ORIGINAL ARTICLE

Susceptibility Profiles of Bacterial Pathogens Causing Community-Acquired Urinary Tract Infections

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ABSTRACT

Introduction: Uncontrolled empirical treatment of urinary tract infections (UTIs) has negative aspect on predicting the emergence of antimicrobial resistance and knowledge of those patterns has become extremely important from time to time. Therefore, the aim of the present study was to check the prevalence and resistance patterns of uropathogens in the community acquired UTIs. **Methods:** A total of 7132 urine samples were combined from male 3131 (43.9%) and female 4001 (56.1%) outpatients suspected of having UTIs, respectively over a three-year period and cultured on routine culture media. The bacteria have been identified using basic biochemical tests, and sensitivity to various antibiotics was determined by the method of disk diffusion. **Results:** Of 7132 urine samples 797 (11.2%) yielded significant uropathogens. Among the bacterial species, *Escherichia coli* was the major causative agent of UTIs for both gender (63.7%), followed by *Klebsiella spp* (20.8%), *Enterococcus faecalis* (5.3%), *Pseudomonas spp* (4.1%), *Proteus spp* (3.1%), *Enterobacter spp* (1.5%), *Candida albicans* (0.6%), *Staphylococcus saprophyticus* (0.5%), *Providencia spp* (0.1%) and *Staphylococcus aureus* (0.1%). The antibacterial sensitivity testing for *E. coli*, to commonly used antibiotics were showed variable resistant as follows: Ampicilln (78%), Amoxicillin (71%), trimethoprim sulfamethoxazole (42%), Amox/clav. (14%) gentamicin (20%), nitrofurantoin (11%), nalidixic acid (22%), ciprofloxacin (20%), Imipenem (16%),Ceftazidim (26%),Cefotaxim (25%),Ceftriaxon (21%),Cefuroxim (33%). **Conclusions:** The findings showed that antimicrobial resistance patterns of uropathogens in variable, and continuous monitoring of resistance patterns by using of antibiotic susceptibility testing in the laboratory is the most appropriate to treat UTIs rather than the choice of UTIs empirical treatment.

Keywords: Antimicrobial resistance, Antibiotics, Community-acquired UTI

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INTRODUCTION

Urinary tract infections (UTIs) affects millions of people each year (1,2).UTIs are commonly found in female than male counterparts (3). Although many different microorganisms can potentially be uropathogens, however, bacteria are still considered the main frequently agents in over 95% of the cases (4). Among these uropathogens, E*scherichia coli, Klebsiella pneumoniae, Proteu smirabilis, Pseudomonas aeruginosa*, and *Enterococcus faecalis* are the main bacterial species involved (5-8). Reducing the course of UTIs requires a prompt diagnosis, which in turn could prevent the ascending infection and other clinical consequences related to complicated UTIs (9).A heightened concern has been raised by many countries on antimicrobial resistance among uropathogens from hospital and community settings (10,11). To improve guidelines on antimicrobial resistance, it is necessary to embark on regular monitoring of resistance patterns especially in the community. However, data on antimicrobial resistance among uropathogens isolated from our community in Libya is lacking. Hence, the current objectives aimed to determine the prevalence and Patterns of resistance to urinary tract pathogens in community-acquired UTIs.

MATERIALS AND METHODS

This study was carried out in the medical bacteriology laboratory of Sabratha Teaching Hospital Libya over a three-year period. The samples were taken from outpatients diagnosed with a suspicion of having UTI by attending physicians. For this study, exemption from ethics approval has been granted by Ministry of Higher Education and Scientific Research, University of Basra College of Dentistry, Iraq because it does not deal with patients directly. All suspected cases were sent to the bacteriology laboratory to identify the pathogen. There were 4001 female samples and 3131 male. The age of the patients started from 1 to 70 years (average age is 30.5 years).A clean catch midstream urine technique was used to obtain the urine samples from adult patients while sterile urine bags were used to collect the samples from children aged less than 2 years old. All urine samples have been cultured on blood agar, MacConkey agar and Sabouraud agar plates, and then incubated overnight at 37°C. The uropathogens was considered significant if the bacterial count was $\geq 10^5$cfu/mL based on the established guideline (12). These significance bacteria were identified by biochemical reaction tests using analytical profile index (API) identification systems (bioMérieux, France).For mixed growth cultures, the two predominant bacteria(those with the two highest counts) were identified. However, bacterial count of$<10^3$cfu/mL were regarded as non-significant growth.

