

RESEARCH ARTICLE

ISOLATION AND ANTIMICROBIAL SUSCEPTIBILITY PROFILES OF MICROORGANISMS CAUSING URINARY TRACT INFECTION AMONG PATIENTS IN ADEN CITY, YEMEN

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Abstract

Urinary tract infection is an infection that usually affects all people around the world, regardless of gender and ages. Therefore, the present study aimed to isolate microorganisms causing urinary tract infection and their antimicrobial susceptibility profiles among patients attending some private hospitals in Aden city, Yemen.

A total of 78 specimens of midstream urinary were collected from patients presenting in some private hospitals and centers located in Aden - Yemen, during the period from January to August 2021. Urine samples were examined macroscopically and microscopically. Also, the samples were cultured in different media and then incubated aerobically for 18-24 h at 37°C. The bacteria were isolated and diagnosed, and then the bacterial isolates were examined and their sensitivity to antibiotics, using the disc diffusion method (Kirby-Bauer).

Of the 78 samples, 57 (73.08%) were positive for the growth of pathogenic bacteria, 3 (3.85%) fungi, while 3 (3.85%) were negative and 15 samples were discarded for incomplete data. A higher rate of isolates was obtained from negative 33 (55%) compared to positive bacteria 24 (40%). The of infection in females was 48 (55%), while the number of males was 12 (20%). The highest infection recorded was *Streptococcus agalactiae* 12/57 (21%), followed by *Pseudomonas aeruginosa* 9 (15.7%), then *Enterococcus faecalis*, *Enterobacter aerogenes* and *Escherichia coli* each 6 (10.5%). The highest sensitivity of 94.7% was recorded by the antibiotics levofloxacin and ciprofloxacin. But the isolated Gram-negative bacteria showed a sensitivity of 100%. Gram-negative bacteria showed moderate sensitivity against tetracycline 58%, gentamicin 42% and ceftriaxone 45.5%. While lower sensitivity rates were observed for Co-Trimoxazole 31.6%, Cefotaxime 15.8%, and Ampicillin/Sulbactam 15.8%. Cephalixin, linezolid and cloxacillin have demonstrated complete resistance to the antibiotics used. There is a high prevalence of bacterial UTIs among women. Levofloxacin is the most effective antibiotic in treating UTIs, followed by amikacin, ofloxacin and ciprofloxacin. Ampicillin / Sulbactam, Co-trimoxazole, Cefotaxime, Tazobactam / Piperacillin. It is considered the most resistant antibiotic of bacteria isolated from urinary tract infection.

The results confirmed the resistance of the isolated urinary tract bacteria to some of the antibiotics used in our study. Accordingly, we recommend our doctors to take into consideration the developments in the use of antibiotics and resistance by bacteria.

Keywords: *Streptococcus agalactiae*, *Pseudomonas aeruginosa*, Urinary tract infection.

Introduction

Microorganisms cause urinary tract infection (UTI) is a common health problem throughout the world. UTI is the most common bacterial infections and account for a significant part of the workload in clinical microbiology laboratories [1]. About 150 million people are diagnosed with UTI each year in worldwide, costing the global economy in excess of 6 billion United States dollars [2].

UTI is defined also as the presence of both significant bacteriuria (10⁵ colony forming unit (cfu/ml) and urinary symptoms [3]. UTI is an extremely common clinical problem which may involve urethra, bladder, uterus and Kidney. Most UTI are caused by bacteria that can live in the digestive tract, the vagina or around the urethra. Infection occurs when bacteria enter the normally sterile urinary system and multiply there [4].

UTI are classified as uncomplicated or complicated [5]. Uncomplicated UTI are subdivided into cystitis and pyelonephritis. Patients with cystitis are presented with dysuria, frequency of micturition, suprapubic pain and may be hematuria [6]. While patients with pyelonephritis are presented with fever, chills, nausea, vomiting, in addition to the other severe symptoms [7]. Complicated UTI occur when there is abnormality in the urinary system or host immunity is suppressed. These patients are mainly predisposed to repeated UTI, renal failure, urea sepsis and death [6].

UTI occurs in all populations and ages [8]. UTI occur in women more commonly than men, this regard to several clinical factors such as, malnutrition factors, hormonal effects and anatomic differences, poor hygiene, low socioeconomic status are associated with UTI and these factors are rife in rural settings [9].

Majority of UTI are ascending infection and begin as cystitis, which results from bacteria entering the bladder through the urethra. If left untreated, these bacteria can spread to kidney causing pyelonephritis, which can result in renal damage. In severe cases, the bacteria can breach the epithelial and endothelial barriers in kidney and invade the bloodstream, leading to bacteremia and systemic sepsis [10].

In Yemen, the resistance to antibiotics becomes more complicated resulting from the availability of antibiotics as over-the-counter-drugs, excess uses, and misuses of antibiotics [11, 12, 13, 14]. Few studies that focused on the surveillance of antibiotic drugs resistance, non-evidence base prescriptions and unsatisfactory data is existed to measure the problem in Yemen [15, 16, 17].

Therefore, the present study aimed to isolate microorganisms causing urinary tract infection and their antimicrobial susceptibility profiles among patients attending some private hospitals in Aden city, Yemen.

