"TO STUDY THE PREVALENCE OF URINARY TRACT INFECTION ITS BACTERIOLOGICAL PROFILE AND THE DRUG RESISTANCE PATTERN OF THE PATIENTS WITH THE MOLECULAR CHARACTERIZATION OF MECA GENE IN MRSA ISOLATES AT A TERTIARY CARE CENTRE IN ANDHRA PRADESH, INDIA".

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Abstract

Introduction: Urinary tract infections (UTIs), one of the main causes of morbidity and comorbidity in patients with underlying conditions, account for the majority of hospital visits globally. The emergence of antibiotic-resistant bacterial strains is a serious problem and greatest challenge in public health care wherein the emergence of methicillin-resistant *Staphylococcus aureus* (MRSA) in the community setting and infections with this pathogen become a prevalent problem among UTI patients.

Aim and Objective: To study the prevalence of urinary tract infection its bacteriological profile and the drug resistance pattern of the Patients with the molecular characterization of MecA gene in MRSA isolates at a tertiary care centre in Andhra Pradesh, India.

Material and Methods: This was a Cross sectional study carried out in the Department of Microbiology at Maharajah Institute of Medical Sciences, Vizianagaram for a period of 1 year i.e, between April 2022 to April 2023. A total of 526 freshly voided mid- stream urine sample were collected in a sterile wide mouth container from the individuals preliminary routine urine tests positive for pus cells and albumin. All the urine samples were processed within one hour after the collection for aerobic bacterial culture. If delayed, samples were refrigerated and processed within 4 - 6 hours. The identification , biochemicals and the AST pattern was done accoding to the CLSI guidelines 2022. The DNA was extracted by using Qiagen DNA Extraction kit, which was further proceeded for the MecA gene detection in MRSA isolates by the conventional PCR.

Results: In the present study a total of 526 urine samples were received out of which 120 (22.8%) urine samples were showing significant growth for UTI. The ratio of females 71 (59.16%) were more as compared to that of the males 49 (40.83%) with the maximum age of 21-30 (40.83%) been affected the most followed by 31-40 (23.33%) years of age and least in the age group above 61 years of age. It was noted that the maximum number of isolates were from the gram negative isolates as compared to the gram positive isolates. It was observed that the maximum number of isolates were from the *E.coli* 56 (46.66%) followed by *Klebsiella pneumonia* 21 (17.5%) and least for *Proteus vulgaris, Acinetobacter baumannii*. Among the gram positive isolates the *S.aureus* (7.5%) was observed to be the maximum with 3 (33.33%) isolates resistant for MRSA . Our study showed a very high rate of resistance (>70%) among *E. coli* isolates to piperacillin. Among *Klebsiella* isolates, no resistance was found for meropenem and low resistance was found for ciprofloxacin (9.52%), norfloxacin(9.52%) , and cefotaxime(23.80%) but high for nitrofurantoin (95.23%) and trimethoprim/sulfamethoxazole (42.85%) . The molecular characterization of MRSA confirmed the detection of MecA gene among the UTI patients.

Conclusion: The phenotypic and genotypic studies are needed to establish and clarify the genetic mechanism behind antibiotic resistance and prevalence. Therefore, the misuse of antibiotics and point to the need for better prescription practices should be followed by regular examinations and thorough adherence to antibiotic stewardship programmes can lower the cost of UTI prevention**.**

Keywords: UTI, Antibiotic sensitivity testing, CLSI, Molecular characterization, Mec A gene

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INTRODUCTION

Urinary Tract Infection (UTIs) are one of the most common bacterial infection in routine clinical practice, clinical presentation of which ranging from asymptomatic to severe sepsis. UTI is one of the most important causes of morbidity in general population, and is the second most important cause of hospital visits. It also contributes as the most common nosocomial infection in many hospitals and accounts for approximately 35% of all hospital-acquired infections [1].

Urinary tract infections (UTIs) are the inflammatory disorders of the urinary tract caused by the abnormal growth of pathogens [2,3]. Urinary tract infection is known to cause short-term morbidity in terms of fever, dysuria, and lower abdominal pain (LAP) and may result in permanent scarring of the kidney [4]. Urinary tract infections can be community acquired or nosocomial. Community-acquired urinary tract infections (CA-UTIs) are defined as the infection of the urinary system that takes place in one's life in the community setting or in the hospital environment with less than 48 hours of admission. Nosocomial urinary tract infections (N-UTIs) are the infection of the urinary tract that occurs after 48 hours of hospital admission, and the patient was not incubating at the time of admission or within 3 days after discharge [5].

Urinary tract infections may be asymptomatic, acute, chronic, and complicated or uncomplicated, and the clinical manifestations of UTIs depend on the portion of the urinary tract involved, the etiologic organisms, the severity of the infection, and the patient's ability to mount an immune response to it [6]. The symptoms of UTIs such as fever, burning sensations while urinating, LAP, itching, formation of blisters and ulcers in the genital area, genital and suprapubic pain, and pyuria generally depend on the age of the person infected and the location of the urinary tract infected [3].

