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Prevalence of Typical Bacterial Load and their Susceptibility to Antimicrobial Agents in Patients with Lower Respiratory Tract Infections in Eastern Nepal

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Abstract:-
Background: The lower respiratory tract infections are a prime cause of morbidity and mortality in our country. A hospital based cross sectional study was fulfilled with an objective to study the bacterial pathogens isolated and susceptibility pattern from patients of lower respiratory tract infections.

Methods: Overall 826 lower respiratory specimens such as sputum, endotracheal aspirates, bronchial aspirate and pleural fluid were cultured aerobically using standard microbiological technique as per Clinical Laboratory Standard Institute guidelines.

Results: Out of 826 lower respiratory specimens, 523(63.3%) bacterial pathogens were recovered. *Streptococcus pyogenes* (65.8%) was the commonest isolate and increase isolation rate was observed in the male 291(55.64%) of 60-79 years age groups. Most of the pathogens showed sensitive to macrolides and aminoglycoisides, however, *Streptococcus pyogenes* and *Staphylococcus aureus* were most susceptible to tetracycline 262(75.9%) and azithromycin 53(73.6%). In case of *Pseudomonas aeruginosa* were sensitive to amikacin 22(64.7%) while *Acinetobactor baumani* were found sensitive to amikacin 16(80%) and *Klebsiella pneumoniae* was sensitive to penicillin 37(71.1%).

Conclusions: For effective treatment of typical bacterial lower respiratory tract infections, bacterial culture and antibiotic sensitivity testing are required.

Keywords:- Lower Respiratory Tract Infections, Bacterial Pathogens, Antibiotic sensitivity.

I. INRTODUCTION

Respiratory tract is the most common site for infection by bacterial pathogens as it is in direct contact with the environment and continuously exposed to microorganisms suspended in the air. In low-income and middle-income countries, a lower respiratory tract infection (LRTI) is the fifth-leading cause of death (1). In some underdeveloped countries, the situation is more complicated and management is often difficult due to the problem associated with inadequate health facilities for the bacterial culture and the antibiotic susceptibility testing in cases requiring antibiotic therapy (3). The most common etiological agents that cause LRTI are *Streptococcus pneumoniae, Streptococcus pyogenes, Haemophilus Influenzae, Staphylococcus aureus, Enterococcus spp, Klebsiella pneumoniae, Pseudomonas aeruginosa, Acinetobacter baumani, Citrobacter spp.*and *Moraxella catarrhalis* (2). In community-acquired pneumonias, the most common bacterial agent *is Streptococcus pneumoniae* (5). Nosocomial pneumonias and pneumonias in immunosuppressed patients have protean etiology with gram-negative organisms and *Staphylococci* as predominant organisms (2).

Atypical bacterial pathogens are Mycoplasma *pneumoniae, Chlamydia spp, Legionella, Coxiella burnetti* and viruses (6). LRTIs are a continuation and a major health problem in the society and antimicrobial resistance is one of the greatest warning to global public health (7, 8). The prevalence of antibiotics resistance in lower respiratory pathogens among patients increases the risk of uncontrollable infections, prolonged hospital stay and increased mortality (9). The bacterial culture and antibiotic sensitivity of lower respiratory bacterial pathogens play a significant role in controlling of empirical antibiotics and hospitalization measures (10, 11, 12, and 13). The aim of our study was to identify the respiratory bacterial pathogens and their antibiotic sensitivity pattern in all patients presented with clinical manifestations of LRTI at a Birat Medical College Teaching Hospital (BMCTH) in Nepal.

