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Research Article

Susceptibility to Colistin of Multi-Resistant *Pseudomonas aeruginosa* Isolated in Douala Laquintinie Hospital, Cameroon

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Abstract *Pseudomonas aeruginosa* is a germ of hospitalism responsible for nosocomial infections; it is naturally resistant to many antibiotics and has a high susceptibility to the acquisition of acquiring new resistance. The observation of strains highly resistant to antibiotics, has led us to look for possible alternative therapeutics. This study was a descriptive and cross-sectional one, conducted from October 2010 to March 2011. All patients hospitalized for at least 48 hours and showing sign of infection were included after obtaining their consent. Forty nine of 150 samples were positive to the cultivation of *Pseudomonas aeruginosa* showing a prevalence of 32.66%. For the antibiotic susceptibility, we obtain amikacin 57.14%, netilmicin 59.20%, ceftazidime 52.60%, imipenem 33%, colistin 97.95%, and ciprofloxacin 51%. Seven strains were resistant to all antibiotics tested other than colistin. One strain was resistant to colistin. Colistin retains high sensitivity to *Pseudomonas aeruginosa*. However, there are some strains multiresistant to antibiotics.

Keywords *Pseudomonas aeruginosa*; hospital infections; multidrug resistance; colistin

1. Introduction

Pseudomonas aeruginosa is a pathogen responsible for nosocomial infections [6]. These infections are now a major public health problem because of their mortality, morbidity, and associated health care costs. It is still poorly controlled health facilities in the world in general and Cameroon in particular.

Pseudomonas aeruginosa ranks fifth among the species responsible for nosocomial infections in the United States [1]. In Europe, it is ranked second after *Staphylococcus aureus* [1].

Nosocomial infections due to *Pseudomonas aeruginosa* are common and occur for the most part in intensive care unit and particularly in immunocompromised patients.

Despite the various therapeutic advances, mortality due to these infections involving *Pseudomonas aeruginosa* remains high at around 30%. These mortality figures are largely explained partly by the severity of the underlying disease of the patients and secondly by therapeutic difficulties caused by this bacterium [4].

Indeed *Pseudomonas aeruginosa* has a combination of natural resistance mechanisms and is able to acquire multiple mechanisms of drug resistance to antibiotic.

Colistin is an antibiotic fairly used in therapy just because of its difficult handling. This difficulty associated with high toxicity [2, 10] has led to difficulties in laboratory study of this drug.

The mastery of the clinical use of colistin, presented at the forefront it use in therapy. This was in order to find out what could be the contribution of colistin in the treatment of multidrug-resistant *Pseudomonas aeruginosa* that we proposed to conduct this study.

2. Patients and method

2.1. Context and type of study

This was a prospective, descriptive, and cross-sectional study, performed at Douala Laquintinie hospital. Patient recruitment and sample analysis were performed in Douala Laquintinie hospital.

The population size was chosen for convenience, and the method of recruitment was standard for a consecutive period of six months.

2.2. Study population

All patients hospitalized in intensive care, surgery, and medicine units for at least 48 hours, with signs of infection and agreeing to participate in the study, were included.

By consent we excluded from the study anyone who had already been taken for microbiological examination and diagnosed positive to *Pseudomonas aeruginosa*. We also

Table 1: Distribution of the population according to the service and the type of sampling.

Services samples	ICU	Surgery				Medicine	ENT	"Petit payant"	Total
		A	B	C	D				
Urinary	23	1	0	1	10	3	0	2	40
Pus	0	14	2	10	7	3	4	1	41
Deep wound	6	12	12	16	3	3	3	6	61
Septicemia	0	0	1	0	0	0	0	0	1
Brulé serious	3	0	0	0	3	0	1	0	7
Total	32	27	15	27	23	9	8	9	150

ENT: ear, nose, and throat. ICU: intensive care unit.

excluded from the study anyone hospitalized for less than 48 hours, all patients not admitted in the Laquintinie hospital or any person who refused participating in the study.

2.3. Process of data collection

The sampling procedure was based on the original sample.

