



Drug Resistant Non-acid Fast Bacteria Pathogens, Isolated from Tuberculosis (TB) Patients with Known HIV Status from the North West Region of Cameroon

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Authors' contributions

This work was carried out in collaboration between all authors. Author IAA did the study design, wrote the protocol, ensured the follow up of standard laboratory procedure, interpreted results and wrote the manuscript. Author DNA proof read the protocol, followed up with the bacteria cultures, interpreted results and made an input in writing the manuscript. Author YFA collected samples and administered questionnaires and did part of the bench work. Author HDM was the main laboratory scientist overseeing the standard procedures in the lab and recorded results for statistical analysis. Author FRFN contributed in statistical analysis and in reading the manuscript. Author VPKT approved the final manuscript.

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ABSTRACT

Aims: To investigate the prevalence of Non-*Mycobacterium tuberculosis* (MTB) bacterial pathogens from TB patients with known HIV status as well as their resistant patterns to commonly used antibiotics.

Study Design: This was a cross sectional laboratory based study.

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Place and Duration of Study: Microbiology/Bacteriology Unit, Bamenda Regional Hospital, from July 2014 to March 2015.

Methodology: We collected sputum from 111 newly diagnosed TB patients who were referred to do a sputum test at the Bamenda General Hospital TB unit. Sixty one (61) were men, 50 women, 70 HIV positive, 41 HIV negative with age ranging from 20-80 years. Data on Socio-demographic factors as well as clinical history were collected using structured questionnaires. One early morning sputum sample was examined microscopically and cultured on blood and chocolate agars. Antimicrobial sensitivity test was then performed for all the isolates using the Kirby Bauer disc diffusion method.

Results: Non-MTB bacterial pathogens were recorded in 43 (38.7%) of the 111 participants in this study. Thirty (42.85%) non-acid fast bacteria was found amongst the 70 HIV positive cases, higher than in the HIV negative group 13/41 (31.70%). These pathogens were also higher in females 24 (55.8%) than in males 19 (44.2%). Bacteria isolated included 28 *S. aureus*, 12 *S. pneumoniae* and 6 *P. aeruginosa*. Although, the prevalence was higher in the HIV positive group and in females, the differences in both cases were not statistically significant ($P = .523, .324$ respectively). Upon antimicrobial sensitivity testing all the isolates showed high susceptibility to Gentamicin (73.9%), Ciprofloxacin (71.1%) and Chloramphenicol (71.7%) but were all resistant to Penicillin (100.0%), Oxacillin (87.3%) and Amoxicillin (96.1%).

Conclusion: HIV patients were more at risk of developing other Lower Respiratory Tract Infection (LRTI) with non MTB bacteria implicated. Therefore in the treatment of tuberculosis, considerations should be made about Non MTB bacterial pathogens and their treatment as well.

Keywords: Lower respiratory tract infection; non MTB bacterial infection; non-acid fast bacteria, MTB; non MTB bacteria; antimicrobial sensitivity; drug resistance pattern; tuberculosis patients.

1. INTRODUCTION

Lower respiratory tract infections (LRTIs) have been a major cause of morbidity and mortality among humans since the dawn of history and are among the most common infectious diseases affecting humans worldwide [1]. They account for over 50 million deaths of each year and occur in both community and health care settings [2].

Tuberculosis is one of the most debilitating bacterial pulmonary infections and it presents with similar signs and symptoms as LRTIs of other aetiologies.

The respiratory system is one of the most common sites for problems in patients with human immunodeficiency virus (HIV) and a majority of lower respiratory diseases are bacterial [3]. Patients with HIV are well recognized to develop pulmonary diseases with a wide range of opportunistic and non-opportunistic processes [4]. Multiple disease processes may affect the respiratory system in HIV infection and multiple pathological processes may occur simultaneously, especially in advanced immunodeficiency. HIV-induced immunosuppression amplifies the risk of bacterial infections, tuberculosis and non-tuberculosis, often involving antibiotic - resistant strains, with severe and / or recurrent potential. About 80% of

these patients are seen to die as a result of such an infection rather than from HIV [5].

The simultaneous appearance of tuberculosis and bacterial infection is not common and is largely unknown. It has been described mainly in patients with acquired immunodeficiency syndrome, presenting with co-infection with tuberculosis and pneumococcal pneumonia [6]. When there is co-infection of pneumonia and tuberculosis in patients with *S. pneumoniae* pneumonia as the underlying disease, undiagnosed could pose health management problems if tuberculosis alone is diagnosed. Bacterial co-infection is associated with deterioration of pulmonary TB patients as shown in an HIV-endemic country in Africa [7]. Preliminary survey has revealed increasing evidences of recurrent lower respiratory tract (LRT) infection among patients attending tuberculosis clinics in Ekpoma suggesting other microbial etiology in addition to *Mycobacterium tuberculosis* [8]. Superadded bacterial infection is an important cause of deterioration during intensive phase among laboratory confirmed TB patients [9].