II. MATERIALS AND METHODS

A hospital based cross sectional study was conducted to determine the lower respiratory typical bacterial pathogens and their antibiotic sensitivity pattern in all patients of all age groups, presented with clinical manifestations of LRTI attended in BMCTH, Nepal from May 2021 to October 2021, the study got ethical clearance (IRC-PA-130) from the Institutional Research committee of Birat Medical College and Teaching Hospital. The early morning sputum samples were collected after giving proper instructions to the patients in a sterile container, the broncho alveolar lavage (BAL) fluid was collected aseptically in a sterile container by aspirating the saline, tracheal secretions received in the laboratory in a Lukens trap and pleural fluid for direct examination and culture. On receiving the specimen were processed for isolation, identification and AST.

A. Inclusion and exclusion criteria

Properly collected specimens of sputum, tracheal aspirate, bronchial alveolar lavage, pleural fluid with completes filled in requisition forms of all LRTI cases attending Birat Medical College and Teaching Hospital were included. The exclusion criteria were inadequate and contaminated specimens and incomplete patient's information.

B. Sample processing

The specimens were subjected to microscopic inspection of Gram stained smears for the presence of bacteria and pus cells, as well as bacterial culture for isolation and identification.

The veracity of the specimen was validated by sputum smears revealing less than 10 squamous epithelial cells and more than 25 leucocytes or pus cells per low power field. Mac Conkey agar, blood agar, and chocolate agar media were used to inoculate specimens. (14).

C. Identification of bacteria

The morphologies of the respiratory pathogens, as well as their cultural and biochemical properties, were used to identify them. The CLSI guidelines were followed for testing the antibiotic susceptibility of the bacterial isolates using the modified Kirby-Bauer disk diffusion method on Mueller-Hinton agar. (15, 16, and 17). Antibiotics were used in antibiotic sensitivity testing for respiratory pathogens such as amoxicillin/clavulanic acid (30µg), ciprofloxacin (5µg), ofloxacin (5µg), levofloxacin (5µg), amikacin (30µg), cefepime (30µg), cefuroxime (30µg), ceftazidime (30µg), ceftriaxone (30µg), cotrimoxazole (1.25/23.75µg), aztreonam (30µg), imipenem (10µg), cefoperazone/sulbactum (75/10µg), and piperacillin/ tazobactum (100/10µg).

For antimicrobial susceptibility testing, Klebsiella pneumoniae ATCC 700603, Escherichia coli ATCC 25922, and Pseudomonas aeruginosa ATCC 27853 were utilized as control strains.

D. Data analysis

The data was imported into Microsoft Excel and then analyzed using SPSS V.20. The proportional differences were analyzed using a two-way analysis. The threshold for statistical significance was established at 0.05.

III. RESULT

Specimens	Total specimen	Significant growth	Insignificant growth
Sputum	370(70.6%)	350(94.6%)	20(5.40%)
Tracheal aspirate	91(17.4%)	89(97.5%)	2(2.19%)
Bronchial alveolar lavage	53(10.1%)	49(92.45%)	4(9.43%)
Pleural fluid	10(1.9 %)	8(80%)	2(20%)

Table 1: Rate of isolation of pathogens in different specimens (n=523)

