ISOLATION AND CHARACTERIZATION OF URINARY PATHOGENS FROM CATHETERIZED PATIENTS IN A TERTIARY CARE HOSPITAL

AMNA BUTT, SANIA BUTT, HASSAN SHOAIB*, HAMID MUKHTAR
[1]*Lecturer Biology, Aspire College Lahore.* [2]*WMO, DHQ Sheikhupura.* [3]*PGR/MO Medicine DHQ Sheikhupura.*
[4]*HOD, Department of Industrial Biotechnology GCU Lahore*
For Correspondence: Dr. Hassan Shoaib, H. No 429 - Gulshan Block, Allama Iqbal Town Lahore. Mob # 0334 4734459*

ABSTRACT
Urinary tract infection (UTI) is the most common nosocomial infection. Indwelling catheters are found to be strongly associated with UTIs. In present study a total of 110 catheterized patients from general wards and ICU of Mayo Hospital Lahore were investigated for their microbial evaluation. Out of 110 urine samples 80 cultured positive of which 25 developed UTI symptoms while 55 were not having UTI symptoms implying 23% acquired catheter associated urinary tract infection (CAUTI) while 50% developed asymptomatic bacterial colonization. Among the isolates 49% *Escherichia coli*, 14% *Psuedomonas aeruginosa*, 7% *Staphylococcus aureus*, 7% *Klebsiella pneumoniae*, 10% *Proteus* species, 5% *Enterobacter aerogens* and 8% fungi and yeast were recovered. Out of 80 patients who were cultured positive 52 were females revealing high susceptibility of females towards acquiring UTI. 74% of patients were married showing higher rate in married. Prevalence of CAUTI was low in this hospital while asymptomatic colonization was 50%, duration of catheterization had strong influence on occurrence of infection as 36% with UTI were catheterized from 3 weeks and only 6% for 1-3 days. Socioeconomic status was not found to have major influence. Patients who needed to be catheterized for longer duration should be checked regularly for UTI symptoms to prevent complications that are life threatening and are difficult to treat.

Key words: UTI, Nosocomial, Catheter associated UTI.

INTRODUCTION
UTI is a condition in which one or more parts of the urinary system (kidneys, ureters, bladder, and urethra) get infected. UTIs are the most common of all infections and affect all age groups. Most cases are caused by bacteria involving lower urinary tract.[1] The greatest risk factor for UTI is catheterization others include kidney stones, menopause, dehydration etc. Catheters were originally designed for short term use but now indwelling catheters are being used in long term settings increasing incidence of UTI.[2] Catheterized patients are more susceptible to UTI with duration of catheterization increasing the risk significantly.[2,3] CAUTIs are the most common type of nosocomial infections accounting for over 1 million cases annually and constituting 80% of all nosocomial UTIs.[4] Timely intervention should be done otherwise it can lead to complications including cystitis, prostatitis, epididymitis, acute or chronic pyelonephritis, bacteremia. Left untreated may lead to urosepsis and death.[5] Isolation and identification of pathogen aids treatment since infectious diseases caused by different pathogen have a variety of clinical courses and consequences. Susceptibility testing of isolates can help in selection of antibiotics for therapy. Recognizing that certain species are being isolated atypically may suggest that a disease outbreak has occurred e.g. from contaminated hospital supplies or poor aseptic technique on the part of hospital personnel.[6] In our country less work has been done on the problems that arise from catheterization. Keeping in view the existing problems of bacterial invasion after catheterization, this study has been designed to investigate the pathogens involved and the impact of various risk factors in CAUTIs.

