine culture in case of emergency for empirical therapy. However data of such susceptibility pattern is not available in Somalia. Therefore, this study aims to estimate the common uropathogen bacteria that cause UTI among outpatients at Shaafi hospital, Mogadishu, Somalia and to determine the antibiotic resistance pattern of \textit{E. coli} isolates among outpatients with UTI infections at Shaafi hospital, Mogadishu, Somalia.

**Methodology**

The study is cross-sectional conducted at Shaafi Hospital, Mogadishu, Somalia, during 2\textsuperscript{nd} April to May 31\textsuperscript{st}, 2019. A total of 128 clean catch-midstream urine samples were collected from outpatients suspected of UTI and then tested through microbiological investigations such as gram staining, microscopic identification, identify colony morphological appearance and biochemical tests following Clinical Laboratory Standard Institute (CLSI) procedure [10]

**Inclusion and exclusion criteria**

All Out-Patients complaining of urinary tract infections according to the attending clinician were included in the study. The study excluded those patients with evidence or suspected of UTI but refused to participate, and patients that were on antibiotic therapy within the week of sample collection.

**Sample procedure**

Midstream urine samples of outpatients with suspected urinary tract infections were collected in sterile containers at the laboratory of shaafi hospital. The samples were well maintained and transported within thirty minutes while storing in 4\degree C at the laboratory department faculty of medicine & health sciences of Jamhuriya University of science and technology for further processing and analysis.

**Culture specimen**

All specimens were directly cultured on standard media of Cystine lysine electrolyte deficiency (CLED) (HiMedia laboratory Pvt ltd in India) agar. A wire loop of (0.001 mL) deep in urine was used to inoculate on the CLED agar media, making a zigzag distribution then, plates were incubated aerobically at 37\degree C for 24 hours. Urine samples with colony $\geq 10^5$ CFu/mL were taken as significant growth (Positive urine culture $= 10^5$ CFU/mL). Significant single colonies were gram stained. The significant growth was identified further using biochemical tests such as Indole, Citrate Utilization and triple Sugar Iron(TSI) [10].

**Antimicrobial Susceptibility testing**

Antimicrobial susceptibility testing was performed on \textit{E. coli} isolates identified from the urine of outpatients using the Kirby-Bauer agar disc diffusion method following the Clinical laboratory standard institute (CLSI) guideline [10]. Briefly, a 3-5 of well-isolated colonies of the bacteria were collected in a sterile wire loop and emulsified into a tube containing 3-5 mL sterile physiological saline. In good light, the turbidity of the suspension was matched to the turbidity of the standard. Using a sterile dry cotton swab, a sample of the inoculums with adjusted turbidity was streaked on the surface of the semi-dried Mueller–Hinton agar plate in well-distributed directions, after removal of the excess by pressing against the sides of the test tube. Using sterile forceps, the selected antibiotics disks of Levofloxacin, Nitrofurantoin, Ciprofloxacin, Ceftriaxone, and Cefixime were placed on the plates and incubated at 37\degree C for 24 hours. The sensitivity and resistance were determined by measuring the diameter of zones of bacterial growth inhibition with a meter rule and then compared against zone diameter breakpoints and interpretative of susceptibility according to CLSI [10].

**Statistical data analysis**

After this information was collected, we used a quantitative method for analyzing and interpreting data. The data were analyzed using the statistical package for the Social Science technique (SPSS) program (version 20.0). Cross-tabulation analysis and odds ratio (OR) with 95% confident interval (CI) was used to determine the association of demographic characteristics and UTI.

**Ethical consideration of the study**

The study obtained approval from the research ethics committee of Jamhuriya University of Science and Technology (Reference: M20181120, Dated: 20\textsuperscript{th} November 2018) for these research activities to be carried out. Besides, Shaafi hospital administration granted permission to access the hospital. The research was conducted respecting the ethical values, confidentiality, and moral expectations of the respondents after their verbal consent.
Results

A total of 128 outpatients suspected of urinary tract infection participated in this study with the age range from 20 to 40 years. Among the study subjects, 101 (79%) were married, 18 (14%) were single, and 9 (7%) were divorced. 91 (71%) were female, and 37 (29%) were male Table 1.