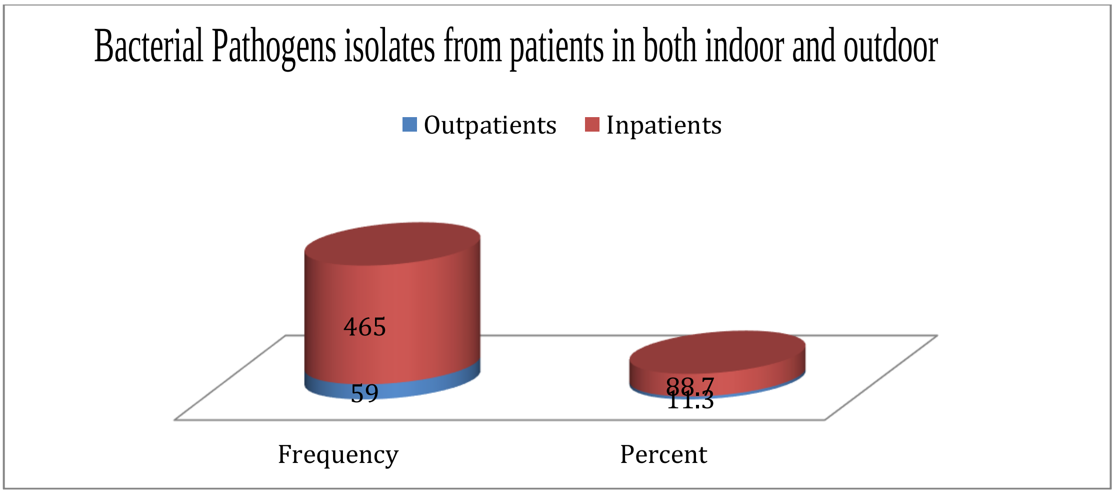

Fig. 1: Bacterial Pathogens isolates from patients in both indoor and outdoor settings (n=523)

