Correlation of drug resistance pattern with Lipase Production in clinical isolates of Klebsiella pneumoniae

Kanimozhi Devanathan (kanimozhidevanathan57@gmail.com)
Mahatma Gandhi Medical College and Research Institute
Umadevi Sivaraman
Mahatma Gandhi Medical College and Research Institute
Pramodhini Subramaniam
Mahatma Gandhi Medical College and Research Institute
Joshy Easow
Mahatma Gandhi Medical College and Research Institute

Research Article

Keywords: Klebsiella pneumoniae, Lipase Production, Drug Resistant, ESBL, Carbapenemase production.

Posted Date: April 20th, 2022

DOI: https://doi.org/10.21203/rs.3.rs-1447332/v2

Abstract

Aim

The aim of our study is to correlate drug resistance patterns with Lipase production in clinical isolates of *Klebsiella pneumoniae.*

Materials and Methods

A total of 150 clinical isolates of *Klebsiella pneumoniae* from various clinical samples collected during the period from July 2020 to December 2020 was included in this study. Lipase production was detected by tween 80 agar and an antibiotic susceptibility test has been performed by Kirby-Bauer disk diffusion method as per CLSI guidelines [10]. Extended Spectrum Beta-lactamase (ESBL) producers were detected by combined disc diffusion method and Carbapenemase production was detected by using the E-test strip method.

Results and Discussion

Out of 150 consecutive non-duplicate isolates of *Klebsiella pneumoniae*, 74 isolates from Exudate, 7 from Blood, and 69 were from urine. Among 150 isolates, 85(56.6%) isolates produced Lipase production, 43(57.3%) were ESBL producers and 20(32.25%) isolates were positive for Carbapenemase production.

Conclusion

Showing virulence in clinical isolates of *Klebsiella pneumoniae,* and the antibiotic susceptibility pattern with lipase production in *Klebsiella pneumoniae* were noticed. If the virulence is increased in *Klebsiella pneumoniae,* a drug-resistant pattern also shows more resistance in number. And finally, to correlate the ESBL and Carbapenemase producer with lipase production, in that also the ESBL and Carbapenemase producer shows more number with lipase production. And also noted, when compared with outpatients, inpatients show more lipase production. And hence ampicillin is an intrinsic resistance to *Klebsiella pneumoniae.* In this study, to correlate the drug-resistant pattern, ESBL, and Carbapenemase producer with lipase production in clinical isolates of *Klebsiella pneumoniae.*

Introduction

Klebsiella pneumoniae is a member of the *Klebsiella* genus of *Enterobacteriaceae* and belongs to the normal flora of the human mouth and intestine. The most pathogenic subspecies of *K. pneumoniae* is *K. pneumoniae* subspecies *pneumoniae*. It is responsible for severe lobar pneumonia, urinary tract infections, meningitis (neonates), septicemia, and pyogenic infections such as abscesses and wound infections [1].

Overuse and misuse of antibiotics lead to antibiotic resistance and it shows a significant public health concern. Due to the expression of virulence factors and occurrence of antibiotic resistance in *K. pneumoniae* is considered as pathogenic bacteria in underlying nosocomial infections. Worldwide, nowadays the emergence of antimicrobial resistance is a rapidly increasing challenge in healthcare institutions. Due to the reduced effectiveness of therapy options pathogenic bacteria like *K. pneumoniae* are quickly developing multidrug resistant (MDR)strains and commonly pose a serious threat to the patients because of an increased fatality rate [2].

Prolonged hospitalization infections, high mortality, and morbidity mainly due to Carbapenemase-producing strains. The many other resistance determinants are usually associated with Carbapenemase and give rise to multidrug resistance [3–5].

The survival of the pathogenic and commensal microbes regularly interfaces with their host. The nutrient acquisition, adherence, and evasion of host antimicrobial defenses facilitate the secreted factors and the production of myriad surfaces of the microbe. lipases secreted constitute a class of bacterial enzymes that plays a significant role in both microbial infection and commensalism. In lipid-rich environments, many microbes express Lipase to break down host-derived lipids into free fatty acids for nutrient acquisition, which promotes bacterial colonization and can lead to disease [6].

Materials And Methods

The proposed study has been carried out in the tertiary care hospital, Pondicherry, India. A total of 150 consecutive, nonduplicate isolates of *K. pneumoniae* were collected from various clinical specimens such as urine, pus, Sputum, ET aspirate, bronchoalveolar lavage, wound swab, tissue, vaginal swab, and blood. This study was done after approval by the Institutional Ethical committee.

Antibiotic Susceptibility Testing

Antibacterial susceptibility of all the isolates of *K. pneumoniae* has been determined by the standard Kirby-Bauer disc diffusion method as per the CLSI guidelines [9–10]. Confirmation of extended-spectrum beta-lactamase detection by combined disc diffusion method.

