https://doi.org/10.33472/AFJBS.3.2.2021.36-41
ISSN: 2663-2187

African Journal of Biological Sciences

Journal homepage: http://www.afjbs.com

Research Paper

Open Access

Antibiotic resistance of bacteria responsible for lower respiratory tract infections seen in the University Hospital of Befelatanana Antananarivo Madagascar

Zafindrasoa Domoina Rakotovao-Ravahatra[1]*, Fidiniana Mamy Randriatsarafara[2] and Andriamiadana Luc Rakotovao[3]

[1]Medical Biology Department of the Faculty of Medicine, University of Antananarivo, Madagascar. E-mail: ravahatradomoina@yahoo.fr
[2]Public Health Department of the Faculty of Medicine, University of Antananarivo, Madagascar. E-mail: fidyrfm@yahoo.fr
[3]Medical Biology Department of the Faculty of Medicine, University of Antananarivo, Madagascar. E-mail: lucdina007@yahoo.fr

Article Info

Volume 3, Issue 2, April 2021
Received : 19 August 2020
Accepted : 12 January 2021
Published : 05 April 2021
doi: 10.33472/AFJBS.3.2.2021.36-41

Abstract

The majority of bacteria responsible for Lower Respiratory Tract Infections (LRTIs) are multiresistant to antibiotics. The objectives of this study are to describe the antibiotic resistance of these bacteria in LRTIs and to describe the factors associated with these infections. It is a prospective study of 54 respiratory samples of patients with LRTIs for a period of six months from July 2019 to December 2019 in the laboratory of the University Hospital of Befelatanana Antananarivo. The microbiological results showed 12 (22%) isolates of nonfermenting gram-negative bacilli, 10 (19%) isolates of streptococci and 6 (11%) isolates of enterobacteria. Regarding the isolates of nonfermenting gram-negative bacilli, the antibiotic resistance, varies from 42% (amikacin) to 100% (cotrimoxazole). Concerning the isolates of streptococci, it varies from 0% (vancomycin) to 80% (penicillin G). And the antibiotic resistance of the isolates of enterobacteria varies from 0% (imipenem and amikacin) to 100% (amoxicillin). Concerning the associated factors, subjects aged 40 and over (58.6%) (p = 0.84; NS), men (71.4%) (p = 0.01) and subjects hospitalized in intensive care units (70.3%) (p = 0.004) are the most affected by LRTIs. In brief, the knowledge of antibiotic resistance of bacteria responsible for LRTIs allows better patient management.

Keywords: *Respiratory samples, Antibiotic resistance, Streptococci, Enterobacteria*

1. Introduction

According to the Global Burden of Disease 2015 study, Chronic Obstructive Pulmonary Disease (COPD) and Lower Respiratory Tract Infections (LRTIs) represent the third and fourth most common causes of death, respectively, after ischemic heart disease and cerebrovascular disease (Global Burden of Disease, 2016). LRTI is a broad terminology which includes different diseases including acute bronchitis, pneumonia, and acute exacerbation of chronic lung diseases such as COPD or bronchiectasis. Annual incidence of pneumonia, one

* *Corresponding author: Zafindrasoa Domoina Rakotovao-Ravahatra, Medical Biology Department of the Faculty of Medicine, University of Antananarivo, Madagascar. E-mail: ravahatradomoina@yahoo.fr*

of the most important LRTIs, is reported to be 24.8 per 10,000 adults. The rates differ based on the age, with higher incidence observed in patients between 65 and 79 years of age (63.0 / 10,000 adults) and >80 years of age (164.3 / 10,000 adults) (CDC EPIC Study Team, 2015). They are usually treated by antibiotics, although guidelines and recommendations suggest reducing the total volume of antibiotic prescription. Likewise, epidemiological studies are generally focused on supplementary investigations and on the prescriptions of antibiotics. While patients' complaints such as fever, cough, sputum and dyspnoea lead to symptomatic treatment prescription by general practitioners (GPs) in addition to antibiotics, little is known about the real magnitude of non-antibiotic therapeutic prescriptions in LRTI (Rahericon *et al*., 2003). To improve the management of patients with LRTIs, the aims of this study are to describe the antibiotic resistance of bacteria responsible for LRTIs and to describe the factors associated with these infections.