UTIs are caused by both Gram-negative and Gram-positive bacteria, as well as by certain fungi. There are many bacteria responsible of causing UTI infection like *E.coli*, *Klebsiella pneumoniae*, *Staphylococcus saprophyticus*, *Enterococcus faecalis*, group B *Streptococcus* (GBS), *Proteus mirabilis*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Candida* spp [7-10].

The incidence of urinary tract infection is increasing because patients are more frequently fitted with various urinary catheters as endourology progresses technologically. In complicated urinary tract infections and hospitalized patients, Gram positive bacteria such as MRSA are comparatively more common [11].

The incidence of urinary tract infection caused by MRSA is increasing because patients are more frequently fitted with various urinary catheter [12]. MRSA is an emerged pathogen that is able to withstand the effect of different antimicrobials especially some strains which produce a biofilm both in hospital admitted patients as well as in the community, so rapid identification of this resistance can help to eliminate the infection effectively. Because of its ability to acquire resistance genes, *S. aureus* becomes resistant to broad types of antibiotics [13,14].

UTI is treated with broad spectrum antibiotics empirically to start with, which are de-escalated to specific antibiotic based on information obtained from the antimicrobial susceptibility pattern of the urinary pathogens [15]. Widespread use of antimicrobial agents has lead to the emergence of antibiotic resistant pathogens also there is increase demand for new drugs. Distribution of uropathogens and their antimicrobial sensitivity patterns may differ regionally so it becomes necessary to study these and compile their data in particular settings [16].

Drug resistance in bacteria has become a global health problem and an emerging threat due to misuse of antibiotics [17,18]. Therefore, the present study was undertaken to study the prevalence of urinary tract infection its bacteriological profile and the drug resistance pattern of the patients with the molecular characterization of MecA gene in MRSA isolates at a tertiary care centre in Andhra Pradesh, India.

MATERIAL AND METHODS

This was a Cross sectional study carried out in the Department of Microbiology at Maharajah Institute of Medical Sciences, Vizianagaram for a period of 1 year i.e, between April 2022 to April 2023. A total of 526 freshly voided mid- stream urine sample were collected in a sterile wide mouth container from the individuals preliminary routine urine tests positive for pus cells and albumin. All the urine samples were processed within one hour after the collection for aerobic

bacterial culture. If delayed, samples were refrigerated and processed within 4 - 6 hours.

The patients presenting or highly suspcious of having UTIs and ready to give consent were included in the study. Any patient who was terminally ill, who fails to give urine samples, with a history of antibiotic administration in the last two weeks and any female who was in their menstruation period were excluded from the study [19].

The patients demographic details including age, gender, tribe, residence, level of education, and history of medical conditions were included in the study.

Microscopic Study

One of the diagnosis criteria of UTI was based on microscopic findings of more than 10 pus cells/ high power field (40×) in urine were included in the study.

Collection and process of urine samples

Mid-stream urine samples were collected in a sterile container and were processed within 2 h of collection time. These urine samples were also centrifuged and urine sediment was used for direct microscopic examination of red blood cells (RBCs), leukocytes, epithelial cell, casts, crystals, and parasites. In the normal urine sediment, a few count of RBCs, pus cells (0–5/high power field), and epithelial cells may present. Epithelial cell count reported as "few," "moderate," or "many" per low-power field.

Isolation and Identification of Uropathogens

The Urine sample was inoculated on a standard culture media Cystine–Lactose– Electrolyte-Deficient (CLED) agar using a calibrated (1 µL) loop.

Culture plates were incubated at 35–37°C ambient air incubator for 18 h. After the allocated time period, the culture plates were visualized for the presence of bacterial colonies. They were reported as significant or non-significant growth on the basis of colony count method. Isolated colonies were further characterised based on cultural characteristics by growing on differential media, such as MacConkeys agar and blood agar [20]. Further, the isolates were identified by cultural, morphological and biochemical tests. The method used in the identification and characterisation of isolated bacteria included Gram staining, motility test and biochemical tests like, TSI and IMViC according to Cheesbrough [21, 22]. Isolated and characterized uropathogens were then preserved in nutrient broth containing 25% glycerol at −20°C.

Following the recommendations of Kass [23] in distinguishing genuine infection from contamination, culture of a single bacterial species from urine sample at a concentration of >105 CFU/ml. Only a single positive culture per patient was included in the analysis.

The plates were incubated at 37°C for 24 hrs and extended to 48 hrs in culture (growth) negative cases. The identification , biochemicals and the AST pattern was done accoding to the CLSI guidelines 2022 [24]. All chemicals required for culture media and reagents were procured from HiMedia laboratories Pvt Ltd., Mumbai.