Detection of ESBL Production

ESBL detection was done by phenotypic test using combined disc recommended by CLSI [9] and the antibiotic discs used were cefotaxime and cefotaxime combination with clavulanic acid. *E. coli* ATCC 25922 and *K. pneumoniae* ATCC 700603 were used as quality control strains.

Figure – 1: Confirmatory Test for ESBL production

Figure – 1 shows the confirmatory test for ESBL

Detection of Carbapenemase Production

The confirmation of Carbapenemase production by using the E-test method strip method as per CLSI guidelines. And the strip used was ertapenem/ertapenem with boronic acid (Hi-Media).

Figure – 2: Confirmatory Test for Carbapenemase production

Figure – 2 shows the confirmatory results for Carbapenemase production.

Detection of Lipase activity

The isolates were spot inoculated on tween 80 agar. After a week of incubation at 37°C, Lipase-producing isolates were found to form an opaque precipitation zone [7]. *K. pneumoniae* ATCC 700603 was used as a quality control strain.

Figure – 3: Lipase producing colonies in tween 80 agar

Figure – 3 shows the confirmatory results for Lipase producers.

Results

In our study 150 nonconsecutive, nonduplicate *Klebsiella pneumoniae* isolates were tested for Lipase production and ESBL& Carbapenemase production. Out of 150 isolates, 74 were from Exudate, 7 were from Blood, and 69 were from urine. From those 85(56.6%) isolates produced Lipase enzyme. The number of ESBL producers and Carbapenemase producers was found to be at respectively 43(57.3%) and 20(32.25%).

Table – 1: Total K. pneumoniae **Isolates from clinical samples**

Type of Sample	No. of Isolates
Pus	35
Wound swab	7
Sputum	11
ET Aspirate	10
Broncho alveolar lavage	2
Blood	7
Urine	69
Tissue	4
Vaginal Swab	3
Pleural fluid	1
Ear swab	1
Total	150

Table – 1 represents the total number of *K. pneumoniae* isolates 150, it is found that the urine sample is the highest and is followed by the pus sample and then other samples.

Antibiotic Resistance Pattern of all Isolates

Table – 2: Antibiotic Resistance Pattern (N = 150)

Antibiotic Discs	Resistant n (%)
Cotrimoxazole	42(28%)
Ceftriaxone/ cefotaxime	75(50%)
Ciprofloxacin/ norfloxacin	62(41.3%)
Gentamicin	44(29.3%)
Amikacin	36(24%)
Imipenem	37(25%)
Meropenem	25(16.6%)
Piperacillin+Tazobactam	9(11.1%)
Cefperazone+Sulbactam	21(14%)
Nitrofurantoin	26(37.68%)
Nalidixic acid	25(36.2%)

To the third-generation cephalosporin antibiotics, resistance is high, followed fluoroquinolones drugs (ciprofloxacin/norfloxacin – 41.3%) is more resistant than aminoglycosides drugs (gentamicin, amikacin – 29.3%) and other drugs.

Figure - S1 shows the antibiotic resistance patterns of *K-P* isolates.

Table – 3: Antibiotics Resistance Pattern of inpatient and outpatient

Antibiotics	Inpatient n=90	Outpatient n=60	P-Value
	Resistant	Resistant	
Cotrimoxazole *	30(33.3%)	12(20%)	0.002
Ceftriaxone/cefotaxime *	57(63.3%)	18(30%)	.00001
Ciprofloxacin/Norfloxacin *	45(50%)	17(28.3%)	.000244
Gentamicin	26(28.8%)	18(30%)	.0997
Amikacin*	28(31.1%)	8(13.3%)	.00047
Imipenem*	26(28.8%)	10(16.6%)	.00374
Meropenem*	18(20%)	7(11.6%)	.01197
Piperacillin+Tazobactam	9(10%)	-	0
Cefperazone+Sulbactam*	17(18.8%)	4(6.66%)	.001933
Nitrofurantoin*	22(24.4%)	4(6.66%)	.0002
Nalidixic acid*	18(20%)	7(11.6%)	.01197

* Significant

Isolates from inpatient samples were found to be more resistant. The resistance pattern significantly differs in cotrimoxazole, ceftriaxone/cefotaxime, ciprofloxacin / Norfloxacin, Amikacin, Imipenem, Meropenem, Cefperazone + Sulbactam, Nitrofurantoin, Nalidixic acid.

Table – 4: - Confirmatory of ESBL production in K. pneumoniae **Isolates**

Total Isolates	Number of third Generation Cephalosporin Resistant isolates	ESBL Positive
150	75	43

Figure - S2 shows the representation of the data from the table, Table – 4.

