IOSR Journal of Dental and Medical Sciences (IOSR-JDMS)
e-ISSN: 2279-0853, p-ISSN: 2279-0861.Volume 19, Issue 2 Ser.3 (February. 2020), PP 35-44
www.iosrjournals.org

Evaluation of Urinary Tract Infections for Microbiological and Antibiogram Studies in The Department of Obstetrics and Gynaecology Of Malda Medical College & Hospital, Malda, West Bengal, India.

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Abstract: *This study was done in Malda Medical College and Hospital, Malda to detect female UTI patients (both pregnant & nonpregnant) and their different parameters. All the urine specimens from suspected UTI cases do not yield growth of microorganism on ordinary culture. A significant number remain sterile on routine culture. A large number of culture-negative urine samples could be screened out earlier by simple microscopic examination. Pregnancy wise analysis of results reveals that incidents of urinary tract infection is more prevalent in pregnant patients. Age wise analysis of data reveals that prevalence of UTI cases is more in the age group between 15-60 years of which highest incidents is observed between 30-45 years. Observation on prevalence of uropathogens isolated reveals that still predominant organisms implicated are enterobacteria of which E. coli predominates. Prevalence of newly implicated uropathogens Staph saprophyticus is not much common in this community. It is found between the age group of 15-30 years in Pregnant patients. Prevalence of different uropathogens in various age groups noted were as follows: E. coli was the predominant pathogenic organism implicated in all the age groups. Prevalence of other different organisms in different age groups noted was as follows:*
maximum Klebsiella spp. were isolated from the patients age groups 15-30 years. Staph aureus was isolated only in the of 45-60 years. S faecalis was isolated from patient of 45-60 years of age group. Contamination of urine specimens occurs in significant numbers of cases. Micrococcus remains most prevalent contaminant followed by Candida spp. Present study reveals that there has been a significant change in the susceptibility pattern of offending organism and they are showing resistance to those earlier antibiotics and also to most of the presently available antimicrobials and remain sensitive to only few members of amino glycosides, eg- Amikacin and third generation Cephalosporin, eg- Cefotaxime and moderately sensitive to Fluroquinolones eg- ciprofloxacin.

Date of Submission: 20-01-2020 Date of Acceptance: 10-02-2020

I. Introduction:

The urinary tract anatomically comprising of the kidneys, the bladder and the urethra having its prostatic part in males, may be considered as a single anatomic unit connected by a continuous column of urine which extends from the urethra to the kidneys. Frequently any portion of the upper or lower part of the tract or whole of the tract may be infected.

Urinary tract infections (UTI) which encompasses a spectrum of clinical and pathological conditions involving various parts of the urinary tract is a very common cause of morbidity and even mortality due to its subsequent complication.

Infection of the urinary tract, which may be acute or chronic, community acquired or hospital acquired is prevalent all over the world and affects different age group of both the sexes with varying frequencies causes of UTI may present with various symptoms like dysuria, lower abdominal pain, loin pain, fever etc or cases may remain asymptomatic making diagnosis difficult.

UTI is associated with the presence and multiplications of microorganisms in the urinary tract. Various microorganisms are implicated in the causation of UTI with varying frequencies which includes different bacteria, some fungi, mycoplasmas or viruses. Of these various offending microorganisms some very frequently met with mostly the coliforms- eg. Escherichia coli, klebsiella spp., Proteus spp., Pseudomonas spp. There are also found other gram-positive cocci like Staph aureus, Staph saprophyticus, Strepto faecalis and some others. Other organisms are rarely found.

Causes of UTI if remain undetected and left untreated may have fatal consequences like septicemia and also eventual death at times or it may cause long term complications like chronic pyelonephritis leading to

hypertension or chronic renal failure.

Presence and persistence of UTI during pregnancy apart from renal complications may aggravate other complications which can be checked by early detection and treatment by administration of appropriate antimicrobial drugs.

But sensitivity of offending microorganisms to a particular antibiotic is not remaining constant. Most of them are showing various degree of resistance to commonly available and used drugs. This change of sensitivity pattern poses particular problem successful is management of UTI cases. Especially nosocomial UTIs which comprises largest single group of Hospital acquired infections caused by more resistant organisms. These resistant patterns vary geographically which precludes the empirical selection of antibiotics for treating UTI cases and magnifies the problem.

II. Brief Review Of Literature:

Historical Background:

Cases of UTI recorded since antiquity, presents clinically in a variety of ways and is associated with the presence and multiplications of the microorganisms in the urinary tract[21,23]. Pasteur (1863) recognized urine as good culture medium for bacteria and Roberts (1880) related symptoms to the presence of bacteria in urine.

