Journal of Biology & Pharmacy and Agricultural Management

RESEARCH ARTICLE

Journal of Biology & Pharmacy and Agricultural Management, v. 14., n. 2, abr/jun 2018

ISSN 1983-4209 revista.uepb.edu.br/index.php/biofarm

INCIDENCE OF NOSOCOMIAL INFECTIONS BY GRAM-NEGATIVE NON-FERMENTERS AND PROFILE OF ANTIMICROBIAL SENSITIVITY IN A HOSPITAL IN CAMPINA GRANDE-PB

Patricia Maria de Freitas e Silva[1], *Ricardo Jefferson Pereira Feitosa*[1], *Mayara Pimentel Couto*[1], *Heronides dos Santos Pereira*[1], *Raïssa Mayer Ramalho Catão*[1], *Lauro Santos Filho*[2]

[1]Departamento de Farmácia, Universidade Estadual da Paraíba, Campina Grande-PB, Brasil.
[2]Departamento de Ciências Farmacêuticas, Universidade Federal da Paraíba, João Pessoa – PB- Brasil
*Corresponding author. E-mail address: patriciaftreitashemoiba@yahoo.com.br

ABSTRACT

The non-fermenting bacilli (BGN-NF) are frequent hospital microorganisms and cause disease by infecting colonization in immunocompromised patients or when they reach sterile body sites through invasive hospital procedures such as introduction of catheters and ventilators, are therefore considered opportunistic bacteria. This work aimed study the epidemiology and antimicrobial resistance of BGN-NF, isolated from infectious process of patients admitted in a public hospital in Campina Grande-PB, in the period April 2009 to March 2011. Of the 1,056 patients analyzed cultures from different clinical specimens, 358 (33.9 %) presented bacterial growth and were classified as cases of community infections (IC) or hospital infections (IH), according to the criteria established by the Law 2616/98 from the Brazilian Ministry of Health, noting 52 (15%) cases of infection by rods BGN-NF. Among these, *Pseudomonas aeruginosa* was most prevalent species with 44 (84.6%) of insulation,especially also for high resistance to antimicrobials. The largest number of *P. aeruginosa*, was detected in secretions collected orotracheal tube with 9 (26.4%) cases, with prevalence in patients above 60 years. Among the hospital enrironments where these microorganisms were more prevalent, the intensive care unit, with 44.2% of cases. There was a high resistance of strains of *P. aeruginosa* from most of the antimicrobials tested. Among those who showed good antimicrobial activity, stood out: imipenen with 2 (5.8%) and ciprofloxacin and amikacin 7 (20.5%) with drug-resistant strains. Not observed difference in resistance between nosocomial and/or communitary strains.

Keywords: Non-fermenter, Hospital Infection, Antibiotic, Resistance.

INCIDÊNCIA DE INFECÇÕES HOSPITALARES POR GRAM-NEGATIVOS NÃO-FERMENTADORES E PERFIL DE SENSIBILIDADE AOS ANTIMICROBIANOS EM UM HOSPITAL EM CAMPINA GRANDE-PB

RESUMO

Bacilos não fermentadores (BGN-NF) são micro-organismos frequentes em hospitais e podem causar doenças infectando pacientes imuocomprometidos ou ainda quando

Journal of Biology & Pharmacy and Agricultural Management

RESEARCH ARTICLE

Journal of Biology & Pharmacy and Agricultural Management, v. 14., n. 2, abr/jun 2018

