EPIDEMIOLOGY, BACTERIAL PROFILE, AND ANTIBIOTIC SENSITIVITY OF LOWER RESPIRATORY TRACT INFECTIONS IN SANA'A AND THAMAR CITY, YEMEN

ABSTRACT

Background and aims:Lower respiratory infections (LRTIs) are the leading cause of death infectious diseases in the world and the fifth leading cause of death in general. The study aimed to identify the general characteristics of LRTI, the causative bacteria and the results of sensitivity to antibiotics. **Subjects and methods**: The study included 555 clinical diagnostic cases as LRTI cases, 328 male and 227 female, aged 3 to 69 years. Clinical and demographic data were collected in the standard questionnaire, and samples included sputum or bronchial lavage (BAL) staining and culture. Samples were cultured in 3 different bacterial media, blood agar and LJ slope, chocolate agar with Co^2; cultures were then examined for possible bacterial pathogens of LRTI. Possible bacterial pathogens were isolated and identified using standard laboratory techniques, and microbial sensitivity testing was carried out by disc diffusion method. **Results**: LRTI was recorded among all age groups and with less frequency in children under 16 years of age. A large number of LRTI (36.2%) was not diagnosed, most in CAP (52.4%), followed by HAP (33.9%) while unidentified cases were lower in AECOPD (22.8%). CAP isolates are *K.pneumoniae* (26.2%), *S.pyogens* (12.3%), and *S.pneumoniae* (9%); in HAP are MSSA (24%), *E.Coli* (12.9%), MRAS (11.1%), *k.pneumoniae* (10.5%) and *P.aeruginosa* (7%); and in AECOPD are *M.catarrhalis* (47.2%), *K.pneumoniae* (17.2%), *H.influnzae* (10.7%) and *P.aeruginosa* (2%). In Gram-positive bacteria, high resistance to ampicillin/sulbactam (100%) and amoxicillin/clavulanate (100%) was recorded, while moderate resistance to amikacin, vancomycin, cefepime and moxifloxacin was recorded. In Gram-negative bacteria, a high resistance to 3^{rd} g Cephalosporin's (68.5%) was recorded, while a moderate sensitivity to the other antibiotics tested was recorded. **Conclusions**: There is a high rate of undiagnosed LRTI in Yemen and this highlights the need for health authorities to develop strategies to diagnose most of the causes of LRTI, including *Mycoplasma*, *Chlamydia*, and viral causes. No antibiotics are completely effective in treating LRTI in our area and antibiotic sensitivity should be performed in all cases.

Keywords: Lower respiratory tract infections (LRTIs), antibiotics, Sana'a City, Thamar city, Yemen**.**

INTRODUCTION

Lower respiratory tract infection (LRTIs) is the leading cause of infectious diseases of death worldwide, the fifth overall cause of death, and the second general cause of disability adjusted life years (DALYs), although they are largely preventable causes of diseases and Death[1]. There have been changes in the epidemiology of LRTIs in the past ten years as there has been a decrease in the number of cases among children under 5 and an increase in infection among older adults as well as an increase in viral infections[1]. Nevertheless, there is no uniform definition of "LRTIs", a fact which has been said to impede the appreciation of its true epidemiological importance[2,3]. From an epidemiological standpoint, most definitions of LRTI include pneumonia, influenza, bronchitis (including acute exacerbations of chronic obstructive pulmonary disease [COPD] [AECOPD]) and bronchiolitis as important diseases [1-3].

The three major bacterial respiratory pathogens are *Streptococcus pneumoniae*, *Moraxella catarrhalis* and *Haemophilus influenza*e. Unfortunately, these causes are spreading and increasing the rate of their resistance to antibiotics worldwide[4,5,6]. The importance of monitoring the progress of this resistance has led to many international, regional and national monitoring programs. However, the results of surveillance studies show wide differences in sensitivity rates, both geographically and over time[7,8]. Bacterial resistance patterns for antibiotics may differ from one region to another depending on the pressure on the antibiotics in that region[9]. Consequently, there is a great need for local resistance spread data in order to guide the experimental prescription and identify areas where new antibiotics with greater effect are needed. In Yemen, data on epidemiology of LRTIs and antibiotic patterns are still rare for bacterial causes. Over the past four years, an increase in mortality has been observed among residents of the capital, Sana'a, due to LRTIs[10]. Therefore, the present study was designed to identify the bacterial profile of lower respiratory tract infections (LRTIs) in Yemen and to determine the antibiotic susceptibility among these pathogens in our areas.

SUBJECTS AND METHODS

This study was conducted on 555 hospitalized patients with LRTI in university hospitals in the cities of Sana'a and Thamar during the period from October 2015 to October 2018. All patients were subjected to full clinical, radiological and relevant laboratory examinations.Clinical sample analyzes were performed in the laboratories of the National Center of Public Health laboratories Sana'a (NCPHL)). The study included 187 patients with community-acquired pneumonia (CAP), 171 patient with hospital-acquired pneumonia (HAP) and 197 patients with acute exacerbation of chronic obstructive pulmonary disease (AECOPD)[5].