Different studies have shown different bacterial pathogens to be implicated to diverse extents but generally the major pathogens encountered are *Streptococcus pneumoniae*,

Haemophilus influenzae, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Staphylococcus aureus*, *Moraxella catarrhalis*, *Streptococcus pyogenes* and some other enteric gram negative rods. These pathogens however differ in prevalence between geographical regions.

Antibiotic resistance is currently the greatest challenge to the effective treatment of infections globally. The consequences of increased drug resistance are far-reaching since bacterial infection of the lower respiratory tract is a major cause of death from infectious disease [10]. Until recently, the global problem of resistance was related mainly to *Staphylococcus* spp., enterobacteria and *Pseudomonas aeruginosa*. Now, a major challenge for the future is the worrying tendency towards increased bacterial resistance among *Streptococcus pneumoniae* and *Haemophilus influenzae*, the key pathogens in community-acquired bacterial infection of the upper and lower respiratory tract [11]. In developing countries the emergence of antibiotic resistance in the management of LRTIs is a serious public health issue. Good quality data are available from more affluent and transitory countries [e.g. in Latin America [12] and Asia [13], but there is little information available from many countries in sub-Saharan Africa, especially from the Central African region [14]. In Cameroon, there are reports on increasing drug resistance to respiratory tract pathogens in Buea [15]. Data on bacterial coinfection in Pulmonary tuberculosis (PTB) patients and drug resistance will provide important information in developing effective strategies for the control, prevention and treatment options in TB patients.

2. MATERIALS AND METHODS

2.1 Study Area and Sampling

This study was carried out in Bamenda, the capital of the North West Region of Cameroon. Bamenda had an estimated population of about 800,000 people as of 2012, and is located 366 km North West of the Cameroonian capital Yaoundé. The TB Centre in the Bamenda Regional Hospital receives suspected cases from all over the region. Patients who tested positive for TB and gave their written or verbal consent were included in the study. Information of their HIV status was gotten from their records. This study population was made up of forty one HIV negatives and seventy HIV positive Patients.

2.2 Study Design

This was a cross sectional laboratory based study involving 111 TB positive patients at the Bamenda Regional Hospital.

Ethical clearance was sought from the Faculty of Health Science Institutional Review Board (FHS-IRB) of the University of Buea. Administrative clearance for specimen collection was obtained from the North West regional delegation for public health and from the director of the Bamenda Regional Hospital.

2.3 Sample Collection and Processing

An early morning expectorated sputum sample was collected in sterile containers from all patients included in the study.

The quality of sputum samples were assessed both microscopically and macroscopically. Watery and non-purulent sputa were considered unsuitable for further processing. All sputa were examined microscopically under the low power objective and the number of epithelial cells and/or polymorphonuclear leukocytes were counted to establish the level of contamination. Sputa that had a majority of epithelial cells with few to no leukocytes were rejected as poorly collected sample. All unsuitable specimens were discarded and new specimens collected.

2.4 Laboratory Procedure

Standard Gram staining was done to investigate bacterial pathogens

2.5 Culture of Sputum

Sputum specimens were inoculated into blood, and chocolate agars. The inoculated plates were incubated at 37°C for 24-48 hours aerobically, except for chocolate agar, in which the plates were incubated for 24-48 hours at 37°C in an atmosphere of 5-10% CO₂ [16]. Cultures with significant growth were presumptively identified based on their cultural and morphological characteristics on selective and differential media [16]. Standard microbiological techniques and biochemical tests were used to further used to confirm bacterial species.

2.6 Antimicrobial Susceptibility Testing

The antimicrobial susceptibility test was performed on all bacterial isolates using the Kirby

Bauer disk diffusion method (Bauer et al., 1966). Antibiotic-impregnated discs used were Cefotaxime (30 µg, Liofilchem s.r.l), Amoxicillin (30 µg, Liofilchem s.r.l), Gentamicin (120 µg, Bioanalyse®), Ciprofloxacin (5 µg, Bioanalyse®), Oxacillin (1 µg, Liofilchem s.r.l) Trimethoprim/Sulphamethoxazole (1.25/23.75 µg, Liofilchem s.r.l), Chloramphenicol (30 µg, Liofilchem s.r.l), Penicillin G (10 IU, Liofilchem s.r.l), Clarithromycin (30 µg, Liofilchem s.r.l) and Erythromycin (15 µg, Liofilchem s.r.l) were placed on the dried plates, and following 18 hours of incubation (growth) at 37°C, the zones of inhibition were measured to the nearest millimetres.