ISSN 1983-4209 revista.uepb.edu.br/index.php/biofarm

alcançam locais estéreis do corpo através de procedimentos hospitalares invasivos como introdução de cateteres e ventiladores, sendo, portanto, consideradas bactérias oportunistas. Este trabalho objetivou estudar a epidemiologia e resistência aos antimicrobianos dos BGN-NF, isolados de processos infecciosos de pacientes internados, em um hospital da rede pública de Campina Grande-PB, no período de abril de 2009 a março de 2011. Dos 1.056 resultados de culturas analisadas, de diferentes espécimes clínicos, 358 (33,9%) apresentaram crescimento e foram classificados como casos de infecções comunitárias (IC) ou hospitalares (IH), de acordo com os critérios estabelecidos pela Lei 2616/98 do Ministério da Saúde, constatando-se 52 (15%) casos de infecção por bastonetes BGN-NF. Dentre estes, *Pseudomonas aeruginosa* foi a espécie mais prevalente com 44 (84,6%) dos isolamentos, destacando-se também pela elevada resistência aos antimicrobianos. O maior número de isolamentos de *P. aeruginosa*, foi detectado nas secreções coletadas do tubo orotraqueal com 9 (26,4%) casos, com predomínio em pacientes acima de 60 anos. Dentre os ambientes hospitalares onde estes micro-organismos foram mais prevalentes, destacou-se a unidade de terapia intensiva, com 44,2% dos casos. Observou-se elevada resistência das cepas de *P. aeruginosa* frente à maioria dos antimicrobianos testados. Dentre os que apresentaram boa atividade antimicrobiana, destacaram-se: impenem com 2 (5,8%) e ciprofloxacina e amicacina com 7 (20,5%) de cepas resistentes. Não se observou diferença no padrão de resistência entre as linhagens nosocomiais e/ou comunitárias.

Palavras-chave: Não fermetador, infecção nosocomial, antibióticos, resistência.

1. INTRODUCTION

The Gram-negative glucose non-fermenting bacilli (BGN-NF) are widely distributed in the environment and can be found in soil, water, food and disinfecting solutions (ANVISA, 2011). In hospitals, they are more frequently isolated from water tap, respirators, suction catheters, antiseptics, serums, sinks, medical equipment and health professionals (PITTET et al., 1999).

They are responsable for serious infections in hospitals. These bacteria can colonize and cause disease after infecting immunocompromised patients or when gain access to sites normally sterile in body through hospital procedures such as invasive introduction of catheters and respirators. They are considered opportunistic micro-organisms (Volk et al., 1999), however, some members are recognized as nosocomial pathogens, with significant role in colonization and infection of patients admitted to hospitals, causing serious complications that can progress to septicemia (FIGUEIREDO et al., 2009; ZANOL; PICOLI; MORSCH, 2010).

They BGN-NF are unable to use carbohydrates as an energy source by fermentation, degrading them by oxidative pathway (MURRAY et al., 2003; SANTOS, 2006). Due to the low metabolic activity compared to enterobacteria, identification becomes more complex, so morphologic, macroscopic and microscopic carachteristics are essential for the identification process that also requires special tests (Silva Filho et al., 2007).

The phenotypic methods (SANTOS, 2006) have low discriminatory power when compared to molecular typing methods (ZANOL; PICOLI; MORSCH, 2010; WOLSKA;

Journal of Biology & Pharmacy and Agricultural Management

RESEARCH ARTICLE

Journal of Biology & Pharmacy and Agricultural Management, v. 14., n. 2, abr/jun 2018

ISSN 1983-4209 revista.uepb.edu.br/index.php/biofarm

JAKUBCAK, 2012), so that phylogenetic studies based on 16S rRNA sequence led to numerous changes in the classification and nomenclature of this group of microorganisms in recent years.

Among the major emerging non fermenting bacteria are: *Pseudomonas, Acinetobacter, Burkholderia, Stenotrophomonas and Chryseobacterium* (BROOKE, 2012). *Pseudomonas aeruginosa* and *Acinetobacter baumannii* pathogens are considered ubiquitous and highly prevalent in hospital settings (VOLK et al., 1999) and are often isolated from blood cultures and samples of hospital patients respiratory tract (DELIBERALI et al., 2011).

The BGN-NF also have high resistance to several antimicrobial agents (FIGUEIREDO et al, 2009; UMADEVI et al., 2011) and are capable of causing serious infections, especially in patients of intensive care units (ICU), undergoing invasive procedures, burned patients and those with cystic fibrosis (CARVALHO; GONTIJO FILHO, 2008; SiLva FILHO et al., 2007).

Among the Gram-negative bacilli isolated from clinical samples, 68-78% are members of the *Enterobacteriaceae* rods while 12-16% are BGN-NF. However, even at lower percentages, these micro-organisms considered as opportunistic, may become severe agents of hospital infections, mainly due to the high level of resistance to antimicrobials commonly used in these environments (MURRAY et al., 2003).

This study aimed studying retrospectively the epidemiology and antimicrobial resistance of Gram-negative glucose non-fermenting (BGN-NF), isolated from nosocomial infections and empiric antibiotic therapy prescribed in a hospital in Campina Grande-PB (Brazil).

