

Content available at: <https://www.ipinnovative.com/open-access-journals>

IP International Journal of Medical Microbiology and Tropical Diseases

Journal homepage: <https://www.ijmmt.org/>

Original Research Article

Detection of ESBL producing *E.coli* and *Klebsiella* spp in urinary tract infection due to long exposure of catheterize condition in Covid positive patient admitted in HDU

Shyam Sundar Bera¹, Traun Kumar Pathak², Arunabha Sarkar³, Devnil Pathak⁴,
Mousumi Mitra⁵, Purna Nandi⁵, Sudeep Mitra⁵, Dilip Kumar Nandi^{5,*},
Bandita Sasmal⁵, Kaberi Bhattacharya⁵, Anita Nanda Goswami⁶

¹Salboni Super specialty Hospital, Midnapore, West Bengal, India²Bankura Sammilani Medical College and Hospital, Bankura, West Bengal, India³North Bengal Medical College and Hospital, Siliguri, West Bengal, India⁴MGM Medical College and LSK Hospital, Kishanganj, Bihar, India⁵Raja Narendra Lal Khan Women's College (Autonomous), Midnapore, West Bengal, India⁶Dept. of BMLT, Raja Narendra Lal Khan women's College (Autonomas), Midnapore, West Bengal, India

ARTICLE INFO

Article history:

Received 15-07-2022

Accepted 25-07-2022

Available online 06-09-2022

Keywords:

ESBL

MDR

CLED

UTI

HDU

ABSTRACT

Introduction: Long exposure of broad spectrum antibiotic can severe causes of extended-spectrum beta-lactamase (ESBL) production within bacteria in UTI patient. Community-acquired infections due to ESBL producing *Escherichia coli* and *Klebsiellae* both are rising worldwide.

Objective: To assess the prevalence and antibiotics resistance patterns of ESBL-producing *Escherichia coli* and *Klebsiella* spp. from covid positive patient admitted in HDU with catheterized condition.

Materials and Methods: A hospital-based cross-sectional study was conducted on Salboni Super specialty Hospital (Covid Level-IV) at West Bengal, India from August 2020 to August 2021 and a total of 1116 urine samples were collected from catheterized patient admitted in male and female HDU due to exposure of covid. Samples were cultured on CLED agar and then Identification of the isolates was done by Gram's staining followed by biochemical tests. Antibiotic susceptibility testing was done by modified Kirby-Bauer disc diffusion method and interpretation was done following Clinical and Laboratory Standard Institute (CLSI) guidelines for detection of ESBL producing *E.coli* and *Klebsiellae* spp.

Result: present study showed 51.6% (n-326) *E.Coli* and 37.2% (n-235) *Klebsiella* spp. from 631 significant growths from both male and female patients. Out of this, total ESBL producing *E.coli* and *klebsiella* spp. seen 55% (n-309) from both male and female patient in different age group. Finally Isolated ESBL producing *E.coli* 45.9 % (n-142) and ESBL producing *Klebsiella* spp. 54 % (n- 167) from both male and female HDU.

Conclusion: Detection of ESBL producing *E.coli* and *Klebsiellae* due to prolonged uses of broad spectrum antibiotic vigorously. The occurrence of multidrug resistance to the third-generation cephalosporins, aminoglycosides, fluoroquinolones, trimethoprim-sulfamethoxazole, and tetracyclines is more common among ESBL producers. It may help to detect other organism in same condition, so that it will be importance in clinical aspect of future treatment for clinician.

This is an Open Access (OA) journal, and articles are distributed under the terms of the [Creative Commons Attribution-NonCommercial-ShareAlike 4.0 License](https://creativecommons.org/licenses/by-nc-sa/4.0/), which allows others to remix, tweak, and build upon the work non-commercially, as long as appropriate credit is given and the new creations are licensed under the identical terms.

For reprints contact: reprint@ipinnovative.com

* Corresponding author.

E-mail address: dilipnandi2004@yahoo.co.in (D. K. Nandi).