The skin samples were obtained using a sterile swab moistened with sterile isotonic solution of 0.9 ‰ NaCl; the process involved rotary movements of the brush on the surface to be sampled. Once the sample is obtained, the swab was returned to its protective case, and labeled.

The deep pus was removed by aspiration using a sterile syringe, labeled and sent to the laboratory.

Urine collection was done by puncture through the tube's wall with a sterile syringe and transferred into a sterile cup.

The samples were rapidly transported to the laboratory where they were analyzed.

2.4. Culture and identification

Culture was done on Mac Conkey agar and cetrimide (Bio-Merieux France, Marcy l'Etoile, France). The identification of colonies followed the following scheme: the oxidase test, motility, and study of biochemical Api 20 E (Bio-Merieux France).

2.5. Susceptibility

The antibiotic susceptibility testing was done according to Kirby Bauer method on Muller Hinton II medium (Bio-Merieux France). The sensitivity assessment was done by measuring the inhibition diameters using a caliper and comparing it to the CA SFM reference.

2.6. Statistical analysis

The use of statistical data was done on Microsoft Excel 2007. The statistical test used was the Fischer test with a significance set at .05.

3. Results

We had 150 patients and collected 150 samples. Men were 102 (68%), with a sex ratio of 2.1. The average age of the subjects was 41.5 ± 12.2 years, the extremes being between 8 months and 72 years.

Table 2: Distribution of the population according to the service and the type of sampling.

Service	Frequency	Percentage (%)
Surgery A	14	28.57
Surgery B	3	6.12
Surgery C	10	20.41
Surgery D	13	26.53
ENT	1	5.2
Medicine	2	4.8
Petit payant*	2	4.8
Intensive care unit	4	8.16
Total	49	—

A: traumatology; B: orthopedics; C: digestive visceral; D: urology.
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3.1. Population and sample distribution

Samples were taken from all the services cited above, but the type was characteristic of each service. Of the 150 patients, 32 (21.33%) were from the intensive care unit (UIC), while 8 (5.33%) were from the ear nose and throat (ENT) department.

The majority of samples were from surgery units, and for type, 142 were from deep wound, pus, and urines were directly link to care and long stay to hospital.

3.2. Frequency of isolation

Of the 150 samples, 49 (32.66%) were culture positive for *Pseudomonas aeruginosa*.

3.3. According to the service

According to the service, surgery units bring out more cases of *Pseudomonas aeruginosa* than *others*. Traumatology presents 14 positives cases against 13 for urology.

3.4. Distribution of strains depending on the type of sample

Of 61 cases of skin wound, 30 were culture-positive of *Pseudomonas aeruginosa* while for the 7 "great burned" 6 were positive for *Pseudomonas aeruginosa*. Skin wound was also associated with high susceptibility of *Pseudomonas aeruginosa*.

3.5. Antibiotic susceptibility profile

Four families of antibiotics were tested; netilmicin 59.2% and amikacin 57.14% had presented a good sensitivity in

Table 3: Distribution of strains depending on the type of sample.

Sample type	Number of samples	Number of positive samples	Percentage (%) of positive type specimen
Urine	40	4	10%
Skin wound	61	30	49.2%
Pus deep	41	8	19.5%
Septicemia	1	1	100%
Great burned	7	6	85.7%
Total	150	49	32.66%

Table 4: Sensitivity of *Pseudomonas aeruginosa* antibiotics.

Antibiotics	Number of sensitive strains	Percentage (%)
Cefixime	0	0.00
Cefpodoxime	0	0.00
Ceftazidime	26	52.60
Imipenem	16	33.00
Amikacin	28	57.14
Netilmicin	29	59.20
Gentamicin	20	40.90
Tobramycin	25	51.00
Ofloxacin	21	43.00
Levofloxacin	22	44.90
Ciprofloxacin	25	51.00
Colistin	48	97.95

Table 5: Resistance profiles to aminoglycosides.