Molecular Detection of MecA Gene by Polymerase Chain Reaction (PCR) in Methicillin resistant *Staphylococcus aureus*

Bacterial DNA was extracted by QIAamp DNA Kit by following manufactures guidelines. *S. aureus* previously extracted DNA was used for the amplification of Mec A gene. The primers were purchased from "**Saha gene'** and was reconstituted with sterile double distilled water based on the manufacturer's instruction.

Figure No.1: The DNA Extraction kit Figure No.2: The Reagents used for the DNA Extraction

Figure No.3: The MecA gene primers from the Saha gene Figure No. 4: Run of Amplified product

All *S. aureus* isolates that were resistant to cefoxitin 30 µg and positive on ORSAB examination were then subjected to a PCR test to detect the presence of the mecA gene [25]. The DNA extraction process was carried out according to the QIAamp DNA Mini Kit protocol, where previously the isolates were purified on MSA (HiMedia Pvt. Ltd, M118) and inoculated on MHA (Oxoid, CM0337).

Gene	Primer	Base Pair	Reference
MecA	F: 5′-AAA ATC GAT GGT AAA GGT TGG C-3′ R: 5′-AGT TCT GCA GTA CCG GAT TTG C-3′.	533	[26]

Table No. 1: Primer used for the MecA gene detection

Molecular Characterization of MecA gene

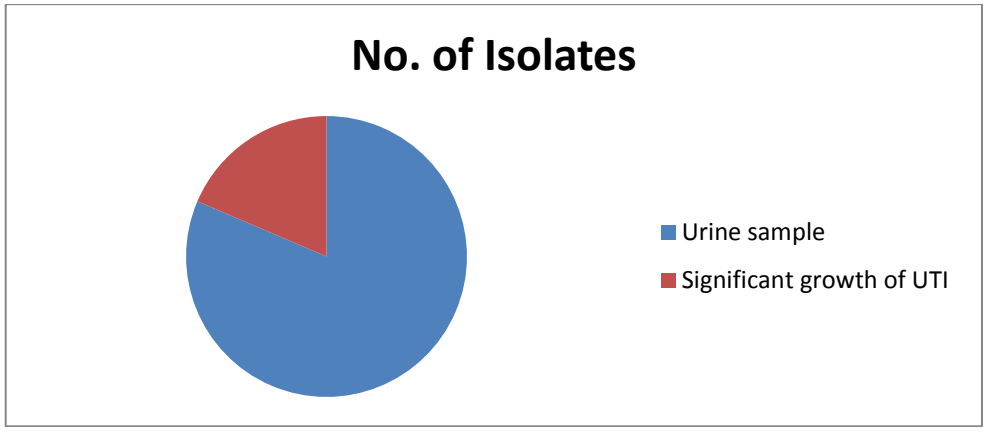
The PCR master mix used GoTaq Green Master Mix (Promega, 9PIM712) which is a ready-to-use solution mixture containing Taq DNA polymerase, dNTPs, $MgCl_2$, and a reaction buffer. DNA was amplified using a Thermal Cycler T100 machine (Bio-Rad, 186-1096) for 40 cycles in 25 µl of the reaction mixture with the following steps: denaturation at 94°C for 30 seconds, annealing at 55°C for 30 seconds, and extension at 72°C for 1 min with a final extension at 72°C for 5 min. A total of 10 µl of PCR product were analyzed by 2% agarose gel electrophoresis, and the gel was visualized under ultraviolet light [27]. A positive test indicated a PCR product in the 533-base pair (bp) band [27].

RESULTS

A total of 526 urine samples were received in the Microbiology Laboratory at Maharajah Institute of Medical Sciences, Vizianagaram , out of which 120 (22.8%) urine samples were showing significant growth for UTI [Table No. 2]. . The ratio of females 71 (59.16%) were more as compared to that of the males 49 (40.83%) [Table no. 3] . The maximum age of 21-30 (40.83%) was affected the most followed by 31-40 (23.33%) years of age and least in the age group above 61 years of age [Table no.4].

