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Research Paper

Assessment of Urine Gram Stain and Urine Culture in the Diagnosis of Urinary Tract Infection

Microbiology

KEYWORDS : Urinary tract infections, Gram staining, Urine culture, Drug resistance

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ABSTRACT

Urinary tract infection is due to pathogen invasion of urinary tract. It is an important health problem affecting millions in a year. In India, incidence of UTI was 36.3% in hospitalized and 16.5% in non- hospitalized outpatient department. Urine culture is considered to be golden standard diagnostic test for UTI, but there is a substantial delay of 24 to 48 hours in obtaining culture results. Hence patients are treated with empirical therapy. But there is a problem of emergence of drug resistance. Gram stain of urine may be an effective method of ruling out UTI in rural health centre patients, thus saving time and money in impoverished settings. The aim of the study is to assess the diagnostic value of gram stain of uncentrifuged urine and urine culture in diagnosing urinary tract infections.50 patients with UTI symptoms were selected from inpatient and outpatient of the hospital. Diagnosis of UTI was made by urine culture regularly. In this study, urine gram stain and urine culture are done for the diagnosis of UTI. Sensitivity, specificity, Positive predictive value, Negative predictive value are calculated. In comparison with urine culture the sensitivity, specificity, PPV, NPV of urine gram stain is 51%, 46%, 48%, 88% respectively. The above findings suggest that urine gram stain can be used as a diagnostic test for UTI.

Introduction:

Urinary tract infection is one of the most common bacterial infections encountered in clinical practice both in community and hospital settings. Urinary tract infection (UTI) is due to pathogen invasion of the urinary tract. The upper or lower tract may be affected, depending on the presence of infection in the kidney, or bladder and urethra. Urinary tract infections present with a varied range of manifestations, including minor symptoms to serious ones that require hospitalization. In the intermediate and long term, recurrent episodes of UTI may lead to chronic renal failure and arterial hypertension. To prevent these complications UTI must be diagnosed and treated early.

Evaluation of suspected UTI includes history, physical examination and laboratory investigation. Urine analysis for presence of pus cells, bacteria and culture are important in the adequate management of UTIs. [1, 2] Bulk of the specimens submitted to clinical microbiology laboratory consists of urine for culture and sensitivity testing. A significant portion of these urine specimens received, fail to grow any pathogen.[1, 3] Besides, there is a substantial delay of 24-48 hours in obtaining culture results. This leads to not only a waste of resources and labor to the lab, but also delayed and ineffective treatment to the patient. There have been conflicting reports about sensitivity and specificity of these screening methods. In this study we tried to evaluate urine GS along with culture because GS has added advantage of guiding the antibiotic therapy by observing the morphology and staining properties.

Materials and methods:

Study population

After obtaining the institutional ethical committee, the study was conducted in a teaching hospital. Patients with clinical features suggestive of urinary tract infection formed the subject of the study. A total of 50 urine specimens collected by clean catch midstream method.

Sample collection

A drop of urine specimen is Gram stained and examined by light microscopy. If gram negative bacteria are observed by Gram stain, cultures are performed in the Microbiology laboratory and the results are noted.

Gram Staining:

Gram staining is of the most common staining procedure developed by Danish Bacteriologist Hans Christian Gram. The procedure is, a drop of urine is applied to glass microscope slide, allowed to air dry. Then it is stained with primary stain gentian violet. After 1 minute it is drained off and washed away. Then the smear is covered with Grams Iodine, a mordant. Then it is washed and treated with acetone, decolorizing agent and rinsed off. Then counter stain usually 5% dilute carbol fuschin is added and washed off after 2-3 minutes. The smear is washed again, air dried and examined microscopically. Gram stains are positive if any organism were present on a survey of 20 oil immersion fields.

Gram staining findings are recorded and correlated with culture results which were considered as gold standard. Urine gram stain was evaluated for its diagnostic accuracy as a screening method by calculation of sensitivity, specificity, positive predictive value and negative predictive value.

