

Non tubercular bacillus etiologic from bacterial lung diseases

Oubayyou AM^{1*}, Gagara Issoufou MA², Mounkaila S³, Traoré AS¹, Moumouni H⁴

(1) Centre de Recherche en Sciences Biologiques Alimentaires et Nutritionnelles, Université de Ouagadougou, 3 BP 7131, Ouagadougou 03, Burkina Faso

(2) Service de Pneumo-physiologie, Hôpital National Lamordé de Niamey, BP: 11653 Niamey-Niger

(3) Faculté des Sciences Techniques, Université d'Agadez, Tél: (+227) 96982186

(4) Faculté des Sciences de la Santé, Université Abdou Moumouni de Niamey, BP: 131/125 Niamey-Niger

*Corresponding Author: Tel: (+227) 96191922; E - mail: mamoudouabdoulaye92@yahoo.fr

ABSTRACT

Etiologic agent of lung diseases can be others bacteria different from tubercular bacillus. Their treatment is based on a probabilistic antibiotherapy. This requires awareness of responsible germs identity in a given region, at all times. With the aim to better care of humane lung infections, a prospective study carried from 1st October 2013 to 30th September 2016. The biologic sample was broncho-alvéolaire fluid, pleural fluid and sputum. At this given period, 247 samples were collected and examined. Bacteriological conventional methods were performed to detect germs in clinical samples. Ninety-one (91) bacteria strains were identified. Their characteristics showed Gram-negative rods group was most represented with 53.86%. *Stenotrophomonas maltophilia* occupy the first rang 14.28% of isolated species in this group. The Gram-positive cocci group was isolated at 46.14%, among which *Staphylococcus aureus* species occupied 24.17%. Diversity of bacteria species in pulmonary infection was showed through the study. That allow to a better chance of success in diagnosis, and probabilistic treatment in the country.

Keywords: Etiology, bacteria, pulmonary disease, in Niamey/Niger

Introduction

Bacterial infections are encountered in lung diseases. Their development has an immediate impact on human health. It is generally tuberculosis that is attributed a central role in this etiology, but enterobacteria (*Klebsiella* sp, *Escherichia coli*, *Yersinia pseudotuberculosis*, etc.) and non-enterobacteria (staphylococci, streptococci, *Haemophilus influenzae*, etc.) have significant morbidity and mortality (Afane et al., 2013).

In Mali, acute respiratory infections represent 8.3% of hospitalizations, and 28.8% in Bobo Dioulasso, Burkina Faso (e-Pilly TroP, 2012).

In Niger, lung diseases remain a concern in public health. In 2013, of all the morbidities observed, cough accounted for 16.50%, pneumonia 9.92% and asthma 0.16% (SNIS, 2013). Pulmonary diseases in general were identified at 23% by Zoumbeye in 2014. Moreover they represented 51% (Koudize, 2013). But the identification of the pathogens is still unknown,

hence the question: what are the bacteria found in pulmonary diseases in Niger? Because it is known to introduce antibiotic therapy the choice is primarily guided by the knowledge of the infecting organism. Ideally, this choice should be made in a targeted way, based on the precise identification of the germ responsible for the infection. If treatment is not initiated in a timely manner, sepsis can also occur and be fatal, especially in the elderly, children and immunocompromised patients. In many cases, the responsible germs can not be identified. In addition, in some serious infections, identification of the causative organ may be impossible to achieve in a sufficiently short time. In an attempt to answer this question, the objective of the study is to identify bacteria isolated from pulmonary samples obtained from patients in health centers in Niamey.

Materials methods

For clinical samples collection, we targeted the two national hospitals that host a large number of patients from the country namely, the Lamordé National Hospital and the National Hospital of Niamey. Added to those, are the other public and private health centers in the area. Biology department of Lamordé National Hospital used us for microbiological manipulation of samples.

This is a prospective cross-sectional study. The data collection was carried out over two (02) years, from October 1st, 2014 to September 30th, 2016.

Biological material consists of bronchoalveolar fluid, pleural fluid and sputum. During this period, 247 samples were collected and examined. The identification of bacterial strains is based on the following phenotypic characters.