2. Materials and methods

It is a prospective study of 54 respiratory samples of patients with LRTIs for a period of six months from July 2019 to December 2019 in the laboratory of the University Hospital of Befelatanana Antananarivo Madagascar. This study includes all respiratory samples of patients who have applied for microbiological examination. These respiratory samples are either sputum, or bronchial aspiration fluids or bronchoalveolar fluids. This study excludes any non-compliant respiratory samples such as salivary specimens and respiratory samples contained in a transport medium other than the laboratory vial (sterile red cap vial). In the laboratory, the respiratory samples were cultured in ordinary agar called Uriselect®, in sheep blood agar prepared from a mixture of Columbia agar® and sheep blood and in chocolate agar. Afterwards, the petri dishes containing agar are incubated in the oven at 37 °C for 48 h. Sheep blood agars and chocolate agars are incubated in an atmosphere rich in CO_2 to facilitate the culture of certain bacteria. Then, a microscope examination is performed after gram staining to quantify epithelial cells and leukocytes and to identify bacteria. After 48 h, the positivity criteria are the presence of bacterial colonies $\geq 10^7$ CFUs (colony-forming units) per mL, the number of epithelial cells ≤ 25 and the number of leukocytes ≥ 25. The bacterial colonies are then identified by the various bacterial identification tests available in the laboratory. Finally, the antibiogram corresponding to the identified bacterial species is carried out. Resistance to antibiotics was determined by the Mueller / Hinton agar diffusion method, according to the recommendations of the "comité de l'antibiogramme de la société française de Microbiologie" (Société Française de Microbiologie, 2019). The dependent variable is constituted by the positivity of the microbiological culture and the results of antibiogram. The independent variables consist of the type of respiratory sample, the gender, the age, the clinical information, the departments, the result of microbiological culture of the pulmonary secretion samples and the results of antibiogram. The antibiotics tested are represented by penicillins (penicillin G, amoxicillin, ticarcillin), amoxicillin-clavulanic Acid (AMC), 3rd generation cephalosporins (3CG), penemes (imipenem), aminoglycosides (gentamycin, tobramycin, amikacin), quinolones (nalidixic acid, ciprofloxacin, levofloxacin), macrolides (erythromycin), lincosamides (lincomycin), cyclins (tetracyclin) , sulfonamides (trimethoprim- sulfamethoxazole : cotrimoxazole) and amphenicols (chloramphenicol). This study was authorized by the director of establishment of the University Hospital of Befelatanana and the Department Head of the laboratory before its implementation. This study respected the notion of anonymity and confidentiality. The data entry and processing was performed on the software Epi-info 3.5.2. The comparison of percentages used the chi-square tests. The statistical significance threshold used was $p = 0.05$.

3. Results

3.1. Microbiological results of the respiratory samples

Among the 54 respiratory samples, the microbiological results showed 12 (22%) isolates of nonfermenting gram-negative bacilli (NFGNB), 10 (19%) isolates of streptococci, 6 (11%) isolates of enterobacteria and 3 (6%) isolates of yeasts (Figure 1). NFGNBs are represented essentially by the species *Acinetobacter baumannii* and *Pseudomonas aeruginosa*. Enterobacteria are represented essentially by the species *Klebsiella pneumonia* and *Escherichia coli*.

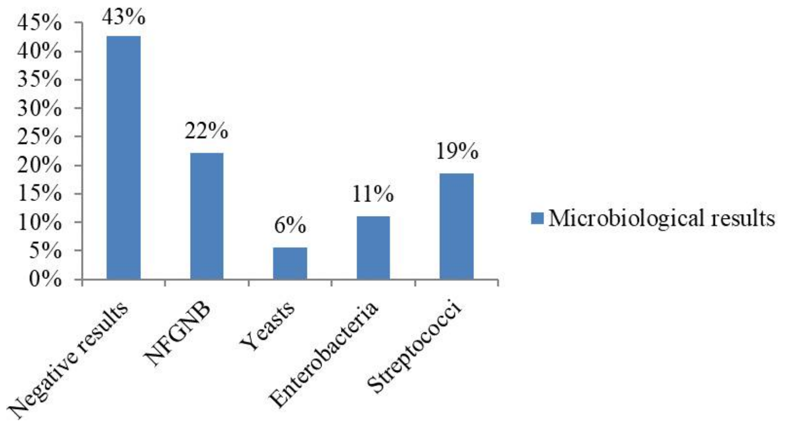

Figure 1: Microbiological results of the respiratory samples

3.2. Antibiotic resistance of bacteria responsible for LRTIs

Regarding the isolates of NFGNB, the antibiotic resistance, varies from 42% (amikacin) to 100% (cotrimoxazole) (Figure 2).