1. Introduction

Extended-spectrum beta-lactamase (ESBL)-producing *E. coli* and *Klebsiellae* have been recognized as a major

multidrug-resistant bacteria implicated in serious hospital and community-acquired infections worldwide, especially in places where poor sanitation, and inadequate hygienic practices are very common.^{1–4} antimicrobial resistance (AMR) is considered a main global threat to human and animal health⁵ AMR is responsible for thousands of human Fatalities annually.⁶ In the last decade, the global growth of livestock has been associated with an increase in antibiotics use.⁵ Extended-spectrum beta-lactamase-producing *Escherichia coli* (ESBL-*E. coli*) represent one of the highest burdens of AMR to public health and have globally spread in both hospital settings and the community.⁷ Extended-spectrum β -lactamases (ESBLs) are class A β -lactamases, a rapidly evolving group of β -lactamases with the ability to hydrolyze and cause resistance to the oxy-imino cephalosporins (cefotaxime, ceftazidime, ceftriaxone, cefuroxime, and cefepime) and monobactams (aztreonam).⁸

ESBL-producing *E. coli* and *K. pneumoniae* are the predominant cause of childhood infections and present significant challenges⁹ such as development of adverse outcomes.,¹⁰ treatment failure due to multidrug resistance, and high morbidity and mortality.¹¹ Empirical and symptomatic (without a diagnosis) use of antibiotics in resource poor settings is responsible for higher incidence of antibiotic resistance among bacteria.¹²

ESBLs are often encoded by genes located on large plasmids, and these also carry genes for resistance to other antimicrobial agents such as aminoglycosides, trimethoprim, sulphonamides, tetracyclines and chloramphenicol.¹³ Recent studies have demonstrated fluoroquinolones resistance mediated by co-transfer of the *qnr* determinant on ESBL-producing plasmids.^{14,15}

Thus, very broad antibiotic resistance extending to multiple antibiotic classes is now a frequent characteristic of ESBL-producing enterobacterial isolates. As a result, ESBL-producing organisms pose a major problem for clinical therapeutics. This review attempts to present a comprehensive picture on the basis of the currently available literature about this diverse, complex and rapidly evolving group of enzymes.

2. Materials and Methods

The current study was carried out in the department of pathology, salboni super speciality hospital, WB, India from August 2020 to August2021 among catheterize patient admitted in ICU for covid purpose. The urine sample were collected from patient in sterile container and cultured on CLED (cystine Lactose Electrolite Deficient) agar, after bacterial growth we further proceed for detection of *Escherichia Coli* and *Klebsiella* spp. through biochemical like Indole, MR,VP, Citrate, Urease, TSI and Mnitol tests.

Antibiotic test was done by the Kirby Bauer Disc diffusion method on Muller Hinton agar. Antimicrobial

agents tested were Ampicillin, Amoxiclav, gentamicin, amikacin, nalidixic acid, norfloxacin, ofloxacin, nitrofurantoin, sulphofurazole, cotrimoxazole, piperacillin-tazobactam, cephalothin, cefixime, cefuroxime, ceftriaxone, ceftazidime, Cefepime, cefotaxime, imipenam, meropenemrecommended by the CLSI 2013 using *Escherichia coli* ATCC 25922 and *Klebsiella pneumonia* ATCC 700603 as a standard strain for routine quality control.

2.1. Phenotyping detection

A phenotypic confirmatory test recommended by the CLSI was done by combined disc diffusion method. ESBLs was detected by the confirmatory method of Clinical and Laboratory Standards Institute (CLSI) using ceftazidime (30 mg) and a disc of ceftazidime plus clavulanic acid (30/10 mg) were placed at a distance of 20 mm on a lawn culture (0.5 McFarland inoculum size) of suspected ESBL producing clinical isolate on Mueller-Hinton Agar (MHA, Hi-Media, Mumbai). *Escherichia coli* ATCC 25922 were used as the negative control and *Klebsiella pneumonia* ATCC 700603 were used as the ESBL positive control. The study was approved by ethics committee of Salboni SSH,WB, India.

2.2. Statistically analysis

Results were analyzed using Pearson's chi-square test, or Fisher's exact test. Avalue of P<0.05 was regarded as statistically not significant (SPSS 16.0, SPSS Inc., Chicago, IL, USA).

3. Result

Total 1116 catheterize urine sample were collected from Male and Female HDU patients during one year of duration (August 2020 to August2021) in covid pandemic situation.