2.7 Statistical Analysis

Excel 2007 was used to enter data and to obtain the general statistics parameters of mean and standard deviation, and to plot charts. The statistical software package, Statistical Package for Social Scientist (SPSS) version 20.0 was used to carry out the Chi square statistic to verify statistical difference of bacteria prevalence among TB patients. A P value of ≤ 0.05 was considered to be significant.

3. RESULTS AND DISCUSSION

A total of 111 pulmonary tuberculosis patients gave their consent to participate in this study. Of these, 61 (55%) were males and 50 (45.0%) females. The age varied from 20-80 years with the mean age of 35.15 \pm 11.87. The PTB patients were grouped into HIV positive and HIV negative cases. A majority of the TB cases were in the age group ranges of 31-40, 20-30 and 41-50 respectively.

Of the 111 sputum samples, 43 (38.7%) had significant growth and were positive and 68 (61.3%) were negative (no growth). From the positive cultures, 40 (93.0%) yielded single isolates and 3 (7.0%) yielded two isolates each giving an overall of 46 isolates from 43 (38.7%) sputum samples. There was higher prevalence of the bacterial infection in females 24 (48.0%) than in males 19 (31.1%) ($P=0.324$). Although prevalence of bacterial LRTIs was not significantly different by age the risk steadily increased with increase in age group ($P=0.088$). The age group of 31-40 had the highest prevalence of bacterial infection 20 (48.7%), followed by the age group of 20-30, 14 (38.7%). Gram positive cocci were the most isolated 40 (87%) compared with gram negative bacilli 6 (13%). Of the gram positives,

Staphylococcus aureus had the highest frequency of isolation with 28 (60.9%) and *Streptococcus pneumoniae* with 12 (26.9%). *S. aureus* was more prevalent in HIV positive patients, 20(66.5%) as compared to HIV negative patients, 8(32.7%). For the gram negatives, *Pseudomonas aeruginosa* was the only isolate with a frequency of 6 (13%). However, HIV seemed to influence the prevalence of these bacterial infections in both males and females with prevalence of 30 (42.9 %) in HIV positive TB patients compared with HIV negative TB patients 13 (31.7 %) [$P=0.523$]. *Staphylococcus aureus* was found to be the predominant isolates in all the cases. Tables 1 and 2 shows the number of positive cultures for all study groups and numbers of bacterial isolates.

Drug susceptibility testing was carried out on all 46 isolates, revealed an overall drug resistance of 57.0 %. The resistance ranged from (0.0-100) % depending on the antibiotics and the species of microorganisms. The lowest susceptibility was observed for all the isolates to Penicillin followed by Oxacillin, Cotrimoxazole and Amoxicillin All the isolates were resistant to at least one antibiotic. Rates of resistance were generally low for Gentamicin, Ciprofloxacin, and Chloramphenicol respectively (Table 3). Overall percentage resistance per isolates was observed to be highest in *Staphylococcus aureus* ranging from 21.4%-100% depending on the antibiotic, whereas *Pseudomonas aeruginosa* was observed to exhibit an overall percentage resistance of 13.0% being the lowest.

3.1 Discussion

We recorded a general prevalence of 38.7% positive non MTB bacterial cultures leaving a greater percentage of negative results 61.3%. This highlights the importance of these bacterial infections which are often not specifically targeted once TB is diagnosed. A recent investigation of non AFB bacteria amongst health seekers suspected with TB in the South West Region, Cameroon, showed a high percentage of 61.4% and that HIV positive also significantly increased the risk of developing LRTIs [17]. In Cameroon the present algorithm for the diagnosis of TB hardly includes gram stain and culture for these other bacterial pathogens which could be the cause of deterioration of patients health especially at the intensive phase as discussed in a study by Waitt et al. [9]. This finding is similar to studies carried out by Akingbade et al. [18], Gauchan et al. [19], who also reported a greater percentage of

negative results. The high bacterial load in the lungs may be due to TB damages, which enhances the multiplication of secondary bacteria invasion and infection. According to Spencer and Philip [20], negative cultures may be attributed to other etiologic agents or patients already on chemotherapy. The frequent use of antibiotics, which may be self-administered by patients or bought over-the-counter from pharmacies as was seen in the questionnaires, may also result in failure to isolate any potential pathogen [21].

