

Existence of bacterial Flora in the Urogenital Tract

M.Anitha, Pratikshia K., Mohamed Sultan A., D.M. Monisha

Department of Microbiology, Shri Sathya Sai Medical College & Research Institute,
Thiruporur, Sri Balaji Vidyapeeth University, Tamil Nadu, India

Abstract: The aim of the study is to detect the normal flora (commensal) from the urine sample. This study explains the normal flora in the urogenital tract which may lead to pathogenic infection. We collected a total of 3243 urine samples and processed in the central microbiology lab as per the standard procedure. Among 3243 samples, 247 (7.6%) samples showed growth of normal flora like *Staphylococcus epidermidis*, *Micrococci* etc., and 2996 (92.4%) of bacterial growth. In our study, the growth of commensals are more prevalent in females 167 (68%) than males 80 (32%). This result concludes that the lack of awareness and improper sample collection which signifies the bacterial contamination.

Keywords: Bacterial flora, Bacterial culture, Urine samples

Introduction: Urine samples are collected for diagnosing variety of infections among which the urinary tract infection is the most common. From the collected sample, Urinary tract infection (UTI) is one of the most important cause of morbidity in the general population, and is the second most common cause of hospital visits (1). Urinary tract infections (UTIs) are more common among women than men, although the prevalence in elderly men and women are similar (2). Urinary tract infection (UTI) defines a condition in which the urinary tract is infected with a pathogen causing inflammation. (3) The adult human body contains 10^{14} cells, of which only 10% compose the body proper and 90% are accounted by members of the micro flora (4). The aerobic introit and urethral flora of patients with recurrent urinary tract infections have been shown to be different from the aerobic flora of healthy

volunteers who do not have urinary tract infections. In that large number of pathogenic bacteria persist on the mucosal surface of the introitus and urethra of those women with recurrent urinary tract infections (5). Although the term 'normal □ flora' is commonly used, it is really a misnomer. Microbial flora has spatial and temporal complexity that differs by individual according to body condition, age, geographical location, health status, diet and type of host.

Even within the same individual, the composition of the microbial flora can vary according to changes in diet, stress, sexual behaviour, medication, hormonal changes and other host-related factors. (6, 7) Even the usefulness of qualitative data is often negatively affected by inappropriate or suboptimal methods of data collection, failure to use appropriate transport systems or enriched media, or a lack of stringent anaerobic technique in the processing and culture of specimens. (8) The present study was conducted to determine the normal bacterial flora of the urine samples and to bring awareness among patients about the practice of mid-stream urine collection.

Material Methods:

Urine samples were collected from a total of 3243 patients of all age groups with urinary tract infections for over a period of 1 year from January 2015 to December 2015.

Sample Collection:

Urine sample were collected from patients and labeled as per the standard procedure.

Culture Methods:

Urine was mixed by rotating the container and was inoculated on Nutrient agar, McConkey agar, Blood agar and UTI chrom agar. The plates were then aerobically incubated at 37°C for overnight. A specimen giving 10⁵ cfu/ml or forming at least 20 colonies were considered as positive for UTI and remaining were considered as commensals. All the bacteria were isolated and identified using morphological, microscopy and biochemical tests following standard procedures. (9, 10)

Results:

In this study, a total number of 3243 urine samples were collected at SSSMC & RI from the period of Jan 2015 to Dec 2015, among which

S. No	Months	Total No. Of Normal Flora	Total No. Of Bacterial Growth
1	January	7 (2.83%)	226 (6.96%)
2	February	7 (2.83%)	226 (6.96%)
3	March	20 (8.09%)	288 (8.88%)
4	April	17 (6.88%)	205 (6.32%)
5	May	13 (5.26%)	263 (8.10%)
6	June	51 (20.60%)	283 (8.72%)
7	July	2 (0.80%)	361 (11.13%)
8	August	6 (2.42%)	203 (6.25%)
9	September	19 (7.69%)	231 (7.12%)
10	October	32 (12.95%)	248 (7.64%)
11	November	26 (10.52%)	294 (9.06%)
12	December	44 (17.81%)	168 (5.18%)
	TOTAL	247 (7.6%)	2996 (92.4%)

Table 1: Showing the prevalence of normal flora and bacterial growth.

