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Bacterial Etiology and Antibiogram of Lower Respiratory Tract Infections at a Tertiary Care Hospital in Kathmandu, Nepal

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ABSTRACT

Background: Globally common, lower respiratory tract infections (LRTIs) are especially difficult to manage in developing countries, where identifying the specific cause and determining the correct antibiotic treatment are major challenges.

Methods: A cross-sectional study was conducted on patients with lower respiratory tract infections (LRTIs). A total of 1912 sputum samples were investigated for bacterial growth using standard microbiological methods. Isolates were identified through cultural, microscopic, and biochemical analyses, and subsequently subjected to antibiotic susceptibility testing.

Results: Overall bacterial prevalence in sputum was found to be 11.7%. K. pnuemoniae was the predominant isolate recovered from 4.6% specimens. Altogether, isolates from 11 different genera were recovered in the study. Bacterial prevalence was more among male (13.8%) and people from age group 76-90 (16.7%). Similarly, bacterial LRTIs were found more prevalent during spring (14.4%) and summer (12.7%) seasons. All these associations were found statistically significant (p-value<0.01). AST pattern revealed most of the first and second-line drugs were ineffective against isolated organisms. For GNB, amikacin among first and second-line was found to be the most effective drug, whereas for S. aureus, ceftazidime and cefepime (100%) were found to be effective drug.

Conclusion: Detection of bacterial agents from sputum and understanding the resistance pattern can be a basis for successful antimicrobial therapy and mapping a way-forward to mitigate the issue of MDR in Nepal.

Keywords: lower respiratory tract infections (LRTIs); etiological agents; MDR; sputum.

INTRODUCTION

Lower Respiratory tract infections (LRTIs) hold a significant share among some of the prevalent diseases worldwide accounting for 20-24% of death of all mortality caused by acute respiratory tract infections.[1] LRTIs refers to conditions such as pneumonia, bronchiectasis, lung abscess and acute exacerbation of chronic bronchitis (AECB).[2] Because the aetiologic agents of LRTIs differ from one region to another, so too will the susceptibility profile.[3,4] To prescribe the right treatment, it is therefore essential to have up-to-date knowledge of the organisms that cause LRTIs and their antibiotic susceptibility profiles. Streptococcus pneumoniae, Haemophilus influenzae, Klebsiella pneumoniae, Pseudomonas aeruginosa, Escherichia coli, Staphylococcus aureus, Bacillus species, Moraxella catarrhalis, Streptococcus pyogenes, and a few other enteric Gram-negative rods like Salmonella choleraesuis and Citrobacter koseri are the common pathogens of RTIs.[5-9] The current study was designed to explore the bacteriological spectrum of sputum from the patients with LRTIs and determine the antibiotic susceptibility pattern of the isolates.

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METHODS

This cross-sectional observational study was conducted at Shukraraj Tropical and Infectious Disease Hospital, Teku, Kathmandu over the course of 18 months from January 2023 to June 2024. A total of 1912 patients visiting hospital during this period for sputum culture were included in this study. Patients were asked for early morning sputum. First, they were advised to gargle mouth with antiseptic before collecting sputum in a clean sterile and leak proof container. The methodology was followed from a similar study done by Regmi et al.[10] A loopful of sample was streaked over Chocolate agar, Blood agar, MacConkey agar, Xylose Lysine Deoxycholate agar and Saboroud Dextrose agar (Hi-Media, India) under aseptic conditions. Blood agar and Chocolate agar plates were kept at CO_2 incubators, whereas the rest of the media were incubated at the aerobic conditions for 24 h at 37 °C. Identification of bacterial isolates was done based on their morphological and biochemical characteristics.[11] Antibiotic susceptibility test was done by modified Kirby Bauer's disc diffusion method following CLSI guidelines (2020).[12] A total of 16 antibiotics for from five different classes were used procured from Hi-Media, India. Isolates resisting any one antibiotic from three different class were categorized as MDR.[13] Each batch of media and reagents was subjected to sterility and performance testing. During antibiotic susceptibility test, quality control was done using the control strains of E. coli ATCC 25922. Data were first collated in Excel spreadsheet 2021 and SPSS V26.0. Chi-square test was performed to find the associations between categorical variables. $p \leq 0.01$ was assigned as having significant association.