Antimicrobial sensitivity Testing

The disk routine method was utilized for the antibiotic sensitivity testing according to the National Committee on Clinical Laboratory Standards (NCCLS) guidelines (13). All antibiotic discs were acquired from Oxoid Limited (Basingstoke, UK). All bacteria were tested against the following antimicrobial agents: ampicillin, amoxicillin, trimethoprim-sulfamethoxazole, amoxicillin/clavulanic acid (amox/clv), gentamicin, nitrofurantoin, nalidixic acid, ciprofloxacin, imipenem, ceftazidime, cefotaxime, ceftriaxone, and cefuroxime.

Statistical analysis

Tests of statistical significance of differences were explained as percentages using the chi-square test.

RESULTS

Bacterial isolation and identification

A total 7132 urine samples were cultured over a three-year period, of which 797(11.2%) had significant bacteriuria. The urine specimens were taken from 4001 female and 3131 male outpatients. The age of all patients started from 1 to 70 years (mean age 30.5 years). The culture positive rate was 14.0% (559/4001) and 7.6% (238/3131) for female and male outpatients, respectively (Table I). Gram negative bacilli were predominantly isolated than Gram-positive cocci [93.5% versus 5.9%] (Table I). Further analysis on the type of uropathogens and gender preponderance revealed that *Escherichia coli, Klebsiella pneumoniae, Enterococcus faecalis, Enterobacter spp, Candida albicans* and *Staphylococcus aureus* were the most predominant bacteria isolated from both gender. However, these bacteria were more frequently isolated in female compared to male outpatients. Surprisingly, the prevalence rates of UTIs due to Pseudomonas aeruginosa and Proteus spp were significantly higher in male than female outpatients (5.88% and 5. 89% versus 3.39% and 1.69%), respectively; p< 0.05). Interestingly, the prevalence rates of UTIs due to Staphylococcus saprophyticus and Providencia spp were significantly higher in female than male outpatients, respectively; (p < 0.05) (Figure1).

Table I: Results of total urine culture (%) and distribution of pathogens (%) in total Positive culture

General characteristics (n=7132)	Frequency n (%)
Number of positive samples	797 (11.17%)
Female	559/4001, 13.97%
Male	238/3131, 7.60%
Non-pathogens	865 (12.12%)
Negative culture	5470 (76.79%)
Total	7132
Distribution of pathogens (%) in total positive culture {797 (11.17%)}	
Pathogens	No. of positive cultures (%) (*n* = 797)
E.coli	508 (63.73%)
Klebs.pneumonia	166 (20.82%)
Strept.faecalis	42 (5.26%)
Pseud.spp	33 (4.14%)
Proteus.spp	25 (3.13%)
Enterobacter.spp	12 (1.5%)
Candida.albicans	5 (0.62%)
Staph.saprophyticus	4 (0.5%)
Providinicea.spp	1 (0.12%)
Staph.aureus	1 (0.12%)

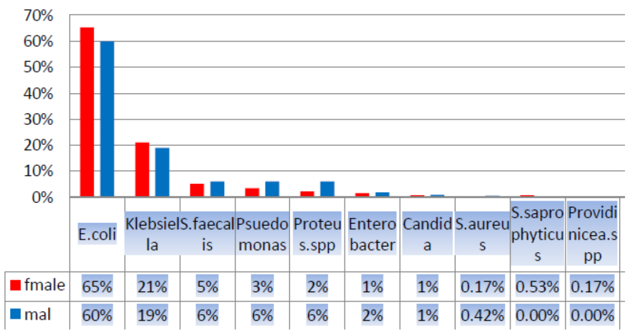
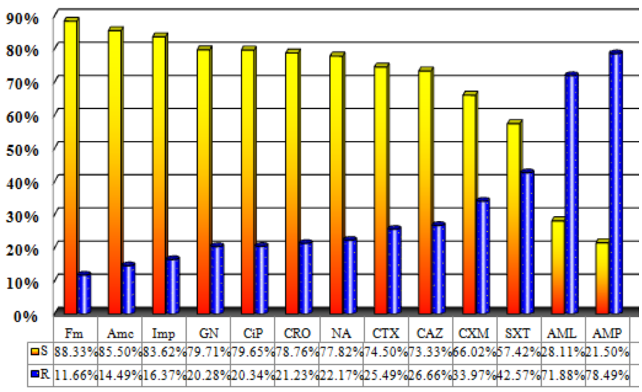