The prevalence of UTI among the study participants was 67.2% (n = 82/122). From the total number of subjects complaining of UTI, 59 (46.1%) were 40 and elder years while 41 (32%) were between the age of 20-29 years and the rest 28 (21.9%) were 30-39 years as shown in Table 1. A cross-tabulation analysis was performed to determine the association of sociodemographic characteristics and UTI. Six sociodemographic variables selected in this study were age, gender, marital status, symptoms of UTI, history of UTI and history of antibiotic use. Concerning the age, the odds of having UTI was 1.197 (95% confidence interval (CI): 0.434, 3.302) higher among the age group of 30-39 years and 1.281 (95% CI: 0.551, 2.976) times higher in age group of 40 years and elder than the reference group of 20-29 years of age. For gender, females have higher odds 1.4 times more than males of having UTI (95% CI: 0.619, 3.211), while the odds of having UTI among single and divorced groups were 2.258 (95% CI: 0.817, 6.238) and 2.823 (95% CI: 0.709, 11.232) times higher than the reference group of married respondents. However, age, gender and marital status were not statistically significant. People with the previous history of having UTI and history of antibiotic use had higher odds of 0.211 (95% CI: 0.080, 0.553) and 0.322 (95% CI: 0.113, 0.917) times more than people with no history of UTI and antibiotic use, respectively. Finally, having symptoms of UTI is strongly associated with UTI (OR = 0.098, 95% CI: 0.011, 0.863) compared to not having symptoms of UTI. Hence, history of UTI, history of antibiotic use and symptoms of UTI are strongly associated with UTI infection, as they were statistically significant

A total of 83 isolates of uropathogens causing UTI among outpatients were identified from the urine samples. Of these, *E. coli* was the predominant microorganism accounting for 34 (41%) of the total

| Characteristics            | Urine culture | Total tested (%) | OR      | 95% CI      |
|----------------------------|---------------|------------------|---------|-------------|
|                            | Positive Total (%) | Negative Total (%) |         |             |
| Age (years)                |               |                  |         |             |
| 20-29                      | 28 (68.3)     | 13 (31.7)        | 41 (32) | Reference   |
| 30-39                      | 18 (64.3)     | 10 (35.7)        | 28 (21.9)| 1.197       |
| Above 40                   | 37 (62.7)     | 22 (37.3)        | 59 (46.1)| 1.281       |
| Total                      | 83 (64.8)     | 45 (35.2)        | 128 (100) | Reference   |
| Gender                     |               |                  |         |             |
| Male                       | 26 (70.3)     | 11 (29.7)        | 37 (29) | Reference   |
| Female                     | 57 (62.6)     | 34 (37.4)        | 91 (71) | 1.410       |
| Total                      | 83 (64.8)     | 45 (35.2)        | 128 (100) | Reference   |
| Marital status             |               |                  |         |             |
| Married                    | 70 (69.3)     | 31 (30.7)        | 101 (79) | Reference   |
| Single                     | 9 (50)        | 5 (50)           | 14 (14)  | 2.258       |
| Divorced                   | 4 (44.4)      | 5 (55.6)         | 9 (7)    | 2.823       |
| Total                      | 83 (64.8)     | 45 (35.2)        | 128 (100) | Reference   |
| Symptoms of UTI            |               |                  |         |             |
| No                         | 1 (16.7)      | 5 (83.3)         | 6 (4.7)  | Reference   |
| Yes                        | 82 (67.2)     | 40 (32.8)        | 122 (95.3)| 0.098       |
| Total                      | 83 (64.8)     | 45 (35.2)        | 128 (100) | Reference   |
| History Of UTI             |               |                  |         |             |
| No                         | 48 (55.2)     | 39 (44.8)        | 87 (67.97)| Reference   |
| Yes                        | 35 (85.4)     | 6 (14.6)         | 41 (32.03)| 0.211       |
| Total                      | 83 (64.8)     | 45 (35.2)        | 128 (100) | Reference   |
| History of antibiotic use  |               |                  |         |             |
| No                         | 7 (41.2)      | 10 (58.8)        | 17 (13.3)| Reference   |
| Yes                        | 76 (68.5)     | 35 (31.5)        | 111 (86.7)| 0.322       |
| Total                      | 83 (64.8)     | 45 (35.2)        | 128 (100) | Reference   |