Number of strains	Resistance profile
6	Gen^R
3	Tob^R
2	Gen^R-Ami^R
1	Gen^R-Tob^R
1	Gen^R-Tob^R-Ami^R
3	Gen^R-Tob^R-Net^R
16	Gen^R-Tob^R-Ami^R-Net^R
17	Gen^S-Tob^S-Ami^S-Net^S

Gen: gentamicin; Tob: tobramycin; Ami: amikacin; Net: netilmicin; R: resistant.

contrast to the 33% with imipenem. Colistin retained a very high sensitivity with 97.95% against 0.0% for cefpodoxime and cefixime as they are *Pseudomonas aeruginosa* naturally resistant.

3.6. Resistance profile to aminoglycosides

We observed different resistance profiles including Gen^R-Tob^R and Gen^R-Ami^R. The following profiles were not observed: Net^R-Ami^R-Tob^R; Net^R-Ami^R-Gen^R; Net^R-Gen^R-Tob^R. A number of 16 strains were resistant to all aminosides and 17 were sensitive. There are no specific profiles.

3.7. Profile of resistance to quinolones

We observe a specific profile for quinolones tested and ciprofloxacin presents the greatest sensitivity. Elsewhere, there is a high rate of resistance of *Pseudomonas aeruginosa* to quinolones.

Table 6: Profiles of resistance to quinolones.

Number of strains	Resistance profile
2	Ofl^R
3	Lev^R-Ofl^R
23	Cip^R-Ofl^R-Lev^R
21	Cip^S-Ofl^S-Lev^S

Ofl: ofloxacin; Lev: levofloxacin; Cip: ciprofloxacin; R: resistant.

Table 7: Profiles of resistance to aminoglycosides and quinolones.

Number of strains	Resistance profile
3	Gen^R
3	Tob^R
1	Ofl^R
4	Gen^R-Ofl^R
1	Gen^R-Cip^R-Ofl^R-Lev^R
1	Gen^R-Tob^R-Ofl^R-Lev^R
3	Gen^R-Tob^R-Net^R-Cip^R-Ofl^R-Lev^R
1	Gen^R-Tob^R-Cip^R-Ofl^R-Lev^R
1	Gen^R-Tob^R-Ami^R-Cip^R-Ofl^R-Lev^R
13	Tob^R-Ami^R-Net^R-Cip^R-Ofl^R-Lev^R
17	Gen^S-Tob^S-Ami^S-Net^S-Cip^S-Ofl^S-Lev^S

Ofl: ofloxacin; Lev: levofloxacin; Cip: ciprofloxacin; Gen: gentamicin; Tob: tobramycin; Ami: amikacin; Net: netilmicin; R: resistant.

Table 8: Multidrug resistance and colistin.

Number of strains presenting multidrug resistance	20/49
Frequency	41%
Number of multidrug-resistant strains sensitive to colistin	20
Frequency	100%

The multidrug resistance is not associated with resistance to colistine.

3.8. Resistance profile to aminoglycosides and quinolones

Analyzing the multi-resistant profiles, 17 strains were sensitive to all antibiotics tested.

3.9. MDR and colistin

A number of 20 (41%) strains of *Pseudomonas aeruginosa* show resistance to all the families of antibiotics tested except that of polypeptides (see Table 7).

4. Discussion

Our study shows an isolation frequency of *Pseudomonas aeruginosa* in nosocomial infection of 32.66%. This prevalence is highly compared with other data from around the world.

In a report of a national survey of prevalence of nosocomial infections in June 2006 in France, *Pseudomonas aeruginosa* implication was 10% [9]. In another study published in France in 2004, this rate was approximately 30% [6]. This variation could be related to the type of recruitment.

From the point of view of sensitivity to antibiotics, colistin has a sensitivity of about 98%, quinolones around 51% for the most active, imipenem of approximately 33%, ceftazidime 52.6% and aminoglycoside molecule most sensitive of 52.2%. The low sensitivity of the strains to imipenem may be due to excessive use of this molecule link to selection of resistant mutans. There is no typical profile for aminoglycosides. Gentamicin seemed to offer more resistance than amikacin which itself is slightly less sensitive than netilmicin.

In a study conducted in 2001 at the Yaoundé Central Hospital, ceftazidime sensitivity was 71.6% while that of imipenem was 90% [8]. For aminoglycosides given sensitivities were respectively 53.3% for tobramycin and 61.2% for gentamicin [8]. The quinolone ofloxacin had a sensitivity of about 66%.