However, simple detection and isolation of these microorganisms in urine specimen does not necessarily prove that they are the offending uropathogen because the isolated organism may merely be contaminants[4]. The crucial steps in the bacteriological diagnosis of UTI was the introduction of the concept of "significant bacteriuria" by Kass who reported that true bacteriuria could be distinguished from contamination by quantitative urine culture[6] and it was accepted that more than 10^5 colonies of single type/ml cultivated from a properly collected and cultured voided urine specimen denotes UTI[4] is diagnosed with 95% certainty[13]. Bacterial count below 10^5 /ml particularly with different types of colonies, signifies contamination[13] but no definite diagnosis can be made from intermediate counts (10^4-10^5/ml).

This criteria for UTI is unreliable for some situations where less than10^5 /ml of bacteria are yielded in MSU despite presence of UTI these are:

I. Patients consuming large amounts of fluid and diluting bladder urine
II. Infant and children
III. Recent antimicrobials therapy
IV. Patients with urinary obstruction which prevents excretion of organisms
V. Patients with early case of pyelonephritis due to hematogenous spread while significant number of organisms does not appear in urine.
VI. Infection with fastidious bacteria
VII. In presence of high urea concentration, low PH and high osmolality, bacterial growth is inhibited which results in diminished number of bacteria[33].
VIII. Antiseptic detergent solution used for cleansing before voiding may cause diminution in the number of organisms in urine.[6]
IX. While the specimen cultured is obtained by suprapubic aspiration or obtained from ureter or renal pelvis[4,33].
X. Urethral syndrome[6,8,33] - a clinical condition with poor pathologic correlation and anatomic precession. In some patient, mostly the sexually active female presenting with dysuria, urgency and frequency of micturition, yields less than 10^5 organism/ml or no bacteria or urine (MSU) culture.

Convert or Asymptomatic Bacteriuria (ASBU)[28,33]:

This is a condition in which apparently healthy asymptomatic patients usually the female of child bearing age or older, more than 10^5 organisms of a single species/ml of MSU are detected on culture[8] and the same finding with repeated urine specimen denotes UTI in 95% of cases. Patient with asymptomatic UTI may be associated with increased mortality rates and may have serious complications.

Aetiology: Different microorganism causes UTI, preponderance of which may vary with UTIs in hospital setting[17] or acquired in community, intervention or manipulation in the urinary tract like catheterization[14], state of the local or systemic defense mechanism of the host[18], presence or absence of other diseases e.g. granulomatous disease[1] or may be age and sex related.

UTI in uncomplicated persons in all ages acquired in community are mostly (80%) caused by Escherichia Coli[2,5,6,7,8,13,15,21] (E. Coli) and is responsible for about 90% of the first UTI in women. A large variety of serogroups of E. coli are implicated[4,7,11]. But 90% of urine specimens of these cases contain only one particular type of subgroup[7]. Besides proteus species[27], often Pseudomonas species (spp) and Klebsiella spp sometimes Enterobacter, Serratia, Streptococcus faecalis account for a number of uncomplicated cases. Staphylococcus saprophyticus[20,24,25], Novobiocin resistant, coagulase negative cocci, causes about 10% to 20% acute

symptomatic UTI in sexually active young female but rarely causes UTI in males[15] or in children[29].

Pathogenesis of UTI: The routes by which the microorganisms invade and infect the urinary tract includes -

i. Ascending infection via urethra to the bladder and then to the kidneys.
ii. Hematogenous descending route - occurrence of UTI via this route is much less.
iii. Lymphatic route - though rare, tubercular infection may follow this route.

Factors influencing occurrence and preponderance of UTI include -

I. Gender, age and sexual activity[21,33].
II. Factors interfering with free flow of urine or increase residual bladder urine volume e.g. presence of calculi, enlarged prostate, tumor etc.
III. Introduction of organism in the urinary tract by indiscriminate catheterization or intervention.
IV. Phimosis and lack of circumcision facilitates UTI[26].

Diagnosis: Diagnosis of UTI remains a complex and perplexing issue in both sexes and in all ages. Clinical symptoms may be suggestive but not diagnostic. UTI may be asymptomatic. Genital infections, symptoms due to other diseases may mimic UTI and in children symptoms may even be atypical. There are some other methods e.g. histopathologic, radiological or other procedures which help in diagnosis but bacteriological examination of the urine remains the mainstay in the diagnosis of UTI caused by bacteria. For other aetiologic agents some other direct or indirect techniques are employed.

