Evaluation of Chromogenic and Uricult Media for Identification of Urinary Tract Infection

Thesis
Submitted for partial fulfillment of
Master degree in
Microbiology

Presented by

Ahmed Mohamed Fkirin
M.B.B.ch
Faculty of Medicine, Menoufia University
2003

Under the supervision of

Prof. Nehal Ibrahim Draz
Professor of Microbiology & Immunology
Faculty of Medicine, Ain Shams University

Dr. Ayman Asaad Ebrahim
Assistant Professor of Microbiology & Immunology
Faculty of Medicine, Ain Shams University

Dr. Essam Abdel Aziz Abdel Mobdy
MD, Internal Medicine and clinical Nephrology
Military Medical Acadamy

Ain Shams University
2010

Acknowledgment

First of all, I thank Allah to whom I relate any success in achieving any work in my life.

I would like to express my deepest gratitude to my eminen.

Prof. Nehal Ibrahim Draz
Professor of Microbiology & Immunology
Faculty of Medicine, Ain Shams University
For giving me the honour of working under her supervision.

also I would like to express my gratitude to
Dr. Ayman Asaad Ebrahim
Assistant Professor of Microbiology & Immunology
Faculty of Medicine, Ain Shams University
For his support, advice and assistance in initiating and completing this work.

Dr. Essam Abdel Aziz Abdel Mobdy
MD, Internal Medicine and clinical Nephrology
Military Medical Acadamy
No words can express my feeling and respect to them regarding their continuous encouragement at every stage of this work.

I would like thank
Dr.Mohamed hassan Shahen
Head of Clinical Pathology-El-Maadi Army Hospital
Military Medical Acadamy

Abstract

Background:- Microbiological examination of urine is the most frequently requested investigation in diagnostic clinical microbiology laboratories. Development of methods for the routine culture of urine for the presence of urinary tract infection in laboratories with very high routine specimen workloads has thought to combine both rapid and efficient techniques with the level of accuracy and reliability that clinical setting demand.

Aim of the work:- Evaluation of chromogenic and uricult media compared to traditional culture methods for presumptive identification of organisms causing urinary tract infections.

Subjects, Material and Methods:- One hundred urine samples were taken from patients attending the urology outpatient clinic of El-Maadi army Hospital. Patients who suffering from symptoms of urogenital infection as dysuria, frequent suprapubic pain, and urgency were selected a brief history was taken as regarded to patient's age and sex. Midstream sample of urine was collected after cleaning external urethra in a sterile universal container. Samples were subjected to complete urine analysis including bacterial count, direct microscopic smears. Isolated colonies from different media were identified by blood, MacConkey's, uricult and chromogenic UTI media.

In this study the age of patient ranged from 12 to 63 years old. The mean age was (39.25 ± 14.45). All samples were showed significant bacteriuria with viable count> I00,000 CFU/mI.

Results:- The isolated organisms from the studied urine samples were as follows :- *E.coli* (74%). *Staph aureus* (7%). *Klebsiella pneumonia* (8%). *Proteus mirabilis* (5%). *Pseudomonas aeruginosa* (2%) and *Candida albicans* (1%). Mixed infections were detected in three samples (3%). The Three cases were (*E.coli* and *Klebsiella pneumoniae* - *Klebsiella pneumoniae* and *Staph aureus* - *E.coli* and *Staph aureus*). UTI was higher among females (67%) more than males (33%). Females under 45 years old (47%) were more affected than older age (20%). For males reverse occurs as It was slightly higher at older ages (24%) compared to younger ages (9%).

Chromogenic UTI medium succeeded in detecting all the urinary tract microorganisms that were detected by conventional methods. All strains developed the expected color. It also had the advantage of improved discrimination of mixed growth and an extended range of direct isolate identification. Its sensitivity, specificity, positive predictive value, negative predictive value and accuracy were 100% of all microorganisms.