Growth characters

- * Culture in the presence or absence of air that is to say of oxygen at partial pressure,
- * Requirement of culture: we can consider as non-demanding a bacterium developing on a so-called ordinary medium (normal agar, peptone water, trypticase agar - soy ...). A demanding bacterium is only grown on enriched medium (blood agar, enriched chocolate agar),
- * Possible cultivation temperatures and optimum temperatures.

Morphological characters

- * The shape of the bacteria: shell, coccobacillus, bacillus, curved bacillus, irregularly shaped bacillus, filamentous bacillus ...
- * Color with Gram stain: Gram + (purple), Gram- (pink).
- * The presence of endospores.

Biochemical characters

- * Presence of enzymes: catalase, oxidase ...,
- * Ability to metabolize a molecule (oses, amino acids, lipids ...),

- * Ability to grow in the presence of an inhibitor, NaCl at various concentrations, antibiotics, antiseptics ...,
- * Synthesis capacity from given carbon sources.

Other criteria

Precise identification may make use of other criteria such as the nature of the antigens (serogrouping).

Data processing and analysis

Databases were designed with Word and Excel 2013 software for results management. Descriptive and comparative analysis of the variables, Epi-Info 6.04 and XLstat.7.1 were used as well as for the Chi-2 and Fisher (LSD) parametric test. To calculate the probability of independence of two variables, a value of p less than or equal to 0.05 is considered significant.

Results

A total of 91 bacterial strains were identified. *Staphylococcus aureus* was the most incriminated species (24.17%) followed by *Streptococcus pneumoniae* (17.58%). Table 1 shows that the Gram-negative bacilli group exceeded half of the isolations (53.86%), the most common species in this group was *Stenotrophomonas maltophilia* (14.28%) and then *Pseudomonas aeruginosa* (13.18%).

Table II: frequency of bacterium species identify (N = 91)

Bacterium species	Effective	Percentage (%)
Gram-positif cocci	42	46,14
▶ <i>Staphylococcus aureus</i>	22	24,17
▶ <i>Staphylococcus epidermidis</i>	4	4,39
▶ <i>Streptococcus pneumoniae</i>	16	17,58
Gram-negative bacilli	49	53,86
Enterobacteria	14	15,44
▶ <i>Klebsiella pneumoniae</i>	4	4,39
▶ <i>Enterbacter cloacae</i>	3	3,29
▶ <i>Escherichia coli</i>	2	2,19
▶ <i>Serratia odorifera</i>	2	2,19
▶ <i>Yersinia pseudotuberculosis</i>	2	2,19
▶ <i>Klebsiella oxytoca</i>	1	1,19
No enterobacteria	35	38,42
▶ <i>Stenotrophomonas maltophilia</i>	13	14,28
▶ <i>Pseudomonas aeruginosa</i>	12	13,18
▶ <i>Haemophilus influenzae</i>	5	5,48
▶ <i>Acinetobacter baumannii</i>	5	5,48
Total	91	100

DISCUSSION

The identified bacteria have belonged either to Gram-positive cocci group or Gram-negative bacilli group.

In the first group, only two bacteria make up of 46.14% the highlighted ones. However, *Staphylococcus aureus* the most predominant, corresponded to 24.17%. The incidence ratio of *Staphylococcus aureus* and its ranking among the germs responsible for pulmonary pathologies varies considerably according to the studies. In Burkina-Faso, a lower value of 13.9% has been reported (Ouedraogo et al., 2010), this difference being the result of the socio-epidemiological situation. But our data were in agreement with those found by authors who estimated this frequency between 16 and 33% (Koulenti et al., 2009, Meyer et al., 2010). The virulence of *Staphylococcus* species is determined by their ability to coagulate blood with coagulase. Coagulase-positive *Staphylococcus aureus* is one of the most ubiquitous and dangerous human pathogens, due to its virulence and ability to develop antibiotic resistance. However, *S. aureus* is a common cause of nosocomial pneumonia and is isolated in 40% of the nosocomial diseases reported in this study. This higher frequency is not in the range of 20 to 30% described by Alp et al. (2004) in nosocomial pneumopathies acquired under mechanical ventilation. The bacterium produces toxins and enzymes that it can excrete and cause specific cell lysis (Parker and Prince, 2012). Pantan-Valentine leucocidin (LPV), described for the first time in 1932, is reported to be carried by less than 5% of *S. aureus* strains in Europe (Holmes et al., 2005).