Type of Clinical Isolates	Number of Isolates	Percentage
Urine samples	526	77.2%
Significant growth for UTI	120	22.8%

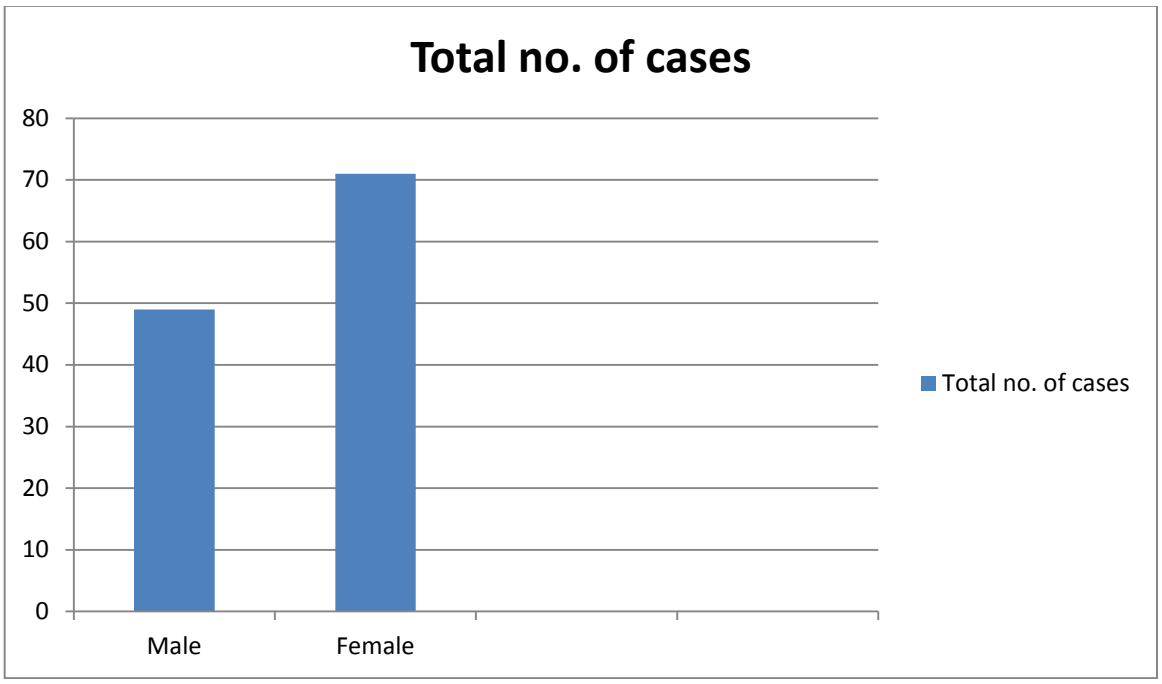
Table No. 2 : Samplewise distribution of the clinical isolates



Graph No. 1: Graphical Representation of Samplewise distribution of the clinical isolates

Gender	Total no. of Cases studies (N=120)	Percentage
Male	49	40.83%
Female	71	59.16%

Table No. 3 : Genderwise distribution of the UTI cases



Graph No. 2: Graphical Representation of Genderwise distribution of the UTI cases

S.N.	Age group (Years)	Male N= 49	Female N= 71	Percentage (%)
1.	0-10	-	-	-
2.	11-20	9	8	14.16%
3.	21-30	17	32	40.83%
4.	31-40	12	16	23.33%
5.	41-50	5	7	10%
6.	51-60	3	5	6.66%
7.	61-70	2	4	5%
8.	≤ 80	1	-	0.83%

Table No. 4 : Agewise distribution of the UTI cases

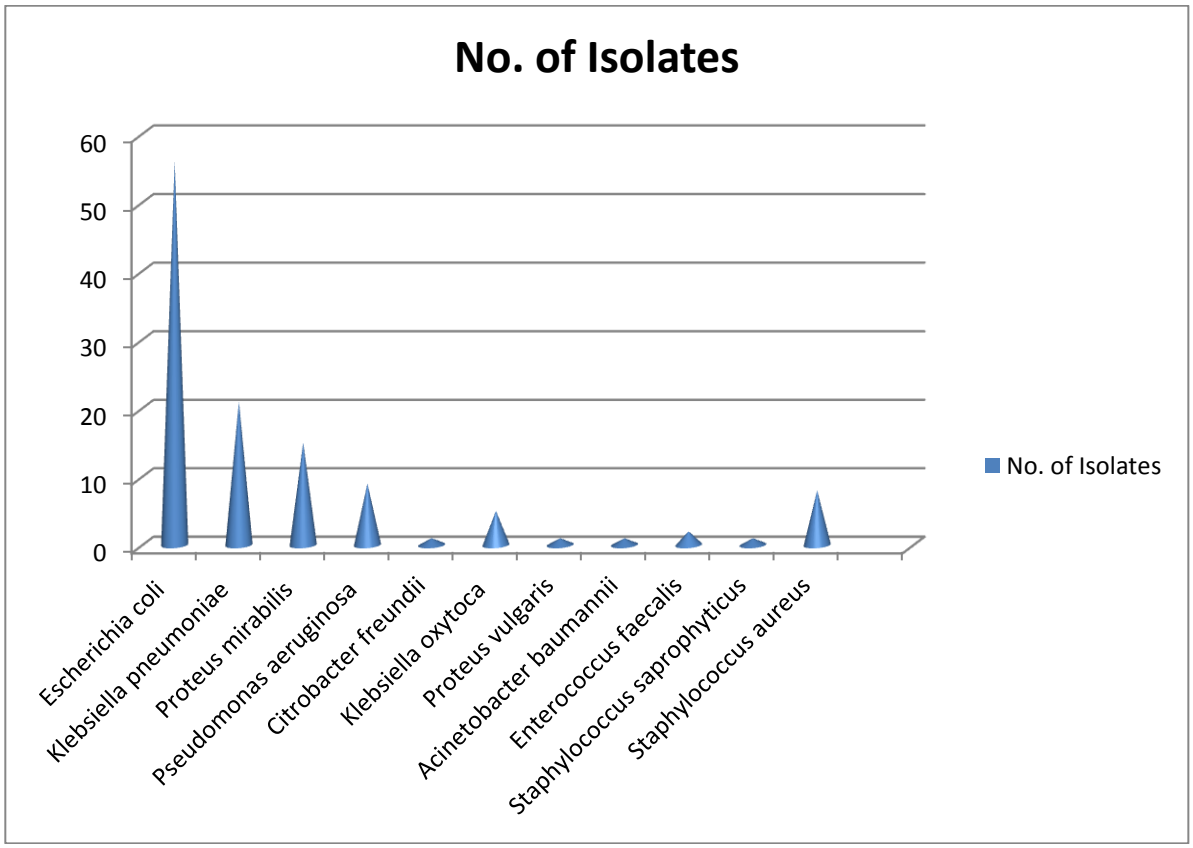
It was noted that the maximum number of isolates were from the gram negative isolates as compared to the gram positive isolates.