Table 1. Non acid fast bacteria isolated from suspected TB patients and HIV status

Bacterial isolates	HIV positive %	HIV negative %	Total %
<i>P. aeruginosa</i>	3 (9.4)	3 (21.4)	6 (13.0)
<i>S. aureus</i>	20 (62.5)	8 (32.7)	28 (60.9)
<i>S. pneumoniae</i>	9 (28.1)	3 (21.4)	12 (26.1)
Total	32 (69.6)	14 (30.4)	46 (100)

Table 2. Status of Non-MTB bacteria patients (HIV, gender and age) compared with their isolation rate

Factor	Status	N°	Prevalence %	
			No growth	Growth
HIV status	Positive	70	40 (57.1)	30 (42.9)
	Negative	41	28 (68.3)	13 (31.7)
Gender	Female	50	26 (52.0)	24 (48.0)
	Male	61	42 (68.9)	19 (31.1)
Age	20-30	31	19 (61.3)	12 (38.7)
	31-40	41	21 (51.2)	20 (48.7)
	41-50	26	18 (69.2)	8 (30.7)
	51-60	4	3 (75.0)	1 (25.0)
	>60	9	7 (77.8)	2 (22.2)

Table 3. Resistance/susceptibility patterns of the Isolates to some antibiotics

Isolates		<i>P. aeruginosa</i> n (%)	<i>S. aureus</i> n (%)	<i>S. pneumoniae</i> n (%)	Total n (%)
Antibiotics					
CFX	R	1 (16.7)	12 (42.9)	8 (66.7)	21 (45.7)
	S	5 (83.3)	16 (57.1)	4 (33.3)	25 (54.3)
P	R	6 (100)	28 (100)	12 (100)	46 (100)
	S	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
AMX	R	6(100)	21 (75)	8 (66.7)	35 (76.1)
	S	0 (0.0)	7 (25)	4 (33.4)	11 (23.9)
C	R	0 (0.0)	8 (28.6)	5 (41.7)	13 (28.3)
	S	6 (100)	20 (71.4)	7 (58.3)	33 (71.7)
COT	R	3 (50)	23 (82.1)	8 (66.7)	34 (73.9)
	S	3 (50)	5 (17.9)	4 (33.3)	12 (26.1)
E	R	5 (83.3)	18 (64.3)	6 (50)	29 (63.0)
	S	1 (16.7)	10 (35.7)	6 (50)	17 (37.0)
GEN	R	0 (0.0)	7 (25)	5 (47.1)	12 (26.1)
	S	6 (100)	21 (75)	7 (58.3)	34 (73.9)
CLA	R	5 (83.3)	10 (35.7)	4 (33.3)	19 (41.3)
	S	1 (16.7)	18 (64.3)	8 (66.7)	27 (58.7)
OXA	R	6 (100)	23 (82.1)	11 (91.7)	40 (87)
	S	0 (0.0)	5 (17.9)	1 (8.3)	6 (13.0)
CIP	R	2 (33.3)	6 (24.1)	5 (41.7)	13 (28.3)
	S	4 (66.7)	22 (78.6)	7 (58.3)	33 (71.7)

Legend: CFX-Cefotaxime; P-Penicillin; AMX-Amoxicillin; C-Chloramphenicol; COT-Cotrimoxazole; E-Erythromycin; GEN-Gentamicin; CLA-Clarithromycin; OXA-Oxacillin; CIP-Ciprofloxacin

The findings in this study recorded a prevalence of bacterial infection in females (48.0%) than males (31.1%) with *Staphylococcus aureus* being the predominant isolates. It is in line with the findings of Theophilus et al. [22] who reported a prevalence of bacterial isolates in TB patients of 48.8% in females and 22.9% in males. This finding also agrees with a study carried out by El-Mahmood et al. [23] and Okesola and Ige [24] who reported a prevalence of 50.9% and 54.7% respectively but also disagrees with other findings carried out by Humphrey et al. [25] and Akingbade et al. [18] who reported a higher prevalence in males than in females. The higher risk in women from our study may be due to lifestyle, in this case, the dominance in the daily burning of wood for cooking in our setting by the women which exposes them more to lower respiratory tract infections and pathogens. We found a higher prevalence of *Staphylococcus aureus* amongst HIV patients. Hong Nguyen et al. [26] reported that *S aureus* is a common course of serious infections in patients infected with HIV.

