Original Article

Isolation and Identification of UTI Causing Agents and Frequency of ESBL (Extended Spectrum Beta Lactamase) in Pakistan

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ABSTRACT

Objective: The objectives of the current study were to isolated and identified UTIs causative agents. To identify the frequency of ESBL producing microorganisms. These findings will help to manage UTIs infections in Pakistani population.

Methods: A total of 1050 outdoor patient's samples were collected by the mid-stram methods. The samples were tested microbiologically by using standard procedure. Antibiotic susceptibility of the isolated pathogens was tested for commonly-used antibiotics by Kirby-Bauer technique according to NCCLS guidelines.

Results: Out of these 225 (21%, $p < 0.05$) samples were found positive for urinary tract infection (UTI). Prevalnace was higher in female 126 (56%, $p < 0.05$) then male 99 (44%, $p \leq 0.01$) with age group of 41- 50 years. Among total 199 gram negative isolates 121 (61%) were ESBL positive *E Coli* 68 (56%) was common followed by *Pseudomonas aerogenosa* 22 (18%), *Klebsiella sp.* 21 (17%) and *Proteus sp*. 10 (8%). Antibiotic susceptibilities of ESBL producing isolates were resistant to Levofloxacin 97 (80%), Tobramycin 72 (60%) and pipemidic acid 73 (60.3%) as compared to ESBL non-producing isolates.

Conclusion: Overall prevalence of UTI observed in our study is 21%. *E. coli* was most prevalent than other organism, most common in female than male age group of 41- 50. ESBL positive were highly resistant to antibiotics as compared to Non-ESBL. This high prevalence of resistant ESBL posing a major clinical crisis of treatment failure with β-lactam antimicrobials. ESBL detection and their antibiotic susceptibility checking should be included in every pathological laboratory to restrict the over and misuse of the

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antibiotics and to improve the treatment and management of UTI.

Keywords: *E. coli*, *Klesiella pneumonia*, Proteus, ESBL, Antibiotics.

INTRODUCTION

Urinary Tract Infections (UTI's) are the most prevalent infections in all the geographical regions of the world causing a great number of morbidity and mortality among all the age groups[1]. In the world about 150 million urinary tract infections are reported per annum and nearly 10% people experience UTI at least once during their lifetime[2]. A worldwide estimate indicated that six million patients visit hospitals for treatment of UTI and about 300,000 are treated in the wards every year[3] UTI treatment is costing the global economy in excess of 6 billion US dollars[4]. The main causes associated with urinary tract infections are malnutrition, poor hygiene and low socio-economic status[5].

Urinary tract infections also cause complication in pregnancy and other diseases as diabetes mellitus, polycystic kidney disease, sickle cell anaemia and renal transplantation. UTI incidence varies with respect to gender; race and age females have 3 to 7 fold more risk of UTI incidence than male[5]. Male babies are more prone to UTI during first year of life while, female babies develop more tendencies to be affected by UTI after attaining age of one year[6]. This infection is more prevalent among middle aged female whereas in men incidences is high after the age of 50.

The urinary tract infection 95% cases due to bacteria which include *Escherichia coli*, *Klebsiella sp, Pseudomonas aerogenosa*, *Proteus sp., Staphylococcus species* and *Acinetobacter, Enterococcus, Morgnella sp. Citrobacter freundii, Corynebacterium urealyticum*[2]. *Escherichia coli* being most frequent causative agent of UTI accounts more than 80% community acquired 50% of nosocomial and more than 80% of cases of uncomplicated pyelonephritis[7]. Proteus infections are predominantly found in males and are also associated with renal stones. *S. saprophyticus* infections are usually found in sexually active young women. *Candida* urinary infection is usually found in diabetic patients and those with immuno-suppression[8].

The new chemical derivatives have been synthesized to fight resistant bacteria which are termed as extended spectrum beta-lactams and the enzymes are known as the extended-spectrum beta lactamases (ESBLs). The first hospital outbreak of an ESBL producing gram-negative organism reported in Germany 1983[9]. It exists in different types of bacteria and causes numerous chronic infections of respiratory tract, urinary tract, skin, blood, gastrointestinal tract, reproductive organs and central nervous system[10]. ESBLs are classified into various groups according to their amino-acid sequence homology[11]. The presence of ESBLs has remarkable clinical significance as antibiotic options in the treatment of ESBL-producing organisms are extremely limited[11].