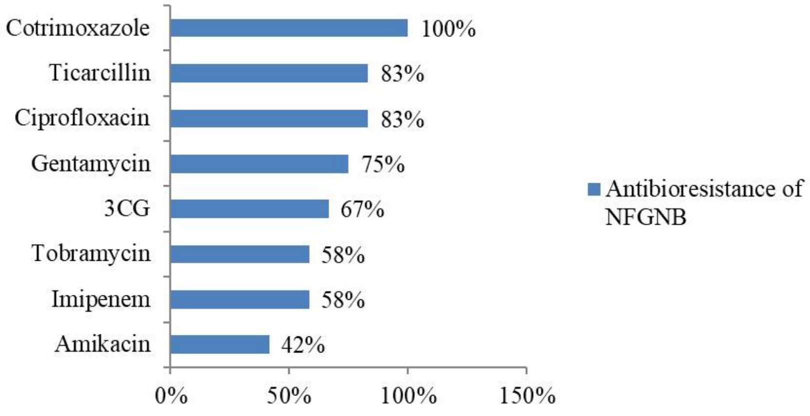

Figure 2: Antibiotic resistance of NFGNB

Concerning the isolates of streptococci, the antibiotic resistance varies from 0% (vancomycin) to 80% (penicillin G) (Figure 3).

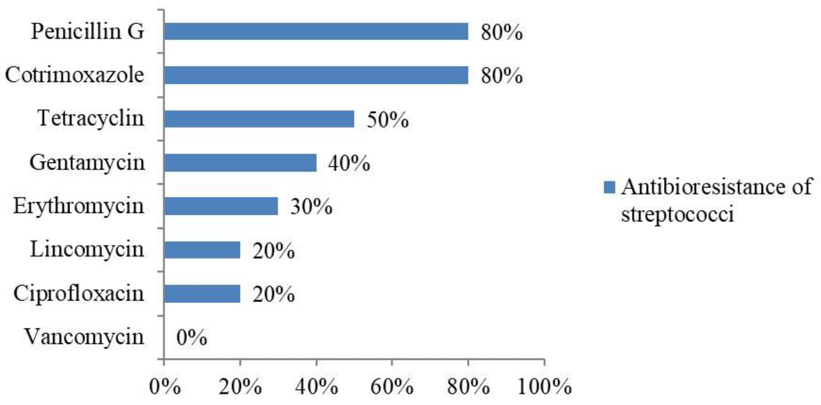

Figure 3: Antibiotic resistance of streptococci

The antibiotic resistance of the isolates of enterobacteria varies from 0% (imipenem and amikacin) to 100% (amoxicillin) (Figure 4).

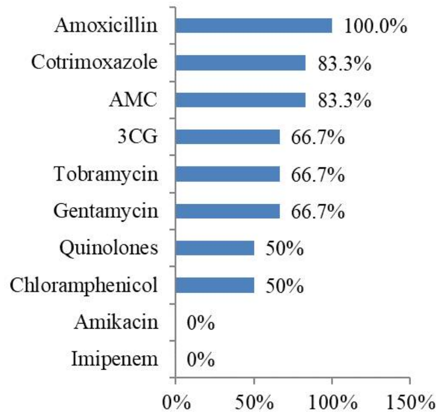

Figure 4: Antibiotic resistance of enterobacteria

3.3. Associated factors with LRTIs

Concerning the associated factors, subjects aged 40 and over (58.6%) ($p = 0.84$; NS), men (71.4%) ($p = 0.01$) and subjects hospitalized in intensive care units (70.3%) ($p = 0.004$) are the most affected by LRTIs (Table 1).

Table 1: Associated factors with LRTIs

	Negative results		LRTIs		Total	*p*-value
	n	%	n	%		
Age (years)						
<40	11	44.0	14	56.0	25	0.84
≥40	12	41.4	17	58.6	29	
Gender						
Female	13	68.4	6	31.6	19	0.01
Male	10	28.6	25	71.4	35	
Departments						
Intensive care units	11	40.7	26	70.3	37	0.004
Other departments	12	66.7	5	29.4	17	

4. Discussion

This study has shown the isolation of NFGNB in 22% of respiratory samples. This result is higher compared to another study carried out in Coastal Karnataka which found 5% of NFGBN (Chawla *et al.*, 2013). Similarly, Malini *et al.* from Kolar in India have documented the isolation of 6.8% (25 of 365) of NFGNB in respiratory samples (Malini *et al.*, 2009). The importance of isolation of non-fermenters has increased in last decade, after more and more reports are correlating them with the either infection outbreaks in hospitals, or healthcare-associated infections (McGowan, 2006). Concerning antibiotic resistance, this study showed a high resistance of isolates of NFGNB to antibiotics. It is 100% for Cotrimoxazole. This antibiotic is improperly prescribed in Madagascar and is sold in small grocery stores. Thus, self-medication reinforces this high resistance. Moreover, the molecules of choice, such as penemes and aminoglycosides, are becoming less and less effective, showing