CAP was defined as acquired pneumonia outside the hospital[11]. HAP was defined as a pneumonia occurring 48 hours or more after admission, which was not developed at the time of admission[12]. AECOPD were defined according to the GOLD guidelines[5]. Patient data were collected using questionnaire including personal data, clinical symptoms, signs, and history of preexisting chronic diseases. Samples included sputum or bronchoalveolar lavage (BAL) for staining and culture. Samples were cultured on 3 bacteriological media. Blood agar aerobically, chocolate agar with Co_2 and LJ slope then cultures were examined for possible bacterial pathogens of LRTI. Possible bacterial pathogens were isolated and identified using standard laboratory techniques, and microbial sensitivity testing was carried out by means of disc diffusion for selected antibiotics.

RESULTS

A total of 555 LRTIs hospitalized patients (328/59.1% male and 227/40.9% female) were enrolled in this study. The most frequent age groups were 30-42 years (26.5%), and age group 43-56 years (22.5%); while children age group was less frequent (8.5%). Bacterial growth yielded on 354 (63.8%) while 201 (36.2%) were negative for bacterial culture (Table 1). A large number of LRTI (36.2%) was not diagnosed, mostly in CAP (52.4%), followed by HAP (33.9%) while lower cases were in AECOPD (22.8%). The isolates in 187 patients with CAP were *K.pneumoniae* (26.2%), *S.pyogens* (12.3%), and *S.pneumoniae* (9%). Isolates in 171 patients with HAP were MSSA (24%), *E.Coli* (12.9%), MRAS (11.1%), *K.pneumoniae* (10.5%) and *P.aeruginosa* (7%). The organisms in 197 patients with AECOPD were *Moraxella catarrhalis* (47.2%), *K.pneumoniae* (17.2%), *H.influnzae* (10.7%) and *P.aeruginosa* (2%) (Table 2). Table 3 shows the frequency of bacterial causative agents of LRTI; the Subtotal Gram positive bacteria were counted for 28.3% from total bacteria isolates, while subtotal Gram positive bacteria was counted for 71.7% from the total bacterial isolates. The most 3 predominant bacteria isolated from LRTIs patients in the study were *K.pneumoniae* 101(18%), *Moraxella catarrhalis* 91(16.8%) and *S.aureus* 60 (10.8%), while others bacteria such as *S.pyogens*, *S.pneumoniae*, *H.influnzae*, *P.aeruginosa*, *E.coli* and *Proteus vulgaris* were less frequent (Table 3). In Gram-positive bacteria high resistance was recorded for ampicillin/sulbactam (100%) and amoxicillin/clavulanate (100%), while a moderate sensitivity rate for amikacin, vancomycin, cefepime and moxifloxacin was recorded. In Gram-negative bacteria, a high resistance to 3^{rd} g of cephalosporins (68.5%) was recorded, while moderate sensitivity to other tested antibiotics was recorded (Table 4). The rates of cure, ICU admission, isolation and death among LRTI cases of positive bacterial growth were almost similar to those of negative culture with slight differences. The mortality rate among total LRTIs was 25%, while for confirmed LRTI cases in bacterial culture it was 22.9%, which is lower among the LRTI cases of negative culture (28.9%) (Table 5).

DISCUSSION

Lower respiratory tract infection (LRTIs) is the leading cause of infectious diseases of death worldwide, the fifth general cause of death, and the second general cause of disability adjusted life years (DALYs), although they are largely preventable causes of diseases and Death[1]. In the current study the mortality rate among total LRTIs was 25%, while for LRTIs cases confirmed for bacterial culture was 22.9%, lower than that among negative culture LRTI cases (28.9%) (Table 5); this rate is higher than that reported by Brown and others in the United States of America where the death rate among community-acquired pneumonia hospitalizations patients was 7.4%[13]. While Global Strategy for the Diagnosis, Management and Prevention of COPD, reported that long-term prognosis following LRTIs was poor, with a 5-year mortality of approximately 50% [14]. The high mortality rate in the current study may be high rates for related factors and include comorbidities especially cardiovascular disease, severity of exacerbations, age, previous hospitalization, low BMI and malnutrition[1].