UTI pathogens have become resistant to most of the therapeutic agents that have been developed against them in recent years, the major contributing factor is the overuse of wide spectrum antibiotics which changed the intestinal flora and induce bacterial resistance[6,12]. Beta lactam antibiotics are among the most frequently prescribed antimicrobial agents worldwide. The production of beta lactamases is the major defense strategy adopted by gram

negative bacteria against beta lactam antibiotics. Among the extensive range of antibiotics, the β-lactams account for 50% of all systemic antibiotics in use. These antibiotics played a pivotal role to cure urinary tract infections. Many of the second and third generation penicillins and cephalosporins were specifically designed to resist the hydrolytic action of major beta lactamases[13].

The major risk factors associated with ESBL producing organisms include long term antibiotic exposure, prolonged ICU stay, nursing home residency, severe illness, older age, diabetes mellitus, catheterisation and recurrent UTI incidences. The other important risk factors are the previous use of fluoroquinolones or cephalosporins, amino penicillins[14]. Prevalence of ESBL in India, Iran and Bangladesh has been reported as 58%, 44.5% and 39.5%, respectively[15]. In Pakistan 40-43% clinical isolates yielded ESBL producing gram negative bacilli[16]. The frequency of ESBL production is considerably higher in children and in old age people due to their weak immune response. Age greater than 60 years has been reported as common risk factor for ESBL infections. Increased ESBL production was seen in males as compared to females[17,18]. Hence, there is an immense need to improve, enhance and utilize the knowledge regarding isolation and identification of UTI, antibiotic susceptibility and frequency of ESBL under geographical conditions of Islamabad. Therefore, the present study was conducted.

EXPERIMENTAL

This study was conducted in Microbiology Department of Pakistan Institute of Medical Sciences, Islamabad, during September to December, 2011-2013. Patients checked by Physician and referred to laboratory for Urine test were included in the study while rest of the population was excluded. The study was included human study and was approved from ethical committee of the hospital. Further, written consent form was also signed from each of the patient before study. This constituted an overall 1050 urine samples that were processed to evaluate the isolation and identification of causative agents of UTI.

Cultural Identification

Urine was mixed by rotating the container and was inoculated on CLED agar allows the growth of both gram negative and gram positive pathogens. The indicator in CLED agar is bromothymol blue and therefore lactose fermenting colonies appear yellow. The plates were then aerobically incubated at 35-37°C for overnight. A specimen giving $\geq 10^5$ cfu/ml or forming at least 20 colonies will be considered as positive for UTI. Commonly isolated pathogens causing UTI were; *Escherichia coli, Klebsiella sp., Proteus sp., Pseudomonas aerogenosa, Candida sp., Staphylococcus aureus* and *Staphylococcus saprophyticus*[19].

Biochemical Identification

Identification of all isolates was performed on the basis of routine biochemical tests recommended by CLSI. For preliminary identification of bacteria the Gram staining, Catalase test, DNase and oxidase tests motility test, indole production, urease production, citrate utilization test and Triple sugar iron reaction, lactose fermentation were performed. Confirmation to the species level was done by using API 20 E and API NE biochemical testing kits[19].

Antimicrobial susceptibility

Antimicrobial susceptibility profile was done by Kirby Bauer disc diffusion method on Muller Hinton agar using antibiotic discs of Oxoid (UK) according to

CLSI 2011 aguidelines including[20], For gram negative bacteria**:** Amoxicillin + clavulanic acid (300 µg), Levofloxacin (5 µg), Ceftazidime (30µg), Ceftrixone (30ug), Imipenem (10 µg), Cefoperazone+sulbactam (105 µg), Piperacillin+tazobactam (110 µg), Tobramycin (10 µg), Polymixin (300 µg), Amikcin (30 µg), Pipemidic acid (30 µg), Nitrofurantoin (30 µg).

ESBL detection

Mueller–Hinton agar plate was inoculated with a suspension of the test organism (adjusted to 0.5 McFarland turbidity standards). A susceptibility disc containing amoxicillin-clavulanate was placed as the inhibitor of beta lactamase in the center of the plate, and ceftazidime, ceftriaxone discs were placed 30 mm (center to center) from the amoxicillin-clavulanate disc. Zones of inhibition around the third generation cephalosporin discs were observed after 18 h incubation at 37°C. If the zone of inhibition around one or more cephalosporin discs was extended on the side nearest to the amoxycillin+clavulanic acid disc, the organism showing this synergy was labeled as ESBL positive (Annexure II)[21,1,12,14]. Following bacterial strains were used for quality control as per CLSI recommendations: *E. coli* ATCC 2599, *Pseudomonas aeruginosa* ATCC 2783. *Staph aureus* ATCC 25923 were used as control strains.