Uricult medium do not need any experience and it was easy to use. But if bacterial growth was mixed, it gave a wrong result. It has sensitivity 85.73%, specificity 98.12%,

positive predictive value 83.75%, negative predictive value 99.55% and accuracy 98.7%.

Conclusion:- It can be concluded that chromogenic UTI is a good alternative for conventional cultures for more rapid and easier identification of urinary pathogens. Uricult is a good screening test for presumptive diagnosis of UTI.

Lists for tables

		Page
Table(1)	Composition of Chromogenic UTI	44
Table(2)	Antimicrobial regimens effective for treatment of acute uncomplicated urinary infection	51
Table(3)	correlation between age and sex	81
Table(4)	Results of conventional culture and Uricult	83
Table(5)	Results of conventional cuture and chromogenic UTI agar	87
Table(6)	correlation between type of organism and sex according to conventional culture media	90
Table(7)	culture of organism on macConkey and CLED.	92
Table(8)	Growth of organisms on Chromogenic UTI.	93

Lists Of Figures

		Page
Figure(1)	RBC's under microscope of urinalysis.	32
Figure(2)	WBC's under microscope of urinalysis.	33
Figure(3)	Renal tubular epithelial cells under microscope of urinalysis.	33
Figure(4)	Hyaline casts	34
Figure(5)	RBC's casts	34
Figure(6)	WBC's casts.	34
Figure(7)	Granular casts.	34
Figure(8)	Crystals under microscope of urinalysis.	35
Figure(9)	procedure of uricult	77
Figure(10)	correlation between age and sex	82
Figure(11)	Comparison between accuracy of uricult in relation to conventional culture	86
Figure(12)	Comparison between accuracy of chromogenic UTI in relation to conventional culture.	89
Figure(13)	correlation between type of organism and sex according to conventional culture media	91
Figure(14)	Uricult With two face:- green CLED media and MacConkey red media	94
Figure(15)	Uricult of E-coli microorganism with yellow CLED media	94
Figure(16)	Comparison between chromogenic UTI(right side) and uricult(left side)	95

Contents

	Page
Acknowledgment	-
Introduction and aim of the work	1
Aim of the work	3
Urinary Tract Infections Overview	4
Route of infection	6
Risk factor	9
Bacteriuria	15
Pyuria	16
Causes of UTI	17
Pathogenesis of UTI	19
Symptoms of UTI	21
Laboratory diagnosis of urinary tract infection	23
Specimen Collection, Transportation, and Processing	23
Nonculture Methods for the Laboratory Diagnosis of UTI	27
Urinalysis	27
Cultures and the Laboratory Diagnosis of UTIs	39
Chromogenic UTI	44
Uricult	47
Treatment of urinary tract infection	50
Prevention of UTI	54
Microorganisms causing UTI	55
Escherichia coli	55
Staphylococcus aureus	61
Klebsiella pneumoniae	65
Enterococcus	67
Proteus mirabilis	69
Pseudomonas aeruginosa	71

Material and Methods	73
Results	81
Discussion	98
Summary and conclution	104
References	106

Abbreviations

ABU	asymptomatic bacteruria
BAP	blood agar plate
Cfu	colony forming units
CHROM UTI	chromogenic urinary tract infections
CLED	cystine lactose electrolyte deficient
E- coli	*Escherichia coli*
E. faecalis	*Enterococcus faecalis*
IL-6	interleukin-6
IL-8	interleukin-8
K. pneumoniae	*Klebsiella pneumonia*
MRSA	methicillin-resistant *Staphylococcus aureus*
NPV	Negative predictive value
P. aeruginosa	*Pseudomonas aeruginosa*
P. mirabilis	*Proteus mirabilis*
PBP	penicillin-binding protein
PCR	polymerase chain reaction
PMNs	polymorphonuclear granulocytes
PPV	positive predictive value
P-value	probability value
RBCs	Red Blood Cells
S. aureus	*Staphylococci aureus*
SP GR	Specific Gravity
TDA	tryptophan deaminase activity
THB	Tamm-Horsfall proteins
TSI	Triple sugar iron
TSS	Toxic shock syndrome
UTI	Urinary tract infections
VISA	Vancomycin-intermediate *Staphylococcus aureus*
VRE	Vancomycin-resistant enterococcus