Table – 5: Confirmatory of Carbapenemase production in K. pneumoniae **Isolates**

Total Isolates	Carbapenem Resistant	MBL Confirmative
150	37	20

Figure - S3 shows Carbapenemase production in *K. pneumoniae* Isolates.

Figure - S4 shows the positive and negative in Lipase Tests.

Table – 6: Lipase Test Positive and Negative results

Lipase Test N=150	Inpatient N=90	Outpatient N=60
Lipase positive (85)	64(75.29%)	21(24.7%)
Lipase Negative (65)	26(40%)	39(60%)

Table – 6 indicates the inpatient and outpatient drug-resistant pattern.

Table – 7: Antibiotic Drug Resistance for Lipase Test

Antibiotics	Lipase test positive resistant N=85	Lipase test negative resistant N=65	P-Value
Cotrimoxazole	23(27.05%)	19(29.23%)	0.237
Ceftriaxone/cefotaxime	35(41.17%)	40(61.53%)	0.694
Ciprofloxacin/Norfloxacin	24(28.23%)	38(58.46%)	0.962
Gentamicin	27(31.76%)	17(26.15%)	0.0580
Amikacin	20(23.5%)	16(24.61%)	0.2202
Imipenem	18(21.17%)	19(29.23%)	0.522
Meropenem	15(17.64%)	10(15.3%)	0.132
Piperacillin+Tazobactam	6(7.05%)	3(4.61%)	0.111
Cefperazone+Sulbactam	10(11.76%)	11(16.92%)	0.530
Nitrofurantoin*	20(23.52%)	6(9.23%)	0.0027
Nalidixic acid	15(17.64%)	10(15.38%)	0.6288

*- Significant

Table-7 indicates the antibiotic-resistant pattern of both Lipase positive and Lipase negative. Further, it is concluded that Lipase positive has more drug resistance patterns when compared to Lipase negative *K. pneumoniae* isolates.

Table – 8: Correlation between ESBL producer and Carbapenemase Producer with Lipase Production

Lipase Test	ESBL producer N=43	Carbapenemase producer N = 20
Lipase Positive	30 (69.7%)	13(65%)
Lipase Negative	13(30.23%)	7(35%)

In Table – 8, it has been shown that number of positive and negative Lipase tests ESBL producers when N = 43 and the number of positive and negative Lipase tests Carbapenem producers when N = 20.

Figure – S5 shows a representation of the number of positive and negative Lipase tests in ESBL and Carbapenem producers when N = 30 and N = 13 respectively.

Discussion

In this study, the proportion of *K. pneumoniae* isolates was 150 of the total clinical bacterial isolates from July 2020 to December 2020. In hospital-acquired infections, *Klebsiella pneumoniae* is the main organism that causes highly contagious outbreaks with increased mortality rates and longer stays. These are all results in inflated health care costs [24].

Most of *K. pneumoniae* in this study were obtained from patients aged 18 to 65 years of age. *K. pneumoniae* isolates were mainly isolated from urine specimens followed by pus. Ashurst and Dawson [25] reported that *K. pneumoniae* colonizes typically in the urinary tract and in invasive infections. In the United States, *Klebsiella pneumoniae is* considered to be the most common cause of hospital-acquired infections. The people from the republic of China reported that the respiratory tract was the main site infection of *Klebsiella pneumoniae* was studied by wang et.al. In comparison, Seifi et al [26] who collected samples from two hospitals in Tehran reported that *K. pneumoniae* samples were isolated from urine, surgical wounds, sputum, and blood with the percentage of

61.7%, 18.1%, 11.7%, and 8.5% respectively. *K. pneumoniae* isolates in inpatients and outpatients were 47 and 18 and in blood 3 and in urine 40 and 42. In a total 0f 58 *K. pneumoniae* isolates, 22 isolates (37.9%) were female and 18 (31%) were from males. In another study 34 (58.6%) isolates from inpatients, which were obtained from intensive care unit 12 (20.7%), pediatrics 7 (12.1%), emergency 7 (12.1%), internal medicine 4 (6.9%), burn 1 (1.7%), surgery 1 (1.7%), ear, nose, and throat department 1 (1.7%) and neurology department 1 (1.7%). About 46.5% of the sample was collected from outpatients and 53.5% were from inpatients.

Table – 9: Lipase Test Percentage

Previous Studies	% Of Lipase Test Positive
Gharrah MM *et al.* (8)	6% & 10%
Alam NG *et al.* (11)	76.9%
Greice H.S. Peil *et al.* (12)	61.90%
Emmanuel M.B.*et al.*(13)	18.8%
Kalaivani.*et al.* (14)	58.2%
Kanimozhi et al	85%

We have compared the results of various authors' findings in this research topic for Lipase production to our study in the form of the table in Table – 9. Results show that our study provides more positive results for Lipase production using the methods we discussed in our paper.

