The Prevalence of Uropathogenic *E. Coli* and to Study its Antibiogram Pattern

Samiksha s Mohod¹, S. N Zodpe²

^{1, 2}Department of Microbiology, Shri Shivaji College of Arts, Commerce and Science Akola 444001 (M. S) India

Abstract: The study was aimed to determine The prevalence of Uropathogenic E. coli and to study the Antibiogram pattern in Akola city. A total 51 urine sample were collected from different pathology Laboratory as well as from Government medical hospital Akola. Total of 22 samples from male and 29 samples from Female were collected. The isolates were characterized using conventionl Microbiological method. The study was carried out in the Microbiology laboratory of the Department of Microbiology of Shri Shivaji college of Arts, Commerce and Science, Akola (MH) India The Prevalence of E. coli in the urine sample of UTI patients showed 51 out of 15 sample were positive for E. coli. Antimicrobial susceptibility was performed on all isolated bacteria by Disc diffusion method the multiple antibiotic resistance (MAR) index of each antibiotic was calculated.

Keywords: Uropathogenic E. coli, Multi drug resistant

1. Introduction

Urinary tract infection (UTI) can be defined as the process by which microorganisms growing actively within the urinary tract continuously UTIs is one of the most serious and contagious infectious in human population all around world. Urinary tract infections (UTIs), the second - ranked infectious diseases, are recognized as a big concern relating to global healthcare systems. Urinary tract infections (UTIs) are common human microbial diseases that affect the urinary tract the kidneys, bladder, urethra, and prostate. UTIs are widespread globally with direct and indirect social and economic effects. Moreover, these diseases are becoming an emergent cause of morbidity. It is estimated that UTI affect about t 150 million people each year in the world. The healthcare costs are over \$6 billion. These human diseases are second only to respiratory tract infections. In the United States. UTIs lead to over 1 million medical examinations in the emergency department and cause over 100, 000 hospital admissions each year. However, UTIs can be clustered into community or nosocomial acquired. The first one, community - acquired urinary tract infections (CA - UTI), occur in community or following less than 48 h of hospitalization (Renner, et al., 2017).

Nosocomial urinary tract infections (N - UTI), instead, appear 48 h after hospital admission or three days after discharge. The UTIs distribution in the population changes depending on age, sex, catheterization, hospitalization, and prolonged use of antimicrobials. Bacteria represent the main cause of UTIs, although viruses, fungi, and parasites may be involved in the development of this infection Gram negative bacteria are responsible for 90% of UTI cases, while gram - positive bacteria are responsible forthe other 10%. Previous studies suggest that the most common cause of UTIs is Escherichia coli, which represent 65 - 90% of infections. Other uropathogens that cause UTIs include Enterococcus species, Klebsiella pneumoniae, Citrobacter species Pseudomonas aeruginosa and Staphylococcus coagulase negative (CONS). (Gupta, 2010).

The urinary tract is the most common site of E. coli infection and more than 90% of all uncomplicated urinary tract infections (UTIs) are caused by E. coli infection (Madappa et al., 2014). particularly in women because of the proximity of the urethra to the anus (Gould, et. al 2010). E. coli UTIs are caused by uropathogenic strains of E. coli. The increase in antimicrobial resistance is a worldwide reality that threatens the prevention and effective treatment of an number of infectionschallenging clinical increasing microbiologists and infectious disease specialists (Bihari DJ, 1995). Two of the most common infections are urinary tract (UTI) and intra - abdominal (IAD) infections caused mainly by Enterobacterales, in particular Escherichia coli and Klebsiella species [Meyer KS 1993]. In the 1980s, extended spectrum beta - lactamase (ESBL) producing Enterobacterales were considered one of the leading causes of nosocomial infections and later also of those acquired in the community [a surveillance study 1996]. These enzymes have the ability to hydrolyze beta - lactam antibiotics, including penicillins, cephalosporins and the monobactam aztreonam but not carbapenems (Shah PM 1991). As a consequence, carbapenems were con - sidered the antimicrobials of choice for the treatment of infections caused by ESBL producers. however the prevalence of carbapenemases, enzymes that inactivate them, continue to in - crease worldwide (Buirma et al., 1991). Hence, the current study was carried out to check the incidences of urinary tract infections, identify the responsible bacteria and to check the antimicrobial profile of organisms.