Coagulase-negative species such as *S. epidermidis* are increasingly involved in nosocomial infections and were found in 4.39% of the isolates in the study. In Cameroon, the rate of *S. epidermidis* is 8% of isolated bacteria (Afane et al., 2013). Their importance is great in pulmonary surgical areas (Afane et al., 2013), because of their ability to form biofilm on medical devices.

Streptococcus pneumoniae (17.58%) was the second most isolated Gram-positive coccus after *Staphylococci*. This bacterium is the cause of lower respiratory infections (bronchi and lungs). Its prevalence varies by country. Some authors have reported higher values: 32.6% by Ouedraogo et al. (2010) in Burkina Faso; 64% by Afane et al. (2013) in Cameroon and up to 70% (e-Pilly 1992). Nosocomial pneumonia with *Streptococcus pneumoniae* was frequent, it represents 20% of registered nosocomial diseases, whereas it is rarely reported in the literature: 0.83% by Chahmout in 2011 in Morocco and 4.1% by Girault et al. (2006) in France. The unequal distribution of pneumococcus responsible for pulmonary diseases can be influenced by the type of microbiological analysis that led to the diagnosis in the various studies, but also by the existence of epidemiological factors (corticosteroids, immunosuppression, cases of patients: medical, surgical or traumatic).

In the second group of bacteria 10 types of Gram-negative bacilli were identified, ie 53.86%. This frequency is similar to 53.5% reported by Ouedraogo et al. (2010) in Burkina Faso. Higher scores (73%) were found by Shahunja et al. (2014) in Bangladesh. The most prominent of Gram-negative bacilli do not belong to Enterobacteriaceae family. They are also involved in nosocomial diseases. These were *Stenotrophomonas maltophilia* (14.28%) followed by *Pseudomonas aeruginosa* (13.18%), *Haemophilus influenzae* and *Acinetobacter baumannii*,

the latter having a frequency of 5.48%. A predominance of *Acinetobacter baumannii* 32.36%, followed by *Pseudomonas aeruginosa* 25.05% was noted by Chahmout, (2011) in Morocco, *Stenotrophomonas maltophilia* constituting only 0.82% of the isolates of his study. Another study on the epidemiology of nosocomial bacterial infections in a Tunisian neonatal and pediatric resuscitation unit in 2006, reported a prevalence of 28.6% of *Pseudomonas aeruginosa* (Ben Jaballah et al., 2006). A lower value (13.5%) of *P. aeruginosa* was found by Dagnra et al. (2003) in Togo. Other bacteria also have a role in pulmonary colonization of this study. These include *Klebsiella pneumoniae*, *Enterobacter cloacae* or more rarely *Escherichia coli*, *Serratia odorifera*, *Yersinia pseudotuberculosis* and *Klebsiella oxytoca*. Our data are close to those of Dagnra et al. (2003) for *Klebsiella* and *Escherichia coli* are respectively 10.4% and 6.3%. The incidence ratio of Gram-negative bacilli and their classification among the germs responsible for pulmonary infections varies considerably according to the studies (Bouyad et al., 2002, Girault et al., 2006, Bennani et al., 2008 and Mehdaoui, 2010).). However, the distribution of germs responsible of pulmonary diseases is influenced by the type of microbiological analysis that led to the diagnosis, but also by the existence of systematic antibiotic therapy, without forgetting status of studied patients (medical, surgical or traumatic) and the existence of comorbidities.

Conclusion

Our results confirm the important place occupied by non-tuberculous bacterial infections in pulmonary diseases etiology in Niger. Several germs have already been reported in the literature. Nevertheless the low frequency of some species isolated does not allow us to make a general statement. It is necessary to determine the antibiogram of these bacterial strains in order to provide therapeutic knowledge and update existing national data to ensure greater chance of successful diagnosis, prevention, surveillance and probabilistic treatment.