Most of K. *pneumoniae isolates were* resistant to various antibiotics with Ampicillin, Ceftriaxone/Ccefotaxime, and Cefepime + Tazobactam being the least effective for *K. pneumoniae* while piperacillin + tazobactam, Cefperazone + Sulbactam, nalidixic acid, and Meropenem had the most favorable profile.

This report is compared by the study conducted by Madahiah et al [28] that found *K. pneumoniae* isolates were 100% resistant to ampicillin and 100% sensitive to amikacin., Ciprofloxacin and Amoxicillin-clavulanic acid showed 38.75% and 36.69% resistance respectively. This finding is similar to Cepas et al [30] that reported 40% of *K. pneumoniae* strains were resistant to ciprofloxacin and amoxicillin-clavulanic acid.

The over use and prescription of antibiotics becomes a commonly known problem for antimicrobial resistance. Many factors such as the use of antibiotics in community, hospital, even in animal production, agriculture, and environment are involving the growth of antibiotic resistance. The antibiotics are used excessively since there is no control and/or restriction in purchasing antibiotics freely without prescription. The main underlying factor in the widespread transmission of difficult to cure antibiotic resistant nosocomial infections is intensive and prolonged use of antibiotics in health setting service.

In our study, in ESBL detection the third-generation Cephalosporin-resistant was 75(50%) and Carbapenem-resistant strains were 37 (25%) in a total number of isolates. After screening, ESBL and Carbapenem-resistant strains were 43 and 20. In that lipase positive has 30 (69.7%) isolates were ESBL positive and 13 (65%) isolates were Carbapenemase positive whereas in lipase negative 13 (30.23%) and 7(35%) isolates produce ESBL and Carbapenemase production.

ESBL s are now a problem in hospitalized patients worldwide. Their prevalence varies from one country to another and from institution to institution was studied by Asma et al. in Kuwait (2006) [15]. First isolated in 1983 in Germany, ESBLs spread rapidly to Europe, the United States, and Asia and are now found all over the world was studied by Kumar et al in India. From India [16], the high prevalence of ESBL producing isolates showed ESBL production was studied by Jain A [17]. In 2002, 68% of gram-negative bacteria were found to be ESBL producers in a study from New Delhi in which 80% of *Klebsiella* were ESBL s was studied by Gupta V in India in 2007[18]. All 117 multidrug-resistant K. *pneumoniae* isolates were cefotaxime resistant. Out of these isolates, 91 isolates were ESBL positive by Ceftazidime clavulanic acid combined disc method, and 95 isolates positive in HI chrome ESBL agar [19].

Faizabad et al in Iran found that 66% of the isolates were Carbapenemase producers [20]. Gupta et al in north India studied meropenem resistance was 6.9% [21] whereas Nagaraj et al 2012 observed 75% of the *K. pneumoniae* isolates were Carbapenemase resistant in their study in South India [22]. Azeem et al stated that 35.3% of K. *pneumoniae* isolates in their study were resistant to Carbapenemase production in 2016 [23].

Carbapenems are not easily hydrolyzed beta-lactamase enzymes because they exhibit great affinity towards penicillin-binding proteins. Through porin channels, they can easily enter the gram-negative bacterial cell.

In our study 85 (56.6%) isolates showed lipase production in similar to our study (58%) isolates produce lipase production was studied by Kalaivani et al in Pondicherry [14]. Gharrah et al encountered 6% and 10% lipase production among their ESBL and non-ESBL producers [8]. Allam et al is reported in 76.9% lipase formation [11].

The survival of the pathogenic and commensal microbes regularly interfaces with their host. They do so through the production of myriad surface and secreted factors that facilitate the nutrient acquisition, adherence, and evasion of host antimicrobial defenses.

Lack of education about infection and antibiotic usage is the major cause of prescribing inappropriate antibiotics. In initial antibiotic therapy, one of the most relevant steps in prescribing antibiotics is an adjustment based on the clinical microbiology result. Therefore, it is necessary to perform antibiotic susceptibility testing. Collecting clinical samples before antibiotic administration is also a critical point. Adjusting the initial antimicrobial therapy based on the clinical microbiology result will diminish the selection pressure to the microorganism in hospital-based infections. Thus, it is of paramount importance for each hospital to have an antibiotic guidance or stewardship program for all pharmacists and physicians based on the most accurate microbiological data. In conjunction with this guidance, a continuous effort in hospital surveillance, infection control, and clinical audits must be conducted to fight against the rapid development of antibiotic-resistant pathogens.