It was observed that the maximum number of isolates were from the *E.coli* 56 (46.66%)

followed by *Klebsiella pneumonia* 21 (17.5%) and least for *Proteus vulgaris, Acinetobacter baumannii, Staphylococcus saprophyticus* with 1 (0.83%). Among the gram positive isolates the *S.aureus* (7.5%) was observed to be the maximum with 3 (33.33%) isolates resistant for MRSA isolates [Table no.5].

Type of Organism Isolated	**No. of Isolates**	**Percentage**
Escherichia coli	56	46.66%
Klebsiella pneumoniae	21	17.5%
Proteus mirabilis	15	12.5%
Pseudomonas aeruginosa	9	7.5%
Citrobacter freundii	3	2.5%
Klebsiella oxytoca	2	1.6%
Proteus vulgaris	1	0.83%
Acinetobacter baumannii	1	0.83%
Enterococcus faecalis	2	1.6%
Staphylococcus saprophyticus	1	0.83%
Staphylococcus aureus	9	7.5%
Total	**120**	**100%**

Table No. 5: The Frequency of bacteria isolated from the UTI cases



Graph No. 3: Graphical Representation of Frequency of bacteria isolated from the UTI cases

In the present study it was also observed that high degree of drug resistance among bacterial isolates was observed . Our study showed a very high rate of resistance (>70%) among *E. coli* isolates to piperacillin. Among *Klebsiella* isolates, no resistance was found for meropenem and low resistance was found for ciprofloxacin (9.52%), norfloxacin(9.52%) , and cefotaxime(23.80%) but high for nitrofurantoin (95.23%) and trimethoprim/sulfamethoxazole (42.85%) .

Antibiotics	***Escherichia coli***	***Klebsiella pneumoniae***	***Proteus mirabilis***	***Pseudomonas aeruginosa***
Ampicillin	53 (94.6%)	21 (100%)	11 (73.33%)	IR
Amoxicillin/clavulanic acid	29 (51.7%)	13 (61.9%)	4 (26.66%)	IR

Piperacillin	50 (89.2%)	20 (94%)	4 (26.66%)	9 (100%)
Piperacillin/tazobactam	19(33.92%)	7 (33.33%)	4 (26.66%)	9(100%)
Cefalotin	42 (75%)	7 (33.33%)	5 (33.33%)	IR
Cefuroxime	6 (10.71%)	7 (33.33%)	5 (33.33%)	IR
Cefoxitin	5 (8.92%)	2(9.52%)	4 (26.66%)	IR
Cefpodoxime	5 (8.92%)	5 (23.80%)	4 (26.66%)	IR
Cefotaxime	2(3.57%)	5 (23.80%)	4 (26.66%)	IR
Ceftazidime	3 (5.3%)	5 (23.80%)	4 (26.66%)	9 (100%)
Cefepime	3 (5.3%)	5 (23.80%)	4 (26.66%)	7 (77.7%)
Meropenem	5(8.92%)	0	0	5 (55.5%)
Amikacin	9 (16.07%)	3 (14.28%)	0	2 (22.22%)
Gentamicin	5 (8.92%)	3 (14.28%)	0	2 (22.22%)
Tobramycin	10 (17.85%)	3 (14.28%)	0	2 (22.22%)
Ciprofloxacin	19 (33.92%)	2 (9.52%)	0	2 (22.22%)
Norfloxacin	19 (33.92%)	2 (9.52%)	4 (26.66%)	2 (22.22%)
Nitrofurantoin	5 (8.92%)	20 (95.23%)	IR	-
Trimethoprim/sulfamethoxazole	30 (53.5%)	9 (42.85%)	4 (26.66%)	IR
Total number of isolates	**56**	**21**	**15**	**9**