2. MATERIALS AND METHODS

2.1 Universe Characterization

The research happened at a hospital in Campina Grande - PB, through a retrospective and quantitative approach. Data were collected from patient formularies and from the hospital microbiology laboratory information, from April 2009 to March 2011.

2.2 Sampling and Methodology

We analyzed the results of 1,056 cultures. Bacterial growths were found in 358 of these cultures from different clinical specimens and were classified as hospital or community-acquired infections, according to the criteria established by the Brazilian law 2616/98 (BRASIL, 1998). Clinical specimens used were: urine, secretions, catheter tip and tracheal tube from patients in different wards of the hospital. Culture media used were blood agar, mannitol salt agar and Eosin Methylene Blue Agar (EMB). The microorganisms were initially identified based on their morphotinctorial features and biochemical use of sugars through the Triple Sugar Iron Agar (TSI). A sign of a non-fermenting bacteria presence was growth in the mentioned medium without change in its original color (SADER et al., 2001).

Journal of Biology & Pharmacy and Agricultural Management

RESEARCH ARTICLE

Journal of Biology & Pharmacy and Agricultural Management, v. 14., n. 2, abr/jun 2018

ISSN 1983-4209 revista.uepb.edu.br/index.php/biofarm

In these cases, additional biochemical tests were used for the identification of NF-BGN, according to the kit for non fermenters used (Probac®) which tested bacterial motility, oxidase, glucose utilization in base medium for oxidation/fermentation (OF), decarboxylation of lysine and arginine, liquefaction of gelatin, urea hydrolysis, DNAse, sensitivity to polymyxin and observation of pigment production. Bacterial identification was made by reading the product numbering system. Verification of the microorganisms susceptibility to antimicrobials (Cefar®) was performed using antibiogram, by disk diffusion method (CLSI, 2006).

2.3 Ethical considerations

This study was approved by the Ethics Committee of the State University of Paraíba (UEPB) by the number: 0020.0.133.000-12.

3. RESULTS AND DISCUSSION

From 1056 bacterial cultures analyzed in this study, 358 showed bacterial growth. 52 (15%) isolated were NF BNG-glucose, 59 (16%) Gram positive cocci and 247 (69%) were Gram-negative fermenting bacilli - Enterobacteria.

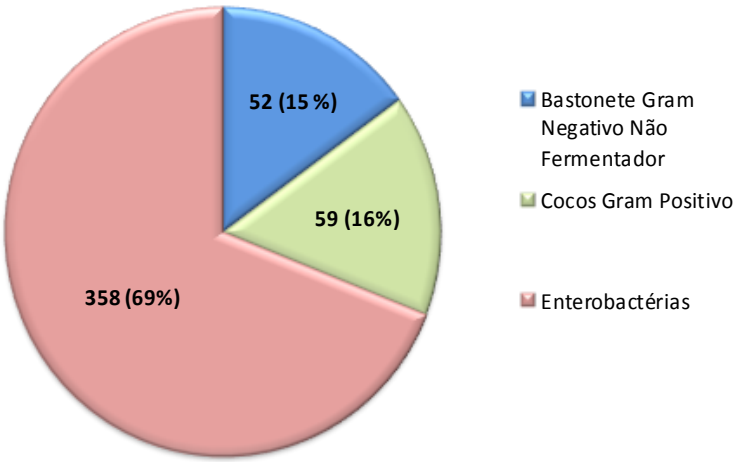


Figure 1: Distribution of bacterial groups isolated from patients cultures at a hospital in Campina Grande-PB, from April 2009 to March 2011

The NF-BGN percentage (15%) in this study was considered high when compared to other authors who found frequencies of 9.4% and 2.18% of NF-BGN in similar studies performed in Brazil (DELIBERALI et al., 2011; MENEZES et al., 2004). The high number of positive cultures for BGN-NF should be an alert to health professionals, because the great presence of these microorganisms reveals the indiscriminate use of antimicrobials in a hospital. However, several authors (MARQUES et al., 2007; PELLEGRINO et al., 2002; SADER et al., 2001; VOLK et al., 1999) emphasize that the prevalence of these isolates

Journal of Biology & Pharmacy and Agricultural Management

RESEARCH ARTICLE

Journal of Biology & Pharmacy and Agricultural Management, v. 14., n. 2, abr/jun 2018

ISSN 1983-4209 revista.uepb.edu.br/index.php/biofarm

can range from one hospital to another and from one study to another within the same hospital.