In this study, the antibiotics like Nitrofurantoin, Nalidixic acid, Cotrimoxazole, Cefotaxime / Ceftriaxone, Ciprofloxacin/Norfloxacin, Amikacin, Imipenem, Meropenem, Cefoperazone, and sulbactam are statistically significant in inpatient when compared with outpatient.

Patients admitted to ICUs are at greatest risk of acquiring nosocomial infections, partly because of their serious underlying disease but also because of exposure to life-saving invasive procedures, prolonged use of in situ invasive devices, therapy with multiple antimicrobials, and extended hospital stays [27–28]. In the lipase test, drug-resistant pattern, the antibiotic nitrofurantoin was only statically significant in lipase positive compared with lipase negative isolates.

Conclusion

In this research paper, it is shown that the Lipase enzymatic production in multidrug-resistant *Klebsiella pneumoniae* isolates are statistically significant in inpatient when compared with outpatient. This study highlights ESBL producer and Carbapenemase producer correlation to Lipase production as pointed out in Table – 8. Further this study highlights the antibiotic resistance pattern from various clinical samples are found to be statistically significant in inpatient when compared with outpatient as pointed out in Table-2. Moreover, the comparative studies of various authors in Lipase production have been listed in Table – 9, whereas our result show higher percentage of Lipase production.

As we have known that detection of virulence factors from the Lipase enzymatic production in this work may help in the antibiotic prescription and use management process, as the presence of virulence factors increases pathogenic ability leading to the therapeutic challenge.

Most of the *K. pneumoniae* isolates as pointed out here showed resistance to a wide range of antibiotics from the Table – 3.

Declarations

Acknowledgements:

Not applicable

Funding:

Not Applicable

Affiliations:

D. Kanimozhi

PhD Scholar , Department of Microbiology,MGMCRI, Sri Balaji Vidyapeeth University, Pondicherry.

S. Umadevi

Professor, Department of Microbiology, MGMCRI, Sri Balaji Vidyapeeth University, Pondicherry.

S. Pramodhini

Professor, Department of Microbiology, MGMCRI, Sri Balaji Vidyapeeth University, Pondicherry.

Joshy M Easow

Head of the Department of Microbiology, MGMCRI, Sri Balaji Vidyapeeth University, Pondicherry.

Contributions:

D. Kanimozhi is the Ph.D. scholar is the mainly responsible person for all the experiments design of Lipase production in twin 80 agar, Carbapenemase production, and ESBL production. Further Ms. Kanimozhi is source of all ideas with regards to the results and data productions and is the corresponding author.

S. Umadevi is Ph.D. thesis supervisor for guidance and is supportive in editing the manuscript

S. Pramodhin is the Ph.D. co-supervisor and is responsible for manuscript editing and suggestion to improve the manuscript

Joshy M Easow is responsible for providing space and laboratory facilities for Ms. Kanimozhi and is further responsible for auditing and validating ethical committee approval

Corresponding Author:

D. Kanimozhi

Ethics Declaration:

Ethics approval certificate applicable and can be provided upon request

Consent of Publication:

All authors are consent-able for this publication

Availability of Materials and Data:

The datasets used and / or analyzed in the current study are available from the corresponding author upon reasonable request.

Competing Interests:

The authors declare that they have no competing interests.