OR: Odds ratio; UTI: Urinary tract infection; CI: Confidence interval.
isolates. The rest of the pathogens were *Staphylococcus aureus* with a prevalence rate of 22 (26.5%) followed by *Pseudomonas aeruginosa* 13 (15.7%), *Klebsiella pneumoniae* 8 (9.6%), and *Proteus spp* 6 (7.2%) respectively as demonstrated in Table 2.

The results of the antimicrobial susceptibility pattern of *E. coli* isolates of outpatients with UTI on five different antibiotic discs that are commonly prescribed for outpatients at Shaafi hospital are presented in Table 3. The majority of the isolates of *E. coli* were susceptible to ciprofloxacin 23 (67.6%) and Nitrofurantoin 29 (85.3%), whereas all isolates of *E. coli* demonstrated high resistance of cephalosporin resistance (ceftriaxone 33 (97.1%) and cefixime 21 (61.8%). Alarmingly, almost all isolates of *E. coli* 30 (88.2%) showed resistance to two or more drugs indicating a high chance of multidrug resistance (MDR) *E. coli* in the clinical isolates.

**Discussion**

Antibiotic resistance was a significant problem among bacteria isolated from UTI in adult patients in Somalia. The study findings likely indicate a high prevalence of UTI among study participants 67.2% (n = 82/122), as this was also reported in a similar study [11]. The risk of UTI among outpatients with the previous history of UTI, history of antibiotic use and having symptoms of UTI were significantly higher than those without a previous history of UTI and antibiotic use; and not having symptoms of UTI. These could indicate there are an association between former variables and urinary tract infections as indicated by other studies [1,12,13]. This could be related to the overuse of antibiotics without microbiological investigations since most clinicians depend on empirical therapy as the first decision or even self-prescription of antibiotics by the patients. Despite the fact that the statistics were not significant between marital status and gender with UTI, yet high percentages of married respondents (n = 70/101, 69.3%) and females (n = 57/91, 62.6%) were in UTI positive group. These were also reported in other studies [14,15].

The most predominant uropathogen that causes UTI among outpatients at Shaafi Hospital was *E. coli*, as it had concurred with previous studies from different geographic areas [14,16-19] However, this was inconsistent with a study from the southwest of Nigeria, which reported *Klebsiella spp* to be the most prevalent uropathogen, and these might have contributed by the differences in the study design, environmental factors, and also patient selection criteria [12]. *S. aureus* being the second most frequent uropathogen was not a unique case only to this study [20] and this might be due to complicated UTI since it is known to cause bacteremia which could disseminate to urinary to urinary tract [20] also pseudomonas aeruginosa is also considered to be among organisms responsible for urinary tract infections [5]. The majority of the respondents of the study were females 91 (71.1%) in comparison to males, which were 37 (28.9%). Showing considerable differences in the prevalence of UTI among outpatient visiting hospital setting, this agrees with a study from Nigeria, which showed that the prevalence of UTI was very high in females attending hospitals for UTI cases.