Table No.6 : Number (%) of common Gram-negative urinary pathogens resistant (R) to antimicrobial agents

Antibiotics	***Citrobacter freundii***	***Klebsiella oxytoca***	***Proteus vulgaris***	***Acinetobacter baumannii***
Ampicillin	IR	IR	IR	IR
Amoxicillin/clavulanic acid	IR	2 (100%)	1(100%)	IR
Piperacillin	3 (100%)	2 (100%)	1 (100%)	0
Piperacillin/tazobactam	1 (33.33%)	2 (100%)	1 (100%)	0
Cefalotin	IR	2 (100%)	IR	IR
Cefuroxime	IR	IR	IR	IR
Cefoxitin	IR	1 (50%)	1 (100%)	IR
Cefpodoxime	IR	2 (100%)	1 (100%)	-
Cefotaxime	2 (66.66%)	2 (100%)	1 (100%)	0
Ceftazidime	2 (66.66%)	2(100%)	1 (100%)	0
Cefepime	1 (33.33%)	2 (100%)	1 (100%)	0
Meropenem	0	1 (50%)	0	0
Amikacin	2 (66.66%)	2 (100%)	0	0
Gentamicin	2 (66.66%)	1 (50%)	0	0
Tobramycin	2 (66.66%)	2 (100%)	0	0
Ciprofloxacin	3(100%)	2(100%)	1 (100%)	0
Norfloxacin	3 (100%)	2 (100%)	1 (100%)	0
Nitrofurantoin	2(66.66%)	1 (50%)	IR	-
Trimethoprim/sulfamethoxazole	1 (33.33%)	1 (50%)	1 (100%)	1 (100%)
Total number of isolates	**3**	**2**	**1**	**1**

Table No. 7: Number (%) of less common Gram-negative urinary pathogens resistant (R) to antimicrobial agents

Antibiotics	***Enterococcus faecalis***	***Staphylococcus saprophyticus***	***Staphylococcus aureus***
Benzyl penicillin	2 (100%)	1 (100%)	9 (100%)
Cefoxitin	-	0	3 (33.33%)
Gentamicin	IR	0	0
Tobramycin	IR	0	0

Levofloxacin	0	0	3 (33.33%)
Clindamycin	IR	0	0
Linezolid	0	0	0
Teicoplanin	0	0	0
Vancomycin	0	0	0
Fosfomycin	2 (100%)	IR	0
Nitrofurantoin	0	0	0
Rifampicin	2 (100%)	0	0
Trimethoprim/Sulfamethoxazole	IR	0	0
Total number of isolates	**2**	**1**	**9**

Table No.8 : Number (%) of common Gram-positive urinary pathogens resistant (R) to antimicrobial agents

The frequency of *Acinetobacter*, *Citrobacter*,*Klebsiella* and *proteus* is mentioned in Table 7 due to their clinical relevance. The number (percentage) of common Gram-negative urinary pathogens resistant (R) to antimicrobial agents is shown in Table 7 and Table 8 refer to the Gram positive isolate .

The DNA Extraction was performed by the Qiagen DNA kit and the DNA was isolated from the samples. From the 9 isolates of *S.aureus*, there were 3 isolates observed positive for MRSA. The DNA Extraction and the gene detection of the isolate 3 isolates was performed for the detection of Mec A gene among the MRSA isolates.