METHODS
Cross sectional study was designed including a population of 110 patients admitted in medical and surgical wards and ICU of Mayo Hospital Lahore who did not have history of sub acute or chronic UTI prior to admission. Urine samples of those patients were taken by disinfecting the wall of catheter at its juncture with the drainage tube, and urine was aspired by a sterile

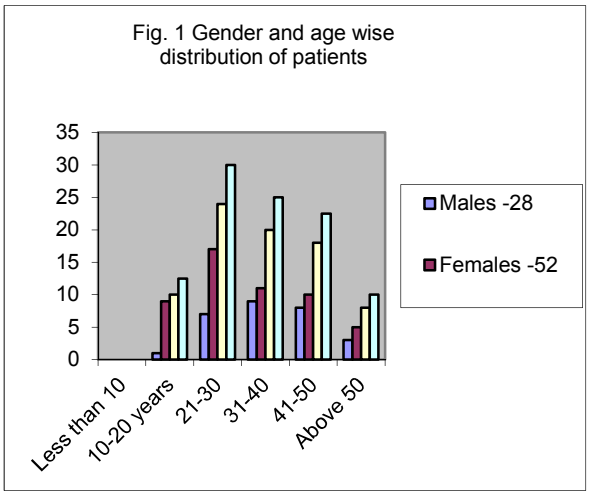
disposable syringe, and labeled according to the sample number. All the samples were taken to the laboratory and processed within 1h of collection under strict aseptic conditions during their handling and processing. Primary Bacterial isolation, from all specimens was attempted on various types of agar media. The samples were streaked on solid media. The inoculated media were incubated at 35-37C for 24 h and examined for evidence of media, which contained mixed culture, first stabs and slants of different media were prepared. Then single colony on the streak tract showing the evidence of pure growth was taken with the help of inoculating loop and subcultured on the appropriate selective, enrichment or differential media and incubated at 37C for 24 h. The growth was examined for purity and processed for identification. Pure cultures of the isolates were obtained on selective, differential and special media for studying their colony characteristics and hemolytic properties. The gross colonial morphology was assessed by observing the form, elevation and margins of the colonies. Gram differential stain was used for dying the bacteria from the cultures growths or specimens. The organisms retaining the color of principal dye and remaining blue or purple in color, after decolorization were considered as gram positive and those decolorized by alcohol and taking up pink or red colors (counter stain) were considered as Gram negative. The specimens were negatively stained using India ink to check the fungal growth. For identification of different bacteria, selective and differential media were used. Pure bacterial cultures were grown on nutrient agar slants and were used for subsequent biochemical tests.

RESULTS

During the present study attempts were made to isolate and identify species of different pathogens from the patients suffering from CAUTI at the Mayo hospital, Lahore, Pakistan. A total of 110 urine samples of catheterized patients from medical and surgical wards of Mayo hospital were collected and processed for microbiological analysis. Out of 110 urine samples 80 samples gave positive culture report so they were considered in the research investigation.

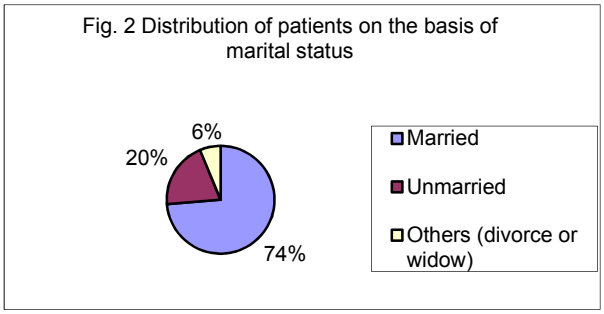
GENDER AND AGE WISE DISTRIBUTION OF PATIENTS

Out of 80 urine samples, 28 (35%) samples were taken from males and 52 (65%) samples were taken from the females. The patients belong to different age groups; among them no one was below 10 years. A total of 10 (12.5%) patients belonged to the age group 10-20 year, 24 (30%) belonged to 21-30 year, 20 (25%) to 31-40 year, 18 (22.5%) to 41-50 year and 8 (10%) were above 51 years of age (Fig- 1).



MARITAL STATUS OF PATIENTS

Among the investigated patients, 59 (74%) were married, 16 (20%) were unmarried and 5 (6%) were widows (3) or divorced (2) (Fig- 2).