Statistical methods

The statistical analysis was performed with the Statistical Package for Social Sciences version 17 for Windows (SPSS Inc.; Chicago, IL, USA) software and Microsoft Excel 2010. Descriptive analysis was done by calculating frequencies and percentages. Chi-square test were applied to evaluated the incidence of disease with gender, to observe the correlation between the prevalence of organism and gender, to observe the correlation between the prevalence of organism and the age groups and T-test was used to analyze the incidence of disease with age. Significance of results were calculated at 95% confidence level ($P \leq 0.05$).

RESULTS AND DISCUSSION

It was a cross sectional hospital based study conducted in Microbiology Department of Pakistan Institute of Medical Sciences, Islamabad. In this study one thousand and fifty outdoor patients's urine samples received for culture and sensitivity during the period September, 2011 to December, 2011 were included. The samples of patients from wards with complicated UTI were processed to evaluate the prevalence of causative agents of UTI and frequency of Extended Spectrum *B*-Lactamase and the patients already on the antibiotic therapy were excluded. Out of total 1050 urine samples which were processed for screening of Urinary tract infection (UTI), 225(21%,$p<0.05$) samples were found positive for UTI. Prevalence of UTI in females (56%) is higher than males (44%) and it is statistically significant ($P < 0.05$) in females. The prevalence of organism is age specific which is significant at ($P < 0.05$) found in age groups ranging from two month infant to hundred years old adult and most of the patients infected were between 41- 50 years old. The incidence of UTI in females was most commonly found from 21- 50 years whereas, in males was higher from 51 – 80 years of age (Fig. 1).

Causative Agents and their Prevalence in UTI

In order to identify the causative agents for UTI, all the samples were inoculated on CLED agar for the detection of microorganism; six microorganisms *viz. Escherichia Coli, Klebsiella pneumonea, Pseudomonas aerogenosa, Proteus sp.,*

Staphylococcus sp. and Candida were isolated from the samples (Table. 1). Majority of the causative agents were gram negative organisms 199(88%) and the rest organisms were gram positive 15(7%) and *Candida sp.* 11(5%) (Fig. 3).

Antibiotic Susceptibility and Frequency of ESBL

The isolates were processed to detect the presence of ESBL and their antibiotic susceptibility pattern. Out of total 199 gram negative isolates 121(61%) were found positive for the ESBL production, whereas 78(39%) isolates were negative for ESBL production (Table. 2; Fig. 2). Of all the ESBL positive 54% were isolated from females and 46 % from males. Among E coli isolates 38(56%) were males and (30)44% females. 14(64%) males and 8(36%) females were infected with ESBL producing *Pseudomonas* strains. ESBL positive *Klebsiella sp*. were predominant in females 13(62%) and males (8)38%. ESBL producing *Proteus sp*. infected 5(50%) females and 5(50%) males (Fig.2).

68(56%) *E. coli* isolates showed ESBL production. *Pseudomonas sp.* were the second predominant ESBL producing organism with 22(18%) isolates, followed by *Klebsiella sp.* with 21(17%) isolates and *Proteus sp.* with 10(8%) isolates. (Table.2; Fig.3).

Antibiotic susceptibility patterns

ESBL Producing and non-ESBL Producing *Escherichia coli*

ESBL producing *E. coli* showed highest sensitivity to PB (87%) followed by IPM (79%), SCF (76%), F (75%) and AK (71%). As far as resistance pattern of ESBL producing E. coli was concerned by, highest resistant was shown to LEV (79%) followed by PIP (61%) and TOB (53%). (Fig.4).

Non ESBL producing *E. coli* isolates showed higher sensitivity trends to all the classes of antibiotics of which IPM was 95% followed by F (93%) and PB (90%) (Fig. 5).

