Contents lists available at ScienceDirect

Asian Pacific Journal of Tropical Biomedicine

journal homepage:www.elsevier.com/locate/apjtb

Document heading

Prevalence of bacteriuria in Jeyaseharan Hospital of South India and their antibiogram

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ARTICLE INFO

Article history:
Received 15 July 2011
Received in revised form 7 August 2011
Accepted 24 August 2011
Available online 10 September 2011

Keywords:
Urinary tract infection
Bacteriuria
Antibiogram
Antibiotics
Organism

ABSTRACT

Objective: To investigate the prevalence of bacteriologically positive urinary tract infection (UTI) among people enrolled in the hospital during January 2010 to June 2010. **Methods:** In the study period, a total of 1 546 patients were screened for UTI and the antibiogram was studied. **Results:** A total of 744 patients were positive. It revealed that females were more vulnerable to bacteriuria than males. Organisms predominantly isolated were *Esherichia coli* followed by *Klebsiella* in both males and females. Among the antibiotics tested against the isolated organisms for sensitivity test, chloramphenicol was more effective followed by amikacin and gatifloxacin for Enterobacteriaceae. For *Pseudomonas aeruginosa*, chloramphenicol was not effective, amikacin was effective. Amoxycillin, amoxycillin/clavulanic acid and first generation cephalosporin were least effective against Enterobacteriaceae and *Pseudomonas aeruginosa*. **Conclusions:** This study would not only help in proper treatment of patients but also discourage the indiscriminate use of antibiotics and prevent development of drug resistance.

1. Introduction

Urinary Tract Infection (UTI) is a serious health problem affecting millions of people each year. Infection of the urinary tract is the second most common infection in the body[1]. UTI causes about 8.3 million people visiting doctors each year. Women are especially prone to UTI for reasons that are not yet well understood[2]. UTI in men is not as common as in women but can be very serious when it occurs[2]. Normally, urine is sterile. It is usually free of bacteria, viruses, and fungi but does contain fluids, salts and waste products. An infection occurs when tiny organisms, usually bacteria from the digestive tract, cling to the opening of the urethra and begin to multiply. The urethra is the tube carrying urine from the bladder to outside the body. Most infections arise from one type of bacteria, *Escherichia coli* (*E. coli*), which normally live in the colon[3]. In many cases, bacteria first travel to the urethra. When bacteria multiply, an infection can occur. An infection limited to the urethra is called urethritis. Bacteria moving to the bladder and multiplying result in a bladder infection, called cystitis. If the infection is not treated promptly, bacteria may then travel further up to the ureters to multiply and infect the kidneys. A kidney infection is called pyelonephritis. Microorganisms called *Chlamydia* and *Mycoplasma* may also cause UTIs in both men and women, but these infections tend to remain limited to the urethra and reproductive system. Unlike *E.coli*, *Chlamydia* and *Mycoplasma* may be sexually transmitted and infection requires treatment of both partners[4].

Not everyone with a UTI has symptoms, but most people show at least some symptoms. These may include a frequent urge to urinate and a painful, burning feeling in the area of the bladder or urethra during urination. It is not unusual to feel tired, shaky, washed out and painful even when not urinating. Often women feel uncomfortable pressure above the pubic bone, and some men experience a fullness in the rectum. The common complain is that despite the urge to urinate, only a small amount of urine is passed. The urine itself may look milky or cloudy, even reddish if blood is present. Normally, a UTI does not cause fever if it is in the bladder or urethra. A fever may mean that the infection has reached the kidneys. Other symptoms of a kidney infection include pain in the back or side below the ribs, nausea, or vomiting. In children, symptoms of a urinary infection may be overlooked or attributed to another disorder. A UTI

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should be considered when a child or infant seems irritable, eats abnormally, has unexplained fever that does not go away, has incontinence or loose bowels, or is not thriving. Unlike adults, children are more likely to have fever without other symptoms[5].