Treatment: Successful therapy depends on administration of appropriate antimicrobial drugs in adequate dosage for optimum period of time[12] along with removal of predisposing factors if possible. Various drugs are widely used which includes Penicillins and its analogues, Tetracyclines, Co-trimoxazole, Aminoglycosides, Cephalosporins, Nalidixic acid and Quinolones, Nitrofurantoin but as bacterial resistance becomes a problem other newer drugs has been introduced and tried[16]. Apart from treating by short and long term therapy for acute and chronic UTI, in some situations prophylactic use of antimicrobials and antiseptics may be effective[3].

III. Aims And Objectives:

For its wide spread prevalence and potential danger, cases of UTIs has drawn persistent attention extensive studies by various workers in different centers in India and abroad have revealed many unexposed aspects of UTIs and literatures are in plenty.

But much is not known about the present UTI situation at Malda, a rural based city, having a medical college and having influx of patients from several adjoining districts of West Bengal, in term of its age and pregnancy wise prevalence in female, clinical picture, the common causative microorganisms and the susceptibility pattern of the offending microorganisms and changes there in, if any.

The present study is therefore undertaken:

I. To determine the prevalence of different microorganisms in cases of UTIs - suspected on the basis of clinical features and or routine urinalysis in different age groups of both the pregnant and nonpregnant cases.
II. To determine the antibiotic susceptibility pattern of the isolated microorganisms to various commonly available and commonly used antibiotics.

A clear idea of which may be helpful in better and effective management of UTI cases of this area.

IV. Materials And Methods:

The present work was carried out in the Department of Obstetrics and Gynaecology of Malda Medical College & Hospital, Malda, west Bengal, India from April, 2016 to October, 2017.

Materials used for studies:

1) Specimen-properly and aseptically collected Mid Stream Urine (MSU), at times catheterized specimens from persons suspected of having UTI and attending Malda Medical College & Hospital.

2) Media: Culture media used the following media, prepared from commercially available dehydrated media in the Microbiology laboratory of Malda Medical College & Hospital:

i. MacConkey's agar plate
ii. Blood Agar Plate (BAP)
iii. Nutrient agar plate for bacteriological culture of urine aerobically. And also, media for antibiotic sensitivity testing- Muller Hinton agar-prepared in Malda Medical College & Hospital laboratory from Dehydrated media.

3) Standard Nichrome wire loop
4) Incubator
5) Microscope

6) Glass wares
7) Different biochemical sets e.g. Sugar sets.
8) Normal saline
9) Refrigerator
10) Staining material for Gram Staining available commercially
11) Forceps
12) Measuring scale
13) Papers
14) Commercially available antibiotic selectivity discs - (Amikacin 30mcg, Ampicillin 10mcg, Chloramphenicol 30mcg, Erythromycin 15mcg, Gentamicin 10mcg, Nalidixic Acid 30mcg, Nitrofurantoin 300mcg, Penicillin G 30mcg, Tetracycline 30mcg, Norfloxacin 1 mcg and Ciprofloxacin 5mcg, Cefotaxime 1 mcg.)

Plan of Study: The proposed study on the urinary tract infection was planned in the following headings:
1. Determination of prevalence of different microorganisms in healthy individuals of different age group of both the pregnant and nonpregnant patients. Healthy individuals were those who at the time of present studies were not found to have any features suggestive of UTI. This comprised the control group.
2. Determination of prevalence of different microorganisms in cases of UTIs was suspected on the basis of clinical features and or routine urinalysis. Attempt was made to determine the prevalence of different uropathogenic microorganisms in different age groups of female patients included both pregnant and non pregnant cases. These different organisms include the commonly encountered uropathogens. Attempt was also made to identify the organisms, the role of which were previously considered insignificant but now received wide spread importance as significant uropathogens. Attempt was also made to find out the incidences of UTI cases in Hospitalized female patients (both catheterized or non-catheterized) or female patients attending the different OPDs directly from the community and receiving treatment.
3. Determination of the antibiotic susceptibility pattern of the isolated microorganisms to commonly available and commonly used antimicrobials.
4. To assess the efficacy role of the preliminary screening procedures by
i. Wet film examination of unspun urine specimen and
ii. Examination of the gram stained smear of the unspun urine specimen as a screening test for detecting UTIs and further corroboration by subsequent semi- quantitative urine culture.

V. Methodology:

Mode of collection and selection of urine specimens:

he specimens of urine were collected from the female patients of Malda Medical College & Hospital, Malda having signs and symptoms suggestive of UTI or routine urinalysis in whom pointed to the diagnosis of UTI or Having undiagnosed febrile illness requiring confirmation of presence absence of UTI a major portion of these female patients were under treatment in different wards of this hospital but specimens were also obtained from a significant number of cases attending different Out Patient Departments (OPDs) of this hospital.