When reviewing the various studies, it is clear that there are some regional differences in the reported etiology of LRTIs, as described by Waterer [15]. This may be related to a number of factors, but it is also important to realize that although LRTIs are not a seasonal disease, many different organisms, including *S.pneumoniae*, *influenza* virus, *Legionella* species infections, and even polymicrobial infections do have seasonal variations[16]. In the current study the most 3 predominant bacteria isolated from LRTIs patients were *K. pneumoniae* 101(18%), *Moraxella catarrhalis* 91(16.8%) and *S. aureus* 60(10.8%), while *S.pneumoniae* and *H. influnzae* were less frequently (Table 3); this result is different from that traditionally, the *pneumococcus* has been reported to be the most common cause of LRTIs[17-19] and the Global Burden of Disease Study analysis of LRTIs (2015)[1] indicated that the *pneumococcus* was the most

common cause of LRTIs among all ages. However, our results is go with repots in which there have been changes noted in the reported etiology of LRTIs, particularly with the use of more sensitive diagnostic tools[19-21]. In general, it is increasingly recognized that viruses appear to play a bigger role in the etiology of LRTIs than has previously been documented[22-25] and cases of infection with more than one pathogen, commonly the association of one or more viruses with one or more bacterial agents are not uncommon [21,22].
For patients with CAP, our results (Table 2) showed bacterial profiles similar to those reported by international studies[6] and regional[26]. This pattern of "local" hegemony should be taken into account when prescribing antimicrobials in our region. When antibiotic sensitivity was considering for bacterial isolates from LRTI patients, in Gram positive bacteria a high resistance was recorded for ampicillin/sulbactam (100%) and amoxicillin/clavulanate (100%), while moderate of sensitivity was recorded for amikacin, vancomycin, Cefepime and moxifloxacin. In Gram negative bacteria a high resistance was recorded for 3[rd] g Cephalosporin's (68.5%), while moderate of sensitivity was recorded for other tested antibiotics (Table 4). Our data revealed high resistance rates for cephalosporins, and the β-lactam-βlactamase inhibitors. These findings are in agreement with the increasing prevalence of resistance of Gram positive bacteria as *S.pneumoniae* to those antimicrobial groups, by regional,[7,27-29] and worldwide [6,7] studies. Moreover, our results highlight the increasing problem of MDR in Gram positive and Gram negative bacteria of LRTIs, a problem that was extensively addressed in the literature[28-30]. This warns us of the need for wise use of different groups of antimicrobials, especially in our resource-poor country. Moreover, this requires greater focus on identifying drivers of resistance relevant and on implementing effective strategies to combat resistance and MDR problems.

For patients with HAP, the problem of antibiotic resistance seems more important; therefore, the situation is more complicated than that in CAP. Nosocomial pneumonias leads to high morbidity and mortality, especially among ICU patients[8,11]. In most clinical cases, there is a need to start empirical antimicrobial therapy before obtaining microbial results. However, the situation is further complicated by the emergence of several beta-lactamase and MDR pathogens[29,31]. Obviously there is a great need to obtain data on the prevalent strains in HAP; along with the sensitivity pattern to help revise antibiotic policy and guide physicians to better manage patients with HAP; especially in developing countries such as Yemen.

The current study revealed the present of MRSA, Gram-negative organisms, and *P.aeruginosa* among patients with HAP. This differs clearly from the results obtained by Goel and co-workers[31] and even those of Ahmed, *et al.*[32], Agmy, *et al.* [33]. Although the later study addressed the problem of HAP in 75 cases of ICU patients, the predominant pathogens were *S.aureus* (32%), *P.aeruginosa* (30%), and *S.pneumoniae* (15%). It is clear that this "regional" difference explains the changing pattern of pathogens that cause over time, even in the same hospital. This underscores the importance of implementing continued local monitoring programs[8]. Also, our data show an alarming high prevalence of MRSA. This coincides with the recent report by Alyahawi, and Al-Safani *et al.*[34,35] who observed that the prevalence of MRSA in invasive isolates from hospitals in Yemen was 23%[34].

CONCLUSION

Lower respiratory infections are still very common and continue to be a major cause of morbidity and mortality in Yemen in children and adults alike, and there are significant changes in the epidemiology of LRTIs in terms of their frequency and infectious pathogens. There is a high rate of undiagnosed LRTI in Yemen and this highlights the need for health authorities to develop strategies to diagnose most of the causes of LRTI, including *Mycoplasma*, *Chlamydia*, and viral causes. The most common bacteria in CAP in Yemen is *K.pneumoniae* while HAP is the *S.aureus* and Gram negative bacteria. For acute exacerbation of COPD, *M.catarrahalis* was the most common. No antibiotics are completely effective in treating LRTI in our area and antibiotic sensitivity should be performed in all cases.

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CONFLICT OF INTEREST

"No conflict of interest associated with this work".

AUTHOR'S CONTRIBUTION

This research work is part of project of the National Center for Public Health Laboratories (NCPHL). The authors performed clinical and laboratory works. The corresponding author (HAA) supervised the laboratory works, and revised and edited the research.

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Table 1: Distribution of age groups, gender and years among LRTI patients and its correlation with bacterial growth outcome.