Materials and Methods.

Study area and period

The present study was conducted at some private hospitals including Al-Buraihy Group International Hospital, Shifa Medical center, AL Madinah Medical Center, and Babel Model Hospital which located in Aden, Yemen during the period of January to August 2021.

Collection of urine samples

About 78 samples of midstream urine was collected from infected UTI patients into a 10-15 mL calibrated sterile screw-capped universal container which was initially distributed to the patients as has been described by [18]. Dysuria is central in the diagnosis of UTI; other symptoms may be variably present, infections attending to different hospitals in Aden City. The specimen was appropriately labeled, transported to the laboratory of Faculty of Science, University of Aden, and analyzed within 2 hours after obtaining.

Macroscopically examination

Urine samples were examined macroscopically for color, its smell, foam and turbidity; and also measurements of its pH, relative specific gravity and osmolality. Urine specimens whose colors showed amber, pale and deep yellow but clear were regarded as normal, while specimens that showed amber to deep yellow but cloudy, colorless and red/blood stained were regarded as abnormal [19]. Urine dipstick test as rapid detectors of bacteriuria and UTI was performed according to [20].

Microscopically examination:

Urine samples were centrifuged at 2000g for 5 minutes. After centrifugation, the supernatant was discarded and a drop or two of the sediment placed on the grease free slide, cover slip applied and examined under the microscope using the high-power field. Reporting system for microscopic identification is at high magnification for pus cells, red blood cells, epithelial cells, casts, crystals, and yeast cells as described by [21].

Isolation and Identification of bacteria

Using calibrated wire loop samples were inoculated in to different medium such as (Cysteine Lactose Electrolyte Deficient (CLED), MacConkey agar, Nutrient agar, DNase agar, Mueller Hinton agar, Nutrient broth, Triple sugar iron agar, Urea agar, Simmon's citrate agar, Mannitol salt agar, Kligler iron agar, Eosine methylene blue agar). After overnight incubation at 37°C for 24–48 hours colonies were counted to check significant growth. Colony counts yielding bacterial growth of 10⁵/ml of urine were regarded as significant for bacteriuria. The identification of isolated bacteria was primarily performed by colony morphology characterization, Gram staining, capsule formation, and selective media.

Also, biochemical reactions such as Catalase test, Coagulase test, Urease test, Triple sugar iron agar Indole, Methyl red, Voges Proskauer, and Citrate utilization was done according to Bergey’s manual of determinative bacteriology [22, 23].

Isolation and Identification of Candida sp.

The isolated Candida sp. was cultured on the surface of Sabouraud Dextrose Agar (SDA) with chloramphenicol (250mg/L). The plates were incubated for 48 h at 37°C. The morphological features for the colony were studied and confirmed by detecting the budding characterization with pseudo-hyphae [24].

The identification of Candida species was performed depending on morphological characterizations on a culture medium, formation of germ tube, and assimilation of carbohydrate test [25].

Antimicrobial susceptibility testing

The antimicrobial susceptibility testing was done on Mueller-Hinton agar (HI Media Pvt. Ltd., India) using disk diffusion (Kirby Bauer's) technique. This method was done according to Clinical and Laboratory Standards Institute (CLSI) guidelines to determine susceptibility of UTI agents [26].

About three to four well isolated colonies were emulsified in 3ml of sterile physiological saline; turbidity of the inoculum was compared with 0.5 McFarland standard and then swabbed on the surface of Muller Hinton Agar plates with sterile sticks. The antibiotics used in the study were belonging to four different groups of antibiotics: β-lactam-β-lactamase-inhibitor (AMC,

AML, and PRL), 5-fluoroquinolons (NOR, OF, and CP), Aminoglycosides (GM, and AK), and Cephalosporins (CTX, and CAZ). The UTI of Gram-negative and gram positive were distributed into different antibiotypes. Multidisc for gram positive isolates: Ampicillin/Sulbactam (AS; 20mcg), Co-Trimoxazole (BA; 25mcg), Cephalexin (PR; 30mcg), Tetracycline (TE; 30mcg), Cefotaxime (CF; 30mcg), Ciprofloxacin (CP; 5mcg), Levofloxacin (LE; 5mcg), Linezolid (LZ; 30mcg), Cloxacillin (CX; 5mcg), Roxithromycin (RF; 15mcg), Lincomycin (LM; 2mcg), Gentamicin (GM; 10 mcg). Multidisc for gram negative isolates: Ampicillin/Sulbactam (AS; 20mcg), Co-Trimoxazole (BA; 25mcg), Cefotaxime (CF; 30mcg), Tazobactam/ Piperacillin (TZP; 110 mcg), Chloramphenicol (CH; 30mcg), Ciprofloxacin (CP; 5mcg), Ceftriaxone (CR; 30mcg), Tetracycline (TE; 30mcg), Ofloxacin (OF; 5mcg), Levofloxacin (LE; 5mcg), Gentamicin (GM; 10 mcg), Amikacin (AK; 30 mcg), (HI Media Pvt. Ltd., India) [27].