In present study all outdoor patients's urine samples received during the four months for culture and sensitivity in the Microbiology Lab of PIMS were included in the study. This constituted an overall 1050 urine samples that were processed to evaluate the prevalence of causative agents of UTI and frequency of Extended Spectrum Beta Lactamase. Patients from wards with complicated UTI and patients already on the antibiotic therapy were excluded. Of all the samples processed for screening of Urinary tract infection (UTI), 225 samples were found positive for UTI with percentage of 21% (Fig.1). High incidence can be attributed to the fact that this data was collected from the OPD patients from different departments who might have been previously infected with UTI at any stage during hospitalization, catheterization or any other chronic disease. Rehman *et al.,* (2009)[22] reported 24.14% UTI prevalence. High indence of UTI ranging from 25%-35% have also been reported by Behroozi *et al* (2010)[9], Modarres *et al* (2009)[23], Ramesh *et al.,* (2008)[7], Rai *et al.,* (2008)[6] and Farajnia *et al.,* (2009)[2] and Nasher *et al.,* (2001)[24]. Omoregie *et al.,* (2010)[25] has reported that 17% and 11.3% samples were positive for UTI in their studies respectively.

The data for UTI was further analysed according to gender and age group which indicated that the incidence of UTI was more in females (56%) as compare to the males (44%) (Fig. 2) statistically the incidence of UTI in females is significant ($P < 0.05$). This might be due to the close proximity of female urethral meatus to the anus, shorter urethra, urothelial mucosa adherence to the muco-polysaccharide lining. In women, fecal-perineal-urethral contamination is the most probable

explanation for infections caused by enteric bacteria[5].

Ejaz *et al.,* (2006)[26] observed that in Pakistan females has 4% higher risk of getting UTI than males. Mehr *et al*., (2010)[27] reported that in Pakistan prevalence of UTI in females is 63% as compared to males 37%. Bano *et al.,* (2012)[3] observed that male to female ratio in Pakistan as 34:81. Hassan *et al.,* (2011)[12] observed in India that urinary tract infection was seen in 70.5% females and 29.5 males. Kashef *et al.,* (2010)[28] reported that among all the patients 82.5% were females. All these studies support our results of female preponderance in UTI.

Most of the patients infected were from 41- 50 years old (Fig. 3). In case of females, UTI was most commonly found in patients from 21- 50 years. The reason for this can be due to the fact that during this age females are sexually active. Whereas, the incidence of UTI was higher in males from 51 – 80 years, statistically the increase incidence of disease with advancing age is significant ($P < 0.05$). Our results are similar with the findings of Jalapour *et al*., (2011)[13] who also showed that UTI is less common in young men below 50 and who did not undergo any genitourinary procedure. Infection tends to rise after the age of 50 in men. Similarly Akram *et al*., (2007)[4] observed that frequency of UTIs was found more in elderly patients (51.04 %). Whereas results observed by Bano *et al.,* (2012)[3] and Roopa *et al*., (2010)[21] are inconsistent with our results who reported higher UTI incidence in patients between 20-30 years. In our results *Klebsiella* infection is more prevalent after the age of 40. Similarly Farajnia *et al*., (2009)[2] also observed that *Klebsiella* infections are more prevalent in the older age groups.

Majority of the causative agents were gram negative organisms 199(88%) and the rest organisms were gram positive 15(7%) and *Candida sp.* 11(5%). Similar results were observed by Akram *et al*., (2007)[4] that gram negative organisms accounted for 92% while gram positive organisms accounted for the remaining 8% of total pathogens causing UTI.

Our results shows that the *E. coli* 109 (48%) is the most prevalent organism followed by *Pseudomonas aerogenosa* 39(17%), *Klebsiella pneumonae* 31(14%), *Proteus sp.* 20 (9%), *staphylococcus sp*. 15(7%), *candida sp*. 11(5%). Similar results were reported by Bano *et al*., (2012)[3], Rai *et al*., (2008)[6], Kumar *et al*., (2011)[18] with *E. coli* being the most common causative agent of UTI with infection percentage from 50%-90%. Ojo *et al.,* (2010)[28] suggests that *E. coli* accounts for 32% of UTI cases. Ejaz *et al.,* (2006)[26] observed 37 % prevalence of E. coli in Pakistan. These studies also support our results. Whereas Adeleke *et al*., (2009)[30] reported that *Staphylococcus aureus* (67.9%) was most common causative agent in children in Nigeria which is inconsistent with our results.