247 (7.6%) samples showed growth of normal flora like *Staphylococcus epidermidis*, *Micrococci*, etc. as shown in Table 1. It also shows 247 (7.6%) normal flora (commensal) and bacterial growth 2996 (92.4%).

The samples were cultured in the central microbiology lab as per the standard procedure.

S. No	Months	(Normal Flora) Male	(Normal Flora) Female	Total No. Of Normal Flora
1	January	5 (6.25%)	2 (1.19%)	7 (2.83%)
2	February	2 (2.5%)	8 (4.79%)	7 (2.83%)
3	March	5 (6.25%)	15 (8.98%)	20 (8.09%)
4	April	10(12.5%)	7 (4.18%)	17 (6.88%)
5	May	3 (3.75%)	10 (5.98%)	13 (5.26%)
6	June	13(16.25%)	38 (22.7%)	51(20.60%)
7	July	0	2 (1.19%)	2 (0.80%)
8	August	1 (1.25%)	5 (2.99%)	6 (2.42%)
9	September	7 (8.75%)	12 (7.18%)	19 (7.69%)
10	October	12 (15%)	20(11.97%)	32(12.95%)
11	November	8 (10%)	18(10.77%)	26(10.52%)
12	December	14 (17.5%)	30(17.96%)	44(17.81%)
	TOTAL	80 (32%)	167(68%)	247(7.6%)

Table 2: Showing the prevalence of normal flora in male and female.

Table 2 describes, from 247 (7.6%) normal flora, males are about 80 (32%) and females 167(68%). In our study, females are having more commensals than male which leads to the pathogenicity and causes various infections in the genital tract. Due to lack of awareness, the improper collection of urine sample leads to bacterial contamination. UTI is one of the most common types of infections in the genital tract. Urine cultures are done to detect organisms that are the causative agents of urinary tract infections. Normally the urinary tract is sterile above the urethra. However, during non-invasive collection techniques, urine is potentially contaminated with normal flora of the urethra and genitourinary tract. For this reason, urine cultures utilize a colony count (quantitation of growth) to aid in determining if dealing with contamination, colonization, or infection. Infections are associated with counts of 100,000 X 10⁵ or more organisms per ml of urine. Hence proper collection of urine samples under aseptic condition will reduce the bacterial contamination during sample processing.

Discussion: From a total of 3243 urine samples, 247 showed normal bacterial flora. In our study, we have focused about the commensals in the urogenital tract while collecting the urine samples. Also we have demonstrated the prevalence of UTI in women and men, from which women were found more vulnerable than men. Growth of bacteria in the flora of the genital tract is stimulated by estrogen. (11) Several lines of evidence support the role for estrogen in increasing the density of vaginal colonization by normal flora organisms (12, 13). Reid et al. (14) suggested an alternate mechanism for control the bacterial flora by the lactobacilli. They found that cell wall fragments of *Lactobacillus* species could block attachment of bacterial uro-pathogens to uro-epithelial cells. It is not clear whether this observation might also apply to vaginal epithelial cells or whether adherence of vaginal microorganisms to the epithelium might be blocked by this mechanism. Colonization of the introits with *Enterobacteriaceae* species is a predisposing factor for urinary tract infection in women. This investigation is correlated with EL-Arabi et al. (15) in which they found the normal flora from the Ewes reproductive tract (uterus, cervix, and vagina). Unlike other studies, our report exposes the growth of normal flora in the urinary tract due to improper collection of urine samples and this will be reduced by providing awareness to the patients about the aseptic midstream urine collection during sampling. Our results disclose that the collection of urine sample in an improper manner leads to bacterial contamination, hence this makes a problem even during diagnosis. This has to be improved by the co-operation of the patients from their clinicians advice.