From the 52 NF-BGN strains found in the hospital studied, 38 (70%) had nosocomial origin (IH) and 16 (30%) were from the community (IC), according to the criteria established by the Brazilian law (BRASIL, 1998).

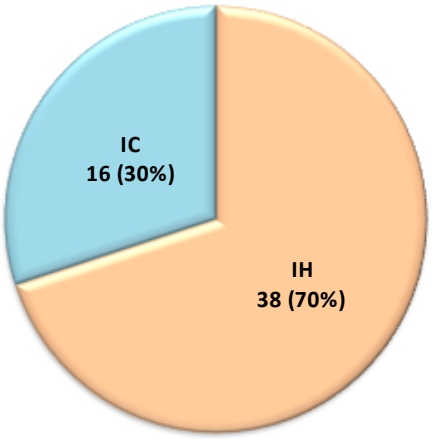


Figure 2: Classification of infections by Gram negative non-fermenters bacilli

Table 1 shows the distribution by age group of patients with infections caused by NF-BNG. The higher frequency of infections occurred in patients above 60 years old, 16 (42.1%). This fact may be related to several factors as physiological changes of aging, decline in immune response and invasive procedures.

Table 1: Distribution by age group of patients with nosocomial infections (IH) caused by Gram negative non-fermenters bacilli (NF-BGN)

Age group of patients with IH	Isolations NF - BGN	
	n	%
0 – 1 year	3	7.8
> 1 – 30 years	2	5.2
> 30 – 60 years	13	34.2
> 60 years	16	42.1
Not informed	4	10.5
Total	38	100.0

Source: Archives of Microbiology Laboratory of the Studied - April 2009 to March 2011.

Rarely *P. aeruginosa* as well as other NF-BGN cause community-acquired infections in healthy individuals infected. In most cases, these bacteria infect individuals with weakened immune system. They are considered opportunistic pathogen, which explains its high prevalence in patients with advanced age.

Journal of Biology & Pharmacy and Agricultural Management

RESEARCH ARTICLE

Journal of Biology & Pharmacy and Agricultural Management, v. 14., n. 2, abr/jun 2018

ISSN 1983-4209 revista.uepb.edu.br/index.php/biofarm

Table 2 demonstrates the diversity of NF-BGN isolated in different hospital departments, with a higher frequency of *P. aeruginosa*, 44 (84.6%). Of these, 34 (77%) had nosocomial origin. The Unidade Carea Unit (ICU) showed the largest number of patients with BGN-NF infection, with 21 (47%) isolates of *P. aeruginosa*. Other areas like oncology and surgical wards, were also considered critical areas respectively with 11 (25%) and 7 (15%) isolates. Just one *Acinetobacter baumannii* was isolated in this study from a community origin patient in the oncology area. Other NF-BGN identified were *Burkholderia cepacia* 3 (5.7%), *Chryseobacterium indologenes* 2 (3.8%), *Flavobacterium oryzihabitaus* 1 (1.9%) and *B. pickett,* 1 (1.9%).

Table 2: Epidemiology of infections caused by Gram negative non-fermenters in inpatients in the hospital departments

Microorganisms	Hospital Departaments											
	ICU		ONC		ALA-C		AMB		PED		TOTAL	
	IC	IH	IC	IH	IC	IH	IC	IH	IC	IH	N	%
Pseudomonas aeruginosas	3 5,8%	18 34,6%	4 7,7%	7 13,5%	0	7 13,5%	3 5,8%	0	0	2 3,8%	44	84,6
Burkholderia cepacia	0	1 1,9%	1 1,9%	1 1,9%	0	0	0	0	0	0	3	5,8
Chryseobacterium indologenes	0	1 1,9%	0	0	0	1 1,9%	0	0	0	0	2	3,8
Acinetobacter baumannii	0	0	1 1,9%	0	0	0	0	0	0	0	1	1,9
Flavobacterium oryzihabitaus	0	0	0	0	0	0	1 1,9%	0	0	0	1	1,9
Burkholderia picketti	0	0	1 1,9%	0	0	0	0	0	0	0	1	1,9
Total	**23 (44,2%)**		**15 (28,8%)**		**8 (15,4%)**		**4 (7,7%)**		**2 (3,8)**		**52**	**100,0**

Source: Archives of Microbiology Laboratory of the Hospital of April 2009 to March 2011. Legend: ICU: Intensive care unit; ONC: Oncology; ALA-C: Surgical Ward; AMB: Outpatient; PED: Pediatrics, IC: Infections Community, IH: Infections Hospital.