References

1. Ananth Narayanan R, Panikkar CKJ. Textbook of Microbiology. 7th Edition of Microbiology. Blackswan. 2006.
2. Levy SB. Impacting on the problem of Antibiotic Resistance. Journal of Antimicrobial Chemotherapy,2002;49(1):25–30.
3. Center for Disease Control and Prevention. Guidance for control of infections with Carbapenemase-resistant of Carbapenemase producing Enterobacteriaceae in acute care facilities. MMWR Morbidity and mortality weekly report. 2009;58: 256–260.
4. Carmeli Y, Akova M, Cornaglia G, Daikos GL, Garau J, Harbarth S, Rossolini GM, Souli M, Giamarellou H. Controlling the spread of Carbapenemase-producing Gram negatives: Therapeutic Approach and Infection control. Clin Microbial Infect, 2010;16: 102–111.
5. Cohen MJ, Block C, Levin PD, Schwartz C, Gross I, Weiss Y, Moses AE, Benson S, Institutional control measures to curtail the epidemic spread of Carbapenemase-resistant *Klebsiella pneumoniae*: a 4- year perspective. Infect Control Hosp Epidemiol. 2011;32: 673–678.
6. Bender J, Flieger A. (2010) Lipases as Pathogenicity Factors of Bacterial Pathogens of Humans. In: Timmis K.N. (eds) Handbook of Hydrocarbon and Lipid Microbiology. Springer, Berlin, Heidelberg. https://doi.org/10.1007/978-3-540-77587-4-246.
7. Chen Xi, Francis III A. Bacterial lipolysis of immune-activating ligands promotes evasion of innate defenses. Proc Natl Acad Sci U S A. 2019; Feb 26; 116(9): 3764–3773.
8. Gharrah MM, E1- Mahadi AM, Barwa RF. Association between virulence factors and Extended-spectrum beta-lactamase-producing *K. pneumoniae* compared to non-producing isolates. Interdisciplinary perspectives on infectious diseases. 2017;1–14. Article ID 7279830.
9. Clinical and Laboratory Standards Institute (CLSI). Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria that grow aerobically. 11th Edition, CLSI, Wayne, PA, USA, M7-A72006; 26.
10. Bauer AW, Kirby WM; Sherris JC, Truck M. Antibiotic susceptibility testing by a standardized single disk method. American Journal of Clinical Pathology. 1966;45(4): 493–96.
11. Alam M, Imran M. Screening and potential of Gram-Negative Bacterial Isolates for their Extracellular Enzymatic Activities Isolated from the Hospital Aquatic Environment. J Basic Clin Pharma. 2018;9: 41–5.
12. Peil GHS, Kuss AV, Rave AFG, Villarreal JPV, Hernandez YML, Nascente PS. Bio respecting of lipolytic microorganisms obtained from industrial effluents. An acid Bras Cinch. 2016; 88:3.
13. Esakkiraj P, Marriappan R, Palavesam A, Immanuel G. *Annals of Microbiology*. Lipase production by *Staphylococcus epidermidis* CMST-Pi 1 isolated from the gut of shrimp Penaeus indicus. 2010;60: 37–42.
14. Kalaivani R, Jenitha JP, Seetha KS. Clinical isolates of *Klebsiella pneumoniae* and its virulence factors from a tertiary care hospital. Indian Journal of Microbiology Research. 2019;6(2): 109–112.
15. Asma M Al-Jasser. Extended- Spectrum β lactamase (ESBL)s: A Global problem review article. Kuwait Medical Journal.2006;38(3):171–185.
16. Kumar MS, Lakshmi V, Rajagopalan R. Occurrence of extended spectrum β-lactamase among *Enterobacteriaceae* spp. isolated at a tertiary care institute. Ind J Med Microbiol.2006,24(3):208–11
17. Jain A, Mondal R. Prevalence of antimicrobial resistance pattern of extended spectrum β lactamase producing Klebsiella species isolated from cases of neonatal septicaemia. Ind J. Med Res.2007; 125:89–94.
18. Gupta V. An update on newer β lactamase. J. Med. Res.2007:126(5):417–27.
19. Kalaivani R, Kali A, Sah Suresh, Pagal S, Prashanth K, Seetha S K. Molecular profile of Emerging Multidrug Resistant *Klebsiella pneumoniae* clinical isolates from Southern India. Journal of Clinical and Diagnostic Research,2019:13(3): DC01-DC06.
20. Faizabad MM, Delfani S, Raji N, Majnooni A, Alogholi M, Shacheraghi F, Parvin M, and Yadegarinia D. Distribution of blab (TEM), blab (SHV), blab (CTX-M) genes among clinical isolates of *Klebsiella pneumoniae* at Labefied Hospital, Tehran, Iran. Iran Microbe Drug Resist. 2013;16: 49–53.
21. Gupta E, Mohanty S, Sood S, Dhawan B, Das K Bimal, Kapil A. Emerging resistance to carbapenems in a tertiary care hospital in north India. Indian J Med Res.2006;124: 95–8.

22. Nagaraj A, Chandran SP, Shamanna P, Macaden R. Carbapenem resistance among *Escherichia coli* and *Klebsiella pneumoniae* in a tertiary care hospital, South India. Indian J Med Microbial. 2012;30: 93–95
23. Azeem R, Shenoy S, Ashish A. Approach to Carbapenemase detection in *Klebsiella pneumoniae* in routine diagnostic laboratories. J. Clin. Diagn. Res. 2016;10: 24–27.
24. Kidd TJ, Mills G, Sa- Pessoa J, Dumigan A, Frank CG, Insua JL, Ingram R, Hobley L, Bengoechea JA. A Klebsiella *pneumoniae* antibiotic resistance mechanism that subdues host defenses and promotes virulence. EMBO Mol Med.2017;9(4): 430–447.doi:10.15252/emmm.20607336.
25. Ashurst JV, Dawson A. *StatPearls.* Treasure Island (FL) 2019. *Klebsiella pneumoniae*.
26. Seifi K, Kazemain H, Heidari H, Rezagholizadeh F, Saee Y, Shirvani F, Houri H. Evaluation of biofilm formation among *Klebsiella pneumoniae* isolates and molecular characterization by ERIC-PCR. Jundishapur J Microbiol.2016;9(1): e30682.doi10.5812/jjm.30862.
27. Jarvis WR, Edwards JR, Culver DH, et al. Nosocomial infection rates in adult and pediatric intensive care units in the United States. Am J Med 1991;91(suppl 3B): S185-91.
28. Donowitz LG, Wenzel RP, Hoyt JW. High risk of hospital-acquired infections in ICU patients. Crit Care Med 1982; 10:355–7.
29. Madahiah BM, Noor US, Abdul S, Dan Ali A Q, *Klebsiella pneumoniae* urinary tract infections associated with long- term catheterization and spinal cord injuries. J. Med Sci. 2002;2: 227–9
30. Cepas V, Lopez Y, Munoz E, Rolo D, Ardanuy C, Marti S, Xeracavins M, Horcajada JP, Bosch J, Soto SM. Relationship between biofilm formation and antimicrobial resistance in gram-negative bacteria. Microb Drug Resist. 2019;25(1):72–9.