RESULTS

During the study period, 1912 sputum samples were collected from which 224 bacterial isolates were recovered. The male to female ratio among the study population was 1:0.8. Average year of the patients enrolled in this study was found to be 44.5±19.2. Age group 16-30 accounted highest number of specimens. The overall growth prevalence was 11.7% in this study. Altogether, 15 species of bacteria belonigng to 11 different genera were isolated. Most prevalent organism was Klebsiella pneumoniae (88) with overall growth prevalence of 4.6% followed by 42 isolates of Pseudomonas aeruginosa (2.2%). Among gram positive organisms, Staphylococcus aureus accounted for 11 (0.6%) isolates. Similarly, Citrobacter spp., were isolated from 29 (1.52%) specimens and 16 (0.8%) isolates of E. coli were also recorded. Of the 223 isolates, only 20.6% isolates were MDR. Among MDR, Klebsiella pneumoniae accounted for 20 (43.5%) followed by E. coli (Figure 1).

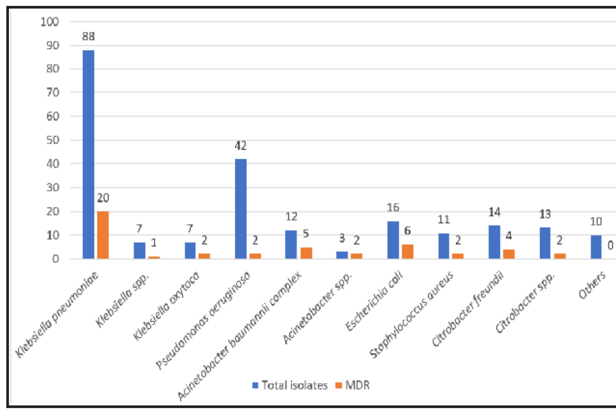

Figure 1. Distribution of total and MDR isolates.

In this study, 1089 participants were male, whereas remaining 823 were female. Of the 223 growth positive samples, highest growth rate was observed in male with 150 (13.8%) returning as culture positive and only 73 (8.8%) sputum were positive among female. The result also turned out to be statistically significant ($p<0.01$). Similarly, highest growth rate was seen in patients from age group 76-90 with 19 (16.7%) samples turning positive, followed by age group 61-75 with 52 (16.5%) growth. None of the specimens from patients aged more than 91 years was found positive. Age group 0-15 also showed least growth rate 2 (6.5%) as compared to other age group. The result was also found statistically significant ($p<0.01$). Similarly, prevalence of bacterial infection was higher in spring season 121 (14.4%) followed by summer/monsoon 42 (12.7%), compared to autumn 16 (10.3%) and winter 44 (7.6%). This finding was also found statistically significant at 1% level of significant (p-value<0.01) (Table 1).

Table 1. Incidence of bacteria and associated factors.

Variables	Frequency (n)	Growth n(%)	p-value
Gender			
Male	1089	150 (13.8%)	0.001*
Female	823	73 (8.9%)	
Age group (years)			
0-15	31	2 (6.5%)	0.000*
16-30	571	43 (7.5%)	
31-45	450	43 (9.6%)	
46-60	422	64 (15.2%)	
61-75	316	52 (16.5%)	
76-90	114	19 (16.7%)	
>91	8	0	
Season			
Spring	843	121 (14.4%)	0.001*
Summer/ monsoon	332	42 (12.7%)	
Autumn	156	16 (10.3%)	
Winter	581	44 (7.6%)	

AST showed that cefixime (62.5%) and ceftriaxone (69.7%) were least effective against Acinetobacter isolates. Similarly, these isolates showed higher resistance towards meropenem (75%) and imipenem (62.5%). Some of the most effective drugs were polymyxin-B (0%), colistin (0%) and amikacin (6.3%). Meanwhile, Klebsiella isolates were found to have higher sensitivity towards meropenem (100%) and ceftriaxone (84%). Ampicillin was, on the other hand, found most resistant drugs with 87%. Against Pseudomonas isolates, piperacillin/ tazobactam (94.4%) and gentamicin (92.9%) were found most effective. All isolates were found to be resistant towards ampicillin. For S. aureus, cefepime and ceftazidime were found 100% effective, whereas cefixime was completely resisted. Overall antibiotic susceptibility pattern of isolates is shown in Figure 2.

PS: AMX: Amoxicillin, COT: Co-Trimoxazole, CIP: Ciprofloxacin, CFM: Cefixime, CTR: Ceftriaxone, IPM: Imipenem, MRP: Meropenem, PB: Polymyxin-B, PTZ: Piperacillin/ Tazobactam, AK: Amikacin, GEN: Gentamicin, AMP: Ampicillin, CL: Colistin, LZ: Linezolid, AMC: Amoxicillin/ clavulanate, CPM: Cefepime CAZ: Ceftazidime, LEVO: Levofloxacin.