There was no statistically significant difference in the prevalence of bacterial infections in the age group though the age group of 31-40 (48.7%) which recorded the highest prevalence followed by the age group of 20-30 (38.7x%). Lower respiratory tract infection is known to be more prevalent in the elderly, showing a shift in the trend to young adults. This is in contrast with the study carried out by Taura et al. [27] where age group ranging from 20-29 years had more isolates 10 (23.5%) followed by 30-39 years 8 (18.6%). The findings of Theophilus et al. [22] showed high prevalence of bacterial isolates among TB patients in the age group of 21-30 (24.1%), followed by 31-40 (15.9%). Members of the age groups of 20-40 are usually the more active and more involved in social and economic activities than those of the other age group which explains the high prevalence of bacterial isolates. This exposes them more to lower respiratory tract pathogens and infections. The numbers of isolates in the age group range of 51-60 years was less than in the range of >60yrs. Members of these age range are usually also involved in social and economic activities and as such are also exposed to respiratory infection, but their levels of exposure is less than those of the age groups of 20-30, 31-40 and 41-50 years and they are usually not immunocompromised. Elderly (>60 years) usually have a weaker immune system due to their age and so are also exposed to respiratory tract infections and may also have

other infections other than HIV which predisposes them to these respiratory tract pathogens and infections (Chan et al., 1995).

Although not statistically significant like the cases of gender and age, a high prevalence of the bacterial infection was recorded among HIV positive cases (42.9%) compared with (31.7%) recorded among HIV negative cases. This result is in line with the findings of Theophilus et al. [22]. The high prevalence among TB/HIV positive cases, might be attributed to the fact that TB and HIV are known immunosuppressive infectious agents which gives route for opportunistic and non-opportunistic pathogens to set in.

Of the 46 bacterial isolates, Gram positive cocci 40 (87%) were the most isolated, compared to Gram negative 6 (13%). Bacterial isolates in our study were *S. aureus* 28 (60.9%), *S. pneumoniae* 12 (26.1%) and *Pseudomonas aeruginosa* 6 (13.0%). *Staphylococcus aureus* (60.9%) were the predominant isolate followed by *Streptococcus pneumoniae* (26.1%). *P. aeruginosa* (13.0%) was the only gram negative bacteria isolated in this study. This findings correlate with earlier study that reported Gram-positive bacteria as being higher than Gram negative bacterial isolates [22]. In another study carried out by Jafari et al. [28], the prevalent Gram positive isolate was *Staphylococcus aureus* (54.1%), followed by *S.pneumoniae* (45.9%). Jafari et al. [28] identified *Pseudomonas sp* 52(27.6%) as the most prevalent bacterial isolate while *Klebsiella sp* 30(16%) ranked third. The findings in this study was in contrast with earlier studies which reported a higher prevalence of gram negatives over gram positive bacterial isolates [18,19]. We may not ascertain the non-isolation of other bacterial isolates from cultures in this present study but speculate that one reason may be because antibiotics are indiscriminately taken in our environment and most patients only come for proper diagnosis after symptoms have persisted [29]. The differences observed in the prevalence of bacterial isolates in studies elsewhere is attributable to age, season, the type of population at risk and under study, the region, and other factors [30]. Studies have identified *S. aureus* as mostly common with patients in intensive care units because it is implicated in nosocomial pneumonia [31,32]. All of our patients were hospitalized and a majority was critically ill. *S. pneumoniae*, though not the most isolated in this study was the most isolated in HIV positive patients. This pathogen has been

reported as the most significant pathogen in lower respiratory tract infections both in studies involving the general population and those among HIV patients [33-35]. In his study involving HIV infected African adults with lung diseases due to common bacteria; Tchamran recorded 81% of infections due to *S. pneumoniae* and stated it to be the most disturbing pathogen in HIV reactive patients [35]. *P. aeruginosa* infection among patients positive for HIV is often associated with considerable mortality. In our study *P. aeruginosa* contributed 13% of all bacterial infections though there was equal distribution among the HIV positive and negative patients. *P. aeruginosa* has been said to be the most common gram negative bacterial pathogen in nosocomial infections among HIV patients with mortality rate up to 23% [36].