Antimicrobial susceptibility testing of Candida sp., was done on Mueller-Hinton agar (HI Media Pvt. Ltd., India) using disk diffusion (Kirby Bauer's) technique. Candida sp., were cultivated on Mueller–Hinton Agar with 2% Glucose and 0.5 µg/mL Methylene Blue.

Results

Of 78 samples, 57/78 (73.08%) were positive for growth of pathogenic bacteria and fungi 3 (3.85%) while 3/76 (3.85%) were negative (Fig. 1). Five teen /78 samples were ignored because data were not completed.

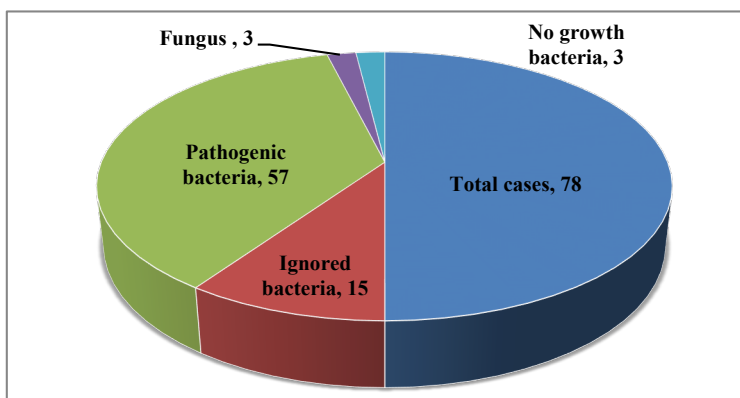


Fig 1: Urinary tract infections (UTI) distribution of pathogenic and non-uropathogenic bacteria.

Macroscopic examination:

Out of the 60 (except Candida case), samples examined, 57 (95%) had positive urine cultures with pathogenic bacteria. The urine colors observed in this study ranged from pale yellow, colorless to red (Tab. 1). The higher percentage 39(65 %) of urine samples were within the normal urine colors, while 21 (35%) had abnormal urine colors of all the samples. A 21 (35%) percentage of samples whose urine colour abnormal had positive UTI culture.

Table 1: Urine color tests in diagnosis UTI.

Urine color	UTI (%)		Total
	Positive	Negative	
Normal	39 (65%)	0	39 (65%)
Abnormal	21 (35%)	0	21 (35%)
Total	60 (100%)	0	60 (100%)

Clinical analysis of urine components:

The study shown an increase in the specific gravity 42 (73.68%). Bilirubin 9 (15.79 %). This study showed the presence of nitrite 9 (15.79 %). The ph value was ranging from 5 to 8.5; Alk.6 (10.53%), Neut. 6 (10.53%) and Acids 45 (78.95 %). Three sample (5.26%), show presence blood (RBC). And some samples show presence of protein 12 (20%), in normal situation. Other components showed normal value such as Glucose and Ketones, test strip results on (Tab. 2).

Table 2: A urine dipstick or test strip results

Chemical Propertied In Urine		Cases N=60
Nitrite		9 (15.79 %)
Specific Gravity		42 (73.68 %)
Protein		12 (21.05 %)
PH	Alk.	6 (10.53%)
	Neut.	6(10.53%)
	Acids	45 (78.95%)
Bilirubin		6 (10.53%)
Urobilinogen		3 (5.26%)
Blood		3 (5.26%)
Ketones		0
Glucose		0

Microscopic characteristics

The percentage of observed microscopically include epithelial cells, bacteria cells, crystals, yeast cells, pus cells and RBC (Tab. 3). The epithelial cells observed in 24 cases (40%), bacterial cells, pus cells and crystals all showed 9 (15%). The study showed 3 (5%) urine specimens which did not show any microscopic feature.

Table 3: Microscopic examination of UTI.

Microscopic results	Urine culture n=60	
	Positive (%)	Negative (%)
Bacterial cells	9 (15%)	0
Epithelial cells	24 (40%)	0
Yeast cells	3 (5%)	0
Pus cells	9 (15%)	0
RBC	6 (10%)	0
Crystals	9 (15%)	0
Total	60 (100%)	0

Among the 60 (100%) UTI isolates, 24 (40%) were Gram-positive, 33 (55%) were Gram-negative and 3 (5%) Candida spp. (Tab. 4). There were 48 (80%) females and 12 (20%) males in patients with urine positive culture.

Table 4: Frequency of isolated microorganisms according to gender.

Isolated bacteria	No (%)	Female(%)	Male (%)
Gram-negative	33 (55%)	24 (40%)	9 (15%)
Gram-positive	24 (40%)	21 (35%)	3 (5%)
Fungi	3 (5%)	3 (5%)	-
Total	60 (100%)	48 (80%)	12 (20%)

Frequency of isolated bacteria

Table 5 shows the frequency of isolated bacterial species that showed that were *Strep. agalactiae* with 12/57(21%), followed *P. aeruginosa* 9 (15.7%), *E. faecalis* 6 (10.5%), *E. aerogenes* 6 (10.5%), *E. coli* 9 (15.8%), *Staph. saprophyticus* 3 (5.3%), *P. mirabilis* 3(5.3 %), *Staph. epidermidis* 3(5.3 %), *K. pneumoniae* 3 (5.3%) and *K. oxytoca* 3(5.3%).