In all the cases requisite and prior instruction regarding proper collection of urine specimens were explained in detail. Both the pregnant and nonpregnant patients were advised to discard the first few ml of normally voided urine and then to collect the next few ml of the stream - the Mid stream Urine (MSU) directly into the container - usually the supplied sterile and clear cotton plugged test tubes.

From the patients having indwelling urinary catheters, specimens of urine were collected from the closed catheter system, urine was not taken from the free tube or opened bag as frequently these are contaminated.

The specimens thus collected and submitted to Microbiology laboratory, Malda Medical College & Hospital, Malda were promptly examined and inoculated onto the appropriate plates or kept preserved in the refrigerator at 4^{o}c for subsequent inoculation to be done soon on that very date. A regularly quite a large number of urine specimens are received by Microbiology Laboratory of Malda Medical College & Hospital. Naturally all the urine samples could not be examined for the present study. Accordingly, we had to select several number of urine specimens regularly for examinations. The selection of specimen was random, but attempts were made to examine specimens encompassing various patients from different wards, different OPDs. Specimen from patients of various age groups in both the pregnant and nonpregnant were considered both normally voided or catheter specimens and macroscopically hazy or turbid, as well as clear urine specimens also were examined.

Examination of urine specimens:

The urine specimens were examined macroscopically for any abnormal color, presence or absence of haziness or turbidity, deposits. Also, the pH of the urine specimen was seen by placing a drop or two of the specimens to the pH paper strip noting the color change, if any and matching against the accompanying pH chart

this was tested because it is known that presence of some particular organisms may cause change in pH of the infected urine.

Preliminary Screening test for Evidence of UTI:

Principle for test infections of the urinary tract usually accompanies "pyuria" (leukocytes[4]) and bacteria, which is detected by microscopy. Yet in some infections due to Mycobacterium tuberculosis, Mycoplasma, Fungus etc. ordinary culture fails to reveal the pathogens, but the presence of pyuria, particularly in symptomatic patients (sterile or abacterial pyuria) suggests UTI and demands for special methods for identifying the uropathogen. Even in presence of organisms in urine, simple microscopic examination by detecting epithelial cells may distinguish contaminated specimen from properly collected specimen with these backgrounds, we performed preliminary screening as an aid to diagnosis.

Examination of the Gram Stained Smear:

A drop of well mixed unspun urine specimen placed with the help of a flamed and cooled sterile Nichrome- wire loop, over a clean glass slide, was spread and allowed to air dry. The smear was fixed and was then gram stained and examined under microscope in oil immersion.

Search was made for detecting gram stained organisms particularly the gram-negative rods and gram-positive cocci. Also, presence or absence of pus cells and squamous cells were noted.

Culture of Urine:

In the present study only aerobic culture utilizing standard loop method was performed[4].

Method: The media, prepared and sterilized properly as stated earlier were made well dried by placing the plates with their lids ajar in the incubator at 37^0c for some time and then taken out to the incubator. Serial numbers of the test specimens were marked over the plates with marking pencils.

The entire length of Nichrome wire loop of standard dimension was made red hot by flaming over the burner. It was allowed to cool down. After removing the cotton plug and warming the trip of the tube, the loop was dipped vertically in the well sharked or mixed urine specimen and again taken out of the specimen slowly and vertically. The wire loop thus charged with and retaining the urine specimen, was touched onto the media in the plates to inoculate by streaking.

Flaming of the loop was made each time inoculating separate specimens in different media the plates thus inoculated were incubated at 37^0c aerobically. Plates were examined after incubation. Presence or absence of growth of organisms were noted, when growth was present, types of colonies present and colony morphology were noted, presence of colonies of more than single types of organisms mixed together denoted contamination and was discarded. Only plates containing pure isolated colonies were examined further.

Identification of the organisms:

For identification of the organisms grown over the culture media, following steps were followed:

Firstly, the colony morphology and pigmentation and abnormal smell if any were noted in the plates presence or absence of hemolysis in BAP was noted. Then the lactose fermenting capacity were observed. Lactose fermenting (LF) colonies are those which producing the pink colonies in MacConkey's agar. Non lactose fermenting (NLF) colonies showed no change in colour.

Then a smear was prepared by taking little amount of the pure and isolated colonies from BAP / MacConkey's agar, dried, fixed and gram stained and examined under microscope in oil immersion and gram staining character of the organism and their arrangement were noted.

For further identification of gram-positive cocci in clusters catalase test and coagulase test were performed. Tube coagulase test was performed in doubtful cases where slide coagulase test was negative.