The sensitivity tests indicated that the isolates were resistant to at least one or more antibiotics, although generally, a low percentage of the isolates were sensitive to the antibiotic tested. The result of the sensitivity test showed that both Gram-positive and Gram negative isolates recorded highest sensitivity to gentamicin, ciprofloxacin and chloramphenicol while high resistance was also recorded for antibiotics such as Penicillin (100%), Amoxicillin (96.1%), Oxacillin (87.3%) and Co-trimoxazole (73.9%). This observation poses a serious health problem in the management of TB. From this study, *Pseudomonas aeruginosa* recorded 0.0% resistance to Gentamicin while *Staphylococcus aureus* recorded a resistance of 25%. The major selective force favoring the emergence of antibiotic resistance is their extensive use [37]. It is noteworthy that gentamicin is probably less abused than other antibiotics because of its mode of administration (solely by injection) and the prohibitive cost of procurement. This result is consistent and in the ranges of the study carried out by Ndip et al. [15]. The susceptibility of Ciprofloxacin and Chloramphenicol to isolates is supported by studies of El-Mahmood et al. [23] and Taura et al. [27], wherein most isolates were sensitive to these antibiotics. The high rate of resistance of isolates to Penicillin (100%), Amoxicillin (96.1%) Oxacillin (87.3%) and Co-trimoxazole (73.9%), which are commonly bought over-the-counter in drug stores, contrasts with the marked levels of susceptibilities of the isolates to Gentamicin, thus suggesting a relationship between antibiotic use and the level of drug resistance encountered in this study as previously suggested in another study [37]. In

Shandong, Wang et al. reported a 47.8% resistance rate for *Staphylococcus aureus* for Erythromycin, Ndip et al. [15] documented 28% in Buea and in Yola, Adamawa State, Nigeria [23] recorded a rate of 8.1%. Compared with 64.3 % reported in this study, the discrepancies noted for the different regions suggest that antibiotic susceptibility patterns vary with time and geographical location as previously documented [37]. These discrepancies may also be due prescribing habits of physicians in different regions. *Staphylococcus aureus* and *Streptococcus pneumoniae* recorded a resistance of 82.1% and 91.7% respectively to Oxacillin. These findings are in the ranges of a report by Wang et al. [38] who reported a rate of resistance of 71.5% and 80.0% respectively. We can speculate that there may be a high level of abuse of Oxacillin in the Bamenda area of Cameroon where the drug is cheap and readily available to the local population, thus leading the emergence of resistant strains of organisms to the drug. All of our isolates demonstrated high levels of resistance to Oxacillin.

4. CONCLUSION

The overall prevalence of non-MTB bacterial pathogens among TB patients in the North West Region of Cameroon is high (38.7%). HIV positivity, age and gender did not significantly affect the prevalence of LRTIs due to non MTB bacteria although there is a higher risk with HIV positivity and increased age. Prevalence was also higher in female. *S. aureus* was the most prevalent pathogen both among HIV positive and HIV negative individuals as well as in age and gender. Bacterial isolates showed high resistance to Penicillin, Oxacillin, Cotrimoxazole, Amoxicillin as well as susceptibility to Ciprofloxacin, Chloramphenicol and Gentamicin. Even though, the results obtained in this study indicated that some of the antibiotics used to treat respiratory tract infections caused by these isolates in this community are still effective, there is a danger of growing drug resistance which needs to be tackled. The findings of this study will help in redesigning diagnostic algorithm for TB suspected cases to include investigation of non-mycobacterial pathogens.

CONSENT

Patients were enrolled into the study only if they gave their written informed consent for both participation in the study and for HIV testing.