Table 3 lists the origin of the clinical specimens in which *P. aeruginosa* were isolated from hospital origin, confirming that the largest number of isolates were secretions from the tracheal tube 9 (26.4%), followed by urines 7 (20.5%) secretions from wounds 6 (16.2%) and surgical wound secretions 3 (8.8%). These results are in agreement with literature reports (BRITO et al, 2003; CARVALHO; GONTIJO FILHO, 2008; HARINGER, 2009; LISBOA et al., 2007).

Journal of Biology & Pharmacy and Agricultural Management

RESEARCH ARTICLE

Journal of Biology & Pharmacy and Agricultural Management, v. 14., n. 2, abr/jun 2018

ISSN 1983-4209 revista.uepb.edu.br/index.php/biofarm

Table 3: Biological specimens used for the isolation of nosocomial *Pseudomonas aeruginosa*

Origin of the material analysed	Samples	
	n	(%)
Endotraqueal tube	9	26,4
Urina	7	20,5
Wound secretion	6	16,2
Surgical wound	3	8,8
Chest drain	2	5,8
Abdominal fistula	2	5,8
Lung secretion	1	2,9
Catheter tip	1	2,9
Hemoculture	1	2,9
Others	2	5,8
Total	**34**	**100,0**

Source: Archives of Microbiology Laboratory of the Studied Hospital - April 2009 to March 2011.

The high number of *P. aeruginosa* in the endothracheal tube occured, probably, because it can colonize the surface of the respiratory tract, releasing proteases that break the mucus mesh rich in fibronectin and exposing the receptors where the bacteria fimbriae bind. Moreover, damaged or irritated tissue due to invasive hospital procedures, facilitates the colonization process of *Pseudomonas* (HARINGER, 2009).

The secretion culture of endotracheal tube was used to predict the development of nosocomial pneumonia in patients with endotracheal breathing, as well as with other patients parameters like chest X-ray. Data used in this study were obtained from the results of cultures count with more than 10^6 colony units formation (CFU), which was the cut to dismiss the possibility of tracheal colonization, characterizing pulmonary infection (HARINGER, 2009). Several authors (CALDERÓN et al., 2011, CARVALHO; GONTIJO, 2008, MARCONI et al, 2008) considers pneumonia as the major cause of nosocomial infection in ICU patients, especially when associated with mechanical ventilation (HARINGER, 2009).

Table 4 shows the susceptibility profile of NF-BGN antimicrobials. Meropenem was the most effective drug against these microorganisms, followed by imipenem. These data agree with the literature reports as to carbapenems being considered the antimicrobial therapy of choice for treatment of serious hospital infections caused by NF-BGN (GALES et al., 2008).

Journal of Biology & Pharmacy and Agricultural Management

RESEARCH ARTICLE

Journal of Biology & Pharmacy and Agricultural Management, v. 14., n. 2, abr/jun 2018

ISSN 1983-4209 revista.uepb.edu.br/index.php/biofarm

Table 4: Profile of antimicrobial resistance of gram-negative bacilli isolated in this study

Antimicrobials	Microorganisms											
	Pseudomonas aeruginosas n=34		*Burkoldera cepacia* n=2		*Chyseobacteriu idologenes* n=2		*Escherichia coli* n=45		*Klebisiella* spp n=20		*Enterobacter* spp n=12	
	n	%	n	%	n	%	n	%	n	%	n	%
AM	7	20,5	0	0	1	50,0	3	6,6	2	10,0	1	8,3
AZ	24	70,5	1	50,0	1	50,0	12	26,6	12	60,0	9	75,0
CE	34	100	1	50,0	1	50,0	21	46,6	13	65,0	11	91,6
CT	34	100	1	50,0	1	50,0	25	55,5	18	90,0	11	91,6
CP	7	20,5	1	50,0	1	50,0	22	48,8	14	70,0	7	58,3
CL	34	100	1	50,0	1	50,0	12	26,6	12	60,0	10	83,3
GE	15	44,1	0	0	1	50,0	15	33,3	12	60,0	8	66,6
TT	34	100	1	50,0	1	50,0	34	75,5	17	85,0	7	58,3
SU	34	100	2	100	1	50,0	31	68,8	17	85,0	9	75,0
AC	34	100	1	50,0	1	50,0	17	37,7	14	70,0	10	83,3
AP	34	100	1	50,0	2	100	37	82,2	17	85,0	12	100
CM	18	52,9	1	50,0	1	50,0	15	33,3	12	60,0	11	91,6
IM	2	5,8	1	50,0	1	50,0	0	0	1	5,0	1	8,3
CF	33	97,0	1	50,0	1	50,0	16	35,5	14	70,0	8	66,6
TO	9	26,4	1	50,0	1	50,0	16	35,5	14	70,0	8	66,6
ME	0	0	0	0	0	0	0	0	0	0	1	8,3
CZ	29	85,2	1	50,0	1	50,0	10	22,2	14	70,0	12	100
CX	34	100	1	50,0	1	50,0	6	13,3	13	65,0	8	66,6