For Paramythiotou et al. [7], the rate of resistance to ceftazidime and imipenem was 10.5%.

Yang et al. [11], in a study of 75 strains of *Pseudomonas aeruginosa*, stated that six strains isolated from six patients were resistant to all classes of antibiotics including colistin.

Of 20 multidrug-resistant strains obtained, colistin has shown a sensitivity of 100%. But one strain isolated in this study shows a single resistance to colistin. As discussed elsewhere [3,5], we also observed that multidrug resistance of *Pseudomonas aeruginosa* is not linked to resistance to colistin in our context.

5. Conclusion

Pseudomonas aeruginosa is highly involved in nosocomial infections at the Laquintinie hospital. Multidrug resistance remains a major phenomenon. Colistin still retains a high sensitivity and could therefore be used as a therapeutic alternative in case of multidrug resistance.

References

[1] J. D. Edgeworth, D. F. Treacher, and S. J. Eykyn, *A 25-year study of nosocomial bacteremia in an adult intensive care unit*, Crit Care Med, 27 (1999), 1421–1428.

[2] M. E. Falagas and S. K. Kasiakou, *Colistin: the revival of polymyxins for the management of multidrug-resistant gram-negative bacterial infections*, Clin Infect Dis, 40 (2005), 1333–1341.

[3] H. K. Johansen, S. M. Moskowitz, O. Ciofu, T. Pressler, and N. Høiby, *Spread of colistin resistant non-mucoid Pseudomonas aeruginosa among chronically infected Danish cystic fibrosis patients*, J Cyst Fibros, 7 (2008), 391–397.

[4] A. Kuikka and V. V. Valtonen, *Factors associated with improved outcome of Pseudomonas aeruginosa bacteremia in a Finnish university hospital*, Eur J Clin Microbiol Infect Dis, 17 (1998), 701–708.

[5] A. Mérens, C. Llanes, C. Pourcel, M. Roussel-Delvallez, G. Vergnaud, and J. D. Cavallo, *Resistance and epidemiology of Pseudomonas aeruginosa isolated from cystic fibrosis patients: a French multi-centre study*, in 19th European Congress of Clinical Microbiology and Infectious Diseases (ECCMID), Helsinki, Finland, 2009.

[6] G. B. Par and R. Ramphal, *Pseudomonas aeruginosa*, in Principles and Practice of Infectious Diseases, G. L. Mandell, J. E. Bennett, and R. Dolin, eds., Elsevier Churchill Livingstone, Philadelphia, PA, 2005, 2587–2615.

[7] E. Paramythiotou, J. C. Lucet, J. F. Timsit, D. Vanjak, C. Paugam-Burtz, J. L. Trouillet, et al., *Acquisition of multidrug-resistant Pseudomonas aeruginosa in patients in intensive care units: role of antibiotics with antipseudomonal activity*, Clin Infect Dis, 38 (2004), 670–677.

[8] J. G. Piéboji, S. Koulla-Shiro, P. Ngassam, D. Adiogo, T. Njine, and P. Ndumbe, *Antimicrobial resistance of Gram-negative bacilli isolates from inpatients and outpatients at Yaounde Central Hospital, Cameroon*, Int J Infect Dis, 8 (2004), 147–154.

[9] J. M. Thiolet, L. Lacavé, H. Tronel, P. Jarno, M. H. Metzger, F. L'Hériteau, et al., *Enquête nationale de prévalence des infections nosocomiales 2006*, Bull Epidemiol Hebd, (2007), 429–432.

[10] E. Wolinsky and J. D. Hines, *Neurotoxic and nephrotoxic effects of colistin patients with renal disease*, N Engl J Med, 266 (1962), 759–762.

[11] K. Yang, H. Zhuo, B. J. Guglielmo, and J. Wiener-Kronish, *Multidrug-resistant Pseudomonas aeruginosa ventilator-associated pneumonia: the role of endotracheal aspirate surveillance cultures*, Ann Pharmacother, 43 (2009), 28–35.