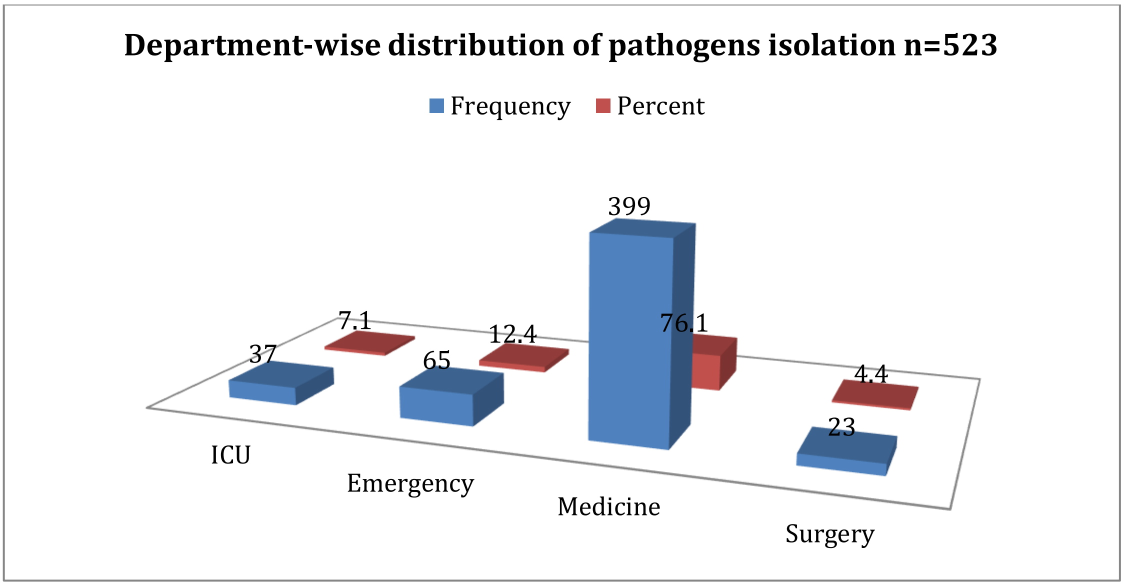

Fig. 2: Department-wise distribution of pathogens isolation n=523

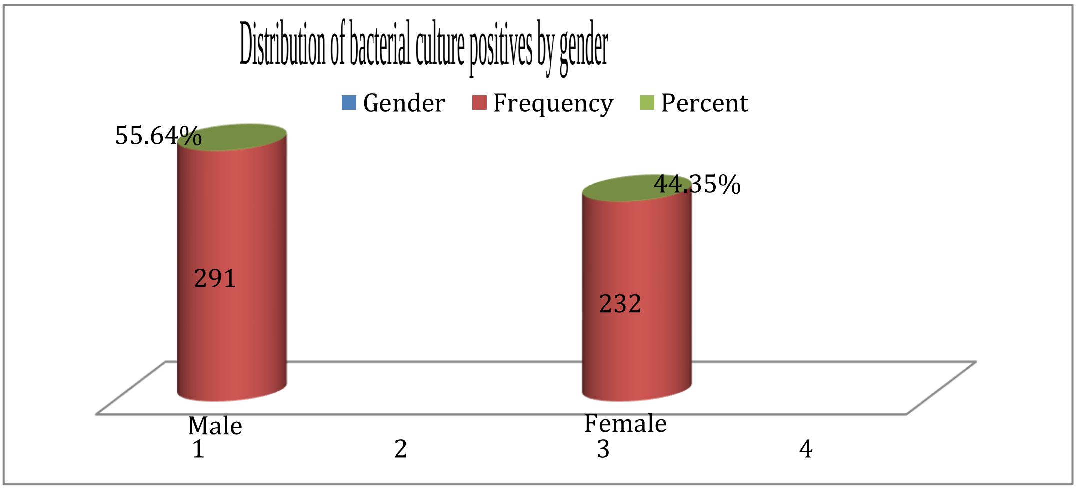

Fig. 3: Distribution of bacterial culture positives by gender (n=523)

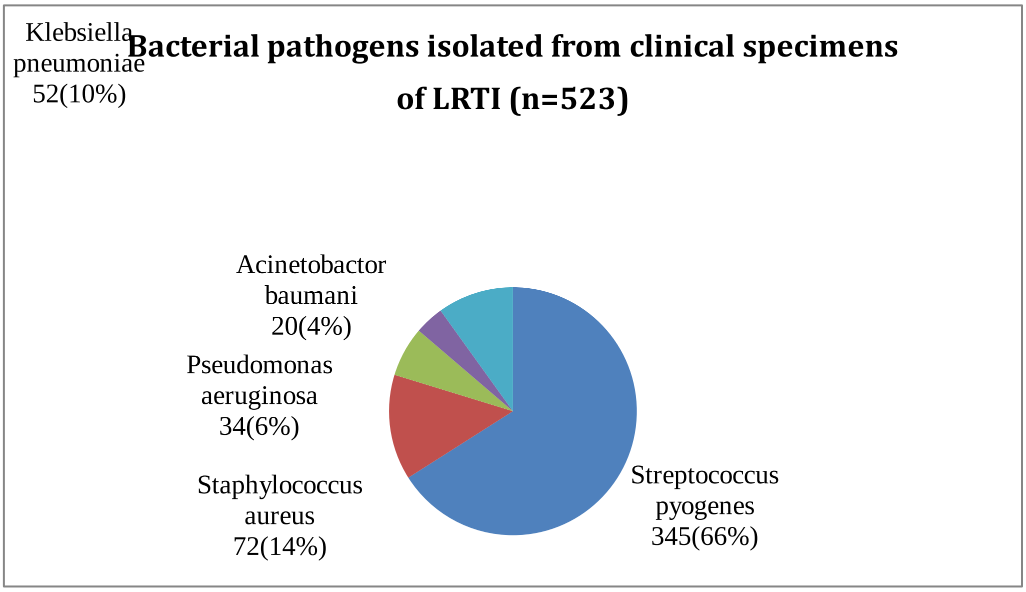

Fig. 4: Age-wise distribution of pathogens isolates from LRTI n=523

Age group	Frequency	Percent
10-19	35	6.7
20-29	49	9.4
30-39	49	9.4
40-49	61	11.6
50-59	87	16.6
60-69	103	19.7
70-79	103	19.7
80+	36	6.9