Legend: AM: Amikacin, AZ: Aztreonam; EC: Cephalexin, CT: Cephalothin, CP: Ciprofloxacin; CL: Chloramphenicol; GE: Gentamicin; TT: Tetracycline; SU: Trimethoprim-sulfamethoxazole; AC: Amoxicillin-Ac. Clavulônico; AP: Ampicillin, CM: Cefepime, CZ: Ceftazidime, IM: Imipenem, CF: Ceftriaxone, TO: Tobramycin, ME: Meropenem, CX: Cefoxitin.

Journal of Biology & Pharmacy and Agricultural Management

RESEARCH ARTICLE

Journal of Biology & Pharmacy and Agricultural Management, v. 14., n. 2, abr/jun 2018

ISSN 1983-4209 revista.uepb.edu.br/index.php/biofarm

The other antimicrobials tested had low or no efficacy. It was observed that 100% of the strains were resistant to amoxacilin + clavulanic acid, ampicillin, cephalexin, cephalothin, cefoxitin, chloramphenicol, sulfamethoxazole-trimethoprim and tetracycline. High percentage of resistant strains, was also found in relation to ceftriaxone 33 (97%), ceftazidime 29 (85.2%), aztreonam 24 (70.5%), cefepime 18 (52.9%) and gentamicin 15 (44,1%). Among the antimicrobials tested against *P. aeruginosa*, ciprofloxacin showed 7(20.5%) resistant strains, behavior similar to aminoglycosides: amikacin and tobramycin, with only 7 (20.5%) and 9 (26.4%) of resistant strains, respectively. However, it was observed higher percentage of resistance in relation to gentamicin with 15 (44.1%) resistant strains. These results are in agreement with others reports (SADER et al., 2001; LÓPEZ-ROJAS et al., 2011) which also showed similar behavior, although with higher percentages: 36.1% and 43.7% respectively for amikacin, and gentamicin (Table 4).

There was some discrepancy in the behavior of two (5.8%) strains of *P. aeruginosa* that were resistant to imipenem, but were susceptible to meropenem. This difference between the two carbapenems was repeated for the other NF-BGN. These data are in agreement with reports of other authors (Gales et al., 2002) who affirm that meropenem has better activity than imipenem against *P. aeruginosa*. However, considering that they are similar drugs, it is important that such discrepancy be confirmed by sending the strains to research accredited agencies (MARQUES et al., 2007).

P. aeruginosa resistant to carbapenems have been detected in several parts of the world and its control is urgent, since they are the drugs of choice for treatment of infections caused by this bacteria (GALES et al., 2002; FIGUEIREDO et al., 2009; PEÑA et al., 2012). The frequency of resistant strains to imipenem (5.8%) in this study was much lower than those documented in other studies that found rates of 14% to 38.34% in some hospitals (PELLEGRINO et al., 2002; TUMITN & PIZZOLITTO, 2003; KALAI et al., 2004). Despite the good efficacy of imipenem in the treatment of infections caused by *P. aeruginosa*, there is a possible risk of resistance during the therapy using this drug (GALES et al., 2002).

The clinical importance of infection by *P. aeruginosa* is characterized by the expression of multiple antibiotic resistance associated with a difficult disease eradication, with high morbidity and mortality (NEVES et al., 2011).