VRSA	Vancomycin-resistant *staphylococcus aureus*
WBCs	White Blood Cells
X^2	Chi-square

Introduction and aim of the work

Urinary tract infections are a serious health problem affecting millions of people each year. Urinary tract infections (UTIs) account for about 8.3 million doctor visits each year. Women are especially prone to UTIs. One woman in five develops a UTI during her lifetime. UTIs in men are not as common as in women but can be very serious when they do occur. **(Griebling, 2005).**

There are two types of UTI; lower UTI which is an infection of the lower part of the urinary tract that includes the bladder and the urethra, and upper UTI which is an infection of the upper part of the urinary tract, which includes the kidneys and the ureters. Upper UTIs are potentially more serious than lower UTIs because there is a possibility of kidney damage. **(Linda and Shortliffe, 2004).**

Bacteria typically enter the urinary tract through the urethra. Once the bacteria are inside the urinary tract they multiply, causing infection. There are a number of different bacteria that are responsible for urinary tract infections, including, *Eschericia coli* (80 to 90 percent), *Proteus, Klebsiella, Enterococcus and Staphylococci*. Occasionally, a UTI may be due to a yeast, such as *Candida albicans*. **(Donald, 2008).**

Most UTIs are easily treated and cause no serious health problems. However, if UTI is left untreated, it is possible that the infection will travel up the urinary tract leading to pyelonephritis that may result in permanent kidney problems. **(Durwood and Neal, 2008).**

Uricult is a culture slide covered on both sides with agar media, one side is covered by green cystine lactose electrolyte deficient (CLED) agar and the other with reddish brown MacConkey medium for detection of microbes causing UTI. **(Thompson, 2005).**

Chromogenic UTI agar consists of a rich nutritive base combining different peptones and 2 chromogenic substrates which enable the detection of activities of specific enzymes. Direct identification of the

bacteria most commonly isolated in urinary tract infections is based on the colouring change of media, for example, *E-coli*: spontaneous coloration (pink to burgundy), *Enterococc*i: spontaneous turquoise coloration of strains*, Proteus*: light brown to dark brown coloration of strains. **(Fallon et al., 2002).**

Aim of the work

The aim of this work is evaluation of chromogenic and uricult media compared to traditional culture methods for presumptive identification of organisms causing Urinary tract infections.

Subjects, Material and Methods

Subjects and patients:-

One hundred urine samples were taken from patients attending the urology outpatient clinic of El-Maadi armed Hospital. Those patients were suffering from symptoms of urogenital infection as dysuria, frequent suprapubic pain, and urgency.

A brief history of each case was taken as regared to patient's age and sex. The samples were taken from patients before receiving any antibiotic treatment.

Collection of samples:-

One urine sample was taken from each patient, careful cleaning and washing of the urogenital area, (the glans penis in males and external genitalia in females), a mid stream urine sample was collected in a sterile container.

The samples were immediately transported to the laboratory, and examined without delay within half an hour of collection.