Table 2

Antibiotics	Sensitive	Resistance
Levofloxacin	83(15.8%)	20(3.8%)
Ofloxacin	155(29.6%)	53(10.1%)
Ciprofloxacin	47 (9 %)	17(3.2%)
Ceftriaxone	254(48.5%)	53 (10.1%)
Cefotaxime	91(17.3%)	31(5.9%)
Cefepime	235(44.8%)	198(37.8%)
Penicillin	266(50.76%)	188(35.9%)
Gentamicin	315(60.1%)	127(24.2%)
Ampicillin	298(56.9%)	148(28.2%)
Azithromycin	349(66.6%)	102(19.5%)
Amikacin	332(63.4%)	136(26%)
co-trimoxazole	269(51.3%)	177(33.8%)
Piperacillin	268(51.1%)	181(34.5%)
Ceftazidime	184(35.1%)	177(33.8%)
Imipenem	261(49.8%)	147(28.1%)
Cefepime	183(34.9%)	184(35.1%)
Amoxycillin	281(53.6%)	110(21%)
Tetracycline	318(60.7%)	85(16.2%)
Erythromycin	224(42.7%)	185 (35.3%)

Table 3: Antibiotics sensitive and resistance pattern of bacterial pathogens from LRTI (n=523)

Antibiotics	*Streptococcus pyogenes n=345*		*Staphylococcus aurus n=72*		*Pseudomonus aeruginosa n=34*		*Acenetobacter baumani n=20*		*Klebsiella pneumoniae n=52*	
	R	S	R	S	R	S	R	S	R	S
Levofloxacin	5(1.5%)	30 (8.7 %)	0(0.0%)	10(13.9%)	3(8.8%)	20(58.8 %)	8(40 %)	3(15%)	4(7.7%)	20(38.5%)
Ofloxacin	30(8.7%)	70(20.3%)	4(5.6%)	36(50%)	6(17.6%)	19(55.9 %)	5(25 %)	2(10%)	8(15.4%)	28(53.8%)
Ciprofloxacin	4(1.2%)	12(3.5%)	0(0%)	5(6.9%)	4(11.8%)	13(38.2 %)	4(20 %)	2(10%)	5(9.6%)	15(28.8%)
Ceftriaxone	31(9%)	182(52.8%)	7(9.7%)	46(63.9%)	5(14.7%)	3(8.8%)	4(20 %)	7(35%)	6(11.5%)	16(30.8%)
Cefotaxime	2(6%)	50(14.5%)	2(2.8%)	6(8.3%)	8(23.5%)	14(41.2 %)	10(50 %)	3(15%)	9(17.3%)	18(34.6%)
Cefepime	132(38.2%)	155(45%)	33(45.8%)	26(36.1%)	10(29.4 %)	18(52.9 %)	6(30 %)	12(60%)	17(32.7 %)	24(46.1%)
Penicillin	136(39.4%)	155(44.9%)	28(38.9%)	39(54.1%)	10(29.4 %)	21(61.7 %)	3(15%)	14(70 %)	10(19.2 %)	37(71.1%)

Gentamicin	91(26.4%)	206(59.7%)	13(18.1%)	51(70.8%)	7(20.6%)	20(58.8%)	7(35%)	8(40%)	8(15.4%)	30(57.7%)
Ampicillin	106(30.7%)	197(57.1%)	21(29.1%)	41(56.9%)	8(23.5%)	20(59%)	1(5%)	13(65%)	11(21.2%)	27(52%)
Azithromycin	72(20.9%)	243(70.4%)	16(22.2%)	53(73.6%)	6(17.6%)	17(50%)	1(5%)	6(30%)	7(13.4%)	29(55.8%)
Amikacin	94(27.2%)	212(61.4%)	18(25%)	46(63.9%)	7(20.6%)	22(64.7%)	4(20%)	16(80%)	13(25%)	35(67.3%)
co-trimoxazole	113(32.8%)	181(52.4%)	27(37.5%)	36(50%)	11(32.4%)	16(47.1%)	8(40%)	10(50%)	17(32.7%)	26(50%)
Piperacillin	122(35.3%)	167(48.4%)	25(34.7%)	37(51.34%)	10(29.4%)	22(64.7%)	8(40%)	11(55%)	16(30.8%)	30(57.7%)
Ceftazidime	114(33.0%)	118(34.2%)	30(41.7%)	23(31.9%)	11(32.3%)	12(35.2%)	1(5%)	9(45%)	21(40.4%)	21(40.4%)
Imipenem	95(27.5%)	170(49.3%)	18(25%)	30(41.7%)	12(35.2%)	18(52.9%)	9(45%)	11(55%)	13(25%)	32(61.5%)
Cefepime	116(33.6%)	127(36.8%)	30(41.7%)	16(22.2%)	11(32.3%)	17(50%)	5(25%)	6(30%)	22(42.3%)	17(32.7%)
Amoxycillin	67(19.4%)	216(62.6%)	14(19.4%)	49(68.0%)	13(38.2%)	3(8.8%)	4(20%)	2(10%)	12(23.1%)	10(19.2%)
Tetracycline	50(14.5%)	262(75.9%)	18(25%)	41(56.9%)	5(14.7%)	1(2.9%)	8(40%)	2(10%)	4(7.7%)	11(21.2%)
Erythromycin	121(35.1%)	174(50.4%)	25(34.7%)	25(34.7%)	14(41.2%)	7(20.6%)	6(30%)	3(15%)	19(36.5%)	14(26.9%)