2. Material and Methods

2.1 Collection of sample

Total 51 urine samples were collected from different pathology laboratory, The age of patients included in the study ranged from 12 to 68 years. The patients on antibiotic therapy were also excluded from the study. Early morning mid - stream urine sample of about ten (10) ml were collected using clean and sterilized plastic bottles with air tight screw cap tops. Each urine sample bottle was labelled with a reference code, age, sex, and time of collection. The samples were placed in a cold box in the laboratory. The urine sample collected from various pathology laboratories such as Deshmukh Clinical Laboratory. Akola, Nidan

Volume 12 Issue 5, May 2023 <u>www.ijsr.net</u> Licensed Under Creative Commons Attribution CC BY Pathology Laboratory Icon Hospital, Akola Government Medical College, Akola, Samadhan Pathology. Akola, Pride Diagnostic Laboratory, Jatharpeth road, Akola, Global Pathology Laboratory Akola

2.2 Isolation and Identification of Urppathogenic E. coli

Isolation of E. coli was conducted according to the method. During the process, a sterile wire loop was deep into the sediments of urine sample of the patients and streaked onto the surface of Eosin - Methylene Blue Agar, MacConkey Agar. The procedure was repeated for all the sample and the plates were incubated 37°C for 24 hours. The colonies which produce pink colour colonies were selected. The presumptive colony of E. coli from each plate was further sub - cultured toobtained pure culture.

2.3 Characterisation of Isolates

Further the isolates were confirmed on the basis of various biochemical tests. Gram staining was done according to method. A thin smear was made by emulsifying an overnight culture of the isolate in normal saline on a well labeled clean glass slide. The smear was air dried and fixed by heat. This is followed by flooding the slide with crystal violet as primary stain for 30 seconds and then rinsed the slide with distilled water. The smear was flooded with Lugol's iodine as a mordant to fix the primary stain and then rinsed with distilled water after 60 seconds. The slide was decolorized using acetone and rinsed immediately. Counter stain with safranin followed and left for 30 before being rinsed off. The stain smear was air dried and observed under microscope.

The isolates were confirmed by carbohydrate fermentation test as well as IMVIC Test

Conventional methods were used for identification of organism. On the basis of this test the organisms were tentatively confirmed.

Antibiotic Susceptibility Test -

- 1) Antibiotic susceptibility testing isolates to 6 antibiotic was performed using dise diffusion method.
- 2) Isolates were enriched in nutrient broth for 24 hours after which each strain of E. coli was uniformly swabbed into the individual plates using sterile cotton swabs.
- 3) The standard antibiotic sensitivity discs were then aseptically placed equidistance on using sterile forceps the plates and allowed to stand for 1 hour.
- Sensitivity pattern of the isolates to Gentamicin (10ug), Amplicillin (30ug), Tetracycline (30ug), Ciprofloxacin (10ug), Erythromycin (30ug), Amoxacillin (30ug)
- 5) The plates were then incubated at 37°C for 24hours. After incubation a clear zone of inhibition was measured and results were (Pitout J. D.2012)