To differentiate between the two coagulase negative staphylococcus - staphylococcus epidermidis and staphylococcus saprophyticus - Novobiocin sensitivity test was undertaken.

Gram positive cocci arranged in short chains could be streptococcus faecalis. The following tests were performed to aid its identification:

I. Biochemical test – a) Mannitol fermentation test b) Voges Proskauer (VP) test
II. Bacitracin sensitivity test
III. Heat resistance test

Gram negative organisms:

Gram stained smear prepared from the growth of organisms in MacConkey's agar plate while examined under microscope in oil immersion revealed gram negative rods. Further tests were performed for identification of the species included a) Motility test b) Oxidase test c) Other appropriate biochemical tests e.g.- Glucose, Lactose, Maltose Sucrose, Indole, Methyl Red, Voges Proskauer (VP), Citrate, Urease, growth in KCN,

Phenylalanine, Triple Sugar Iron Agar (TSI), Ornithine, etc.

VI. Observations And Results:

The result of this present study on urinary tract infections was obtained and noted. A total of 200 specimens of urine collected from patients of different age groups of both the pregnant and nonpregnant were examined of which 86(39.5%) were from nonpregnant patients, 114(57%) were from pregnant cases.

Control Group:

A total of 200 specimens were examined from symptomatic and apparently healthy persons comprising 100 from nonpregnant and 100 from pregnant of the age group of 24-50 years mean group being 32 years.

Macroscopic and Microscopic Examination:

Urine was turbid or slightly hazy in 176 specimens and 24 specimens were apparently clear. Amongst the turbid specimens 61 were from nonpregnant patients and 115 were pregnant cases.

PH examination of urine specimens revealed Acidic. PH in 193 cases and just alkaline PH in 7 cases out of total 200 specimens. Occasional pus cells (2-3) or more HPF detected in total 110specimens that from pregnant cases it was 71 and from nonpregnant cases it was 39. Organisms were detected in 79 specimens out of which 30 were from nonpregnant cases and 49 were from pregnant cases. Squamous cell was detected in 31 specimens from pregnant cases. All the specimens in the control group were clear. PH in all the control group was in the acidic range.

Few pus cell 1-2/HPF was present in one specimen from nonpregnant patients and from three samples from pregnant controls. No organism was detected in urine specimen of either pregnant or nonpregnant control group. Presence of Ca-oxalate was detected in one specimen from nonpregnant and squamous cells in one specimen from nonpregnant case.

Analysis of results are as follows:

Table IA

Showing number of urine specimens yielding growth of organisms

NO. OF SPECIMENS EXAMINED	NONPREGNANT	PREGNANT	NO. OF SPECIMENS SHOWING GROWTH OF ORGANISMS	SPECIMEN SHOWING NO GROWTH
200	86	114	79	121
PERCENTAGE	43%	57%	39.5%	60.5%

TABLE IB

SHOWING PRESENCE OF GROWTH OF ORGANISMS ON CULTURE

TOTAL NO OF SPECIMEN TESTED	TOTAL SPECIMEN YIELDING GROWTH OF ORGANISMS	UROPATHOGENIC MICROGANISMS ISOLATED	CONTAMINATED GROWTH IN SPECIMENS
200	79	65	14
PERCENTAGE	39.5%	32.5%	7%

TABLE II

SHOWING THE NUMBER OF GROWTH IN SPECIMEN FROM HOSPITALIZED PATIENTS AND PATIENT ATTENDING THE DIFFERENT OUT PATIENT DEPARTMENTS (OPD)

CATEGORY OF PATIENT	TOTAL NUMBER OF SPECIMEN TESTED	UROPATHOGENIC MICROORGANISM ISOLATED	CONTAMINATED GROWTH IN SPECIMEN	NO GROWTH
CATHETERIZED (A)	17	8	7	2
NON-CATHETERIZED (B)	64	26	4	34
TOTAL Hospitalized (A+B)	81	34	11	36
OPD (C)	119	31	3	85
ALL (A+B+C)	200	65	14	121

TABLE IIIA

SHOWING PREVALENCE OF DIFFERENT URO PATHOGENIC ORGANISMS

No of specimen yielding pure growth of pathogenic organisms	Uropathogenic organisms isolated								
	E. coli	Klebsiella	Proteus	Enterobacter	Pseudomonas	Staph aureus	Staph saprophyticus	Strepto faecalis	others
65	51	11	0	0	0	1	1	1	0
32.5%	25.5%	5.5%	0%	0%	0%	0.5%	0.5%	0.5%	0%