SOCIOECONOMIC STATUS OF PATIENTS

From a total of 80 infected cases, 35 (44%) patients were belonging to the labor class, 20 (25%) patients were businessmen and 25 (31%) patients were Government employees (Fig- 3). Among these patients, 25 (31%) had monthly income less than 5,000 rupees, 34 (43%) patients had monthly income between 5,000-10,000 rupees, 17 (21%) patients were earning between 11,000-20,000 rupees monthly and 4 (5%) patients were earning more than 20,000 rupees monthly (Fig- 4).

REASONS OF CATHETERIZATION

Reason of catheterization was trauma in 14 (18%) patients, major surgery in 32 (39%) patients, renal failure in 19 (24%) patients and 15 (19%) patients were catheterized because of multiple reasons as dibetes, hypertension and paralysis (Fig- 5).

Fig. 3 Distribution of patients on the basis of occupation

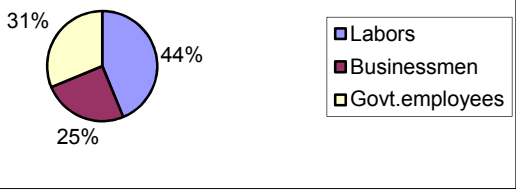


Fig. 4 Distribution of patients on the basis of monthly income

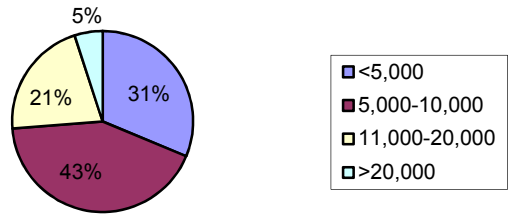
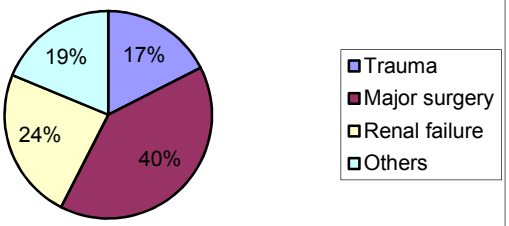


Fig. 5 Reasons of catheterization of patients



TYPE OF CATHETER USED

Depending upon type of catheter used, patients were divided in to two categories. 78 (97%) patients were catheterized via ordinary catheter and only 2 (3%) patients were catheterized via silicon catheter (Fig- 6).

DURATION OF CATHETERIZATION

Among a total of 80 patients, 5 (6%) patients were on catheterization from 1-3 days, 11 (14%) patients were on catheterization from one week, 17 (21%) patients were on catheter from two weeks, 29 (36%) patients were on catheter from three weeks and 18 (23%) patients were on catheterization from a month or more (Fig- 7).

Fig. 6 Type of catheter used in patients

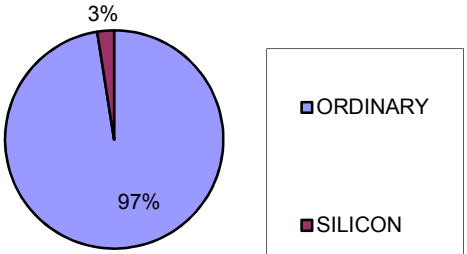
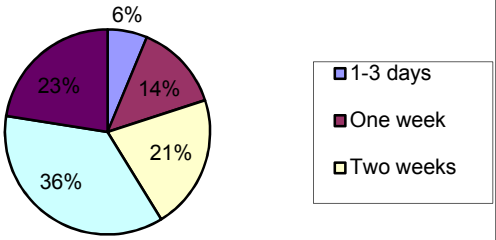