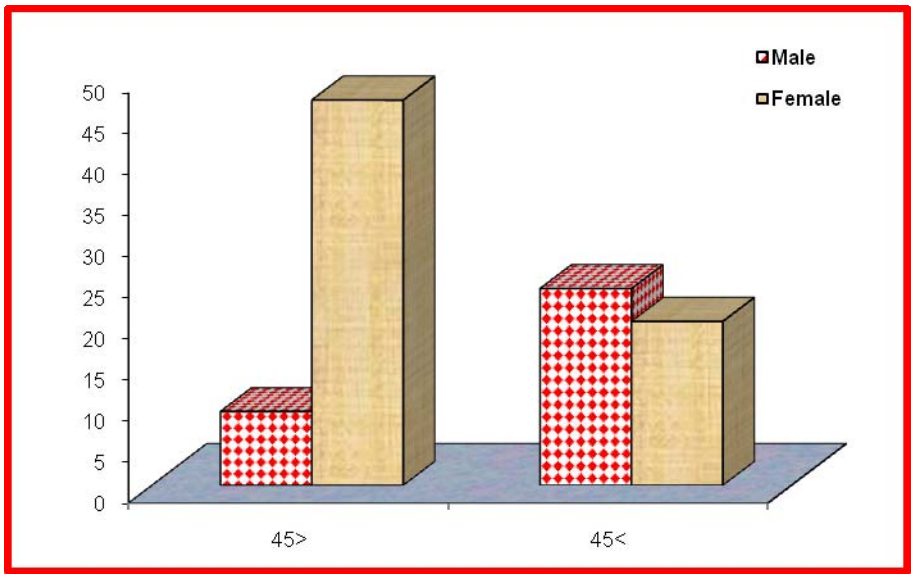
Each sample was subjected to the following:-

1) ***Complete urine analysis:-*** with physical, macroscopic and direct microscopic examination of an unstained smear for presence of pus cells, casts, crystals and epithelial cells.
2) ***Viable bacterial count :-*** The samples which gave > 100000 (CFU/mL) were included in the study and those which gave < 100000 (CFU/mL) were excluded .
3) ***Culture, isolation and identification:-*** the urine specimens were centrifuged and the sediment was cultured on (Blood, MacConkey, Chromogenic UTI and Uricult agar).

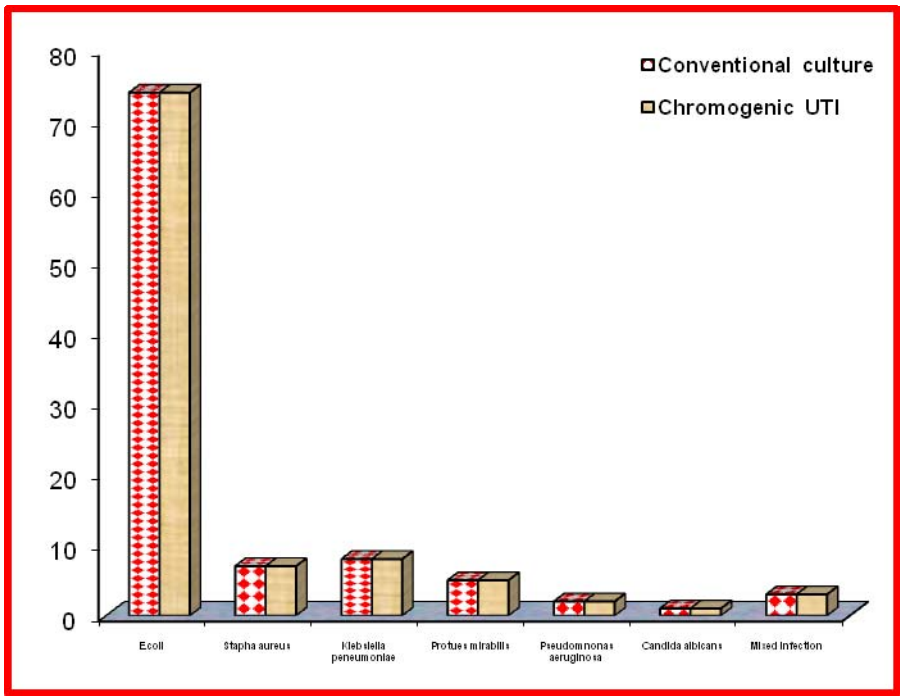
RESULTS

This study included 100 patients suffering from dysuria and pain in the loin. These samples were collected from El-Maadi army hospital.

In this study the age of patients ranged from 12 to 63 years old. The mean age was (39.25 ± 14.45), they were 33 males and 67 females. All of the samples were demonstrated to have a significant bacteriuria with viable count> I00000 CFU/mI.



Correlation between age and sex.



Comparison between accuracy of chromogenic UTI in relation to conventional culture.