58% (imipenem) and 42% (amikacin) of resistance. These NFGNBs are represented essentially by the species *Acinetobacter baumannii* and *Pseudomonas aeruginosa*. Similarly, another study in Iran showed resistance *Acinetobacter baumannii* with cefixime (99%), ceftazidime (99%), ciprofloxacin (98%), meropenem (99%), trimethoprim-sulfamethoxazole (99%), imipenem (91.5%), ceftriaxone (99%), levofloxacin (96.5%), amikacin (70 %) and gentamycin (85%) (Goudarzi *et al*., 2013). The emerging challenges of multidrug resistance, both intrinsic and acquired, among NFGNB, are of serious concern to the treating physician. Improved antibiotic stewardship and infection control measures will be needed to prevent or slow down the emergence and spread of multidrug-resistant NFGNB in the healthcare setting. Identification of NFGNB and monitoring their susceptibility patterns will help in improving the empirical therapy (Baruah *et al*., 2015).

Concerning the streptococci isolates, this study has shown 19% isolates in LRTIs. This result is higher compared to another study which found 8% of streptococci in LRTIs (Lieberman *et al*., 2002). These isolates are also highly resistant to Cotrimoxazole like the NFGNB isolates. Likewise, they are highly resistant to tetracycline and penicillin G. Indeed, these three classes of antibiotics are used frequently by the population. Cotrimoxazole and tetracycline are sold in small grocery stores. Misuse of these antibiotics increases antibiotic resistance. However, the streptococci isolates are less virulent than the NFGNB isolates because they are sensitive to other antibiotics in the majority of cases. All isolates tested in this study were susceptible to vancomycin. These results are consistent with findings reported in other countries (Camara *et al*., 2013; SENTRY Surveillance Program, 2008). These data suggest that glycopeptides (teicoplanin and vancomycin) could be an effective alternative choice in streptococci infections especially in LRTIs.

Concerning other bacteria, this study found 11% of enterobacteria isolates in LRTIs. These enterobacteria are represented essentially by the species *Klebsiella pneumonia* and *Escherichia coli*. This result is lower compared to another study carried out in Nepal which found 36 (17.9%) isolates of *Klebsiella pneumonia* and 26 (12.94%) isolates of *Escherichia coli* (Khan *et al*., 2015). These isolates are also highly resistant to Cotrimoxazole like the streptococci isolates. Likewise, they are highly resistant to beta-lactams like amoxicillin, AMC and 3CG. The high consumption of beta-lactams by the population may be responsible for this antibiotic resistance (Randriatsarafara *et al*., 2015). However, all enterobacteria isolates tested in this study were susceptible to amikacin and imipenem. Another study in Douala also found that imipenem (1.3% of resistance) and amikacin (12.9%) are the most effective antibiotics against enterobacteria (Ebongue *et al*., 2015). These data suggest that penems (imipenem, meropenem) and aminoglycosides especially amikacin could be an effective alternative choice in enterobacteria infections especially in LRTIs. The other aminoglycosides like gentamicin and tobramycin are less effective because they are used frequently in the hospital resulting in the antibiotic resistance of enterobacteria.

Concerning the associated factors, men were the most affected by respiratory infections with a significant difference. This is probably due to the toxic habits of men, especially tobacco, which promotes lung infections. Likewise, subjects hospitalized in intensive care units are the most affected by LRTIs with a significant difference. In fact, LRTIs, in particular pneumonia associated with healthcare, is the most common infection in intensive care (Koulenti *et al*., 2017). This infection includes two different entities including Ventilator Associated Pneumonia (VAP) and severe pneumonia acquired in the hospital. The incidence of VAP is between 1.9 and 3.8 per 1,000 days of mechanical ventilation in the United States and exceeds 18 per 1,000 days of mechanical ventilation in Europe (Koulenti *et al*., 2017).

5. Conclusion

In brief, the bacteria responsible for LRTIs have been represented by NFGNB, streptococci and enterobacteria. Among these bacteria, NFGNB are the most resistant to antibiotics. These bacteria are common in intensive care units. Thus, In view of the antimicrobial resistance of NFGNB, detection in a patient must be the subject of an official declaration of the circulation of this strain at Coordination center for the fight against nosocomial infections.

References

Baruah, F., Hussain, A.N., Kausalya. and Grover, R.K. (2015). Antibiotic resistance profile of non-fermenting Gram-negative bacilli isolated from the blood cultures of cancer patients. *Journal of Global Infectious Diseases*. 7(1), 46-47.