Table 5: Frequency of bacterial species isolated from urine specimens.

Bacteria	Frequency	Rate (%)
<i>Streptococcus agalactiae</i>	12	20
<i>Pseudomonas aeruginosa</i>	9	15
<i>Enterococcus faecalis</i>	6	10
<i>Enterobacter aerogenes</i>	6	10
<i>Escherichia coli</i>	6	10
<i>Staphylococcus saprophyticus</i>	3	5
<i>Proteus mirabilis</i>	3	5
<i>Proteus vulgaris</i>	3	5
<i>Staphylococcus epidermidis</i>	3	5
<i>Klebsiella pneumoniae</i>	3	5
<i>Klebsiella oxytoca</i>	3	5
<i>Candida spp.</i>	3	5
Total	60	100

Among males, 12/60 (20%) UTI cases were found (Tab. 6). of *Strepto. agalactiae* (6 female group *P. aeruginosa*, 6 male and female *E. faecalis*, 6 females *E. aerogenes*, 3 female *Staph. saprophyticus*, 3female *Staph. epidermidis*, 3 female *P. mirabilis*, 3 female *K. oxytoca* and 3 male *K. pneumoniae*).

Table 6: Frequency of isolated microorganisms of UTI in relation to gender.

Type of isolated	Male No. (%)	Female No. (%)	Total No. (%)
Gram positive bacteria			
<i>S. agalactiae</i>	3	9	12 (20%)
<i>E. aerogenes</i>	0	6	6 (10%)
<i>Staph. saprophyticus</i>	0	3	3(5%)
<i>Staph. epidermidis</i>	0	3	3 (5%)
Total	3	21	24
Gram negative bacteria			
<i>P. mirabilis</i>	0	3	3 (5%)
<i>P. vulgaris</i>	0	3	3 (5%)
<i>K. pneumonia</i>	3	0	3 (5%)
<i>K. oxytoca</i>	0	3	3 (5%)
<i>E. coli</i>	0	6	6 (10%)
<i>P. aeruginosa</i>	3	3	9 (15%)
<i>Enterococcus faecalis</i>	3	3	6 (10%)
Total	9	21	30
<i>Candida spp.</i> ,			
<i>Candida spp.</i>	0	3	3 (5%)
Total	0	3	3

Bacterial and fungal biochemical test:

Using a loop of well-mixed un-centrifuged urine was inoculated on blood and MacConkey agar. Then, bacterial colony **Table 7** was sub-cultured onto the other mentioned above media, were inoculated on biochemical tests. The carbohydrate assimilation tests for *C. albicans* are presented in table 7. It shows that the test was positive for Xylose, Sucrose and Glucose.

Table 7: Biochemical test of bacteria isolated from urine samples:

Test Type of bacteria	Gram Staining	Capsule	DNase	Urease test	Oxidase test	Motility test	Methyl red	Voges Proskauer	Hemolysis	Catalase test	Simmons citrate test	Glucose	Lactose	Sucrose	Fructose	Xylose	Mannitol
<i>P. aeruginosa</i>	-	-		-	+	+	-	-	-	+	+	-	-	-	+	-	+
<i>S. agalactiae</i>	+	+	ND	-	-	-	-	-	+	-	ND	+	+	+	+	-	-
<i>E. faecalis</i>	+	ND	-	-	-	-	ND	+	+	-	-	+	+	+	+	-	+
<i>P. mirabilis</i>	-	+	-	+	-	+	+	-	ND	+	+	+	+	-	ND	+	-
<i>P. vulgaris</i>	-	-		+	-	+	+	-	-	+	-	+	-	-	ND	ND	-
<i>E. coli</i>	-	+	-	-	-	+	+	-	-	+	-	+	+	-	-	+	+
<i>S. saprophyticus</i>	+	+	-	+	-	-	-	-	-	+	-	+	+	+	+	+	+
<i>K. pneumoniae</i>	-	+	-	+	-	-	-	+	ND	+	+	+	+	+	-	+	+
<i>E. aerogenes</i>	-	+	-	-	-	+	-	+		+	+	+	+	+	+	+	+
<i>S. epidermidis</i>	+	+	-	+	-	-	-	+	-	+	-	+	+	+	+	-	-
<i>k. oxytocin</i>	-	+	-	+	-	-	-	+	-	+	+	+	+	+	ND	+	+
<i>Candida albicans</i>	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	+	-	+	-	+	-

Fungus isolated from UT infection:

Among 60 UTI patients tested 5% three case positive for fungus infection, in this study period the patient was woman, *Candida albicans* was responsible for 5% of UTI. The same case was growth on blood agar. Symptoms such as fever, dysuria, and flank pain was absent in these patients.

Antibiotic susceptibility pattern of UTI isolates:

The isolated uropathogens were high sensitive to the various antibiotics. Among the isolated pathogens showed the highest degree of sensitivity 94.7% towards Levofloxacin and Ciprofloxacin, but showed 100% sensitivity for gram negative. Followed by Amikacin, Ofloxacin and Chloramphenicol which showed 100%, 91%, 81.8% sensitivity for gram negative isolates. Low

sensitivity rates were observed to Tetracycline 58%, Gentamicin 42% and Ceftriaxone 45,5% For gram negative. While low sensitivity rates were observed to Co-Trimoxazole 31.6%, Cefotaxime 15.8%, Ampicillin /Sulbactam 15.8%, Roxithromycin, and Lincomycin

which showed 25%, 12.5% sensitivity. For gram positive, Tazobactam/Piperacillin 9.1% for gram negative however the Cephalexin, Linezolid, Cloxacillin they showed complete resistance **table 8**.