Conclusion: In this study, we explain the normal flora in the urogenital tract which may lead to

pathogenicity causing infection due to bacterial contamination in the urogenital tract. Therefore aseptic mid-stream urine (MSU) collection will help to identify and diagnose a UTI accurately. The problem with the first-flush urine is that it can wash bacterial flora and debris of the skin and into the sample, leading to false-positive or inaccurate result. Hence the study highlights the professional responsibility to educate and make the patient understand the importance of the proper collection of the urine sample in order to diagnose UTI precisely and prevent bacterial commensal contamination.

Acknowledgements: We extend our heartfelt gratitude to the Hospital and Management for their support to carry out this project.

References:

- 1) Kolawale, A.S., Kolawale, O.M.,Kandaki-Olukemi, Y.T., Babatunde, S.K., Kplawale, C.F. Prevalence of urinary tract infections among patients attending Dalhatu Araf specialist hospital, Lafia, Nasarawa state, Nigeria. (2009); *Int. J. Med. Sci.*, 1(5): 163-167.
- 2) C.Manikandan and A.Amsath. Antibiotic susceptibility pattern of *Escherichia coli* isolated from urine samples in Pattukkottai, Tamilnadu, International journal of current microbiology and Applied Science(2014); 3(10) 449-457
- 3) Sree Bhushan Raju, SC Tiwari, Urinary tract infection – A suitable approach, Journal, Indian Academy of Clinical Medicine Vol. 2, No. 4 October-December (2001).
- 4) Hooper LV, Bry L, Falk PG, Gordon JI. Host-microbial symbiosis in the mammalian intestine: exploring an internal ecosystem. *BioEssays* (1998); 20: 336-43.
- 5) Stamey, T. A., M. Timothy, M. Millar, and G. Mihara. Recurrent urinary infection in adult women. The role of introital Enterobacteria. *Calif. Med.* (1971); 115:1-19.
- 6) Salminen S, Isolauri E, Onnela T. Gut •flora in normal and disordered states, *Chemotherapy* (1995); 41: 5-15.
- 7) Keane FE, Ison CA, Taylor-Robinson D. A longitudinal study of the vaginal •flora over a menstrual cycle. *Int J STD AIDS* (1997); 8: 489-94.
- 8) Bryan Larsen I and Gilles R. G. Monif. Understanding the Bacterial Flora of the Female Genital Tract, *Flora of the Female Genital Tract x CID* (2001):32 (15 February).
- 9) Sharma, K. Manual of Microbiology: Tools and Techniques, 2nd Ed. Ann Books Gopaljee Enterprises, Delhi, India (2008).
- 10) El-Arabi A. A., Taylor D. J., Logue D. N. and Benothman M. Isolation and Identification of Bacterial Flora from Reproductive Tracts of Normal Ewes in Glasgow *J. Veterinart. Advances.*, (2013); 3(10): 275-280
- 11) Gujjar, P, Finucane M, Larsen, B. The effect of estradiol on *Candida albicans* growth. *Ann Clin Lab Sci* (1997); 27:151-6.
- 12) Larsen B, Markovetz AJ, Galask RP. Quantitative alterations of the genital microflora of female rats in relation to the estrous cycle. *J Infect Dis* (1976); 134:486-9.

- 13) Weinstein L, Howard JH. The effect of estrogenic hormone on the Hion concentration and the bacterial content of the human vagina, with special reference to the Doderlein bacillus, Am J Obstet Gynecol (1939); 37:698–703.
- 14) Reid G, Servin AL, Bruce AW, Busscher HJ. Adhesion of three *Lactobacillus* strains to human urinary and intestinal epithelial cells. Microbios (1993); 75:57–65.