	NO	% (Total)	Bacterial growth outcome					X^2	*P*
			Growth			**No growth**			
			No	%		**No**	%		
Age category								4.913	0.0296
(3-16)	**47**	8.5%	**26**	55.3%		**21**	44.7%		
(17-29)	**119**	21.4%	**76**	63.8%		**43**	36.2%		
(30-42)	**147**	26.5%	**92**	62.6%		**55**	37.4%		
(43-56)	**125**	22.5%	**85**	68%		**40**	32%		
≥ 57	**117**	21.1%	**75**	64.1%		**42**	35.9%		
Gender								4.940	0.029
Male	**328**	59.1%	**218**	66.5%		**110**	33.5%		
Female	**227**	40.9%	**136**	60%		**86**	40%		
Data								19.124	<0.0001
2015	142	25.6%	90	63.4%		52	35.6%		
2016	178	32.1%	124	69.6%		54	30.4%		
2017	159	28.6%	100	62.9%		59	37.1%		
2018	76	13.7%	40	52.6%		36	47.4%		
Total	**555**	**100.0%**	**354**	**63.8%**		**201**	**36.2%**		

Table 2. Bacterial profile of lower respiratory tract infections in Yemen.

Common Bacterial pathogens (No/%)		
CAP (n=187/33.7%)	**HAP (n=171/30.8%)**	**AECOPD (n=197/35.5)**
S. pneumoniae (17 /9%)	MRSA (19/11.1 %)	*H. influnzae* (21/10.7%)
K. pneumoniae (49 /26.2%)	*k. pneumoniae* (18/10.5%)	*K. pneumoniae* (34/17.2%)
St. pyogens (23/12.3%)	*E. Coli* (22/12.9%)	*M. catarrhalis* (93/47.2%)
	P. aeruginosa (12/7%)	*P. aeruginosa* (4/2%)
	MSSA (41/24%)	
	Proteus vulgaris (1/0.6%)	
No Bacterial growth (98/52.4%)	No Bacterial growth (58/33.9%)	No Bacterial growth (45/22.8%)

CAP: Community-acquired pneumonia; HAP: Hospital-acquired pneumonia; AECOPD; Acute exacerbations of chronic obstructive pulmonary disease; MRSA: Methecillin-resistant *Staphylococcus aureus*; MSSA: Methecillin-sensitive *Staphylococcus aureus*.

Table 3: The frequency of bacterial causative agents of LRTI

Isolated Bacteria	**No (%)**	**% Total n=555**
Gram Positive		
S. *pneumoniae*	17 (17)	3
S. aureus	60 (60)	10.8
S. *pyogenes*	23 (23)	4.1
Subtotal Gram positive	**100 (28.3)**	**18**
Gram Negative		
K. pneumoniae	101 (39.8)	18
H. influenzae	21 (8.3)	3.8
P. aeruginosa	16 (6.3)	2.9
Proteus vulgaris	1 (0.4)	0.18
Moraxella catarrhalis	93 (36.6)	16.8
E. coli	22 (8.7)	4
Subtotal Gram negative	**254 (71.7)**	**45.8**
Total positive culture	354	63.8
Total negative culture	201	36.2
Fungi		
C. albicans colonization	**159**	28.6

Table 4: . Antibiotic sensitivity and resistance rates (percentages) of gram positive and gram negative bacteria in 354 patients with LRTI in Yemen

Antibiotics	Test	Bacteria	
		Gram positive percentage	Gram negative percentage
Vancomycin	S	69.2	ND
	I	11	
	R	19.8	
Moxifloxacin	S	47.5	71
	I	14	7
	R	38.5	22
3[rd] g Cephalosporin’s	S	8	22.5
	I	13	9
	R	79	68.5
Ciprofloxacin	S	37	82
	I	12	3.2
	R	51	14.8
Cefepime	S	46	65.4
	I	19	13.1
	R	35	21.5
* Aampicillin/sulbactam	S	0	48.2
	I	0	11.2
	R	100	40.6
*Amoxicillin/clavulanate	S	0	67
	I	0	11
	R	100	22
Amikacin	S	58.2	80
	I	19	9
	R	22.8	11

*Not done for *P.aeruginosa,* ND= not done

Table 5: The output of LRTI cases with bacterial infections in comparison with LRTI cases caused by other agents

Outcome	**LRTI with bacterial infections (n=354)**		**LRTI cases with non-bacterial agents (n=201)**		**Total (n=555)**	
	No	**%**	**No**	**%**	**No**	**%**
Cure	273	77.1	143	71.1	416	75
ICU	22	6.2	31	15.4	53	9.5
Isolation	1	0.3	3	1.5	4	0.72
Death	81	22.9	58	28.9	139	25
Total n=555	354	63.8	201	36.2	555	100