Table 8: Antibiotic sensitivity test Kirby-Bauer method. (-) Resistance, (+) Low sensitive, (++) High sensitive.

Bacteria		Antibiotic																
		AS	BA	PR	TE	CF	CP	LE	LZ	OF	CX	RF	LM	GM	TZP	CH	CR	AK
<i>P. aeruginosa</i>	R/S	-	-		+	+	++	++		+				+	-	+	+	++
<i>E. faecalis</i>	R/S	-	+	-	+	-	++	++	-		-	++	+	+				
<i>P. mirabilis</i>	R/S	++	++		++	-	++	++		++				++	-	++	-	++
<i>P. vulgaris</i>	R/S	-	-		++	-	++	++		++				-	-	++	-	++
<i>E. coli</i>	R/S	-	-		-	-	++	++		++				++	-	+		++
<i>K. pneumoniae</i>	R/S	-	++		++	-	++	++		++				++	-	++	-	++
<i>k. oxytocolin</i>	R/S	-	-		-	-	++	++		++				-	-	++	-	++
<i>S. agalactiae</i>	R/S	-	+	-	++	+	++	++	-		-	+	-	+				
<i>E. aerogenes</i>	R/S	+	+		++	+	++	++		++				++	+	++	++	++
<i>S. saprophyticus</i>	R/S	-	-	-	-	-	++	++	-		-	-	-	-				
<i>S. epidermidis</i>	R/S	-	++	-	-	-	++	++	-		-	-	-	-				

Pseudomonas aeruginosa showed the highest rates of sensitivity to Amikacin, levofloxacin and Ciprofloxacin 100%, and the low sensitivity to Gentamicin and Cefotaxime (33.33%), the Ampicillin / Sulbactam, Co-Trimoxazole and TZP they showed complete resistance. *Escherichia coli* showed the highest rates of sensitivity to Ofloxacin, Gentamicin, Amikacin, Levofloxacin and Ciprofloxacin 100%, the Ampicillin/Sulbactam, Co-Trimoxazole, Cefotaxime, TZP and Tetracycline they showed complete resistance. *Enterococcus faecalis* showed high sensitivity for Ciprofloxacin, Levofloxacin and Roxithromycin 100%, and showed complete resistance to Ampicillin/ Sulbactam, Cephalexin, Cefotaxime, Linezolid and Cloxacillin. *Proteus mirabilis* showed sensitivity to Ampicillin/ Sulbactam, Co-Trimoxazole, Chloramphenicol, Ciprofloxacin, Tetracycline, Ofloxacin, Gentamicin, Amikacin and levofloxacin and showed resistance to Cefotaxime, Tazobactam/ Piperacillin and Ceftriaxone. *Proteus vulgaris* showed sensitivity to Chloramphenicol, Ciprofloxacin, Tetracycline, Ofloxacin, Amikacin and levofloxacin and showed resistance to Ampicillin/ Sulbactam, Co-Trimoxazole, Cefotaxime, Tazobactam/Piperacillin, Gentamicin and Ceftriaxone. *Klebsiella pneumonia* showed sensitivity to Co-Trimoxazole, Chloramphenicol, Ciprofloxacin, Tetracycline, Ofloxacin, Gentamicin, Amikacin and levofloxacin and showed resistance Ampicillin/ Sulbactam, Cefotaxime, Tazobactam / Piperacillin, and Ceftriaxone. *Klebsiella oxytocolin* showed sensitivity to Ciprofloxacin, Chloramphenicol, Ofloxacin, Amikacin and levofloxacin and showed resistance to Ampicillin/Sulbactam, Co-Trimoxazole, Cefotaxime

Tazobactam/Piperacillin, Ceftriaxone, Tetracycline and Gentamicin.

According to UTI bacterial species isolated, *Strepto. agalactiae* as the predominant cause of UTI showed the highest rates of sensitivity to Tetracycline, Ciprofloxacin and Levofloxacin 75%, the Ampicillin /Sulbactam, Cephalexin, Linezolid, Cloxacillin and Lincomycin the showed complete resistance. *Enterobacter aerogenes* showed high sensitivity to Chloramphenicol, Ciprofloxacin, Ceftriaxone, Tetracycline, Ofloxacin, Gentamicin, Amikacin and levofloxacin 100% and followed by Ampicillin/ Sulbactam, Co- Trimoxazole, Cefotaxime and Tazobactam /Piperacillin 50%. *Staph. saprophyticus* showed sensitivity to Ciprofloxacin and Levofloxacin and resistance to Ampicillin/Sulbactam, Co-Trimoxazole, Cephalexin, Tetracycline, Cefotaxime, Cloxacillin, Roxithromycin, Lincomycin, Gentamicin and Linezolid. *Staphylococcus epidermidis* showed sensitivity to Ciprofloxacin, Levofloxacin and Co Trimoxazole, and showed resistance to Ampicillin/Sulbactam, Cephalexin, Tetracycline, Cefotaxime, Cloxacillin, Roxithromycin, Lincomycin, Gentamicin and Linezolid.