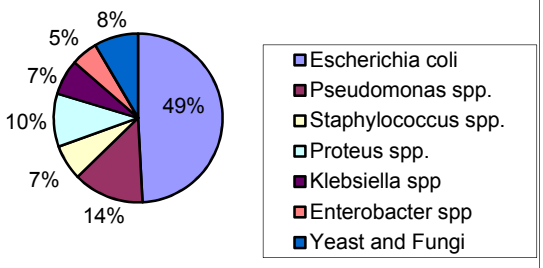
Fig. 7 Duration of catheterization



BACTERIOLOGICAL STUDIES

On bacteriological examination of 80 samples, 58 (49%) isolates of *Escherichia coli*, 16 (14%) isolates of *Pseudomonas* spp, 8 (7%) *Staphylococcus* spp, 12 (10%) isolates of *Proteus* spp, 8 (7%) isolates of *Klebsiella*, 6 (5%) isolates of *Enterobacter* and 10 (8%) isolates of yeasts and fungi were recovered and identified (Fig- 8).

Fig. 8 Types and no. of isolates in urine samples



DISCUSSION

The prevalence of CAUTI found in this setting is 25% and asymptomatic bacterial colonization is 50%. This finding is supported by investigation of Richards, *et. al.,* (1998)[6], Sedor and Mulholland (1999)[7], Tambyah and Maki (2000)[8], Danchaivijitr, *et. al.,* (2005)[9] and Esposito, *et. al.,* (2008)[10]. Wang *et. al.,* (1997)[4] reported that in USA CAUTI is the most common type of nosocomial infection accounting for over 1 million cases annually and over 40% of all nosocomial infections in hospitals and nursing homes and constitute 80% of all nosocomial UTIs. Out of 80 positive culture sample 28 (35%) were taken from males and 52 (65%) were taken from females (Fig-1). It indicates that percentage of CAUTI is more in females than in males. This finding is supported by the investigation of Stamm (1991)[11] who reported that females are more prone to urinary tract infections than males.

Among the patients 74% were married and 20% were unmarried so the rate of infection is higher in married patients (Fig-2). There was not enough previous literature to support these findings but it may be due to their involvement in sexual activity. Socioeconomic status of patients was not found to have significant effect on occurrence of CAUTI (Fig-3, 4). The income chart of the patients shows that 31% of patients were earning less than 5,000, 43% patients were earning between 5,000-10,000, 21% patients were earning between 10,000-20,000 and only 5% of patients were earning more than 20,000 (Fig-4). These findings might seem to have an impact but taking into account that the study was conducted in Government Hospital, where a high percentage of patients belong to lower middle class so these findings don't give a sound relation with the infection rate.

Reason of catheterization was found to be trauma in 14 (18%) patients, major surgery in 32 (39%) patients, renal failure in 19 (24%) patients and 15 (19%) patients were catheterized because of multiple reasons as diabetes, hypertension and paralysis (Fig-5). These findings are supported by investigations of Zhan, *et. al.,* (2009)[12] who reported that the major surgery is responsible for high rates of CAUTI. Because patient is in critical condition with low immunity so he is more prone to infections. Erben, *et. al.,* (2009)[13] reported that renal failure, unconsciousness, multiple trauma, surgery, central vascular catheter, tracheostomy, mechanical ventilation and peritoneal dialysis were observed more frequently in patients who developed the CAUTI, which also resembles with our findings. Wald, *et. al.,* (2008)[14] reported that the likelihood of having an indwelling urinary catheter at the time of discharge after major surgery is strongly related with occurrence of CAUTI as indicated in present study.

Depending upon type of catheter used patients were divided into two categories. 78 (97%) patients were catheterized via ordinary catheter and only 2 (3%) patients were catheterized via silicon catheter (Fig-6). As ordinary catheters are used mostly in Government Hospitals of Pakistan so there was not sufficient data for analyzing the effect of type of catheter used in relation to infection. Although there is significant findings of researchers on the type of catheter used. Brosnahan, *et. al.,* (2004)[15] reported that the use of silver alloy indwelling catheters for catheterizing hospitalized adults short-term reduces the risk of CAUTI. Catheters coated with a combination of minocycline and rifampin may also be beneficial in reducing bacteriuria in catheterized patients but further testing is required. There was not enough evidence to suggest whether or not any standard catheter was better than another in terms of reducing the risk of urinary tract infection in hospitalized adults catheterized short-term.