References

- Afane Ze E, Djifack TN, Vouking M. 2013. Étiologies des Pneumopathies Bactériennes non tuberculeuses en milieu pneumologique à Yaoundé. Health Sci. Dis, 14 (3): 1-4.
- Alp E, Guven M, Yildiz O, Aygen B, Voss A, Doganay M. 2004. Incidence, risk factors and mortality of nosocomial pneumonia in intensive care units: a prospective study. Ann. Clin. Microbiol. Antimicrob, 3: 17.
- Ben Jaballah N, Bouziri A, Kchaou W, Hamdi A, Mnif K, Belhadj S, Khaldi A, Kazdaghli K. 2006. Épidémiologie des infections bactériennes nosocomiales dans une unité de réanimation néonatale et pédiatrique tunisienne. Med. et Mal. Infect, 36 (7): 379-385.
- Chahmout S. 2011. Pneumopathie nosocomiale a *Acinetobacter Baumannii* en réanimation. Thèse Pharm, N0 72, Université Mohammed V, Raba, 179 pages.
- Dagnra AY, Awessob B, Prince-Davida M, Tidjania O. 2003. Nature et sensibilité aux antibiotiques des bactéries isolées des pleurésies purulentes à Lomé. Med. Mal. Infect, 33 (6): 327–330.

e-Pilly TroP. 2012. Association des professeurs de pathologie infectieuse et tropicale: Maladies infectieuses tropicales. Edition alinéa plus, 975 pages.

e-Pilly TroP. 1992. Association des professeurs de pathologie infectieuse et tropicale: Maladies infectieuses et tropicales. 12e ed, 679 pages.

Girault C, Tamion F, Beduneau G. 2006. Évaluation des soins et pneumopathies nosocomiales en réanimation. *Rev. Mal. Respir*, 23: 4S27-4S43.

Holmes A, Ganner M, McGuane S, Pitt TL, Cookson BD, Kearns AM. 2005. Staphylococcus aureus isolates carrying Panton-Valentine leucocidin genes in England and Wales: frequency, characterization, and association with clinical disease. *J. Clin. Microbiol*, 43: 2384-2390.

Koudize A. 2013. Pleuro-pneumopathies au service de médecine interne de l'hôpital national de Niamey. Thèse de méd, N0 2072, université Abdou Moumouni de Niamey, Niger, 148 pages.

Koulenti D, Lisboa T, Brun-Buisson C, Krueger W, Macor A, Sole-Violan J. 2009. Spectrum of practice in the diagnosis of nosocomial pneumonia in patients requiring mechanical ventilation in European intensive care units. *Crit. Care Med*, 37: 2360-2368.

Le Minor L, Véron M. 1982. Bactériologie médicale. Paris: Flammarion.

Meyer E, Schwab F, Gastmeier P. 2010. Nosocomial methicillin resistant Staphylococcus aureus pneumonia. *Eur. J. Med. Respir*, 15: 514-2425.

Ouédraogo SM, Toloba Y, Badoum G, Ouédraogo G, Boncounou K, Bambara M, Ouédraogo EWM, Zigani A, Sangaré L, Ouédraogo M. 2010. Aspects épidémiocliniques des pneumopathies aiguës bactériennes de l'adulte au CHU Yalgado Ouedraogo. *Mali Med*, Tome XXV (3): 15-18.

Parker D, Prince A. 2012. Immunopathogenesis of Staphylococcus aureus pulmonary infection. *Semin. Immunopathol*, 34: 281-297.

Shahunja KM, Mohammed AS, Tahmeed A, Pradip KB, Shafiqul AS, Hasan A, Abu S-GF, Md Iqbal Ho, Md Munirul I, Sumon KD, Sharifuzzaman A-S, Mohammad S Bin S, Ehsanul H, Mohammad HRS, Mohammad JC. 2014. Bacterial Isolates from Tracheal Aspirates and their Anti-microbial Susceptibility in Mechanically-Ventilated Children with Pneumonia Admitted to an Urban Critical Care Ward. *Bangladesh Crit. Care J*, 2 (2): 60-64.

SNIS. 2013. Annuaire des statistiques sanitaires du Niger, 325 pages. URL: www.snis.cermes.net consulté le 01/02/2014.

Zoumbeye BH. 2014. Pneumonies aiguës communautaires d'allure bactérienne dans le service de pneumo-phtisiologie de l'hôpital national Lamordé de Niamey. Thèse de Med, N0 2187, université Abdou Moumouni de Niamey, Niger, 108 pages.