Antifungal susceptibility pattern

Treatment was often based on a single culture without documenting the site of infection inside the urinary tract and without a follow-up urine culture performed. Fluconazole was the most frequent antifungal used followed by Voriconazole. The other antifungal antibiotic were resistance to the, Nystatin, Clotrimazole, Miconazole and Metronidazole.

Table 9: Antifungal susceptibility pattern of *C. albicans* isolates:

Fungi	NS	VRC	FLC	CC	MI0	MIC
Candida spp.	R	S	S+	R	R	R

Discussion

Health care associated infections are the leading cause of morbidity and mortality [28]. The high overall prevalence of UTI in the present study was 73.08%. This is similar to that find in Iraq 70% [29], but UTI was higher than other studies from Indian 49.19% [30], Libya 13.9% [31]. The reason for such variable results may due to the differences in the sample size and number, hospital site and season.

Among the relative prevalence of uropathogens isolated in this study, (55%) belonged to gram negative bacteria, (40%) belonged to gram positive bacteria, while (5%) belonged to fungi. Such findings were nearly come in agreement with result of [32], who found that the percentage of Gram negative bacteria, Gram positive constituted (34.14%), (26%), respectively. The gram negative bacteria were more prevalent (55%) in this study which is in agreement with previous studies [33]. Gram negative bacteria are the dominant bacteria that cause UTI [34]. This could be due to the presence of unique structure in gram negative bacteria (pilus adhesins and capsular polysaccharide) which help these bacteria for attachment to the uroepithelial cells and prevent bacteria from urinary lavage, allowing for multiplication and tissue invasion in invasive infection and pyelonephritis.

In laboratories used fresh, uncentrifuged urine dipstick as specimens. Many urinary pathogens will convert nitrates in the urine to nitrites. As such, the presence of nitrites in the urine is highly suggestive of infection, this study and the other study points to that [35]. Some pathogens do not induce this reaction. The leukocyte esterase, which produced by WBC, its presence in the urine raises the doubt of a UTI, but the test is often positive in the absence of a UTI. However, if this test negative, the chance is low of the patient having a UTI.

More girls (80%), than boys (20%), were having UTI. This is similar to that find in Hawassa, Southern Ethiopia were females are more affected than males 58.5% versus 41.5% [36], the same results were found in Yemen were females are more affected than males 40.3% versus 29% [17]. High prevalence of UTI among female may be due to lack of prostatic fluid, moist urethra or females have wider urethra. Previous researches has shown that female patients have much higher predisposition to UTI than males [37, 38], but this results was different from the study done in Nepal report high percentage of males with UTI 69.3% [39].

The distribution of UTI among patients' age groups, pointing to the higher incidence of UTI was seen in adults and elderly 80%, especially sexually actives, followed by then among children 20%. In contrast to the other research, UTI among elderly, adults and children were 58.7%, 36.2%, and 5.1%, respectively [40]. While, UTI was found in 16.55% of elderly women [41]. The differences results in the type and distribution of uropathogens show a discrepancy from area to another due to many factors as environmental conditions, health practices, patient conditions, personal hygiene, number of patients examined, and laboratory procedures. The possible causes of UTI in elderly may be attributed to many factors including urinary tract anomalies, urinary and fecal incontinence, malnutrition, functional disability, diabetes, problems in the immune system, prostate enlargement in males and post-menopausal hormonal changes in females [41].

The study showed some our isolates have a capsular polysaccharide such as (*K. oxytoca*, *E. aerogenes* and *Strepto. agalactiae*), this is similar to the find [42], most capsules function in microbial pathogenesis by protecting the microbe against host immune mechanisms, although for some the capsular structures can serve as adhesions.

In the present study, the most prevalent isolated bacterial uropathogens was *Strepto. agalactiae*, with an isolated rate of 20%, this finding is higher than the study of Nadia and Zainb [43], (7.75%). The rarity of UTI by *Strepto. agalactiae* is in contrast to the high frequency with which the organism colonizes the normal urethra, and come opposite with previous studies conducted in *E. coli* which was the most in Nepal 74% [44], Bangladesh 48.39% [45], but higher than our study (10%). The second predominant isolate were *P. aeruginosa* with overall isolated rate of 15%, which is similar with previous study in Rea-Raisin 16% [46]. While previous study done in Iran reported lower percentage of *P. aeruginosa* 4.9% [47], and in Yemen were 3.1% [32].