Pseudomonas aerogenosa (17%) is the second most prevalent organism in our study. It is inconsistent to the most of the previous studies. Bano *et al.,* (2012)[3] from Pakistan reported *Klebsiella pneumonia* being the second most prevalent organism with percentages as 18 and 16 respectively. Rehman *et al.,* (2009)[22], Kumar *et al.,* (2011)[18], Irajian *et al.,* (2010)[31], Moyo *et al.,* (2010)[31] all reported *Klebsiella* (11% - 37%) being second most prevalent organism causing UTI. In contrary to our findings Qureshi, (2005)[8] from Pakistan observed that *E. coli* (1%) is the least common causative agent of UTI. Similar finding of least incidence of *Pseudomonas sp.* as a causative agent of UTI that is in contrary to our results was observed by Farajnia *et al.,* (2009)[2]. The percentage distribution of *Proteus sp.* (9%), *staphylococcus sp*. (7%), *candida sp*. (5%) shown in our results is similar to the previous studies. Bano *et al.,*

(2012)[3] observed prevalence of *S. aureus* (12.04%), *Candida spp.* (4.81%) in Pakistan. Results reported by Hassan *et al*., (2011)[12] from India are similar to our results showing that *Staphylococcus aureus* was the commonest Gram- positive isolate (1.5%). Kashef *et al.,* (2010)[28] observed that 12.4% *Proteus sp*. caused UTI. Kumar *et al.,* (2011)[18] reported that in their study only single strain of *Candida sp.* was isolated. 7% occurrence of staphylococcus sp. reported by Akram *et al.,* (2007)[4] and Dytan *et al.,* (1999) [33] is similar to our results. Our results indicated that average prevalence of Staphylococcus is more common in males, similar results were observed by Oladeinde *et al*., (2011)[5].

One hundred and twenty one (61%) gram negative bacilli showed the production of extended spectrum beta lactamase (ESBL) whereas 78(39%) isolates were non ESBL producers (Table. 2; Fig. 3). Overall prevalence of ESBL production is 61%. Previously, Jabeen *et al*., (2005)[34] observed in Pakistan that (40%) of the isolates tested were found to be ESBL producing. Taneja *et al.,* (2008)[35] reported that in India prevalence of ESBL is 36.5%. Mathur *et al*., (2002)[36] reported that of all the isolates tested 68% were ESBL producers. Similar results were observed by previous studies. Baral *et al.,* (2012)[1] revealed prevalence of ESBL as 42%, 45%, 44% and 55% respectively. Ramesh *et al.,* (2008)[7] observed that 71% isolates were ESBL producing which is higher than the percentage observed in present study. Ramazanzadeh, (2010)[37] and Irajian *et al.,* (2010)[31] reported ESBL production as 14.5% and 28% respectively which is lower as compared to the present study.

Our results showed that highest ESBL production was observed in *E. coli* isolates 68(56%), followed by ESBL producing *Pseudomonas sp.* 22(18%), *Klebsiella sp.* 21(17%) and *Proteus sp.* 10(8%) (Table 2 and Fig.3). Similar results were reported by Hassan *et al*., (2011)[12] in Pakistan that 54% *E. coli* were ESBL producers.

Our results showed that all ESBL producing isolates were resistant to amoxicillin-clavulanic acid and to third generation cephalosporins such as ceftazidime and ceftriaxone. Similar results were shown by Bourjilat *et al*., (2011)[38], Taneja *et al.,* (2008)[35], Ramazanzadeh, (2010)[37], Chaudhary *et al.,* (2004)[14], and Bradford, (2001)[39]. Our results revealed that ESBL producing *E. coli* showed highest sensitivity to PB (87%) followed by IPM (79%), SCF (76%), F (75%) and AK (71%). As far as resistance pattern of ESBL producing *E. coli* was concerned by, highest resistant was shown to LEV (79%) followed by PIP (61%) and TOB (53%) (Fig. 4).

ESBL producing *Klebsiella sp.* showed highest sensitivity to both PB and IPM (86%) followed by SCF (81%) and AK (76%) followed by SCF (81%) and AK (76%). ESBL producing *Klebsiella sp.* showed (71%) resistance to LEV, followed by TOB (62%) and PIP (62%). (Fig. 5).

ESBL producing *Pseudomonas sp*. was highly sensitive to PB (86%), IPM (82%) and SCF (73%). The pattern of highest resistance shown by these organisms was LEV (91%), PIP (77%), TOB (68%) and TZP (55%) (Table.3, Fig.22). Non-ESBL producing *Pseudomonas* isolates were highly sensitive to SCF (100%), AK (94%), PB (88%), TZP (88%) and IPM (76%).Highest resistance was against PIP (71%). (Fig. 5).