[Table/Fig-8]: Antibiogram of lower respiratory pathogens (n=523)

R-Resistance, S-Sensitive

IV. DISCUSSION

The second most common cause of hospital-acquired illnesses is respiratory tract infections. The microbiological diagnosis of LRTIs can be difficult since the specimen collection for the study can be contaminated by bacteria that live in the upper respiratory tract, necessitating the use of invasive methods. Despite breakthroughs in treatment and preventive strategies, developing resistance to antimicrobial drugs is a growing worry for doctors around the world. Antimicrobial overuse has also contributed to the formation of resistance, which may turn out to be a main cause of sickness and mortality in poor nations. The current research looks at the incidence and antibiogram of Lower Respiratory Pathogens isolated from LRTI patients at Birat Medical College in Eastern Nepal. *Streptococcus pyogenes* (65.8%) was the predominant organism, followed by *Staphylococcus aureus* (13.7%), in contrast to the finding of Thapa S et al (2017), that the most prevalent bacterial pathogen of LRTI in patients was Streptococcus pneumoniae (16.6%). (18). In a previous study the most prevalent isolate was *Pseudomonas aeruginosa* (34%) (19). Other studies also reported that the most common organism isolated was Klebsiella pneumoniae (41.95 %), followed by Pseudomonas aeruginosa (26.84 %) (20). In a study from, North Kerala, India S. pneumoniae, H. influenzae, and K. pneumoniae were the most common LRTI pathogens (21). Some studies reported that K. pneumonia (40 %), P. aeruginosa (21.33 %), E. coli (14.66%), Acinetobacter calcoaceticus baumannii complex (17.33%), C. freundii (2.66%), K. oxytoca (1.33 %), C. koseri (1.33 %), and Enterobacter spp.(1.33%) were among the bacteria recovered from the samples (22). The medical wing (general medicine, emergency, surgery, etc.) had the highest isolation rate 399 (76.1%), 65 (12.4%), and 23 (4.4%), respectively, followed by the intensive care units (ICU) with 37 (7.1 %). This could be related to patients who came to the general medicine or respiratory medicine departments with respiratory tract infections. The medical wing (general medicine, pulmonary medicine, dermatology, etc.) had the highest isolation rate (75.50 %) in the previous study, followed by the intensive care units (12.42 %) (20). In this study distribution of lower respiratory tract infections among indoor and outdoor patients, higher in inpatients department 465(88.7%) and lower in outpatients department 59(11.3%). In this study, the distribution of lower respiratory tract infections among indoor and outdoor patients was higher in the inpatients department 465(88.7%) and lower in the outpatients department 59(11.3 %). Previous study was showed distribution of lower respiratory tract infections in indoor and outdoor patients were 267(49.7%) and 266(49.9%) (23). In our study total of 853 specimens from lower LRT were processed according to the standard microbiological methods. Specimen processed in this study were sputum 370(70.6%), Tracheal aspirate 91(17.4%), Bronchial aspirate 53(10.1%), and Pleural fluid 10(1.9 %). The significant growth were 350(94.6%), 89(97.5%), 49(92.45%) and 8(80%). In previous study sputum (n=1081), endotrachial secretion (n=61) and bronchi al washing (n=20), only 497 showed significant growth (44.4%) (23) .The age-specific data presented in this study revealed a higher prevalence in the 60-69 and 70-79 age groups 103. (19.7%) (24, 25). The senior population may be more vulnerable due to age-related physiological and immunological changes, as well as various co-morbidities.