A study of Aamal [48], found that the percentage *Staphylococci spp.*, of positive culture of urine specimen collected from UTI among pregnant women in Al-Diwaniya city district only about (4.63%) that gave agreement with our result. In contrast, the prevalence of *Staph. saprophyticus* and *Staphy. epidermidis* (5%) in the current study is lower than the 16.8% reported by [49]. The prevalence of *E. aerogenes* was (10%) in this study, this close with the results of [32], whose showed that *E. aerogenes* was 7.8%, while Angoti *et al.*, [37], found that *E. aerogenes* was 29.61%. *Enterococcus faecalis*, 10% isolated which is lower than the findings of [45], (29.11%), and higher than the study of [37], (2.3%). The prevalence of *P. vulgaris* was (5%) in this study, this lower than the study of [39], whose showed that *P. vulgaris* was 17.9%, While the *P. mirabilis* 5.1%, that gave agreement with our study 5%. The prevalence of *k.*

oxytoca and *K. pneumoniae* (5%) in this study, is also comparable with the study reported by Richa *et al.*, [39], which was *K. oxytoca* (7.7%), and *K. pneumoniae* (2.6%). The similarities and differences in the type and distribution of UTI may result from geographical variability, host factors, and practices of people such as healthcare and education, socioeconomic and hygiene standards in different countries.

Regarding the distribution of UTI among patients' age groups, an evidence of higher prevalence of UTI showed in 19-50 years of adults. This finding is similar to study conducted by Seifu *et al.*, [38] and Enayat *et al.*, [49]. Prevalence difference has been also observed among various age groups. This difference suggests that age is one risk factor associated with UTI. The high incidence of UTI amongst the old age group could be due to genitourinary atrophy and vaginal prolapse after menopause in female which in turn increases the risk of bacteriuria by increasing vaginal pH and decreasing vaginal *Lactobacillus* thereby allowing gram-negative bacteria to grow and act as uropathogens. Because of wide spread of antibiotic, especially in developing countries, the resistance profile of microorganism to antimicrobial agents is increasing frequently Foxman, [50].

This study showed that the highest susceptibility rate (94.7%) for all uropathogens isolates were to Ciprofloxacin. This finding is comparable to other reports but relatively higher than the reported of [39, 51]. Amikacin was highly active (100%) for gram negative, this finding is similar to previous studies in the other countries [31, 37, 52]. However, this sensitive rate to amikacin is higher than 67.2% prevalence reported from South India [53]. Levofloxacin was highly active (100%) too, which contradicted with the study of [54]. The possible explanation for high level susceptible to Levofloxacin, Amikacin and Ciprofloxacin may be due to infrequent prescriptions, also they are not easily accessible. Thus, Levofloxacin, Amikacin and Ciprofloxacin could be considered as alternative options in the treatment of UTI.

The activity of Co-Trimoxazole against Gram negative isolates was (13.6%), this result is come negatively with that shown by Tsegay, [55] in Ethiopia, Co-Trimoxazole was reported to showed 58.4% activity, while Raya, [56], found that 67% of the isolates were resistant, and cefotaxime was active against (15.8%) of isolates. This result agreed with that of Raya, [56], found that 96% of the isolates were resistant, while Mohammed *et al.*, [31] who showed that high sensitivity of isolates to Cefotaxime in UTI. Cefotaxime against *E. coli* isolates was (0%), While Eshetie *et al.*, [57] found that the cefotaxime showed low resistance against 21.4% of the isolates.

Ampicillin /Sulbactam had low activity (15.8%) to Gram negative isolates in this study, this finding is similar to

some studies conducted in Yemen showed that Ampicillin was resistance 97% of Gram negative pathogens. Ampicillin/Sulbactam is often used to treat polymicrobial bacterial infections [58, 59, 60]. Its use, as well as the use of other β -lactam- β -lactamase inhibitor antibiotics, will likely increase in hospitals as a result of the high prevalence of antibiotic-resistant organisms. Although ampicillin/Sulbactam has traditionally had good activity against *E. coli* [61, 59].

Gentamicin was active against (42%) of isolates. Gentamicin was active against (42%) of isolates. Gentamicin was active against (42%) of isolates. This result come negative with that of [62] (80.6%), and [32] (50.3%), who reported that the isolates had higher sensitive to Gentamicin. Tetracycline was active against (58%) of isolates, will the study of Kahiry *et al.*, [32] who showed that low sensitivity of isolates to Tetracycline was 19.1% in Yemen, Mohammad, [31] who reported that the resistance to tetracycline was (71%).

Pseudomonas aeruginosa showed the highest rates of sensitivity to amikacin, levofloxacin and Ciprofloxacin 100%, followed by Chloramphenicol, Ceftriaxone, Tetracycline and Ofloxacin 66.7 %, and the low sensitivity to Gentamicin and Cefotaxime (33.33%). The Ampicillin /Sulbactam, Co-Trimoxazole and Tazobactam / Piperacillin they showed complete resistance, in critical care settings with a high rate of antibiotic use, colonization pressure and non-antibiotic exposures may be the crucial factors for *P. aeruginosa* acquisition, whereas Fluoroquinolones may actually decrease its likelihood. For the acquisition of strains resistant to Piperacillin/Tazobactam, Fluoroquinolones and multiple drugs, exposure to amikacin may be more relevant than previously recognized, Trigueros *et al.*, [63].