Figure No. 5: The DNA Extraction

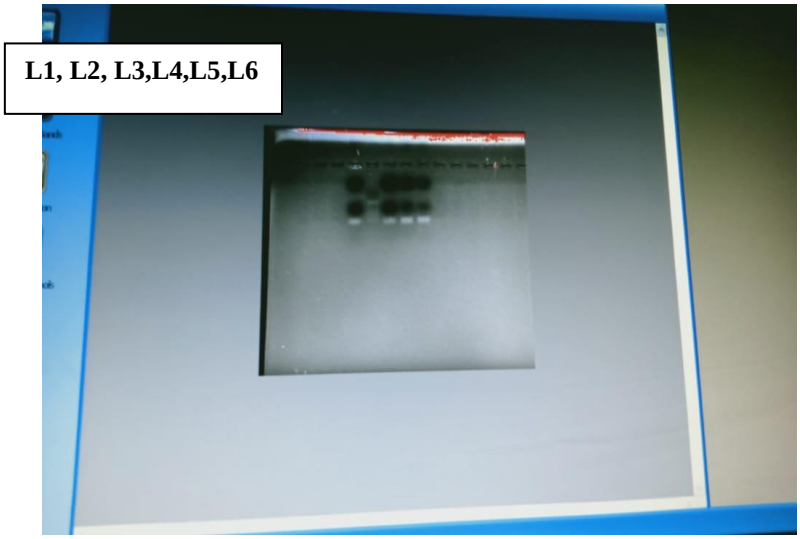


Figure No. 6: Photograph of amplified MecA gene in MRSA; the amplified DNA band size was obtained 533bp, L1 corresponds to the Negative control and L2 corresponds to the Positive control , L3 corresponding to 100bp ladder used ;Lane 4-6 are the sample positive for MecA

In the current study it was observed that there were 3 isolates found positive for the MecA gene.

GTTGTAGTTGTCGGGTTTGGTATATATTTT
TATGCTTCAAAAGATAAAGAAATTAATAA
TACTATTGATGCAATTGAAGATAAAAATTT
CAAACAAGTTTATAAAGATAGCAGTTATA
TTTCTAAAAGCGATAATGGTGAAGTAGAA
ATGACTGAACGTCCGATAAAAATATATAA
TAGTTTAGGCGTTAAAGATATAAACATTC
AGGATCGTAAAATAAAAAAAGTATCTAAA
AATAAAAAACGAGTAGATGCTCAATATAA
AATTAAAACAAACTACGGTAACATTGATC
GCAACGTTCAATTTAATTTTGTTAAAGAAG
ATGGTATGTGGAAG

TCTAAAAAGCATGTAAAAGAATTTGCGAC
CAGATTGCAAAATCTGCAACGAGCTTTGG
GTTTACTCCCCCCGGTGGAGATGGATATA
AAAATGCTCAAAAAAGTACCACCACTATA
TTTTCCTAAGAAGCTATCAAATAATTATAA
TCA

Figure No.7: Obtained gene sequences of MecA gene in *S. aureus*

DISCUSSION

Urinary tract infections (UTIs) are amid the most critical infections observed globally. UTIs include varieties of disorders, such as urethritis, cystitis, and pyelonephritis. Reports showed that 50% of women had a history of UTIs in their lives. UTIs are thoughtful health issues that concluded 150 million individuals globally yearly [28]. Reports showed that bacteria are the most common cause of UTIs. However, the *Staphylococcus aureus* is not documented as a major pathogen responsible for the occurrence of UTIs, but its prevalence has been increased in recent investigations [29].

In the present study the prevalence of UTI was found to be 22.8%. This finding was similar to the study performed by the other authors Ahmad S et al and Suhail A. et al., where the prevalence was found to be 20.54% and 32% respectively [30,31] .

In the current study the maximum number of isolates were from the Females 71 (59.16%) as compared to that of the males 49 (40.83%). This study was similar to the study by Suhail A. et al, and Martin Odoki et al., in 2019 where the ratio of females was more as compared to the males [31] [32].

It was noted that the maximum number of isolates were from the gram negative isolates as compared to the gram positive isolates. It was observed that the maximum number of isolates were from the *E.coli* 56 (46.66%) followed by *Klebsiella pneumonia* 21 (17.5%) and least for *Proteus vulgaris, Acinetobacter baumannii* with 1 (0.83%). Among the gram positive isolates the *S.aureus* (7.5%) was observed to be the maximum with 3 (33.33%) isolates resistant for MRSA. Similar study was performed by the other research workers where among 206 bacterial isolates obtained from 417 urine samples, majority of the isolates (99%) were Gram negative bacteria which

included *Escherichia coli* (56.79%), *Klebsiella sps* (19.9%), *Pseudomonas sps* (6.3%), *Proteus sps* (5.8%), *Enterobacter sps* (3.8%), *Citrobacter sps* (1.4%), *Enterococcus sps* (0.9%), and other *NFGNB* (4.8%) [33].

In the present study it was found that antimicrobial resistance was seen both in Gram-positive and Gram-negative bacteria. Multiple resistances were high among the isolated urinary pathogens.

Staphylococcus aureus is a significant human pathogen responsible for most cases of nosocomial and hospitalacquired infections. It is responsible for the occurrence of several diseases, including UTIs, respiratory and soft tissue infections, endocarditis, osteomyelitis, and endocarditis . The bacterium has an emergence of severe antimicrobial resistance. Clinical experiences showed that around 50% of the *S. aureus* isolates harbored complete resistance toward penicillins and cephalosporins groups of antimicrobials which called them methicillin-resistant *S. aureus* (MRSA). MRSA strains caused complicated diseases for a more extended period with a higher economic burden due to hospitalization and treatment [34].