UTIs are treated with antibacterial drugs. The choice of drug and course of treatment depend on the patient's history and the urine tests that identify the offending bacteria. The sensitivity test is especially useful in selecting the most effective drug. The drugs most often used to treat routine, uncomplicated UTIs are trimethoprim (Trimpex), trimethoprim/sulfamethoxazole (Bactrim, Septra, Cotrim), amoxicillin (Amoxil, Trimox, Wymox), nitrofuantoin (Macrodantin, Furadantin) and ampicillin (Omnipen, Polycillin, Principen, Totacillin). A class of drugs called quinolones includes four drugs approved in recent years for treating UTI[6]. These drugs include ofloxacin (Floxin), norfloxacin (Noroxin), ciprofloxacin (Cipro), and trovafloxin (Trovan). Often, a UTI can be cured with 1 or 2 days of treatment if the infection is not complicated by an obstruction or other disorder. Still, many doctors ask their patients to take antibiotics for a week or two to ensure that the infection has been cured. Single-dose treatment is not recommended for some patients, for example, those who have delayed treatment or have signs of a kidney infection, patients with diabetes or structural abnormalities, or men who have prostate infections. Longer treatment is also needed for patients with infections caused by *Mycoplasma* or *Chlamydia*, which are usually treated with tetracycline, trimethoprim/sulfamethoxazole (TMP/SMZ), or doxycycline. Kidney infections generally require several weeks of antibiotic treatment. Researchers at the University of Washington found that 2-week therapy with TMP/SMZ was effective as 6 weeks of treatment with the same drug for women with kidney infection that did not involve an obstruction or nervous system disorder. In such cases, kidney infections rarely lead to kidney damage or kidney failure unless untreated. Various drugs are available to relieve the pain of a UTI. The difficulty is increased by the variations in sensitivity patterns of different population. The degree of exposure of a population to specific antibiotic could play a role in this variation[7]. UTIs pose a serious health threat with respect to antibiotic resistance and high recurrence rates.

Microorganisms responsible for UTI such as *E.coli* and *Klebsiella* spp. have the ability to produce Extended-Spectrum β-lactamase (ESBLs) in large quantities. These enzymes are plasmid borne and confer multiple drug resistance, making urinary tract infection difficult to treat. *E. coli* is the most frequent urinary pathogen isolated from 50%-90% of all uncomplicated urinary tract infections. Antibiotics are usually given empirically before the laboratory results of urine culture are available. To ensure appropriate therapy, current knowledge of the organism that cause UTI and their antibiotic susceptibility should be educated. Much of the data is available for community acquired infections. This may be different from that of hospital acquired infections. Since patterns of antibiotic resistance in a wide variety of pathogenic organisms may vary even over short periods and depend on site of isolation and on different environments, periodic evaluation of antibacterial activity is needed to update this information[8].

This study was carried out on hospitalized patients with UTI and those attending the outpatient department with UTI. Clinical laboratory records of UTI cases were studied for the spectrum of bacterial isolates and their antibiotic susceptibility results were analyzed during January 2010 to June 2010.

2. Materials and methods

2.1. Sample collections

Specimen "Clean-catch", midstream urine were collected in sterile wide mouthed, screw-capped bottle after very thorough preliminary cleansing of external genitalia with soap and water. The number of organisms was estimated by calibrated loop method to evaluate the clinical significance of a "positive" urine culture.

2.2. Inoculation by calibrated loop method

A 4 mm platinum loop which delivers 0.01 mL is used. MacConkey ' s agar was inoculated by quickly transferring one loopful of the diluted or undiluted well-mixed, uncentrifuged urine specimen, and then touching the loop to 3 or 4 places on the MacConkey ' s agar. The blood agar was inoculated, by 4 area-streaking, from one area to the next, and not going over previously streaked areas. This should give adequate isolation of colonies in the fourth area of streaking.After overnight incubation of all plates at 37℃, the number of each colony was counted, and the number of viable bacteria present in 1.0 mL undilution urine was also calculated. The number of colonies was multiplied by 100 if undiluted urine was used in inoculation; by 1 000 if a 1:10 dilution of urine was used; by 10 000 if a 1:100 dilution was used. The significance of a positive urine culture is most reliably assessed in terms of the number of viable bacteria present in the urine.

2.3. Culture techniques

Aseptically collected samples were plated on suitable culture medium like blood agar and MacConkey´s agar. Blood agar serves as an enriched medium and a differential medium for haemolytic organisms. It is useful to differentiate various strains of *Streptococci*. MacConkey´s agar is a selective as well as differential media. It is used for the detection of coliforms and pathogenic species of *Enterobacteriaceae*. Bile salt mixture inhibits gram positive organisms. Lactose fermenting coliforms produce red colonies and non lactose fermentors produce colourless colonies on the medium.

2.4. Identification of organisms

Bacterial isolates were further identified by staining

techniques and conventional biochemical tests like indole test, methyl red test, citrate utilization test, triple sugar-iron agar and mannitol motility test. Species were identified based on Bergy ' s Manual of Systemic Bacteriology.

2.5. Susceptibility test by Kirby Bauer method

Microbial susceptibility was tested by the disc diffusion method of Kirby Bauer. Antibiotic impregnated discs release antibiotic into the surrounding medium when placed on the surface of solid agar (Muller Hinton agar) containing uniformly inoculated and actively growing organism. A clear zone formation around the antibiotic disc, which inhibits the growth of the organism, indicates the susceptibility of the organism.