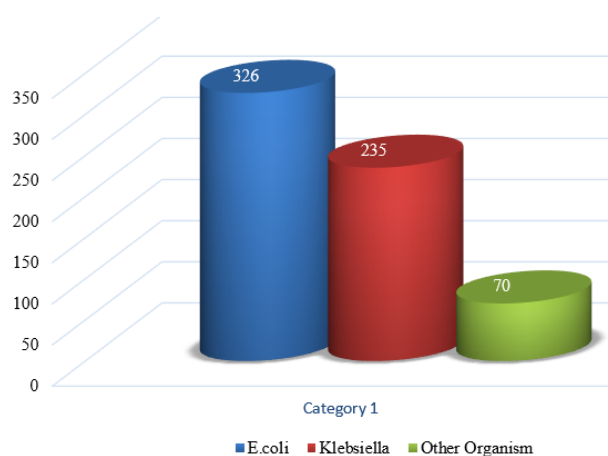


Fig. 1: Analysis of different isolated organism from bacterial growth

Table 1: Statistical analysis of bacterial growth

Age group	Female HDU patient		Male HDU patient	
	Growth	Non-Growth	Growth	Non-Growth
Years	Numbers(N) with Percentage%	Numbers(N) with Percentage%	Numbers(N) with Percentage%	Numbers(N) with Percentage%
30-39	110(52%)	99(48%)	102 (53%)	89 (47%)
40-49	119(54%)	100(46%)	100 (52%)	91 (48%)
50-59	100(65%)	52(35%)	100 (64%)	54 (36%)
P-value	7.0226,P=0.0298,P<0.05 Significant		7.4182, P=0.0244,P<0.05 Significant	

Table 2: Analysis of different isolated organism from bacterial growth

Age Group	<i>E.coli</i> (326)		<i>Klebsiella</i> spp. (235)		P value
	Numbers(n)	Percentage (%)	Numbers(n)	Percentage (%)	
Years					6.1666
30-39	115	54%	95	44%	P=0.0458
40-49	111	51%	90	41%	P<0.05
50-59	100	50%	50	25%	Significant

Table 3: Statistically analysis of ESBL production from male and female patients

Age group	ESBL (309)		Non-ESBL (252)		P value
	Numbers(n)	Percentage (%)	Numbers(n)	Percentage (%)	
Years					0.9522 P=0.62121
30-39	105	50%	92	43%	P<0.05
40-49	99	49%	84	39%	Not-Significant
50-59	105	70%	76	30 %	

Table 4: ESBL producing *E.coli* and *Klebsiella* spp. in between male and female

Age group	ESBL Producing <i>E.Coli</i> (142)		ESBL Producing <i>Klebsiella</i> (167)		P-Value
	Numbers(n)	Percentage (%)	Numbers(n)	Percentage (%)	
30-39	63(44.3%)		56 (33.5%)		3.9354 P=0.139775 P<0.05 Not-Significant
40-49	42(29.5%)		62 (37.1%)		
50-59	37 (26%)		49 (29.3%)		

Studied in three different age group (30-39, 40-49, and 50-59) in between male and female (Table 1). In present study showed significant bacterial growth (n=329, P=0.0298) in female and in male (n=302, P=0.0244). After identification of bacterial, growth showed (n=326) 51% *E.coli* growth and (n=235) 37% *Klebsiella* growth and (n=70) 11% other organism causing urinary tract infection. Furthermore, total ESBL producing *E.coli* and *Klebsiella* spp. seen (n=309) 55% (P=0.62121) in between male and female patient. After that finally ESBL producing *E.coli* seen 45% (n=142) and ESBL producing *Klebsiella* Spp. seen 54% (n=167) in respect to age group (30-39, 40-49, and 50-59 in both male and female HDU patient.

4. Discussion

In this study the first description of ESBL producing uropathogenic *E. coli* and *Klebsiella* spp. involved in major public health cases of urinary tract infections in our country. Urinary tract infections are the most common infections in adults' patients and *E. coli* and *Klebsiella* being leading pathogenic agent in these infections; it was our matter of interest is to excess ESBL Producing *E.coli* in catheterized

patient admitted in HDU in covid situation.

Urinary tract infections (UTIs) are a severe public health problem and are caused by a range of pathogens and prevalence varies with the age and sex. In this present study, it was noted that the significant bacteriuria was found in high prevalence (P-value = 0.0298) of UTI positive clinical sample in male and female in the age groups of 40-49 years. Similarly, study was observed Parveen RM¹⁶ et al, 2011 in Puducherry, India and 39 isolates investigated, 37 (94.8%) were found to be resistant to at least one of the third generation cephalosporins. Among these 37 isolates, 36 (97.2%) were found to be ESBL positive by phenotypic testing from blood. And others study was observed Shakya Pet al,¹⁷ 2017 in Nepal and showed significant bacteriuria with 365 (80.9%) *E. coli*, 17 (3.8%) *Klebsiella pneumoniae* and 3 (0.7%) *Klebsiella oxytoca*.