Figure 1: Frequency of isolated bacteria from positive urine samples according to patient gender

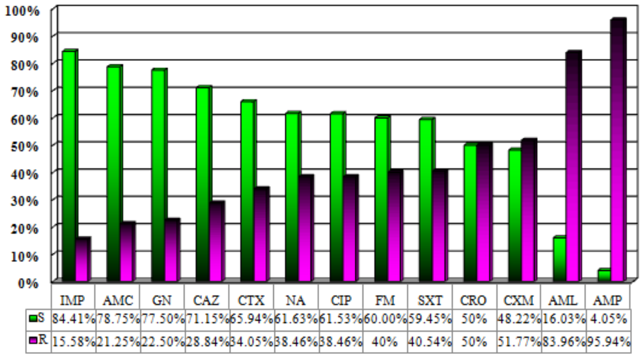
Antimicrobial sensitivity profiles

Being the most predominant uropathogen of UTI in this study, *E. coli* showed variable percentages of resistance rates to antibiotics as follows (in descending order): ampicillin (78.4%), amoxicillin (71.9%), trimethoprim-sulfamethoxazole (42.6%), cefuroxime (33.9%), ceftazidime (26.6 %), cefotaxime (25.4%), nalidixicacid (22.1%), ceftriaxone (21.2%), ciprofloxacin (20.3%), gentamicin (20.2%), imipenem (16.3%), amox/clv (14.4%) and nitrofurantoin (11.6%) (Figure 2). *Klebsiella spp* were the second predominant uropathogen that exhibited variable resistance to the antibiotics. Majority of *Klebsiella sp*p exhibited resistance to ampicillin (95.9) and amoxicillin (83.9).While 84.4%,78.7,77.5 and 71.1 of *Klebsiella spp* were susceptible to imipenem, amox/clv, gentamicin and ceftazidime, respectively (Figure 3). Among these uropathogens, *P. aeruginosa* and *Proteus spp* both exhibited the highest rate of resistance to most antibiotics with high rates of susceptibility to imipenem (85%) and gentamicin (81.4%), respectively (Figure 4, 5). *Enterococcus faecalis* was accounted for a low percentage of UTIs (5.26%) with identical halves of sensitive and resistance to the antibiotics used in this study (figure 6). Although the following isolates like *Enterobacter spp, Candida albicans, Staphylococcus saprophyticus, Providencia spp* and *Staphylococcus aureus* have been isolated from UTIs, they however represent very low percentage rates of urinary infections, 1.5%, 0.62% , 0.5%, 0.12%, and 0.12%, respectively, that they did not submit to antimicrobial susceptibility testing due to the absence of their statistical values.



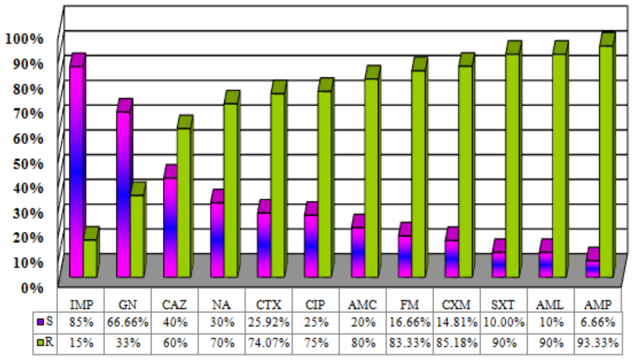
	Fm	Amc	Imp	GN	CiP	CRO	NA	CTX	CAZ	CXM	SXT	AML	AMP
S	88.33%	85.50%	83.62%	79.71%	79.65%	78.76%	77.82%	74.50%	73.33%	66.02%	57.42%	28.11%	21.50%
R	11.66%	14.49%	16.37%	20.28%	20.34%	21.23%	22.17%	25.49%	26.66%	33.97%	42.57%	71.88%	78.49%

Figure 2: Distribution of antibiotic susceptibility patterns of *E. coli* in community-acquired UTIs in the present study. AMP, Ampicillin; AML, Amoxicillin; SXT,Trimethoprim/sulfamethoxazole; CXM, Cefuroxim; CAZ, Ceftazidim; CTX, Cefotaxim; NAL, Nalidixic acid ; CRO, Ceftriaxon; CIP, Ciprofloxacin; GEN, gentamicin, IMP, Imipenem; AMC, amoxi/clav; FM, Nitrofurantoin.