3. Results

In this study, a total of 1 546 patients were screened for UTI, out of which 744 patients were positive. The incidence of UTI was 48.1%. Out of 744 positive patients 675 sample showed more than 100 000 colonies per mL, 57 samples showed colonies between 50 000 and 100 000 per mL and 12 sample showed colonies between 50 000 per mL. The various conventional bio-chemical tests were done to identify the causative organisms which were presented in Table1. Staining reaction, motility and growth characteristics of isolated bacterial species were observed on media like blood agar and MacConkey ' s agar. It showed 10 bacterial species were identified based on the Bergy ' s Manual of Systemic Bacteriology. The percentage contributions of each uropathogen were presented in Figure 1. A total of 43% males were recorded UTI positive against 58.1% females. It signifies that females were more prone to UTI than males. The incidence of *E.coli* infection among male and female were 72.1% and 82.7% respectively. The result revealed that incidence of infection due to *E.coli* was higher in female. The incidence of infection due to *Klebsiella sps*, *Citrobacter diverses*, *Enterobacter aerogenies*, *Proteus mirabilis*, *Enterococcus faecalis* were higher in females than males. *Pseudomonas aeruginosa* showed the higher infection in male than female (Figure 2). The results were grown in Table 2. The isolated 10 bacterial species from all samples were screened with different appropriate commercially available antibiotic disc. The percentage sensitivity of different antibiotics with most commonly isolated bacterial were shown in Table 2. This survey showed that chloramphenicol was more effective on the prominently isolated organism *E.coli* and *Klebsiella sp.* followed by amikacin and gatifloxacin. It was found that amikacin was more effective on *Pseudomonas aeruginosa* comparing with their antibiotics. Majority of the antibiotics tested for *Pseudomonas aeruginosa* were found less effective.

Table 1
Biochemical tests results of bacterial isolates.

Organisms	Motility	Manital	Trible sugar Iron Agar	Indole	Citrate	Urease	Mehtyl red	Voges proskauer Test
Escherichia coli	+	+	++gas	+	−	−	+	−
Klebsiella species	−	+	++ gas	−	+	NT	−	+
Pseudomonas aeruginosa	+	−	−−	−	+	NT	NT	NT
Citrobacter diverses	+	+	−+	+	+	NT	NT	NT
Enterobacter aerogenes	+	+	++	−	+	−	−	+
Proteus mirabilis	+	−	−+H2S gas	−	NT	+	NT	NT
Proteus morgani	+	−	−+ gas	+	−	+	NT	NT
Citrobacter freundi	+	+	−+ H2S	−	+	−	NT	NT
Enterococcus faecalis	−	NT	NT	NT	NT	NT	NT	NT
Beta haemolytic streptococci	−	NT	NT	NT	NT	NT	NT	NT

Table 2
Percentage of sensitivity of most commonly isolated organisms with commercial antibiotic discs (January 2010 to June 2010).

S.No.	Organisms	Sensitivity	Percentage sensitivity															
			Amoxycillin 10 mcg	Amoxycillin / lavulanic acid 30 mcg	Chloramphenicol 30 mcg	Cephalexin 30 mcg	Cefotaxime 30 mcg	Cefuroxime 30 mcg	Co-trimoxazole 25 mcg	Gentamicin 10 mcg	Amikacin 30 mcg	Norfloxacin 10 mcg	Ciprofloxacin 5 mcg	Ofloxacin 5 mcg	Gatifloxacin 5 mcg	Nitrofurantoin 300 mcg	Penicillin 10 units	Erythromycin 15 mcg
1	*Escherichia coli*	S	7.0	9	78	25	41	33	37	45	72	37	32	36	64	30		
		MS	2	4	4	12	33	8	1	3	12	2	6	3	16	33		
		R	91	87	18	63	56	59	62	52	16	61	62	61	20	37		
2	*Klebsiella species*	S	6	7	72	33	44	41	46	51	72	50	42	53	69	18		
		MS	7	10	4	10	4	4	3	5	10	3	6	–	10	17		
		R	87	83	24	57	52	55	51	44	18	47	52	47	21	65		
3	*Pseudomonas aeruginosa*	S	–	–	6	–	3	–	–	16	39	16	16	19	29	–		
		MS	–	–	6	–	16	–	3	–	3	6	13	3	13	–		
		R	100	100	88	100	81	100	97	84	58	78	71	78	58	100		
4	*Citrobacter diverses*	S	–	–	64	–	29	7	43	29	43	43	36	43	71	–		
		MS	–	7	–	7	21	7	–	7	14	7	7	21	14	–		
		R	100	94	36	94	50	86	57	64	43	50	57	36	14	100		
5	*Enterobacter aerogenes*	S	7	–	50	7	43	14	43	50	57	64	57	57	57	–		
		MS	7	7	7	21	14	43	–	–	7	–	7	–	14	14		
		R	86	93	43	72	43	43	57	50	36	36	36	43	29	86		
6	*Proteus mirabilis*	S	20	20	40	20	30	30	20	40	30	50	30	40	50	–		
		MS	–	–	20	10	–	10	–	–	20	10	10	–	10	–		
		R	80	80	40	70	70	60	80	60	50	40	60	60	40	10		
7	*Enterococci faecalis*	S	100	100	40	10	50	50	20	–	–	–	–	–	20	–	20	10
		MS	–	–	20	30	10	10	–	–	–	10	10	20	30	20	20	20
		R	–	–	40	60	40	40	80	100	100	90	90	80	50	80	60	70

S- Sensitive; MS - Moderate Sensitive; R - Resistant.