Concurrently, significant (P=0.0458) growth of *E.coli* and *Klebsiella* spp. was seen for both male and female patient, similarly study was observed on Kayastha et al,¹⁸ 2020 and showed *E. coli* (n = 79), *Klebsiella pneumoniae* (n = 18), and *Klebsiella oxytoca* (n = 6) were isolated from different clinical specimens.

We showed non-significant of ESBL producing *E.coli* and *Klebsiella* ($P=0.139775$) from both male and female (Table 4) patient dividing age group of 30-39, 40-49, and 50-59, similarly the study was done by Abayneh M et al,¹⁹ 2018 in Canada and study showed ESBL-producing phenotypes were detected in 23% (n - 17) of urinary isolates, of which *Escherichia coli* accounts for 76.5% (n - 13) and *K. pneumoniae* for 23.5% (n - 14).

5. Conclusion

Use of broad-spectrum antibiotic during long period can cause of multiple drug resistant (MDR) *E.coli* and *Klebsiellae* spp. detected catheterize urine sample in covid patients.

The present study concludes that, high prevalence of *E. coli* and *Klebsiella* associated with UTI and illustrates the maximum number of organisms from female patients of catheterize condition in age group i.e. 25-44 years. Most of the *E. coli* and *Klebsiella* isolates showed multi drug resistance. The most effective drug was found to be Nitrofurantoin followed by Amikacin, Gentamicin and Imipenem. Amikacin, Gentamicin and Imipenem are the parental drugs but only Nitrofurantoin which showed good activity is an oral drug. So, Nitrofurantoin remains the only available alternative with high susceptibility rate among oral agents for UTI. To provide optimum use and to avoid misuse and over use of these drug, culture and susceptibility testing is the need of hour to preserve it for next generation.

Hence, forth concluded that microbiological diagnosis is very essential for better treatment so that appropriate antibacterial drug can be initiated on time in order to limit the worsening of condition and to prevent complications and should not take antimicrobial drugs vigorously without clinician advise.

6. Conflict of Interest

None.

7. Source of Funding

None.