ETHICAL APPROVAL

All authors hereby declare that all experiments have been examined and approved by the Faculty of Health Sciences Institutional Review Board, University of Buea and have therefore been performed in accordance with the ethical standards laid down in the 1964 declaration of Helsinki. The Faculty of Health Sciences Institutional Review Board Project number is 2015/345.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Carroll KC. Laboratory diagnosis of lower respiratory tract infections: Controversy and conundrums. *Journal of Clinical Microbiology*. 2002;40(9):3115-3120.
2. Zafar A, Hussain Z, Lomama E, Sibiie S, Irfan S, Khan E. Antibiotic susceptibility of pathogens isolated from patients with community-acquired respiratory tract infections in Pakistan- The active study. *Journal of Ayub Medical College Abbottabad*. 2008;20(1):7-9.
3. Shailaja VV, Pai LA, Mathur DR, Lakshmi V. Prevalence of bacterial and fungal agents causing lower respiratory tract infection in patients with HIV infection. *Indian Journal of Medical Microbiology*. 2004;22:28-33.
4. Dufour V, Cadranel S, Wislez M, Lavole A, Bergot E, Parrot A, Rufat P, Mayaud C. Changes in the pattern of respiratory diseases necessitating hospitalization of HIV-infected patients since the advent of highly active antiretroviral therapy. *Lung*. 2004;182(6):331-41.
5. George J, Hamida A, Das AK, Amarnath SK, Rao RS. Clinical and lab profiles of 60 patients with AIDS: A South Indian study. *Southeast Asian Journal of Tropical Medicine and Public Health*. 1996;27:686-90.
6. Schleicherg G, Feldman C. Dual infection with *Streptococcus pneumoniae* and *Mycobacterium tuberculosis* in HIV-seropositive patients with community acquired pneumonia. *International Journal of Tuberculosis and Lung Diseases*. 2003; 7:1207-1208.
7. Furumoto A, Taniguchi T, Mogol-Gustilo L, Telan E, Dimaano EM, Villarama B, Shimazaki T, Suzuki M, Yoshida LM, Ariyoshi K. The influence of bacterial co-infection on mortality among hospitalized patients with pulmonary tuberculosis in the Philippines. (1) *Clinical Medicine*, Nagasaki University Institute of Tropical medicine, Nagasaki City, Japan, (2) Okinawa Prefectural Chubu Hospital, Uruma City, Japan, (3) San Lazaro Hospital, Metro Manila, Philippines; 2012.
8. Ihongbe J, Agwu E, Inyang N. Pneumococcal pneumonia complicates presentation of pulmonary tuberculosis and pseudomembranous candidiasis, predictive of unknown HIV infection in Ekpoma Nigeria. *The International Journal of Microbiology*. 2007;5:2.
9. Waitt CJ, Peter KBN, White SA, Kampmann B, Kumwenda J, Heyderman RS. *Journal of Infectious Disease*. 2011; 204(3):358-62.
10. Kumari HBV, Nagarathna S, Chandramuki A. Antimicrobial resistance pattern among aerobic gram-negative bacilli of lower respiratory tract illness in the community. *Thorax*. 2007;56:109-114.
11. Adam D. Global antibiotic resistance in *Streptococcus pneumoniae*. *Journal of Antimicrobial Chemotherapy*. 2002;50:1-5.
12. Gauchan P, Lekhak B, Sherchand JB. The prevalence of lower respiratory tract infection in adults visiting Tribhuvan University Teaching Hospital. *Journal of Institute of Medicine*. 2006;28(2):10-14.
13. Sader HS, Jones RN, Gales AC, Silva JB, Pignatari AC. SENTRY Participants Group (Latin America). SENTRY antimicrobial surveillance program report: Latin American and Brazilian results for 1997 through 2001. *Brazilian Journal of Infectious Diseases*. 2004;8:25-79.
14. Song JH, Chang JY, Suh KS, Ko SI, Jung WS, Oh KR, Peck NY, Lee Y, Yang A, Chongthaleong N, Aswapokee CH, Chiu MK, Lalitha J, Perera TT, Yee G, Kumararansinghe F, Jamal A, Kamarulazaman N, Parasakthi PH, Van T, So TK. Ng and on behalf of the ANSORP Study Group. Macrolide resistance and genotypic characterization of *Streptococcus pneumoniae* in Asian countries: a study of the Asian Network for Surveillance of Resistant Pathogens (ANSORP). *Journal of Antimicrobial Chemotherapy*. 2004;53:457-463.