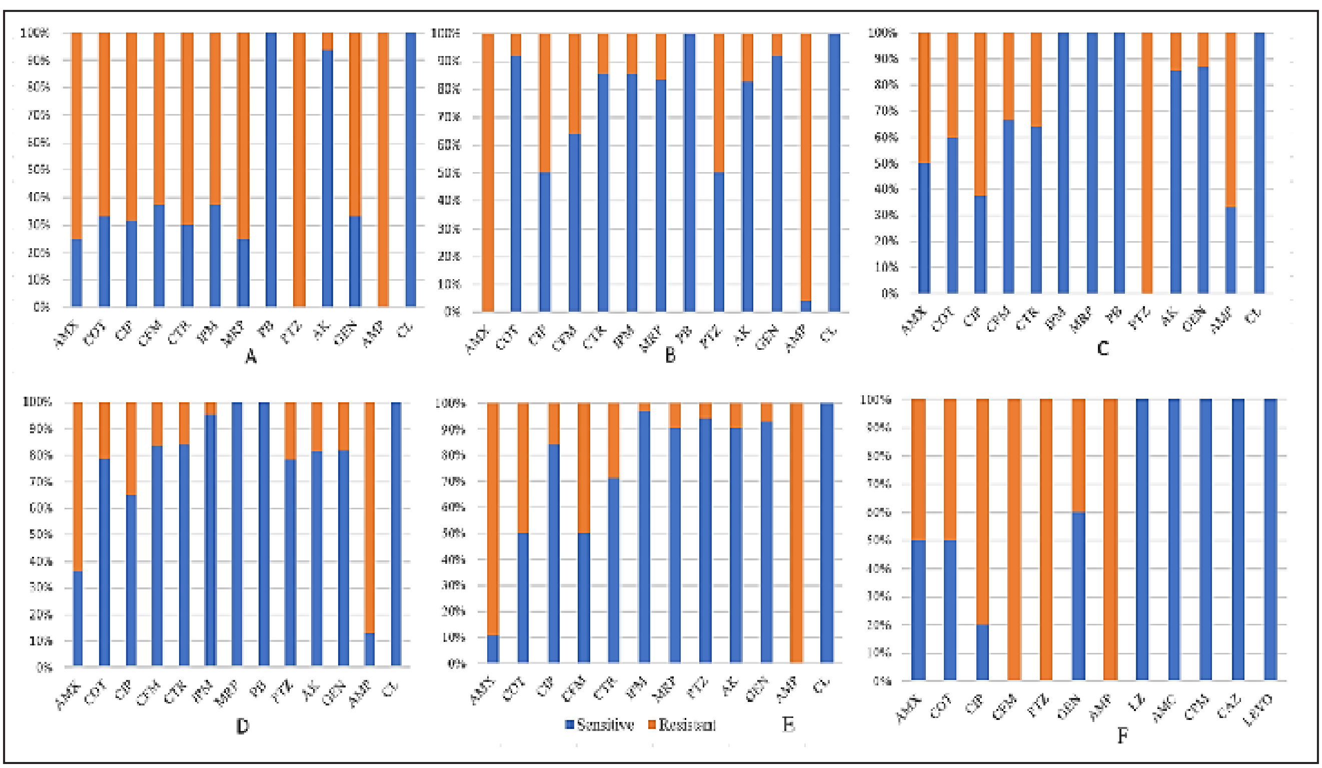

Figure 2. Antibiotic susceptibility pattern of A: Acinetobacter Spp., B: Citrobacter Spp., C: E. coli, D: Klebsiella Spp., E: P. aeruginosa, F: S. aureus.

DISCUSSION

The current study was carried out to investigate the etiological factors of LRTIs and observe antibiotic resistance pattern of recovered isolates. Bacterial infections are among the most common infections in low-and middle-income nations like Nepal. These infections are made more complex by the bacteria's acquisition of different resistant genes. This study observed overall bacterial prevalence of 11.7% among the people with LRTIs. Similar result was also reported in a study conducted in Kathmandu, where the bacterial prevalence was 10.9%, whereas another study done in same city reported a slightly lower prevalence rate (5.1%).[14,7] In contrast, various studies conducted in different parts of Nepal have reported much higher bacterial prevalence. Previous studies done in Bharatpur reported prevalence as high as 47.3% and 28.4%.[10,15] Meanwhile, similar study done in Pokhara reported a bacterial prevalence of 29.1%.[16] Similar varying prevalences have been reported in other studies conducted worldwide as well.[4-6,17] This variation in prevalence from city to city and hospital to hospital might be due to variation in sample size and different methods employed for the collection and processing of the samples in different places.