Among a total of 80 patients 5 (6%) patients were on catheterization from 1-3 days, 11 (14%) patients were on catheterization from one week, 17 (21%) patients were on catheter from two weeks, 29 (36%) patients were on catheter from three weeks and 18 (23%) patients were on catheterization from a month or more (Fig-7). These findings are supported by investigation of Pigrau and Rodriguez, (2008)[16] who reported that duration of catheterization has influence on CAUTI, Danchaivijitr, *et. al.,* (2005)[9] who reported that incidence of CAUTI increase after two weeks of catheterization. Nicolle and Lindsay (2005)[18] reported that long term catheterization increase the risk of infection, which also goes with our findings.

On bacteriological examination of 80 urine samples, 58 (49%) isolates of *Escherichia coli*, 16 (14%) isolates of *Pseudomonas aeruginosa*, 8 (7%) *Staphylococcus aureus*, 12 (10%) isolates of *Proteus spp*, 8 (7%) isolates of *Klebsiella pneumoniae*, 6 (5%) isolates of *Enterobacter aerogenes* and 10 (8%) isolates of yeasts and fungi were recovered and identified (Fig-8). These finding are supported by investigation of Lee, *et. al.,* (1996)[17], Nicolle and Lindsay (2005)[18] , Milan and Ivan, (2009)[19] who isolated *Escherichia coli*, *Pseudomonas aeruginosa, Staphylococcus aureus* , *Klebsiella pneumoniae* and yeast from the catheterized patients. Their findings suggested that *Escherichia coli* is major cause of CAUTI as indicated in the present study.

Although pathogens isolated by the different researchers are more or less same and goes with our findings but the percentages of the pathogens except

Escherichia coli vary slightly from one another and different scientists reported the different prevalence of these pathogens. The difference is because different environmental conditions where the scientists worked, as uropathogens are usually nosocomial microorganisms so it depends on the hospital environment. The immunity of the patients belonging to the different countries also varies so this might be a factor in difference in percentages of different pathogens.

CONCLUSIONS

In the present investigation the isolation of various types of pathogens from the urine samples of catheterized patients included *Escherichia coli*, *Staphylococci aureus*, *Pseudomonas aeruginosa*, *Enterobacter aerogenes*, *Klebsiella pneumoniae*, *Proteus vulgaris*, *Proteus mirabilis* and yeast and fungi. *Escherichia coli* were found in high percentage hence found to be most prevalent cause of CAUTI. Females were found to be more prone to CAUTI. Among the different risk factors studied reason major surgery, long-term duration of catheterization, marital status of patient and type of catheter used have a strong influence on the occurrence of CAUTI. Socioeconomic status of patient was not found to have strong relation with CAUTI. In order to reduce incidence of CAUTI unnecessary catheterization should be avoided, where required it should be done under aseptic measures. When used for long duration, catheter should be replaced on weekly basis so that the chances of colonization of pathogen on catheter tip can be reduced. Silicon catheter should be preferred over ordinary catheter as it reduces the chance of infection.

ACKNOWLEDGEMENTS

The authors would like to acknowledge Infection Control Department of Mayo Hospital and Industrial Biotechnology Department of GCU Lahore for providing research studentship.
Funding: No funding sources
Conflict of interest: None declared
Ethical approval: None required