All the Non-ESBL producing gram negative isolates included in our study were more susceptible to all the classes of antibiotics as compared to ESBL positive strains. Non-ESBL producing strains showed highest sensitivity to Nitrofurantoin (F), Imipenem (IPM), Polymixin (PB), and aminoglycosides which include Amikacin

(AK) and Tobramicin (TOB). However, high resistance to Pipemidic acid (PIP) was shown by all Non-ESBL producing isolates. Similar results were observed by Roshan *et al.,* (2011)[40] in Pakistan. Ullah *et al*., (2009)[15] found that Resistance was high in the ESBL positive strains as compared to the ESBL negative strains. A statistically significant difference was found in the susceptibilities of flouroquinolones, amikacin, cefoperazone/sulbactam, pipe-racillin/tazobactam, and meropenem for ESBL positive and ESBL negative isolates which also supports the results of present study. Findings of Sader *et al*., (2001)[41] also supports our results that showed that the carbapenems showed excellent activitiy against these pathogens whereas he observed that the fluoroquinolones were very active against ESBL-producing organism which is inconsistent with our results.

CONCLUSIONS

Overall prevalence of UTI observed in our study is 21%. *E. coli* (48%) is the most prevalent organism followed by *Pseudomonas aerogenosa* (17%), *Klebsiella pneumonae* (14%), *Proteus sp.* (9%), *Staphylococcus sp*. (7%), *Candida sp*. (5%).

UTI is more prevalent in females (56%) as compared to males (44%). The UTI is more common in the age group 41-50. Out of total 199 gram negative isolates 121(61%) were found positive for the ESBL production. ESBL production was most common in *E Coli* 68(56%) followed by *Pseudomonas aerogenosa* 22(18%), *Klebsiella sp.* 21(17%), *Proteus sp*. 10(8%). Our results suggest that all ESBL positive isolates are resistant to amoxicillin + clavulanic acid and to third generation cephalosporins i.e. ceftazidime and ceftriaxone. As compared to Non-ESBL producing isolates ESBL producing ones are more resistant to fluoroquinolones, aminoglycosides and pipemidic acid. However, ESBL positive organisms are sensitive to imipenem, polymixin and nitrofurantoin, therefore these can be the treatment of choice for such highly resistant organisms.

Future Recommendations

It is quite alarming to note the high prevalence of extensively resistant ESBL producing isolates in our setup, which is posing a major clinical crisis of treatment failure with β-lactam antimicrobials. So, screening for ESBL detection and their antibiotic susceptibility checking should be included in the routine monitoring of every pathological laboratory to restrict the over and misuse of the antibiotics to limit the evolvement of highly resistant strains and to improve the treatment and management of UTI. Furthermore, there is a need for large scale studies on prevalence and genetic recombinations of ESBL producing strains all over the country to improve the knowledge regarding such fastidious organisms.

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Table 1. Organisms isolated from urine samples

S. No	Isolated organisms	Isolates No.	%	Male		Female	
				No.	%	No.	%
1	*E. coli*	109	48	52	48	57	52
2	*Klebsiella sp*	31	14	9	29	22	71
3	*Pseudomonas*	39	17	20	51	19	49
4	*Proteus sp*	20	9	9	45	11	55
5	*Staph sp*	15	7	5	34	10	66
6	*Candida sp*	11	5	4	36	7	64
	Total	225	100.0	99	44	126	56

Table 2. ESBL producing and non ESBL producing isolates of gram negative bacilli

S. No.	Organism	ESBL		Non ESBL	
		No.	%	No.	%
1	*E. coli*	68	56	41	53
2	*Klebsiella sp*	21	17	10	13
3	*Pseudomonas sp*	22	18	17	22
4	*Proteus sp*	10	8	10	13
	Total	121	100	78	100