Amoxycillin and amoxycillin /clavulanic acid were found to be more effective on *Enterococcus faecalis*.

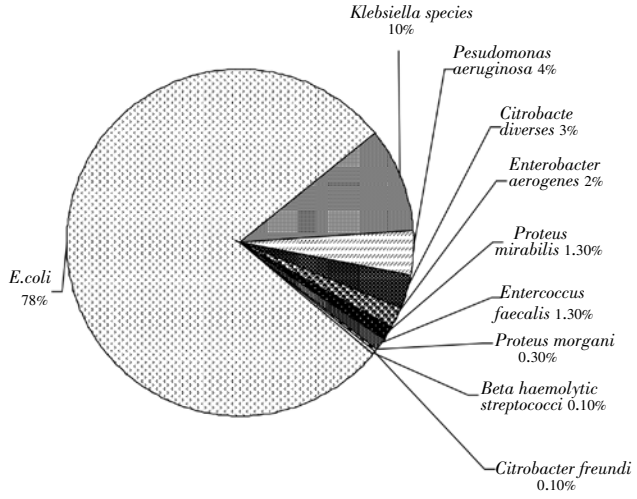

Figure 1. Percentage incident of bacterial species among total isolates.

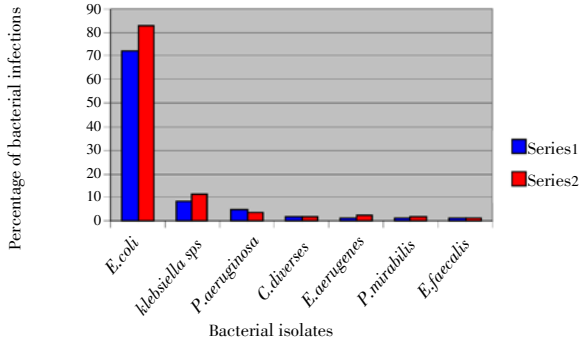

Figure 2. Percentage incidents of various UTI pathogens among male and female.

4. Discussion

This study clearly indicates that females are more prone to UTI. The incident of *E.coli* infection of male and female are 72.1% and 82.7%, respectively. Infection due to *Klebsiella sp*, *Citrobacter diverses*, *Enterobacter aerogenies*, *Proteus mirabilis*, *Enterococcus faecalis* showed the higher percentage in females than males. Gupta *et al* investigate uropathogenic strains from Inpatient and Outpatient Departments of the Government Medical College and Hospital, Chandigarh[9]. It showed that *E. coli* (64%) predominated amongst the indoor as well as outdoor patients. *K. pneumoniae* (24.8%) was the second commonest in the indoor patient group, followed by *P. aeruginosa* (10%), *Acinetobacter* and *Enterococcus faecalis* (6.4%). Gupta *et al* supports the results of this study, in which *E. coli* (78%) predominated the presence of *Klebsiella species* (10%), *P. aeruginosa* (4%), *Enterobacter aerogenes* (2%) at Jeyasekaran Hospital[9]. In another study by Bajaj *et al* at the Department of Microbiology, Government Medical College, *Aurangabad*, *Klebsiella sp* (84.7) was predominant followed by *E.coli* (78.1%) and *Pseudomonas sp* (71.9%) which was contradictory to the present study[10]. Yengkokpam *et al* (2007) in the Department of Microbiology, Regional Institute of Medical Sciencnes (RIMS), Impahal, Manipur, India, reported that *E.coli* was the predominant (72.8%) followed by *Klebsiella sp* (14.4%) and *Pseudomonas species* (3.5%)[11]. Based on the antibiotic sensitivity test, chloramphenicol is more effective against the prominently isolated organism *E.coli* and *Klebsiella sp*. This is identical to the study by Bajaj *et al*[10]. The sensitivity pattern of different UTI bacterial isolates showed high percentage of resistant towards most commonly used antibiotics. It clearly indicates that multi drug resistance is quite common to all the infectious pathogens recently. In view of the emerging drug resistance amongst bacteria, therapy should only be advocated, so far as possible, after culture and sensitivity has been performed. This would not only help in proper treatment of patients but would also discourage the indiscriminate use of antibiotics and prevent development of drug resistance.

Conflict of interest statement

We declare that we have no conflict of interest.

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