TABLE IIIB

SHOWING PREVALENCE OF CONTAMINATED SAMPLE AND CONTAMINANTS

Total no of specimens examined	Total no of contaminated specimen	Individual contaminated isolated			
		Candida	Micrococcus	Staph epidermidis	Mixed growth
200	14	2	5	1	6
Percentage	7%	1%	2.5%	0.5%	3%

TABLE IV

Showing age wise incidence of pathogenic microorganisms isolated in the urine specimens from UTI cases

Age in yr	E. coli	Klebsiella spp	Proteus spp	Enterobacter	Pseudomonas	S. Aureus	S. Saprophyticus	S. Faecalis	Total no of pathogens isolated
10 – 15	7	1	0	0	0	0	0	0	8
15 - 30	14	4	0	0	0	0	1	0	19
30 - 45	19	3	0	0	0	0	0	0	22
45 - 60	6	2	0	0	0	1	0	1	10
60 - 75	5	0	0	0	0	0	0	0	5
75 -85	0	1	0	0	0	0	0	0	1

TABLE V

Showing pregnancy wise prevalence of total pathogens isolated in urine specimens for UTI cases

	E. coli	Klebsiella spp	Proteus spp	Enterobacter	Pseudomonas	S. Aureus	S. Saprophytic as	S. Faecalis	Total no of pathogens isolated
Non pregnant	22	5	0	0	0	0	0	0	27
Pregnant	29	6	0	0	0	1	1	1	38

Antibiogram:

Organism tested	Total no of isolated	Amikacin		Ampicillin		Cephalexin		Chloramphenicol		Cefotaxime		Ciprofloxacin		Erythromycin		Gentamycin		Nalidixic acid		Nitrofurantoin		Norfloxacin		Penicillin		Tetracycline	
		S	R	S	R	S	R	S	R	S	R	S	R	S	R	S	R	S	R	S	R	S	R	S	R	S	R
E. coli	51	51	0	15	36	17	34	14	37	30	21	21	30	-	-	29	22	13	38	39	12	21	30	-	-	-	-
	%	100	0	29.4	70.6	33.3	66.7	29	71	58.8	41.2	41.2	58.8	-	-	56.7	43.3	25	75	24	76	41.2	58.8	-	-	-	-
Klebsiella	11	8	3	1	10	5	6	3	8	6	5	4	7	-	-	4	7	2	9	-	-	4	7	-	-	-	-
Spp.	%	72.7	27.3	9	91	45.5	54.5	27.3	72.7	54.5	45.5	36	64	-	-	36	64	18	82	-	-	36	64	-	-	-	-
Staph	1	1	0	1	0	0	1	1	0	1	0	0	1	0	1	0	1	-	-	1	0	0	1	0	1	0	1
Saprophyticus	%	100	0	100	0	0	100	100	0	100	0	0	100	0	100	0	100	-	-	100	0	0	100	0	100	0	100
Staph	1	1	0	0	1	1	0	1	0	1	0	1	0	0	1	1	0	0	1	1	0	1	0	0	1	0	1
Aureus	%	100	0	0	100	100	0	100	0	100	0	100	0	0	100	100	0	0	100	100	0	100	0	0	100	0	100
Strepto	1	1	0	0	1	-	-	1	0	-	-	1	0	0	1	0	1	0	1	1	0	1	0	0	1	0	1
Faecalis	%	100	0	0	100	-	-	100	0	-	-	100		0	100	0	100	0	100	100	0	100	0	0	100	0	100

VII. Discussion:

The present study aimed in detecting the UTI cases in terms of its age and pregnancy wise prevalence and the prevalence of its various causative microorganisms, to note the susceptibility pattern of the isolated uropathogens to the commonly available and prescribed antimicrobial drugs in Malda. Attempt was also made to evaluate the diagnostic potential of the preliminary microscopic examination or Urine specimen.

The present study involved bacteriological examination of 200 specimen of urine from patients clinically suspected of having UTI, to confirm or exclude presence of UTI out of which 121(60.5%) failed to yield growth of any organism by routine culture indicating absence of urinary infection caused by common aerobic organisms.

In a study of Hyderabad by Vigg et al. 1991[32.] It was 50.5% of the total specimens tested. It establishes that symptomatology alone cannot always confirm diagnosis, even it may be misleading as symptoms due to genital infection or other disease may mimic that of UTIs.[21]

The present study attempted in evaluating the role of preliminary screening examination employed. In this study out of 200 specimens macroscopically 176 specimens were turbid but microscopy of them 66 specimens failed to reveal presence of pus cell or microorganisms and subsequent culture were sterile. On the other hand, 110 specimens on microscopy revealed presence of pyuria and or microorganisms. Culture yielded growth of organisms. Microscopy showed that specimens which on culture yielded either pure growth of organism or contaminated growth had pyuria and/or organism. Specimens which on culture yielded no growth of organisms had absence of pyuria or microorganism on preliminary microscopy. Few samples from pregnant cases showed presence of pyuria only with accompanying squamous cells indicating contamination probably with vaginal secretion and which on culture showed either no growth of organism or contaminated growth.