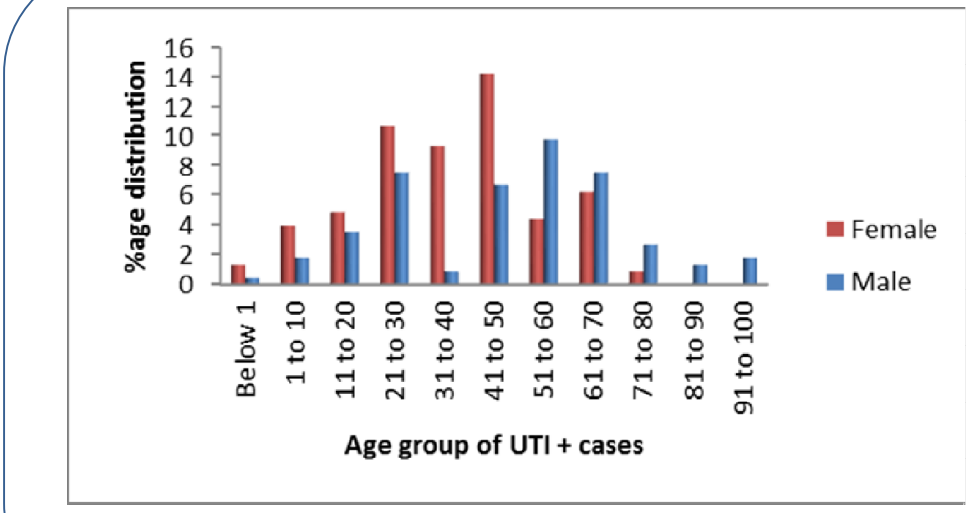

Figure 1. Age wise and gender wise correlation of prevalence of UTI

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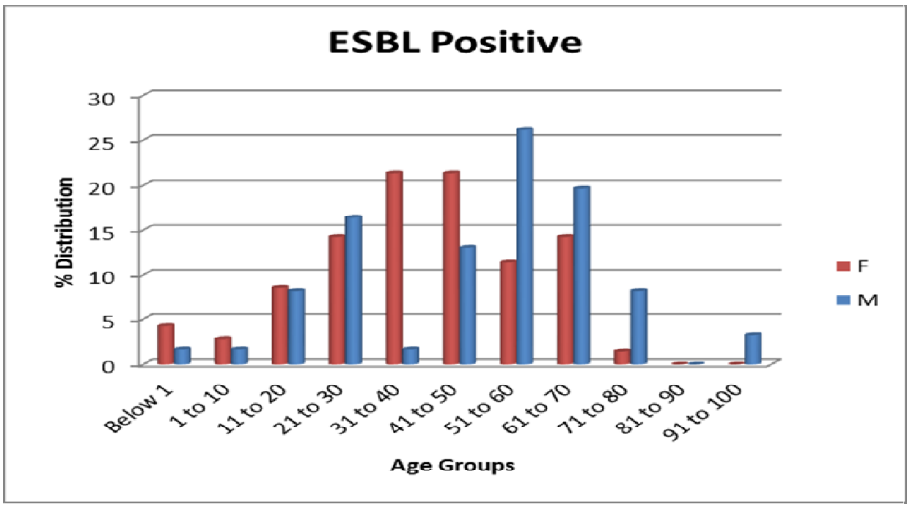

Figure 2. Prevalence of ESBL with reference to age

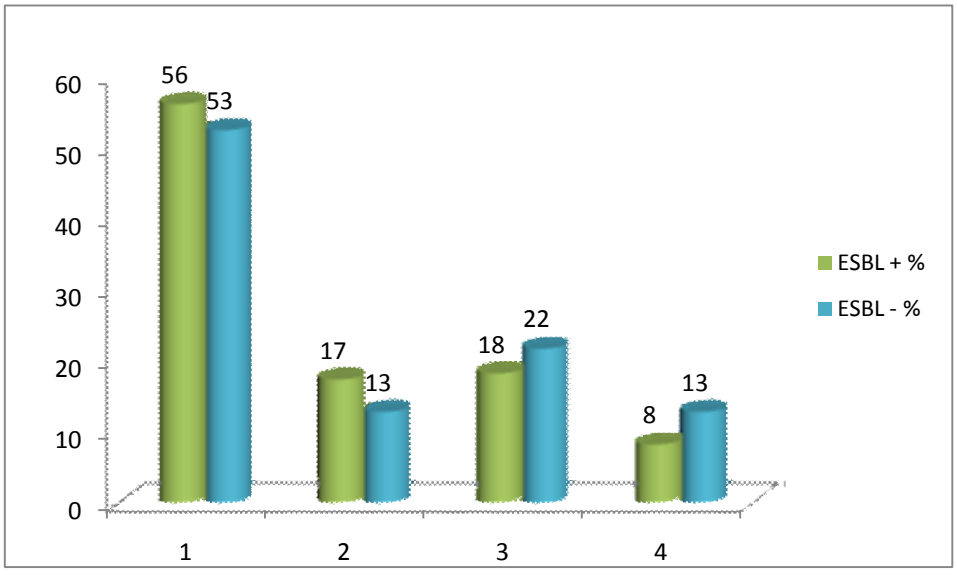

Figure 3. ESBL producing and non ESBL producing isolates of gram negative bacilli