This study observed Klebsiella pneumoniae (39.5%) to be the most predominant isolates followed by Pseudomonas aeruginosa (18.8%). The result is in tune with past studies carried out in Shukraraj Tropical and Infectious Disease Hospital, Manmohan Memorial Medical Hospital and BP Koirala Memorial Cancer Hospital as they found Pseudomonas aeruginosa as chief isolate followed by Klebsiella spp.[14,15,18] In our study, E. coli and Acinetobacter spp accounted for 7.2% and 6.7% growth. Contrary to this, one study conducted by Bhatta et al in Pokhara has reported Acinetobacter spp. As the predominant organism in LRTI.[16] Similarly, S. aureus was only gram-positive cocci that has been isolated in this study. Regmi et al. and Adhikari et al. have also reported significant amount of S. aureus in sputum from Bhartpur city.[10,18] In earlier researches by Gauchan et al., Sharma et al., and Mishra et al., Haemophilus influenzae has been reported as the most prevalent isolate.[19-21] Despite being a leading cause of LRTIs, Haemophilus influenzae was not isolated in the present study.

The male to female ratio in this study was found to be 1:0.7. Similarly, higher prevalence of bacterial infection was recorded from male (13.8%) as compared to females (8.8%) in this study. This finding is in tune with some of the past studies conducted by Adhikari et al (98.9%), Parajuli et al (42.6%), Gaire et al. (15.4%) and Lamichhane et al. (51.6%).[18,22,14,15] This also resonates with several studies conducted worldwide.[17,21] Contrary to this, a study done in Nigeria has reported higher prevalence rate among females (52.8%) as compared to male.[4] In LMICs like Nepal, where men predominate, men are more likely than women to be exposed to community and hospital settings and are therefore more likely to become infected. Also, male are more likely to be involved in smoking and alcohol consumption, which are believed to be risk factors for LRTIs.[23] This study reported higher bacterial prevalence in age group 76-90 (16.7%) and 61-75 (16.5%) as compared to other age groups. The result was also found to be statistically significant ($p<0.01$). This finding is in alignment with one of the past studies conducted in Bharatpur hospital where the growth rate was 22.6% in age group 51-60 and 19.4% in 61-70.[15] Another study conducted in Shukraraj Tropical and Infectious Disease Hospital reported prevalence of LRTIs to be 25.4% in people aged more than 60 years.[14] Meanwhile, another study done in Bharatpur showed contrasting result as it reported a higher bacterial incidence in age group 31-45 (56%).[10] Patients with 60 years of age and older had a significantly higher prevalence of LRTIs. The high prevalence found in this study may be explained by the fact that people in this age group may have weakened immunity as a result of aging or other conditions that affect the immune system and their exposure in community keep getting increased.[24] Prevalence of LRTIs was likely to be higher in spring (14.4%) and monsoon (12.2%). Both of these seasons are referred to as warm seasons in Nepal. A study conducted in Hanzhong, China, revealed bacterial infection being more prevalent in spring and summer.[25] Another study done in western part of Nepal has also reported a significant increase of bacterial infection of lower respiratory tract during warm season.[26]

Some of the effective antibiotics in this study were aminoglycosides and carbapenems. Cefixime and ceftriaxone were moderately effective drugs for isolates belonging to Enterobacteriaceae group except Acinetobacter spp. This finding is contrary to the study conducted in Kathmandu, where most of the GNB isolates were resistant to these antibiotics.[14] Similar study conducted in Bharatpur showed higher resistance towards carbapenem drugs, which is also contrary to our findings. While the resistance towards amikacin in each study was low, higher resistivity was observed towards gentamicin.[14] Except amikacin, none of the first and second-line drugs were effective against Acinetobacter spp., although all isolates were sensitive towards polymyxin-B and colistin. This finding is in tune with a study done by Gaire et al.[14] but in contradiction with the one conducted by Lamichhane et al.[14,15] Only 20.6% isolates recovered in this study were found to be MDR. This finding is similar to that of a researc carried out in Kathmandu (20.7%), whereas the rate is lower when compared to other studies done in different parts of Nepal where MDR rate varies from 46.9% to 55.6% to 79.6%.[14,15,18] MDR is a wicked problem that has prompted the relevant authorities to take drastic action to stop its spread. The number of MDR isolates in hospital settings is rapidly rising, and the burden varies by region. Lower middle-income countries (LMIC) are facing challenges because of a variety of factors ingrained in the practices, policies, and features of the health system.[27]

CONCLUSIONS

This study found an overall bacterial prevalence of 11.7% in sputum among the patients with LRTIs. Discovery of multidrug resistant bacteria including resistance to some of the last-line drug is worrisome and concerned authorities should be more alert to abate their incidence and dissemination.

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