### Table 2. Distribution of bacterial Uropathogens isolated from urine culture of urinary tract infected outpatients (N = 83) at Shaafi hospital, Mogadishu, Somalia during April 2nd to May 31st, 2019.

| Organism Isolated       | Total (%) |
|-------------------------|-----------|
| *Escherichia coli*      | 34 (41)   |
| *Staphylococcus aureus* | 22 (26.5) |
| *Pseudomonas aeruginosa*| 13 (15.7) |
| *Klebsiella pneumonia*  | 8 (9.6)   |
| *Proteus spp*           | 6 (7.2)   |
| **Total**               | 83 (100)  |

### Table 3. Antibiotic resistance pattern of *E. coli* isolated from urine culture of urinary tract infected outpatients (N = 34) at Shaafi hospital, Mogadishu, Somalia during April 2nd to May 31st, 2019.

| Antimicrobials | Pattern  | Total (%) |
|----------------|----------|-----------|
| Ciprofloxacin  | Sensitive| 23 (67.6) |
|                | Resistance| 11 (32.4) |
| Nitrofurantoin | Sensitive| 29 (85.3) |
|                | Resistance| 5 (14.7)  |
| Levofoxacin    | Sensitive| 17 (50)   |
|                | Resistance| 17 (50)   |
| Ceftriaxone    | Sensitive| 1 (2.9)   |
|                | Resistance| 33 (97.1) |
| Cefixime       | Sensitive| 13 (38.2) |
|                | Resistance| 21 (61.8) |

287
111 (74%) compared to males 39 (26%) [14,15] as well as study conducted in Iran reported high proportion of UTI among outpatients were females (85.2%) [4]. These results indicated that females were prone to UTI as these were attributed to the nature of urogenital tract of the females; it is proximity to the anus and also lacking the bacteriostatic nature of prostatic secretions of male [14,15,20]. In this study it revealed that the age group with the highest prevalence of UTI was above 40 years (62.7%). In females this could be associated with vaginal prolapse following menopause, which could escalate the risk of bacteriuria as the vaginal pH increases due to decreased lactobacillus in the birth canal as this gave a chance for other uropathogens to colonize. Other risk factors that could be associated with UTI include genitor-urinary atrophy, catheterization, and recurrent UTI [1,14].

Due to the high resistance strains of E. coli, the study further investigated antimicrobial susceptibility and demonstrated that from the five antimicrobial tested against E. coli, ciprofloxacin, and nitrofurantoin had the highest susceptibility of 67.6% and 85.3% respectively, agreed by various previous studies [1,2,4,14,17]. However, in contrast to the present study, some studies showed high resistance of E. coli against nitrofurantoin [7,21]. In this study, high resistance was exhibited by E. coli against ceftriaxone and cefixime 97.1% and 61.8%, respectively, as these agree with previous studies [22]. Therefore, the high prevalence of resistance against third-generation cephalosporins, a first-line antibiotic therapy against urinary tract infections caused by gram-negative bacteria in the study area were escalating, though they were the preferred antimicrobials in the study setting. Similarly, it also demonstrated in other related studies. High resistance rate was observed 30 (88.2%) of E. coli isolates against two or more commonly used antibiotics in the study setting, these were also reported in previous studies with similar situations [2,12,16] as these may be associated with different variables such as incomplete duration of drugs, easy availability of drugs, self-medication practices, lack of strict laws of on drugs that punishes for drug misuses hence all this contributed to high emerging rate of drug resistance [11,13]. Therefore, strong policy measurements of antibiotic usage were to be put in to place to curb these misuse of drugs, and also regular monitoring of the emerging of multi-drug resistance uropathogens have to be carried out to ensure the golden age of antibiotic are maintained.

The limitation of this study was the use of outpatient of one hospital, which may not be an accurate representation of a general population in this setting. Also, the bias method of antibiotic selection was based on the five commonly prescribed antibiotics for urinary tract infections at Shaafi hospital.

Conclusion
The results indicate high rates of antibiotic resistance against E. coli among outpatients with UTI, as well as shattering the light of the commonly used antibiotics against urinary tract in this context. We urge the physicians to consider the local susceptibility patterns of urinary pathogens of E. coli obtained in this study for determining empirical therapies. There is an urgent need for regular surveillance and monitoring of antibiotics to be aware of the trend and also tackle antibiotic resistance.

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