3. Results and Discussion

 Table 1: Collection of Urine Samples for Isolation of Uropathogenic E. coli

pathogenic E. coli								
Sr. No.	Sample Collected	Collection Site	Sex	Age				
1	U1	Deshmukh Pathology, Akola	Male	42				
2	U2	Deshmukh Pathology, Akola	Male	38				
3	U3	Deshmukh Pathology, Akola	Male	33				
4	U4	Deshmukh Pathology, Akola	Female	28				
5	U5	Nidan Pathology Lab. Akola	Male	24				
6	U6	Nidan Pathology Lab. Akola	Female	18				
7	U7	Nidan Pathology Lab. Akola	Female	23				
8	U8	Nidan Pathology Lab. Akola	Female	15				
9	U9	Nidan Pathology Lab. Akola	Female	19				
10	U10	Nidan Pathology Lab. Akola	Male	27				
11	U11	Nidan Pathology Lab. Akola	Male	63				
12	U12	Nidan Pathology Lab. Akola	Male	57				
13	U13	Nidan Pathology Lab. Akola	Female	26				
14	U14	GMC, Akola	Male	33				
15	U15	GMC, Akola	Male	53				
16	U16	GMC, Akola	Female	19				
17	U17	GMC, Akola	Female	21				
18	U18	GMC, Akola	Female	61				
19	U19	GMC, Akola	Male	24				
20	U20	GMC, Akola	Female	24				
20	U20	GMC, Akola	Male	39				
21								
	U22	Samadhan Clinical Lab. Akola	Male	68				
23	U23	Samadhan Clinical Lab. Akola	Male	65				
24	U24	Samadhan Clinical Lab. Akola	Male	46				
25	U25	Samadhan Clinical Lab. Akola	Female	27				
26	U26	Samadhan Clinical Lab. Akola	Female	22				
27	U27	Samadhan Clinical Lab. Akola	Female	37				
28	U28	Samadhan Clinical Lab. Akola	Male	56				
29	U29	Pride Diagnostic Lab., Akola	Female	35				
30	U30	Pride Diagnostic Lab., Akola	Female	45				
31	U31	Pride Diagnostic Lab., Akola	Female	7				
32	U32	Pride Diagnostic Lab., Akola	Male	32				
33	U33	Pride Diagnostic Lab., Akola	Female	5				
34	U34	Pride Diagnostic Lab., Akola	Female	39				
35	U35	Pride Diagnostic Lab., Akola	Female	55				
36	U36	Pride Diagnostic Lab., Akola	Female	62				
37	U37	Pride Diagnostic Lab., Akola	Male	41				
38	U38	Pride Diagnostic Lab., Akola	Female	43				
39	U39	Global Pathology Lab., Akola	Female	58				
40	U40	Global Pathology Lab., Akola	Male	62				
41	U41	Global Pathology Lab., Akola	Female	23				
42	U42	Global Pathology Lab., Akola	Male	50				
43	U43	Global Pathology Lab., Akola	Male	40				
44	U44	Global Pathology Lab., Akola	Female	26				
45	U45	Global Pathology Lab., Akola	Female	28				
46	U46	Global Pathology Lab., Akola	Female	18				
47	U47	Global Pathology Lab., Akola	Female	12				
48	U48	Global Pathology Lab., Akola	Male	24				
49	U49	Global Pathology Lab., Akola	Female	31				
50	U50	Global Pathology Lab., Akola	Female	16				
51	U51	Global Pathology Lab., Akola	Male	26				

Table 2: Antibiotic Susceptibility Test against E. coli Isolates

Isolates	Zone of Inhibition in mm					
	Gentamicin	Ampicillin	Tetracycline	Ciprofloxacin	Erythromycin	Amoxiclave
U1	19		21	32	10	
U2	20		12	24	30	
U3	18		10	30	31	
U4	19		16	24	29	

Volume 12 Issue 5, May 2023

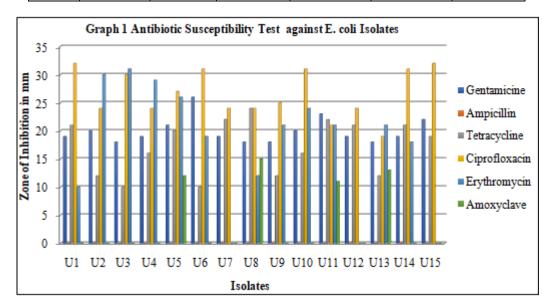
<u>www.ijsr.net</u>

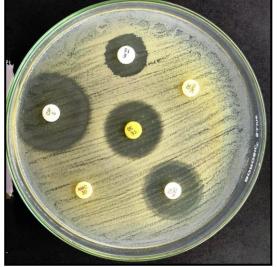
Licensed Under Creative Commons Attribution CC BY

International Journal of Science and Research (IJSR) ISSN: 2319-7064 SJIF (2022): 7.942

U5	21	 20	27	26	12
U6	26	 10	31	19	
U7	19	 22	24		
U8	18	 24	24	12	15
U9	18	 12	25	21	
U10	20	 16	31	24	
U11	23	 22	21	21	11
U12	19	 21	24		
U13	18	 12	19	21	13
U14	19	 21	31	18	
U15	22	 19	32		

XZ - - R





Antibiotics Susceptibility test against E. coli

During the current research work total 51 urine sample were collected form different Pathology Laboratories as well as from Government Medical Hospital Akola (Table No.1). All the samples were collected using clean and sterilized Plastic bottles with air tight screw cap tops. The samples were collected and classified as per the sex and age of the patient. Total 22 samples from male and 29 samples from female were collected. Similarly samples were collected for low age group of 7 years to 68 years age group, to check the prevalence of *E. coli* organism. Similar studied was carried out by Zorc et al., (2005). They reported that many human diseases are as result of infections caused by pathogenic bacteria, either internal or external of the human host. One

of such bacterial infection is urinary tract infection (UTI), involving the presence of bacteria in the urinary tract (UT) which is naturally sterile.