Out of 200 specimens examined - 193 showed acidic PH. Only 7 of them showed just alkalinity. On culture of these specimens growth of Klebsiella spp., and Staph Saprophyticus were observed. All of these organisms have the ability to split urea by urase and to render urine alkaline.

Visual appearance and macroscopic examination of the urine specimens remains a moderate guide to the diagnosis of UTIs. Presence of macroscopic turbidity may be due to presence of pus cells or organism from infected urine or from vaginal contamination. Again, it may be due to presence of crystals. Likewise, presence of UTIs have been detected in clear urine specimens or hematuria because it is known that presence of less than 10^5 organism/ml of urine may not appear turbid to the eye[13]. Yet presence of turbidity haziness raise suspicion for presence of UTI. This observation is consistent with the observation made by Flanagan et al (1989)[10.] Although preliminary screening tests (eg. Microscopic examination of unspun urine, gram staining), may be helpful to the diagnosis of UTI yet the culture remains the mainstay for the diagnosis of UTI.

Pregnancy wise analysis of data during this study revealed. Out of 200 specimens examined - 32.5% specimen yielded growth of uropathogen of which 41.5% were from nonpregnant patients and 58.5% were from pregnant patients.

This strongly suggest that UTI is more prevalent in pregnant patients.

Age wise splitting of data obtained from this study reveals that incidence of UTI increases with increasing age group of 15-30years, 30-45 years and 45-60 years of which 30-45 age group predominated. The age group is likely to lead active sexual life.

The reason for high incidence in this age group is not clear, it may be related to pattern of sexual life or of hormonal change or increased intervention to relive obstruction.

The present study shows that the proportion of UTI cases is more in hospitalized patients than the patients attending the different OPDs.

Due to lack of proper and reliable treatment history no definite inference was drawn regrading UTI cases recceing drug treatment.

Data obtained from this study on prevalence of different microorganisms causing UTI in this locality revealed that E. Coli (25.5%) remains the most common organism causing UTI. klebsiella was next in order (5.5%). Other organisms were S. aureus (0.5%), S. faecalis (0.5%). The incidence of S. saprophyticus as uropathogen in this present study was not significant. It was only 0.5%.

According to a study at Hyderabad by Vigg et al (1991)[32] reveals that E. coli was the predominant pathogen 16.2% and Klebsiella aerogenes was the next in order 4.3%. Ps. aeruginosa was 2.7%, Proteus mirabilis in 1%, Strepto faecalis in 2.0% and S. aureus in 1% cases and also few Citrobacter.

The present study reveals the predominance of E. coli, which is consistent with the findings of other studies[9,21] but prevalence of other organism noted in this study was different from other results.[9,34]

As in this study no Proteus, Pseudomonas, Enterobacter was detected. It was also noted that Klebsiella remain the second in order of frequency in this community which is consistent with the data from Vigg et al (1991)[32.]

The data on incidence of specimen contamination and prevalence of contaminants reveals that incidence of contaminated sample was 7%, majority of them were from the nonpregnant patients.

The pattern of leading sexual life may have any relation to it. Of the contaminated specimens 6 yielded mixed profuse confluent growth of organisms having significant colony count 8 other specimens yielded growth of pure colonies of contaminants majority of which was Micrococcus (2.5%) mostly from nonpregnant patients and candida spp (1%), from the pregnant patients occupied the second in order. Due to lack of paired sample follow up of the contaminated specimens could not be done.

The present study revealed the following antibiotic susceptibility pattern of the uropathogens. E. coli showed following sensitivity pattern: Amikacin (100%), Cefotaxime (58.8%), Gentamicin (56.7%). Sensitivity to other drugs are not striking eg- Ciprofloxacin (41.2%), Norfloxacin (41.2%) and shows partial resistances to Ampicillin, Chloramphenicol, Cephalexin, Nalidixic acid, Nitrofurantoin. Klebsiella spp shows sensitivity to Amikacin (72.7%), Cefotaxime (54.5%). It shows partial resistance to Cephalexin, Gentamicin, Norfloxacin, Ciprofloxacin and showed 82% resistance to Nalidixic acid and 90% resistance to Ampicillin.

Staph aureus showed sensitivity to Amikacin, Cephalexin, Chloramphenicol, Ciprofloxacin, Norfloxacin, Nitrofurantoin and resistance to Ampicillin, Erythromycin, Nalidixic acid, Tetracycline.