1. E. coli, 2. Klebsiella, 3. Pseudo, 4. Proteus.

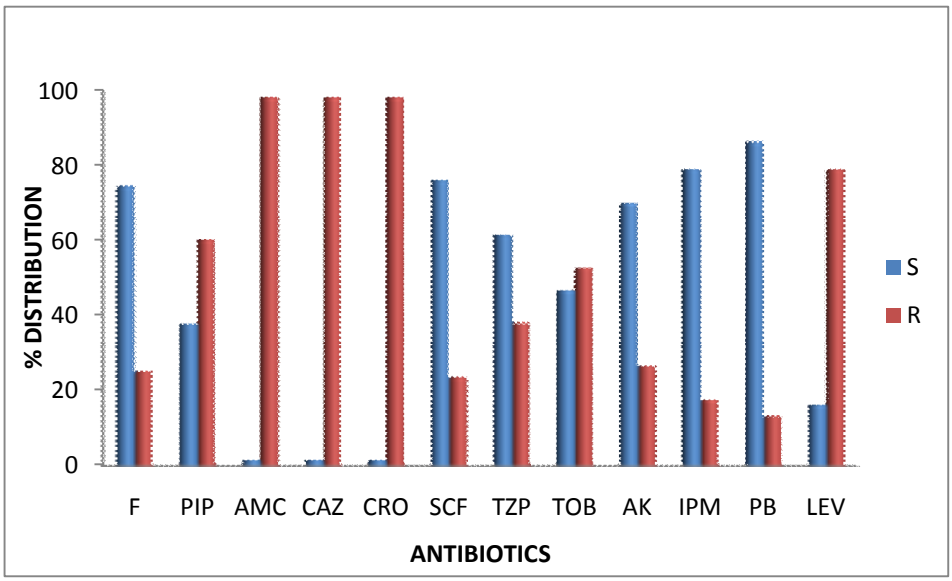

Figure 4. Antibiotic susceptibility pattern of ESBL producing *E. coli*

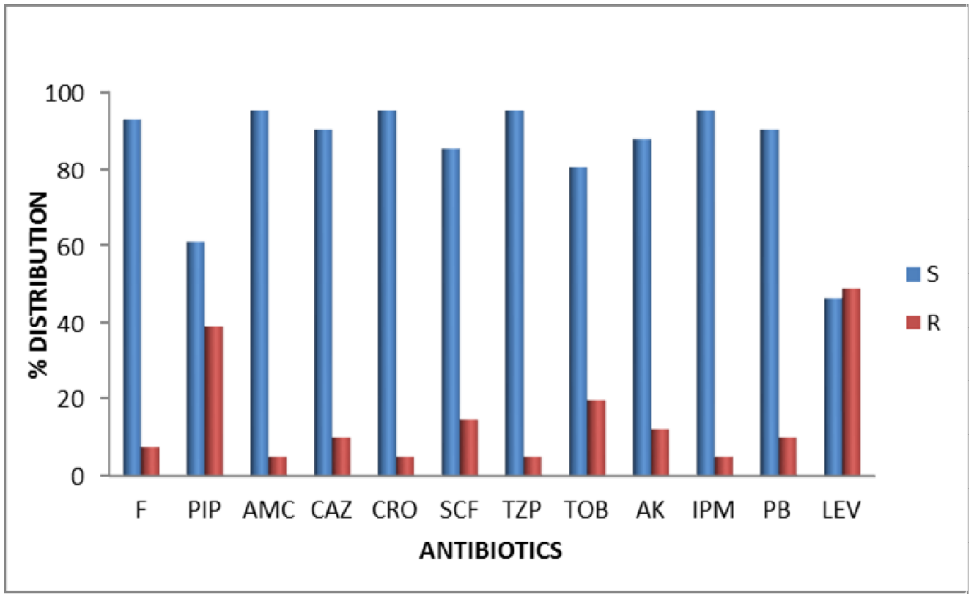

Figure 5. Antibiotic susceptibility pattern of non ESBL producing *E. coli*