REFERENCES

1. Azzarone, G., S. Liewehr and K. Connor. 2007. Cystitis. Pediatr Rev, 28(12): 474-6.
2. Jacobsen, S.M., D.J Stickler., H.L.T. Mobley and M.E. Shirtliff. 2008. Complicated Catheter-Associated Urinary Tract Infections Due to *Escherichia coli* and *Proteus mirabilis* .Clinical Microbiology Review, 21:26-59
3. Kunin, C. M., S. Douthitt, J. Dancing, J. Anderson, and M. Moeschberger. 1992. The association between the use of urinary catheters and morbidity and mortality among elderly patients in nursing homes. *Am. J. Epidemiol*. 135:291-301.
4. Wang, L. P., H. Y. Wong, and D. P. Griffith. 1997. Treatment options in struvite stones. *Urol. Clin. N. Am*. 24:149-162.
5. Wang, X., J. F. Preston III, and T. Romeo. 2004. The pgaABCD locus of Escherichia coli promotes the synthesis of a polysaccharide adhesin required for biofilm formation. *J. Bacteriol.* 186:2724-2734.
6. Richards, M.J., J.R. Edwards, D.H. Culver and R.P. Gaynes. 1998. Nosocomial infections in intensive care units in the United States: National Nosocomial Infections Surveillance System. *Crit Care Med.,* 27:887--92.
7. Sedor, J., S.G. Mulholland. 1999. Hospital-acquired urinary tract infections associated with the indwelling catheter. *Urol Clin North Am*, 26(4):821-8.
8. Tambyah, P. A., and D. G. Maki. 2000. Catheter-associated urinary tract infection is rarely symptomatic: a prospective study of 1,497 catheterized patients. *Arch. Intern. Med*. 160:678-682.
9. Danchaivijitr, S., C. Dhiraputra, R. Cherdrungsi, D. Jintanothaitavorn and N. Srihapol. 2005. Catheter-associated urinary tract infection. *J Med Assoc Thai*, 10:S26-30.
10. Eposito, S., S. Noviello and S. Leone. 2008. Catheter-associated urinary tract infections: epidemiology and prevention. *Infez J Med,* 16(3):130-43
11. Stamm, W.E. 1991. Catheter-associated urinary tract infections: epidemiology, pathogenesis, and prevention. *Am J Med*, 91(3B): 65S-71S.
12. Zhan, C., A. Elixhauser, C.L. Richards, Y. Wang, W.B. Baine, M. Pineau, N. Verzeier, R. Kliman and D. Hunt. 2009. Identification of Hospital-Associated Urinary Tract Infections from Medical Claims: Sensitivity and Positive Predictive Value. Med Care, 47(3): 364-9.
13. Erben, N., S.N. Alpat, E.D. Kartal, I. Ozgunes and G. Usluer. 2009. Analysis of the risk factors in nosocomial urinary tract infections and effect of urinary catheter use on distribution of the causative agents. *Mikrobiyol Bul.* 43(1):77-82.
14. Wald, H.L., A.M. Epstein, T.A. Radcliff and A.M. Kramer. 2008. Extended use of urinary catheters in older surgical patients: a patient safety problem? *Infect Control Hosp Epidemiol.* 29(2): 116-24.

15. Brosnahan, J., A. Jull and C. Tracy. 2004. Types of urethral catheters for management of short-term voiding problems in hospitalized adults. *Evid Based Nurs.* 7(3): 85.
16. Pigrau, c. and M.D. Rodriguez-Pardo. 2008. Infections associated with the use of indwelling urinary catheters. Infections related to intrauterine devices.
Enferm Infecc Microbiol Clin, 26(5): 299-310.
17. Lee, Y.L., L.D. Thrupp, R. Lee, S. Nothvogel, N. Farsad and T. Cesario. 1996. Methicillin-resistant Staphylococcus aureus (MRSA) in a Malaysian Hospital. *Med. J. Malaysia,* 49(1): 24-28.
18. Nicolle and E. Lindsay. 2005. Catheter-Related Urinary Tract Infection. *Drugs and aging,* 22(8): 627-639.
19. Milan, P.B. and I.M. Ivan. 2009. Catheter-associated and nosocomial urinary tract infections: antibiotic resistance and influence on commonly used antimicrobial therapy. *Int Urol Nephrol*, 41(3): 461-4.