Figures

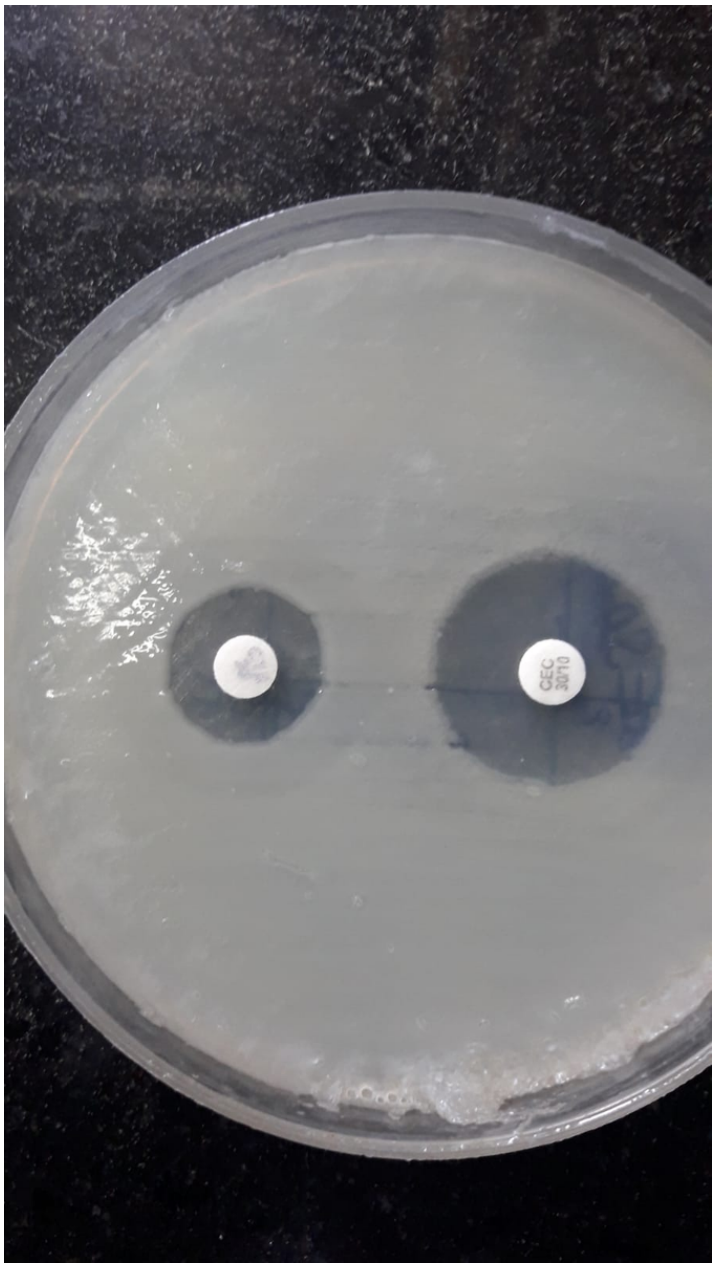

Figure 1

Confirmatory Test for ESBL production

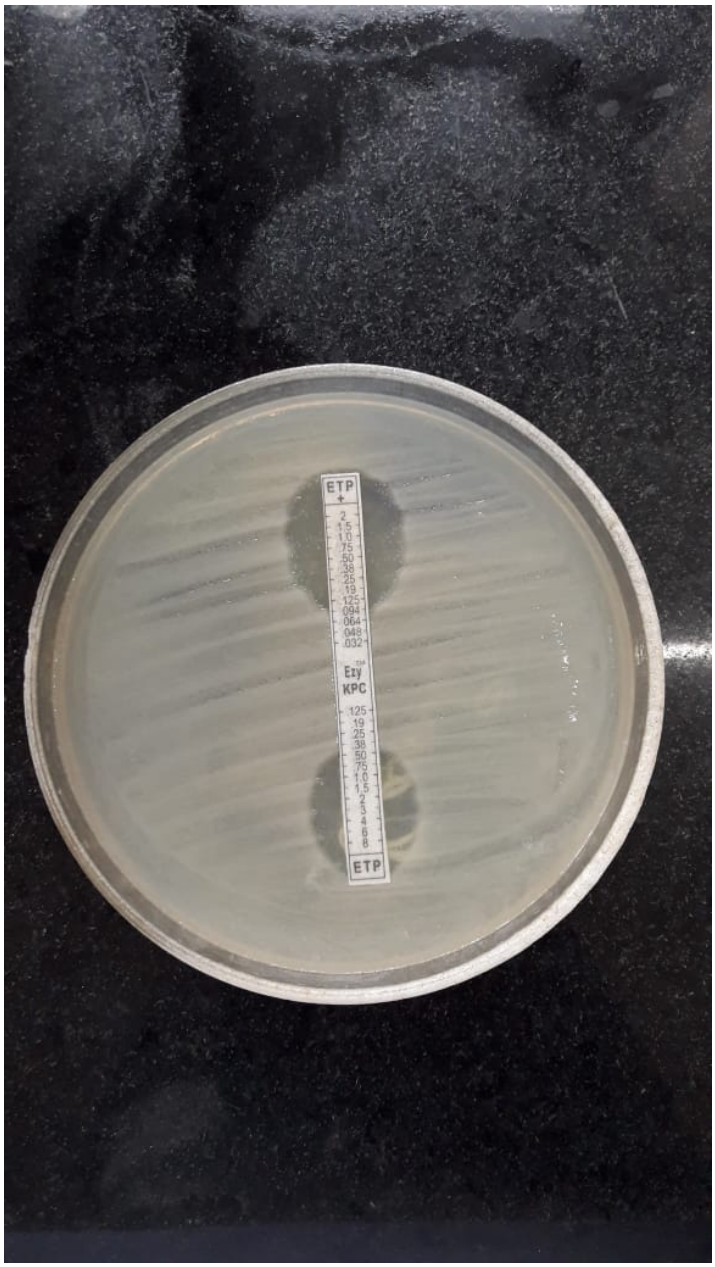

Figure 2

Confirmatory Test for Carbapenemase production

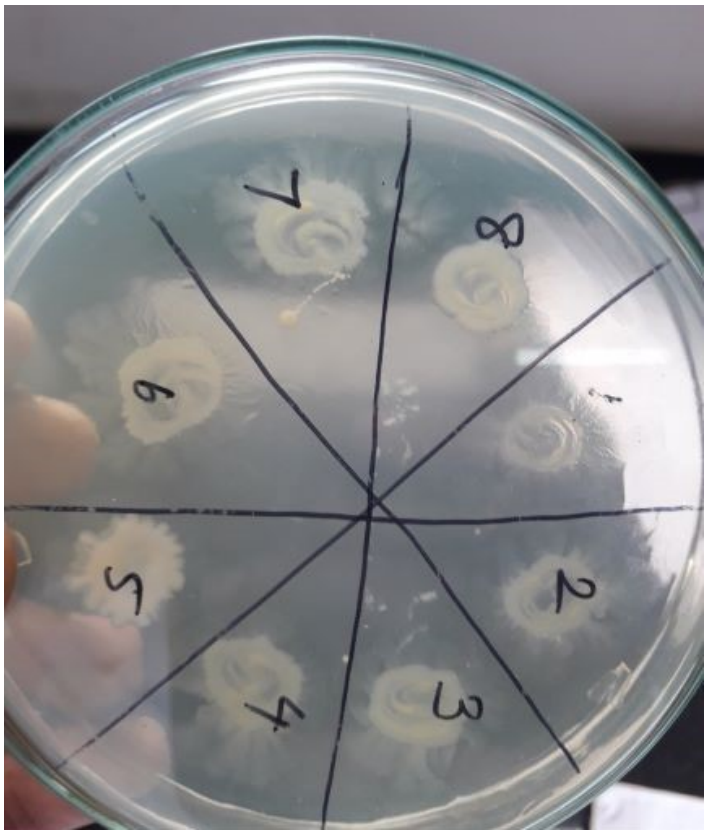

Figure 3

Lipase producing colonies in tween 80 agar

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- CorrelationofdrugresistancepatternwithLipaseProductioninclinicalisolatesofKlebsiellapneumoniaeKanimozhiSuppFigure.docx