After collection of the sample the study was proceed for the isolation of the organism *E. coli* from urine sample. During the study isolates were grown on various selective media such as EMB Agar and differential media such as MacConkey Agar was used. On EMB agar the greenish Metallic Sheen colour colonies were observed whereas, on MacConkey Agar Pink coloured colonies were observed.

The result for identification of E. coli were observed. The isolates were identified on the basis of Cultural and Morphological Characteristics as well as on the basis of Biochemical characteristics which includes sugar fermentation as well as IMViC Test. In sugar fermentation test, of Glucose, Lactose and Mannitol were checked whereas, four different test were performed for IMViC i. e. Indole, MR, VP, Citrate. Almost all the organism shows acid and gas production positive with the fermentation of Glucose and lactose but it was found to be negative for Mannitol fermentation. In case of most of the isolates shows positive for Indole and Methyl Red test were found to be negative for VP test and Citrate Test.

The antibiogram profile of *E. coli* were studied to check the sensitivity and resistance pattern of *E. coli* (Table no.2). During the study different antibiotic were check such as ciprofloxacin, Tetracycline, Ampicillin, Gentamicin, Erythromycin and Amoxiclave. All the isolates were tested against all the antibiotics and it was observed Ampicillin

Volume 12 Issue 5, May 2023 www.ijsr.net Licensed Under Creative Commons Attribution CC BY shows complete resistant towards the *E. coli* organism, followed by Ampicillin Amoxiclave shows most of the resistant towards the *E. coli*, whereas, Gentamicin, Tetracycline, Ciprofloxacin, Erythromycin showed zone of inhibition and Gentamycin, Tetracycline, Ciprofloxacin and Erythromycin shows sensitivity towards the isolates. Gentamycin shows the maximum zone of 26 mm for U6 followed by Tetracycline show the highest zone of 24 mm for isolates U8, whereas, the list zone of only 10 mm showed by isolate number U3. U1 isolates showed 32 mm of zone against the Ciprofloxacin, were as the list zone of 18 mm was shown by isolate Np. U13. Erythromycin shows the highest zone of 31 mm for isolate U3, whereas the list zone of only 10 mm showed by isolate number U1.

The study was aimed to determine the prevalence and antibiotic Susceptibility pattern of *Escherichia coli* isolated from urinary tract infection patients in Akola city. The judicious use of antibiotic is recommended which will help to limit the increasing rate of drug resistance in the Pathogens.

4. Conclusion

In This Study it has been founded that Gram Negative Bacteria are the commonest Organism isolated from UTI Patient. *E. coli* are one of the leading causes of the urinary tract infection in humans, the finding of this study reviled that E. Coli was observed as the most common etiologic agent of UTI. the Prevalence of UTIs was High in Female then in Males. Antibiotic susceptibility pattern showed that many isolates showed the multiple drug resistance pathogens.

References

- [1] Renner LD, Zan J, Hu LI, Martinez M, Resto PJ, Siegel AC, et al. Detection of ESKAPE bacterial pathogens at the point of care using isothermal DNA - based assays in a portable degas - actuated microfluidic diagnostic assay platform. Applied and Environmental Microbiology.2017; 83 (4): e0244916.
- [2] Gupta et al., 2010 R. K. Gupta, Z. Arany, P. Seale, R. J. Mepani, L. Ye, H. M. Conroe, Y. A. Roby, H. Kulaga, R. R. Reed, B. M. Spiegelman
- [3] Gould D. Causes, prevention and treatment of Escherichia coli infections. Nursing Standard.2010; 24 (31): 50–6.
- [4] Madappa T. Escherichia coli infections. (accessed 29 January 2015).2014. Nicholas D Maynard, Robert C Mason, David J Bihari, Mark N Smithies, R Neil Dalton Chest 108 (6), 1648 - 1654, 1995
- [5] Nicholas D Maynard, Robert C Mason, David J Bihari, Mark N Smithies, R Neil DaltonChest 108 (6), 1648 -1654, 1995
- [6] A surveillance study 1996
- [7] John Bycroft, Rizwan Hamid, Helen Bywater, Prasad Patki, Michael Craggs, Julian Shah Neurourology and urodynamics 23 (3), 252 - 256, 2004
- [8] RJA Buirma, AM Horrevorts, JHT WagenvoortScand J Infect Dis 78 (Suppl), 35 - 44, 1991