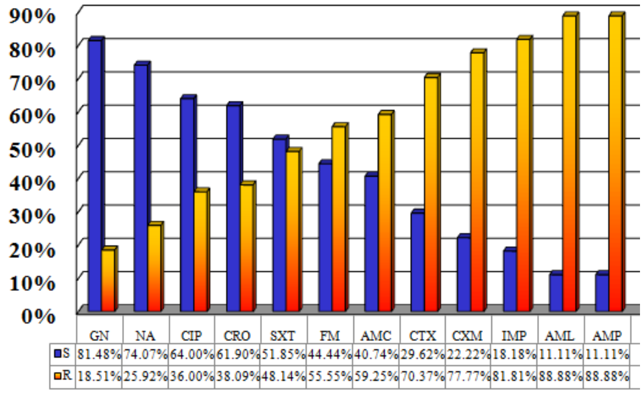
	IMP	AMC	GN	CAZ	CTX	NA	CIP	FM	SXT	CRO	CXM	AML	AMP
S	84.41%	78.75%	77.50%	71.15%	65.94%	61.63%	61.53%	60.00%	59.45%	50%	48.22%	16.03%	4.05%
R	15.58%	21.25%	22.50%	28.84%	34.05%	38.46%	38.46%	40%	40.54%	50%	51.77%	83.96%	95.94%

Figure 3: Distribution of antibiotic susceptibility patterns of *Klebsiella spp* in community acquired UTIs in the present study.



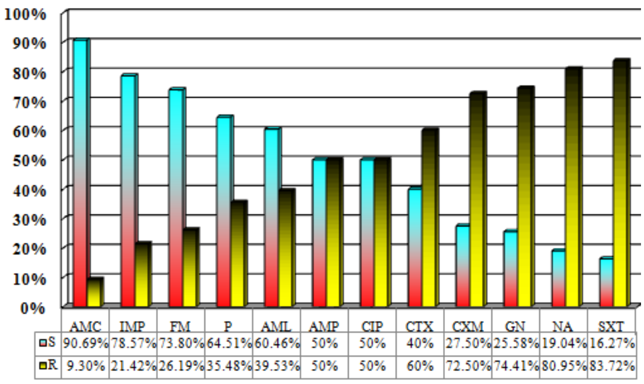
	IMP	GN	CAZ	NA	CTX	CIP	AMC	FM	CXM	SXT	AML	AMP
S	85%	66.66%	40%	30%	25.92%	25%	20%	16.66%	14.81%	10.00%	10%	6.66%
R	15%	33%	60%	70%	74.07%	75%	80%	83.33%	85.18%	90%	90%	93.33%

Figure 4: Distribution of antibiotic susceptibility patterns of *Pseudomonas aeruginosa* in community acquired UTIs in the present study



	GN	NA	CIP	CRO	SXT	FM	AMC	CTX	CXM	IMP	AML	AMP
S	81.48%	74.07%	64.00%	61.90%	51.85%	44.44%	40.74%	29.62%	22.22%	18.18%	11.11%	11.11%
R	18.51%	25.92%	36.00%	38.09%	48.14%	55.55%	59.25%	70.37%	77.77%	81.81%	88.88%	88.88%

Figure 5: Distribution of antibiotic susceptibility patterns of *Proteus spp* in community acquired UTIs in the present study



	AMC	IMP	FM	P	AML	AMP	CIP	CTX	CXM	GN	NA	SXT
S	90.69%	78.57%	73.80%	64.51%	60.46%	50%	50%	40%	27.50%	25.58%	19.04%	16.27%
R	9.30%	21.42%	26.19%	35.48%	39.53%	50%	50%	60%	72.50%	74.41%	80.95%	83.72%