References

1. Aruna K, Mobashshera T. Prevalence of extended spectrum beta-lactamase production among uropathogens in south Mumbai and its antibiogram pattern. *EXCLI J*. 2012;11:363–72.
2. Hussain A, Shaik S, Ranjan A, Nandanwar N, Tiwari SK, Majid M, et al. Risk of Transmission of Antimicrobial Resistant *Escherichia coli* from Commercial Broiler and Free-Range Retail Chicken in India. *Front Microbiol*. 2017;8:2120. doi:10.3389/fmicb.2017.02120.
3. Hussain A, Shaik S, Ranjan A, Suresh A, Sarker N, Semmler T, et al. Genomic and functional characterization of poultry *Escherichia coli* from India revealed diverse Extended-spectrum β -lactamase-producing lineages with shared virulence profiles. *Front Microbiol*. 2019;10:2766. doi:10.3389/fmicb.2019.02766.
4. Pitout JDD, Nordmann P, Laupland KB, Poirel L. Emergence of Enterobacteriaceae producing extended-spectrum β -lactamases (ESBLs) in the community. *J Antimicrob Chemother*. 2005;56(1):52–9. doi:10.1093/jac/dki166.
5. Wall BA, Mateus A, Marshall L, Pfeiffer D, Lubroth J, Ormel HJ, et al. Food and Agriculture Organization of the United Nations. Drivers, Dynamics and Epidemiology of Antimicrobial Resistance in Animal Production; Food and Agriculture Organization (FAO): Rome, Italy; 2016.
6. IACG. No Time to Wait: Securing the Future from Drug-Resistant Infections. [Accessed on 23 July 2020]. Available from: <http://www.who.int/antimicrobial-resistance/interagency-coordination-group/final-report/en>.
7. Doi Y, Iovleva A, Bonomo RA. The ecology of extended-spectrum β -lactamases (ESBLs) in the developed world. *J Travel Med*. 2017;24(1):S44–51. doi:10.1093/jtm/taw102.
8. Peirano G, Pitout JDD. Molecular epidemiology of *Escherichia coli* producing CTX-M β -lactamases: the worldwide emergence of clone ST131 O25:H4. *Int J Antimicrob Agents*. 2010;35(4):316–21. doi:10.1016/j.ijantimicag.2009.11.003.
9. Bakshi P, Walia GSJ. Prevalence of extended spectrum β -lactamase in multidrug-resistant strains of gram negative bacilli. *J Acad Indus Res*. 2013;1(9):558–60.
10. Upadhyay A, Parajuli P. Extended spectrum β -lactamases (ESBL)-producing *Klebsiella* species isolated at National Medical College and Teaching Hospital Nepal. *Asian J Pharma Clin Res*. 2013;6(4):161–4.
11. Thenmozhi S, Moorthy K, Sureshkumar B, Suresh M. Antibiotic resistance mechanism of ESBL producing enterobacteriaceae in clinical field: a review. *Int Pure App Biosci*. 2014;2(3):207–26.
12. Neupane S, Pant ND, Khatiwada S, Chaudhary R, Banjara MR. Correlation between biofilm formation and resistance toward different commonly used antibiotics along with extended spectrum β -lactamase production in uropathogenic *Escherichia coli* isolated from the patients suspected of urinary tract infections visiting Shree Birendra Hospital. *Antimicrob Resist Infect Control*. 2016;5:5. doi:10.1186/s13756-016-0104-9.
13. Paterson DL. Recommendation for treatment of severe infections caused by Enterobacteriaceae producing extended-spectrum β -lactamases (ESBLs). *Clin Microbiol Infect*. 2000;6(9):460–3. doi:10.1046/j.1469-0691.2000.00107.x.
14. Mammari H, Van De Loo M, Poirel L, Martinez-Martinez L, Nordmann P. Emergence of plasmid-mediated quinolone resistance in *Escherichia coli* in Europe. *Antimicrob Agents Chemother*. 2005;49(1):71–6. doi:10.1128/AAC.49.1.71-76.2005.
15. Wang M, Sahn DF, Jacoby GA, Hooper DC. Emerging plasmid-mediated quinolone resistance associated with the *qnr* gene in *Klebsiella pneumoniae* clinical isolates in the United States. *Antimicrob Agents Chemother*. 2004;48(4):1295–9. doi:10.1128/AAC.48.4.1295-1299.2004.
16. Parveen RM, Khan MA, Menezes GA, Harish BN, Parija SC, Hays JP, et al. Extended-spectrum β -lactamase producing *Klebsiella pneumoniae* from blood cultures in Puducherry, India. *Indian J Med Res*. 2011;134(3):392–5.
17. Shakya P, Shrestha D, Maharjan EE, Sharma V, and RP. Causing Urinary Tract Infection: A Hospital Based Study. *Open Microbiol J*. 2017;11:23–30. doi:10.2174/1874285801711010023.
18. Kayastha K, Dhungel B, Karki S, Adhikari B, Banjara MR, Rijal KR, et al. Extended-Spectrum β -Lactamase-Producing *Escherichia coli* and *Klebsiella* Species in Pediatric Patients Visiting International Friendship Children's. *Infect Dis (Auckl)*. 2020;13:1178633720909798. doi:10.1177/1178633720909798.
19. Abayneh M, Tesfaw G, Abdissa A. Isolation of Extended-Spectrum β -lactamase- (ESBL-) Producing *Escherichia coli* and *Klebsiella pneumoniae* from Patients with Community-Onset Urinary Tract Infections in Jimma University Specialized Hospital. *Can J Infect Dis Med Microbiol*. 2018;p. 4846159. doi:10.1155/2018/4846159.

Author biography

Shyam Sundar Bera, Microbiologist

Traun Kumar Pathak, Medical Superintendent cum Vice Principal

Arunabha Sarkar, HOD

Devnil Pathak, PGT

Mousumi Mitra, College Teacher

Purna Nandi, Research Scholar

Sudeep Mitra, Research Scholar

Dilip Kumar Nandi, HOD

Bandita Sasmal, College Teacher

Kaberi Bhattacharya, College Teacher

Anita Nanda Goswami, Student

Cite this article: Bera SS, Pathak TK, Sarkar A, Pathak D, Mitra M, Nandi P, Mitra S, Nandi DK, Sasmal B, Bhattacharya K, Goswami AN. Detection of ESBL producing *E.coli* and *Klebsiella* spp in urinary tract infection due to long exposure of catheterize condition in Covid positive patient admitted in HDU. *IP Int J Med Microbiol Trop Dis* 2022;8(3):237-241.