Amikacin, meropenem and gentamicin were the most effective antibiotics against the two strains of *Burkolderia cepacia* and *Chryseobacterium idologene*. Meropenem was the only drug in which the strains of non-fermenting Gram-negative bacilli showed no resistance. Considering the small number of isolates (n = 2) of the *B.cepaciae* and *Chryseobacterium idologene*, it was not possible to have a good analysis of the level of resistance of these species (Table 4). Regarding the behavior of *C. idologene*, uncommon human pathogen (CALDERÓN et al., 2011).

It was also found that 20.5% of *P. aeruginosa* strains were resistant to ciprofloxacin. However, this value can be considered low compared to other studies where authors (MENEZES et al., 2004; PAVIANE; STADNIK; HEINEK, 2004) found 58.0% and 37.0% of resistant strains.

Imipenem has good efficacy in the treatment of infections caused by *P. aeruginosa*, despite the risk of resistance during treatment (GALES et al., 2002). The percentage of strains resistant to imipenem (5.8%) found in this study was much lower than those

Journal of Biology & Pharmacy and Agricultural Management

RESEARCH ARTICLE

Journal of Biology & Pharmacy and Agricultural Management, v. 14., n. 2, abr/jun 2018

ISSN 1983-4209 revista.uepb.edu.br/index.php/biofarm

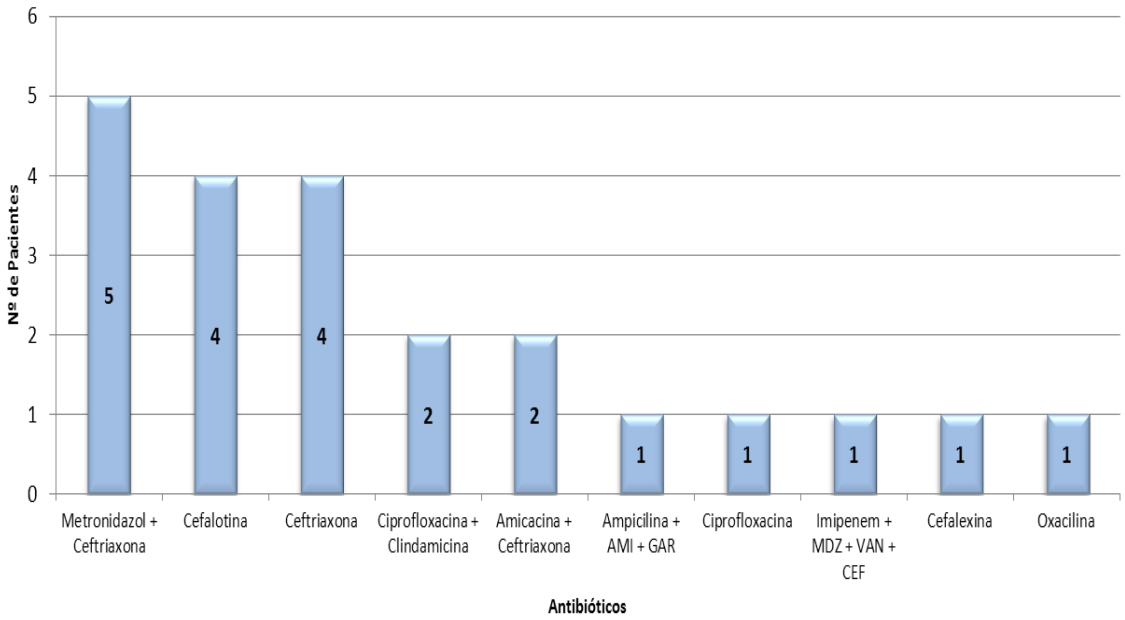
reported by other authors who found values between 14% and 20.4% (TUMITAN; PIZZOLITTO, 2003; Kalaii et al., 2004; TAKAGI et al., 2009).

All *P. aeruginosa* were resistant to cefoxitin, cefalexin and cephalothin (Table 4). Reports of first and second generation resistant cephalosporins strains are common since *P. aeruginosa* has intrinsic resistance to these antimicrobials (SADER et al., 2001; Peña et al., 2012). We also observed high percentages of third generation cephalosporins resistant strains: 29 (85.2%) resistance strains to ceftazidime and 33 (97%) resistant strains to ceftriaxone, cephepime, a fourth generation cephalosporin, was the less resistant drug against *P. aeruginosa*, 18 (52.9%). Other authors (PELLEGRINO et al., 2002; LEISER; TOGNIM; BEBENDO, 2007; PIRES et al., 2009) found the same inversion of resistance as to cephalosporins generations.