In the present study it was observed that the rate of MRSA among the UTI cases were 2.5%. This study was in support with the study performed by the other author Khaleel R et al., [35] where 7.7% of the urine specimens of hospitalized patients who suffered from UTIs were positive for the MRSA strains. MRSA isolates displayed a boost resistance rate toward erythromycin, ceftaroline, penicillin, gentamicin, and ciprofloxacin antimicrobial agents. Additionally, MRSA isolates harbored a boost distribution of beta lactamase gene MecA antimicrobial resistance-encoding genes. It seems that the antimicrobial-resistant MRSA isolates may be an emerging cause of UTIs in Andhra Pradesh. Similarly, Lunacek et al [36] labelled that the MRSA prevalence amongst urine specimens in Austria was 4.06%. They disclosed that MRSA isolates were resistant toward cephalosporin, aminopenicillin, penicillin G, carbapenem, and β-lactamase antimicrobial agents. They also presented that catheter utilization is the most critical risk factor for MRSA occurrence in UTIs. An Irish survey [37] described that the prevalence of MRSA strains was 27.9%. Besides, MRSA isolates of the urine specimens displayed the uppermost resistance rate toward flucloxacillin (100%), co-amoxiclav (100%), and ciprofloxacin (98%).

Our study showed a very high rate of resistance (>70%) among *E. coli* isolates to piperacillin. Among *Klebsiella* isolates, no resistance was found for meropenem and low resistance was found for ciprofloxacin (9.52%), norfloxacin(9.52%), and cefotaxime(23.80%) but high for nitrofurantoin (95.23%) and trimethoprim/sulfamethoxazole (42.85%) . This resistance is most likely due to the massive use of third-generation cephalosporins and fluoroquinolone antibiotics in UTIs patients. The high resistance in trimethoprim/sulfamethoxazole susceptibility pattern may be due to non-judicious use and over-the-counter selling of this antibiotic [31].

The antibiotic susceptibility of uropathogenic bacteria is known to change with time and is inconsistent in different regions [38]. Here, we have described the impact of the best antimicrobials with low resistance rate (overall resistance %) against the uropathogens in this study. The best antimicrobials for Gram-negative organisms was meropenem, amikacin , gentamicin , tobramycin , and cefepime and moderate resistance rate were ciprofloxacin , cefotaxime , cefoxitin , norfloxacin , ceftazidime , cefpodoxime , piperacillin/tazobactam ,
and cefuroxime It was noteworthy that high resistance rate was found to be against cefuroxime , trimethoprim/sulfamethoxazole , nitrofurantoin , amoxicillin/clavulanic acid piperacillin and ampicillin .

The current finding is similar to other reports which suggest that gram negative bacteria, particularly *E. coli* was the commonest pathogens isolated from patients with UTI [39,40]. The incidence of *E. coli* in our study was higher when compared with the Nigerian studies reporting 42.10% [41] and 51% [42]. Most of the studies conducted in Africa and Arab countries showed less than 50% isolation of *E coli* from the UTI patients but reported a higher percentage (29%) of *S aureus* as second most frequently isolated bacteria from UTI cases. Reports from other developing or developed countries were the isolation of Gram positive bacteria as uropathogen is very low <10% [43,44].

In contrast, the antimicrobial sensitivity pattern of antimicrobials for Gram-positive organisms shows linezolid , teicoplanin , vancomycin , cefalotin

screen , moxifloxacin , nitrofurantoin , and levofloxacin were sensitive however there were 33.33% resistance observed for cefoxitin.

The overall proportion of MRSA among isolated *S. aureus* in this study was elevated from previous studies conducted in Iran (25.8%) [45] Nepal (30.8%) [46] Uganda (33.3%) [47] and Nigeria (13%) [48].This increased proportion of MRSA might be due to differences in geographic area, MRSA becoming a global nosocomial pathogen with rapid spread to health care as well as community and urinary tract infection associated factors may also play an important role in increasing the prevalence of MRSA in the community

The etiology of bacteria causing UTI as well as their susceptibility to antimicrobials continue to vary over time period and it is different among different countries [49].

To successfully treat the patients who are suffering from UTI, it is crucial to accurately identify the causative pathogen. Failure to do so will not only prolong the disease and will render the patient to complications but will also promote negative consequences of bacterial resistance due to a non-judicious use of inappropriate antibiotics.

CONCLUSION

The drug resistance in bacteria has become a global health problem and an emerging threat due to misuse of antibiotics. Hence, it is necessary to be aware of the changes in the spectrum of drug resistance to ensure appropriate treatments.

The Effective methods of infection prevention control should be utilized to help reduce the high prevalence of *S. aureus* and MRSA infections. Further phenotypic and genotypic studies are needed to establish and clarify the genetic mechanism behind antibiotic resistance and prevalence. However, further surveys should perform to assess other epidemiological features of MRSA in UTIs.

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