Camara, M., Dieng, A. and Bouh Boye, C.S. (2013). Antibiotic susceptibility of streptococcus pyogenes isolated from respiratory tract infections in dakar, senegal. *Microbiology Insights.* 6, 71-75.

CDC EPIC Study Team (2015). Community-acquired pneumonia requiring hospitalization among US adults. *The New England Journal of Medicine.* 373, 415-427.

Chawla, K., Vishwanath, S. and Munim, F.C. (2013). Nonfermenting Gram-negative Bacilli other than *Pseudomonas aeruginosa* and *Acinetobacter* spp. causing respiratory tract infections in a tertiary care center. *Journal of Global Infectious Diseases.* 5(4), 144–148.

Ebongue, C.O., Tsiazok, M.D., Nda Mefo'o, J.P., Ngaba, G.P., Beyiha, G. and Adiogo, D. (2015). Evolution of antibiotic resistance of Enterobacteriaceae isolated at the Douala General Hospital from 2005 to 2012. *The Pan African Medical Journal.* 20, 227.

Global Burden of Disease (2016). Mortality and Causes of Death Collaborators. Global, regional, and national life expectancy, all-cause mortality, and cause-specific mortality for 249 causes of death, 1980-2015: A systematic analysis for the global burden of disease study 2015. *Lancet.* 388, 1459-1544.

Goudarzi, H., Douraghi, M., Ghalavand, Z. and Goudarzi M. (2013). Assessment of antibiotic resistance pattern in *Acinetobacter baumannii* carrying bla oxA type genes isolated from hospitalized patients. *Novelty in Biomedicine.* 1, 54-61.

Khan, S., Priti, S. and Ankit, S. (2015). Bacteria etiological agents causing lower respiratory tract infections and their resistance patterns. *Iranian Biomedical Journal.* 19(4), 240-246.

Koulenti, D., Tsigou, E. and Rello, J. (2017). Nosocomial pneumonia in 27 ICUs in Europe: perspectives from the EUW VAP/CAP study. *European Journal of Clinical Microbiology & Infectious Disease.* 36(11), 1999-2006.

Lieberman, D., Korsonsky, I., Ben-Yaakov, M., Lazarovich, Z., Friedman, M.G., Dvoskin, B., Leinonen, M., Ohana, B. and Boldur, I. (2002). A comparative study of the etiology of adult upper and lower respiratory tract infections in the community. *Diagnostic Microbiology and Infectious Disease.* 42(1),21-28.

Malini, A., Deepa, E.K., Gokul, B.N. and Prasad, S.R. (2009). Nonfermenting gram-negative bacilli infections in a tertiary care hospital in kolar, Karnataka. *Journal of Laboratory Physicians.* 1(2), 62-66.

McGowan, J.E. (2006). Resistance in nonfermenting gram-negative bacteria: multidrug resistance to the maximum. *The American Journal of Medicine.* 119(6 Suppl 1), S29-36.

Raherićon, C., Poirier, R., Daurès, J.P., Romand, P., Grignet, J.P., Arsac, P., Tartavael, J.M., Touron, D. and Tayard, A. (2003). Lower respiratory tract infections in adults: non-antibiotic prescriptions by GPs. *Respiratory Medicine.* 97(9), 995-1000.

Randriatsarafara, F.M., Ralamboson, J., Rakotoarivelo, R., Raherinandrasana, A. and Andrianasolo, R. (2015). Antibiotic consumption at Antananarivo University Hospital: prevalence and strategic challenges. *Santé Publique.* 27(2), 249-255.

SENTRY Surveillance Program (2008). Comparative in-vitro activity of daptomycin against Grampositive microorganisms: SENTRY surveillance Program, Spain (2002–2006). *Enfermedades Infecciosas y Microbiologia Clinica.* 26(8), 489-494.

Société Française de Microbiologie (2019). Tables of critical concentrations for the interpretation of MICs and critical diameters of inhibition zones. In : *CASFM / EUCAST : Société Française de Microbiologie Ed*, 38-94.

Cite this article as: Zafindrasoa Domoina Rakotovao-Ravahatra, Fidiniana Mamy Randriatsarafara and Andriamiadana Luc Rakotovao (2021). Antibiotic resistance of bacteria responsible for lower respiratory tract infections seen in the University Hospital of Befelatanana Antananarivo Madagascar. *African Journal of Biological Sciences.* 3(2), 36-41. doi: 10.33472/AFJBS.3.2.2021.36-41.