Staph saprophyticus showed sensitivity to Amikacin, Chloramphenicol, Ciprofloxacin, Norfloxacin and Nitrofurantoin.

In a study by Saini Santosh et al (1983)[25] the isolated coagulase negative Staphylococcus showed following resistance - 58% to penicillin, 60% to cloxacillin, 14% ampicillin, 36% to erythromycin, 16% nitrofurantoin.

In a study by Uttley Anne HC et al (1988)[31] the isolated enterococcus showed resistance to penicillin, erythromycin, Tetracycline, chloramphenicol and also to vancomycin.

In one study Ramani et al (1988)[22] at Manipal university sensitivity of klebsiella isolated to different drugs was Ampicillin (3%), Co-trimoxazole (11%), Mandelamine (11%), Nalidixic acid (68%), Terramycin (1%), Chloramphenicil (26.32%), Erythromycin (2.11%), Furacantin (30.53%), Gentamicin (41.05%), Kanamycin (40%), Streptomycin (20%).

In another study a Hyderabad Vigg et al 1991[32] Nalidixic acid was the most effective drug. Next antibiotic of choice was Nitrofurantoin and Cephalexin remained the third. For the gram-positive isolates, Ampicillin remained most sensitive followed by Nitrofurantoin, Tetracycline and Cephalexin.

In either of the studies no sensitivity data to Quinolones in available, which is available in our study but present study reveals there has been a significant change in the susceptibility pattern of offending organism and they are showing resistance to those earlier antibiotics and also to most of the presently available newer antimicrobial. eg - member of Fluroquinolones, Cephalosporin and remain sensitive to only few members of amino glycosides, eg- Amikacin and third generation Cephalosporin, eg - Cefotaxime.

The reason for this change of sensitivity pattern and emergence of resistance to common cheap drugs is not fully understood. This may be plasmid mediated or due to development of mutants variants or may be due to geographic variation. Other factors may also be responsible. Improper use of antibiotics may be an underlying cause. The results of sensitivity test emphasized the problem of multi drug resistance in our community.

In the present study only 200 specimens of urine were examined. It appears that if a larger number of urine specimens could have been tested, the results would have been more effective. In the present study diagnosis only up to the species was done and due to lack of resources and time serotyping could not be performed.

The present study could not include the detection or isolation of Mycobacterium Tuberculosis and anaerobic organisms. Due to lack of facilities no test for virus or fastidious organisms or other rare or uncommon pathogens could be undertaken. Also, it is expressed that though essential test with paired specimens or test for follow up of the cases could not be undertaken due to lack of facilities. In the absence of these pitfalls the result would have been more yielding and informative.

VIII. Conclusion:

Urinary tract infection still remains an important cause of morbidity and significant health problem. Still most urinary infections are caused by Enterobacteria of which Escherichia Coli still predominate. Other enterobacteria mainly Klebsiella account for most other cases of UTI. Other uropathogens have detected. eg - Staph saprophyticus and their role identified as the of UTI in this community.

During past decades the offending uropathogens remained susceptible to commonly available antimicrobial (eg- Tetracycline, chloramphenicol etc) permitting empirical therapy to start.

But present study reveals that there has been a significant change in the susceptibility pattern of offending organism and they are showing resistance to those earlier antibiotics and also to most of the presently available antimicrobials and remain sensitive to only few members of Aminoglycosides eg- Amikacin and third generation Cephalosporin eg- Cefotaxime and moderately sensitive to Fluoroquinolone eg- Ciprofloxacin. Present Study reveals that if situation demands for urgent therapy for UTI cases of this area, if not contraindicated, empirical therapy with Amikacin can be started.

IX. Recommendation:

Accordingly, it is essential that the UTIs detected at an earlier stage, the uropathogens identified and appropriate therapy can be instituted promptly to avoid complication. But indiscriminate and improper use of antimicrobials is to be avoided, so as to minimize the possibility of emergence of multidrug resistant organisms.

Selection of antimicrobials should be made after in vitro susceptibility tests.

The task is not easy. However, the well-equipped microbiology laboratory, the microbiologists, the laboratory personnel, all must play a major and sincere role in the proper detection of all the etiologic pathogens and appropriate therapeutic programme of the urinary tract infection in patients utilizing common and also sophisticated laboratory techniques. Also, there must be a proper collaboration and communication between the clinician and the personnel of the microbiology laboratory, both of which ideally should be complementary/supplementary to one another's job by providing adequate, relevant and timely information for proper detection and effective treatment of UTI cases.

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