Results:

A total of 50 urine specimens were processed. Predominant age group was of 60-70 years. Males were 29(58%) and females 21(42%). Of 50 urine specimens, 18 yielded growth and 32 did not grow any pathogen. Gram staining of 17 urine specimens did not show any bacteria in the smear, of which 15 were culture negative, which was in agreement with Gram stain finding. Analysis of Gram stain and culture results was done according to age and sex depicted in **Table-1&2**.Sensitivity of Gram stain was calculated to be 51%, specificity 46%, PPV 48% and NPV 88% from the formulae above.[**Table 3**]

Discussion:

Urinary tract infections are one of the most common presentations encountered in medical practice. Often, the causative agents are gram negative bacteria (Enterobacteriaeceae) followed by coagulate negative staphylococci (CONS). [2, 3] Patients are usually treated by empirical antibiotic therapy. However there is a problem of emergence of drug resistance and also ineffective treatment when using empirical antibiotic treatment. This study evaluates GS of urine as a screening test for urine culture and hence, as a guide to empiric antibiotic therapy.

GS of the urine provides invaluable information about culture results and empiric antibiotic selection. [4] Seeing one or more organisms in the uncentrifuged GS urine correlates with significant bacteriuria. More importantly, it helps in guiding choice of empiric antimicrobial agents at the time of initial clinical evaluation based on GS observation and morphology. [3] However, clinician should consider local sensitivity patterns of the possible pathogen. [4]

In the present study sensitivity of gram stain was calculated to be 51%, specificity 46%, PPV 48%, NPV 88%. Similarly a Turkish cross-sectional study reported that Gram stain had high sen-

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sitivity, specificity, PPV and NPV and was recommended a rapid tool to rule out the diagnosis of UTI in clinical and laboratory settings Furthermore a study in Japan concluded that quantitative unspun urine microscopy with confirmatory oil immersion as simple, accurate diagnostic method to evaluate significant bacteriuria and is useful because of its quickness.[5-8]

In the present study, PPV was 48% and NPV 88% which state that urine Gram staining is very reliable in interpretation of urine cultures and can be used for empirical treatment of patients presenting with UTIs. There were seventeen false positives and two false negative results in our study. False positives could be due to bacteria which failed to grow in culture either because of antibiotic effect or they were fastidious organisms. False negatives could be attributed to fewer numbers of bacteria in the urine which could not be detected by GS but grew on culture.

Our study had a few limitations. Study was conducted in a teaching hospital which receives patients referred from various hospitals in the region. Hence, the patients who received antibiotic when they reached the hospital could not be identified.

One major limitation of the microscopic method reported in this study is its lower sensitivity for detecting bacteriuria in urine specimens containing < 10^5 CFU/ml, a level that may be present in the acute dysuria syndrome in women and in children. But in urine specimens containing > 10^5 CFU/ml, usually associated with asymptomatic patients, patients with acute pyelonephritis, and patients with acute cystitis, a GS may be used as an accurate and inexpensive screening method.

Conclusion:

Urine culture is the current golden standard diagnostic procedure for UTI with high sensitivity and specificity. The main drawback is delay in result and expensive. On the other hand Gram stain of uncentrifuged urine is a very sensitive and specific screening test for diagnosis of UTI. It is a quick and reliable substitute to culture.Results of Gram staining can be used in the selection of antibiotics for empirical therapy. However the clinician should also consider the local sensitivity pattern of the probable pathogen while prescribing empiric therapy. In conclusion, Urine gram stain may be a good alternative as a diagnostic test for UTI, especially in health centers with limited sources.

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Table 1 Analysis with age

AGE	5-10	11-20	21-30	31-40	41-50	51-60	61-70	71-80	>80
BOTH CULTURE AND SMEAR POSITIVE	2	1	2	1	1	2	4	3	-
SMEAR POSITIVE CULTURE NEGATIVE	1	2	2	3	3	2	4	1	-
CULTURE POSITIVE SMEAR NEGATIVE	–	-	1	-	-	-	1	-	-
BOTH CULTURE AND SMEAR NEGATIVE	2	-	3	-	5	3	1	-	-
TOTAL	5	3	8	4	9	7	10	4	-

Table 2 Analysis with Sex

	CULTURE AND SMEAR +VE	SMEAR +VE CULTURE -VE	CULTURE +VE SMEAR -VE	CULTURE AND SMEAR –VE
MALE	9	11	1	8
FEMALE	7	6	1	7

Table 3 Comparison of Gram stain Finding and Culture results

GRAM STAIN FINDING	NO. OF URINE CULTURE RESULTS		
	POSITIVE	NEGATIVE	TOTAL
POSITIVE	16^{TP}	17^{FP}	33
NEGATIVE	2^{FN}	15^{TN}	17

TP- True Positive **FP- False Positive**

FN- False Negative **TN- True Negative**

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