14. Vlieghe E, Phoba MF, Tamfun JJM, Jacobs J. Antibiotic resistance among bacterial pathogens in Central Africa: A review of the published literature between 1955 and 2008. *International Journal of Antimicrobial Agents*; 2008.
15. Ndip RN, Ntiege EA, Ndip LM, Nkwelang G, Akoachere TK, Nkuo AT. Antimicrobial resistance of bacterial agents of the upper respiratory tract of school children in Buea, Cameroon. *Journal of Health Population and Nutrition*. 2008;26:397-404.
16. Cheesbrough M. *District Laboratory Practice in Tropical Countries Part II*. Cambridge University Press, Cambridge. 2006;183(2):166-167.
17. Ngekeng S, Thumamo Pokam B, Dilonga Meriki H, Longdoh Njunda A, Nguedia Assob JC, Ane Anyangwe I. High prevalence of bacterial pathogens in sputum of tuberculosis suspected patients in Buea. *British Microbiology Research Journal*. 2016;11(5):1-8.
18. Akingbade OA, Ogiogwa JI, Okerentugba PO, Innocent-Adiele HC, Onoh CC, Nwanze JC, Okonko IO. Prevalence and antibiotic susceptibility pattern of bacterial agents involved in lower respiratory tract infections in Abeokuta, Ogun State, Nigeria. *Report and Opinion*. 2012;4(5): 25-30.
19. Gauchan P, Lekhak B, Sherchand JB. The prevalence of lower respiratory tract infection in adults visiting Tribhuvan University Teaching Hospital. *Journal of Institute of Medicine*. 2006;28(2):10-14.
20. Spencer RC, Philip JR. Secondary respiratory infections in hospital patients: Effect of antimicrobial agents and environment. *British Medical Journal*. 1974;2:359-62.
21. Santanam P, Morenzoni G, Kayder FH. Prevalence of antimicrobial resistance in *Haemophilus influenzae* in Greece, Lebanon and Morocco. *European Journal of Clinical Microbiology and Infectious Diseases*. 1990;9:818-20.
22. Theophilus KCU, Moses J, Uzoechina A, Ameh EJO, Chigozie NO. Microbial aetiologic agents associated with pneumonia in immunocompromised hosts. *African Journal of Infectious Diseases*. 2010;4(1):1-6.
23. El-Mahmood M, Isa H, Mohammed A, Tirmidhi AB. Antimicrobial susceptibility of some respiratory tract pathogens to commonly used antibiotics at the Specialist Hospital, Yola, Adamawa State, Nigeria. *Journal of Clinical Medicine and Research*. 2010;2(8):135-142.
24. Okesola AO, Ige OM. Trends in bacterial pathogens of lower respiratory tract infections. *Indian Journal of Chest Disease and Allied Science*. 2008;50:269-272.
25. Humphery H, Newcombe RG, Entone J, Smyth ET, McIlvenny G, Davis E, Spancer R. Four country health care-associated infection prevalence survey: Pneumonia and lower respiratory tract infections. *Journal of Hospital Infections*. 2010;74(3): 266-270.
26. Hong Nguyen M, Carol A. Kauffman, Richard P. Goodman, Cheryl Squier, Robert D. Arbeit, Nina Singh, Marilyn M. Wagener, Victor L. Yu. Nasal carriage of and infection with *Staphylococcus aureus* in HIV-Infected Patients. *Ann Intern Med*. 1999;130(3):221-225.
27. Taura DW, Hassan A, Yayo AM, Takalmawa H. Bacterial isolates of the respiratory tract infection and their current sensitivity pattern among patients attending Aminu Kano Teaching Hospital Kano-Nigeria. *International Research Journal of Microbiology*. 2013;4(9):226-231.
28. Jafari NJ, Ranjbar R, Haghi-Ashtiani MT, Abedini M, Izadi M. The study of prevalence and antimicrobial susceptibility of tracheal bacterial strains isolated from paediatric patients. *Pakistan Journal of Biological Science*. 2009;12(5):455-458
29. Ndip RN, Titanji VPK, Akenji TN, Mutanga AM, Mbacham WF, Ndip LM. Antibigram of *Klebsiella pneumoniae* isolates from Buea, Cameroon. *Central African Journal of Medicine*. 2001;47:173-6.
30. Collee JG, Watt B. Bacterial infection of respiratory tract. In Topley and Wilson's *Principles of Bacteriology, Virology, and Immunity*. 1990;8(2).
31. Hageman JC, Uyeki TM, Francis JS. Severe community-acquired pneumonia due to *Staphylococcus aureus*, 2003–04 influenza season. *Emerging Infectious Diseases*. 2006;12:894–899.
32. Gillet Y, Issartel B, Vanhems P. Association between *Staphylococcus aureus* strains carrying gene for Panton-Valentine leukocidin and highly lethal necrotising pneumonia in young immunocompetent patients. *Lancet*. 2002;359:753–759.

33. Patel SN, McGeer A, Melano R, Tyrrell GJ, Green K, Pillai DR, Low DE. Susceptibility of *Streptococcus pneumoniae* to fluoroquinolones in Canada. *Antimicrob Agents Chemother*. 2011;55(8):3703-3708.
34. Ndiaye AG, Boye CS, Hounkponou E, Gueye FB, Badiane A. Antimicrobial susceptibility of select respiratory tract pathogens in Dakar, Senegal. *J Infect Dev Ctries*. 2009;3(9):660-666.
35. Tchamran M. Bacterial lung disease from common bacteria during HIV infection in African adults hospitalized in Abidjan. *Bull Soc Pathol Exot*. 1997;90:370-372.
36. Franzetti F, Grassini A, Piazza M, et al. Nosocomial bacterial pneumonia in HIV-infected patients: Risk factors for adverse outcome and implications for rational empiric antibiotic therapy. *Infection*. 2006;34:9-16.
37. Ndip RN, Akoachere J-FT, Mokosso DK, Ndip LM, Anyangwe IN. Carriage of *Vibrio* species by shrimps harvested from the coastal waters of South West Cameroon. *East African Medical Journal*. 2002;9:146-9.
38. Wang Y Zhang, Li WR, Feng Y, Leng T. Serious antimicrobial resistance status of pathogens causing hospital-acquired lower respiratory tract infections in North China. *The Journal of International Medical Research*. 2009;37:899-907.

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