The previous study showed an increased prevalence in the 61-80 years age group (50%) (20). In a prior study, the prevalence of respiratory disease was found to be higher in people aged 61 to 80 (50%). It is documented that males were more isolated than females, with 291 (55.64 %) experiencing isolation 232 (23.83 %). In previous study, males were more likely than females to be isolated (76.17 %), indicating that males are more susceptible to LRTIs than females (23.83%) (20). In our study, higher number of resistance in macrolides and aminoglycisides. In previous study, In contrast to Nidhi Goel et al. (26), amikacin resistance was higher in Acinetobacter baumanii (61.9%), followed by Klebsiella pneumonia (34.3%), Citrobacter koseri (10%), and Escherichia coli (7.2%), and Pseudomonas aeruginosa (7.2%). (13.5 %). The respiratory bacteria are becoming more resistant to antibiotics, empirical treatment with standard medicines has become more difficult, and a clear bacteriological diagnosis and susceptibility testing would be required for effective LRTI care.

In the current investigation, antimicrobial susceptibility tests for 5 bacterial isolates revealed that the most effective antibiotics against Gram-positive bacteria *Streptococcus pyogenes* and *Staphylococcus aureus* were Tetracycline 262 (75.7%) and Azithromycin 53 (73.6%). Penicillin and Ceftazidime were the least effective, with resistance rates of 136 (39.4%) and 30 (41.7%), respectively. In the instance of Pseudomonas spp, Amikacin 22(64.7 percent) was the most effective antibiotic, whereas Tetracycline 1(2.9%) was the least effective. Penicillin sensitivity was highest in Klebsiella pneumonia 37(71.1%), while Amikacin sensitivity was highest in Acinetobacter spp.16 (80%). Cefotaxime resistance was found in 10 (50%) of Acinetobacter spp. Gauchan et al. found that 100% of Gram-negative bacteria were susceptible to chloramphenicol but only 20.6 percent to co-trimoxazole in a similar investigation. Similarly, for Gram-positive bacteria, ciprofloxacin was shown to be the most effective antibiotic (79.2% sensitivity), whereas co-trimoxazole was the least effective. (19). In previous study, gentamicin (100%) was the most effective (100% sensitivity) antibiotic against Gram-positive bacteria and penicillin the least effective one (100% resistance). In the instance of Pseudomonas spp., carbenicillin (100%) was the most effective (100%) antibiotic, whereas piperacillin was the least effective (29%) antibiotic. Amikacin sensitivity was highest in Klebsiella pneumoniae (94%), while chloramphenicol sensitivity was highest in Acinetobacter spp (70%). Ceftazidine resistance was found in (100%) of Acinetobacter spp. (70%) of E. coli were amikacin-sensitive, while all of them were ampicillin-resistant. Enterobacter spp. were found to be completely resistant to all antibiotics tested(27).

V. CONCLUSION

This study shows that LRTI is caused by a variety of bacteria, and antibiotic resistance has become a substantial public health issue. To minimize infection rates and prevent the transmission of resistance, a longitudinal surveillance program, the implementation of infection control methods, and the sensible use of antibiotics are all strongly recommended.

Conflict of interest none declared

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