Figure 6: Distribution of antibiotic susceptibility patterns of *Enterococcus faecalis* in community acquired UTIs in the present study

DISCUSSION

Most antimicrobial susceptibility surveillance studies of UTIs are more inclined toward hospitalized patients, but now issues on the antibiotic resistance possess a great concern among community-acquired UTIs (14). The present study involved 7,132 outpatients with a suspicion of UTIs, but only 11.17% had uropathogens in their urine samples. These data in terms of the positivity rates compared to other studies were lower detection percentages (15). This could probably be explained by the non-specific clinical manifestations of UTIs especially among children less than 2 years old (4), or however, most urine specimens in this study were taken from outpatients who had a simple lower UTIs, and most clinical diagnoses were made by general physicians rather than by a urologist. The main strengths of our survey are the number of much samples involved; the overall frequency of pathogens involved was as expected and confirms the validity of the survey with gender group or with other studies. The predominant species isolated was *E. coli*, is a well known of UTI pathogenis, exceeding 63.73% of proven infections in both genders. The results also confirmed that *E. coli* was at the top of the list compared to other bacterial pathogen of urinary bacterial infections and it was in agreement with other studies (16), as well as the recurrence rates in women more than in men (Table I and figure 1), the explanation may be due to the larger number of female urine taken or particularly the female at risk of developing UTIs because of their short urethra (17). Although the spread of bacteria in different region of the world is sometime similar, however, the antimicrobial resistance patterns from different parts are varying considerably and antibiotics resistance is rising. In this study, the frequency of antibiotic susceptibility with *E.coli* has been shown to change over time; the resistance rates were found highest percentages especially for ampicillin (78.49%), amoxicillin (71.88%) and trimethoprim-sulfamethoxazole (57.42%), but many are still sensitive with highest percentages to nitrofurantoin (88.33%), amoxi/clav(85.5%), imipenem (83.62%), gentamicin (79.71%), ciprofloxacin (79.65%) (Figure 2); these results are somewhat similar to other studies (18- 24).

Also it was found that *Klebsiella* occupied the second causative agent of urinary tract infections among the bacteria isolated in this study (Table I and Figure 1) and it showed a similar sensitivity to *E.coli* as shown in Figure 3. In line with other studies, *Klebsiella spp* also have shown the similar resistant to ampicillin (95.94%) and amoxicillin (83.96%), whereas many are still sensitive to imipenem (84.41%), amc/clv. (78.75%) and gentamicin (77.5%) (25,26), As a further matter in figure 4, *P. aeruginosa*, is normally related to urinary catheters but surprisingly to have among the community, it was also found with a high rate of resistance to antibiotics similar to other studies (27). While, in the case of proteus spp, which normally this bacterium is isolated from UTIs, it was totally resistant to ampicillin, amoxicillin, imipenem, cefuroxime and cefotaxime, (Figure 5).These resistance basically were found higher than other reports (28). Lastly *E. faecalis* was also isolated from urinary tract infections with low percentages and half of them were resistant to ampicillin,ciprofloaxcine, cefotaxime, cefuroxime, gentamicin, nalidixic acid and trimethoprim-sulfamethoxazole (Figure 6), which is worrying compared with other findings (29). This may be due to the fact that the excessive use of antibiotics without following medical prescriptions has led to the spread of bacterial resistance in our regions compared to other regions, which comply with the application of health guidelines. The resistance of different antibiotics can be explained based on regional areas and methods of using antibiotics also; in addition to that, the influence of inappropriate use of antibiotics may development of antibiotic-resistant between the strains. Moreover, appropriate diagnostics and optimized antibacterial therapy are vital in order to limit this escalating antibacterial resistance. To this extent, continuous surveillance of antimicrobial susceptibility at the hospitals as well as in private laboratories remains important for CA-UTIs.

CONCLUSION

The results of this study emphasized the imipenem, amox/clav and nitrofurantoin were found to be the most appropriate drugs for treatment of complicated UTI in Libya community. This study recommends that urine culture and antimicrobial sensitivity testing should be performed in order to treat suspected bacterial UTIs instead of empiric therapy or directing physicians for the final decisions of treatment due to variation in antibiotics resistance. Therefore, monitoring of antibiotic resistance must be continuous to decrease the development of resistance.

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