There are more than 100 species of *Candida* (yeast) in nature, but only a few species are recognized causes of disease in humans. *C. albicans* is the most common medically significant *Candida* species identified [64]. Antifungal drugs are effective in the treatment of *C. albicans*. Fluconazole appeared to be the best drug for the treatment of *C. albicans*. Our results in line with the study of [65] who points to the Azole antifungals group (fluconazole, voriconazole), are recommended as the first-line treatment in all guidelines. imidazoles, triazoles demonstrate greater target specificity, are more resistant to metabolic degradation, and have superior potency [66]. Azoles ordinarily have long half-lives at approximately 30 h, with the exception of voriconazole which has a half-life of 6 h [67].

Conclusion

Of the total 78 patients complains of UTI, the overall rate prevalence of UTI was 73.08%. The specimens diagnosed based on patient's clinical symptoms, presence of leukocytes, and bacteria in the urine. Among the causative agent of UTI Gram negative bacilli bacteria, Gram positive cocci bacteria and fungi wear constituted of (55%), (40%) and (5%), respectively. The predominant uropathogens isolated wear *Strepto. agalactiae* with 20%, followed by *P. aeruginosa*. The prevalence of UTI was high in female than male 80% and 20% respectively and high in age group 19_60 years of adults than other age group. The levofloxacin was the most effective antibiotic for treatment of UTI followed by Amikacin and Ofloxacin, Ciprofloxacin. Ampicillin/Sulbactam, Co-trimoxazole, Cefotaxime and Tazobactam/Piperacillin, wear the most antimicrobial agent to which the isolates developed resistance in UTI.

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عزل وتقييم حساسية المضادات الحيوية للكائنات الحية الدقيقة المسببة التهاب المسالك البولية بين المرضى بمدينة عدن، اليمن

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المُلخَص

عدوى المسالك البولية هي عدوى تصيب عادة جميع الناس في جميع أنحاء العالم، بغض النظر عن الجنس والأعمار. لذلك هدفت الدراسة الحالية إلى عزل الكائنات الحية الدقيقة المسببة لعدوى المسالك البولية وملاحح حساسيتها لمضادات الميكروبات بين المرضى الذين يترددون على بعض المستشفيات الخاصة في مدينة عدن، اليمن.

تم جمع 78 عينة من مجرى البول من المرضى الموجودين في بعض المستشفيات والمراكز الخاصة الموجودة في عدن - اليمن، خلال الفترة من يناير إلى أغسطس 2021. تم فحص عينات البول بالعين المجهرية والميكروسكوبية. كما تم استنبات العينات في أوساط مختلفة ثم حضنت هوائياً لمدة 18-24 ساعة عند 37 درجة مئوية. تم عزل وتشخيص البكتيريا ومن ثم فحص العزلات البكتيرية ومدى حساسيتها للمضادات الحيوية باستخدام طريقة الانتشار القرصي (كيريبي- باور).

من 78 عينة كانت 57 (73.08%) موجبة لنمو البكتيريا الممرضة و 3 (3.85%) فطريات بينما 3 (3.85%) كانت سلبية و 15 عينة تم استبعادها لعدم اكتمال البيانات. تم الحصول على معدل أعلى من العزلات سالبة الصبغة 33 (55%) مقارنة بالبكتيريا الموجبة الصبغة 24 (40%). وبلغت نسبة الإصابة عند الإناث 48 (55%) بينما كان عدد الذكور 12 (20%). كانت أعلى إصابة مسجلة هي *Streptococcus agalactiae* 57/12 (21%)، تليها *Pseudomonas aeruginosa* 9 (15.7%)، ثم *Enterococcus faecalis* و *Enterobacter aerogenes*، و *Escherichia coli* الكل من 6 (10.5%). سجلت أعلى حساسية 94.7% بالمضادات الحيوية الليفوفلوكساسين والسيبروفلوكساسين. لكن البكتيريا سالبة الجرام المعزولة أظهرت حساسية بنسبة 100%. أظهرت البكتيريا سالبة الجرام حساسية معتدلة ضد التتراسيكلين 58%، الجنتاميسين 42% و سيفترياكسون 45.5%. بينما لوحظ انخفاض معدلات الحساسية لكل من Co-Trimoxazole 31.6%، Cefotaxime 15.8%، Ampicillin / Sulbactam 15.8%. أظهر سيفالكسين ولينزوليد وكلوكساسيلين مقاومة كاملة للمضادات الحيوية المستخدمة. هناك انتشار كبير للعدوى البكتيرية في المسالك البولية بين النساء. الليفوفلوكساسين هو المضاد الحيوي الأكثر فعالية في علاج عدوى المسالك البولية، يليه أميكاسين، أوفلوكساسين وسيبروفلوكساسين. أميسيلين / سولباكتام، كو-تريموكسازول، سيفوتاكسيم، تازوباكتام / بيراسيلين. يعتبر المضادات الحيوية الأكثر مقاومة للبكتيريا المعزولة من عدوى المسالك البولية. أكدت النتائج مقاومة بكتيريا المسالك البولية المعزولة لبعض المضادات الحيوية المستخدمة في دراستنا. وبناءً على ذلك، نوصي أطبائنا بمراعاة التطورات في استخدام المضادات الحيوية ومقاومة البكتيريا للمضادات.

الكلمات المفتاحية: *Streptococcus agalactiae*، *Pseudomonas aeruginosa*، عدوى المسالك البولية.

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