Resistance to ceftriaxone, ceftazidime and cefepime were also reported in different hospitals (SADER et al., 2001; PIRES et al., 2009; MARQUES et al., 2007; LEISER; TOGNIM; BEBENDO, 2007), but at levels below the percentage found in this study. Considering these results, it is evident that *P. aeruginosa* has high capacity of developing different mechanisms of resistance to cephalosporins.

Figure 3 shows the antimicrobials that were being used by patients before the microbiological cultures. From the 44 cases of *P. aeruginosa* infection, 22 (50%) patients were already using empirical treatment, with some type of antimicrobial prescribed by the doctors of the hospital. It was found that the most commonly prescribed antibiotic was the association between ceftriaxone and metronidazole (5 cases), followed by cephalothin and ceftriaxone (4 cases each).

Figure 3: Antibiotics used by medical prescription before the identification of *P. aeruginosa*



Source: Archives of Microbiology Laboratory of the Studied Hospital - April 2009 to March 2011

Journal of Biology & Pharmacy and Agricultural Management, v. 14., n. 2, abr/jun 2018
ISSN 1983-4209 revista.uepb.edu.br/index.php/biofarm

It was found that all patients who were being treated empirically with cephalothin, and cephalexin had resistant strains to these antimicrobials, considering the three patients who received ciprofloxacin alone or in combination, one demonstrated resistant strains to this antimicrobial. From the 12 patients who used ceftriaxone, either in combination with other antimicrobials, 8 (66.6%) also showed drug-resistant strains to this antimicrobial. These data suggest therapeutic failures and/or development of resistance during antibiotic therapy.

Metronidazole, was prescribed for patients with infections caused by *Pseudomonas aeruginosa*, it wasn´t a good antimicrobial choice, since metronidazole has antibacterial activity against Gram-positive sporulated microorganisms and anaerobic ones, not against *Pseudomonas*. Likewise, clindamycin and oxacillin have better indication for Gram-positive and Gram-negative treatment, never for *P. aeruginosa.*

4. CONCLUSIONS

The high number of antimicrobials associations has been observed in some cases, for example, the association between imipenem, metronidazole, vancomycin and ceftriaxone used to treat one patient. In this case, therapy might have been effective due to the use of imipenem. It is relevant to note that imipenem should be the last choice therapy in a hospital setting to prevent the emergence of resistant strains. So the routine cultures with antibiogram should be mandatory, thus aiming effective and less harmful therapeutic choices to the hospital epidemiology.

P. aeruginosa demonstrated high resistance to antimicrobials used in hospital routine, showing, however, good sensitivity only to carbapenems (imipenem and meropenem), followed by ciprofloxacin and amikacin.

Considering the fact that several antimicrobials used empirically in routine hospital were not suitable for the treatment of patients with nosocomial infection, it is evident the need for standardization and routine performance of tests for bacterial identification and antibiogram, to ensure therapeutic success and increased patient survival, helping to ensure the rational use of antimicrobials in the hospital.

REFERENCES

ANVISA - Agência Nacional de Vigilância Sanitária. *Manual de Microbiologia Clínica Para o Controle de Infecção em Serviços de Saúde.* Disponível em http://bvsms.saude.gov.br/bvs/publicacoes/manual_microbiologia_completo.pdf. Accessed: December 1, 2011.

BRASIL (1998). Ministério da Saúde. Diretriz e Normas para a Prevenção e Controle das Infecções Hospitalares. Portaria Nº 2.616, de 12 de maio de 1998.

BRITO, D.V.D; OLIVEIRA, E.J; DARINI, A L.C; ABDALLAH, V.O.S; GONTIJO-FILHO, P.P. (2003). Nosocomial outbreaks due to *Pseudomonas aeruginosa* and A*cinetobacter baumannii* in a neonatal intensive care unit (nicu) of the Uberlândia Federal University Hospital Brazilian. *J. Microbiol.* 34 (1), 27-28.

Journal of Biology & Pharmacy and Agricultural Management, v. 14., n. 2, abr/jun 2018
ISSN 1983-4209 revista.uepb.edu.br/index.php/biofarm

BROOKE, J, S. (2012). *Stenotrophomonas maltophilia*: an Emerging Global Opportunistic Pathogen. *Clin. Microbiol. Rev*. 25:2-41. doi: 10.1128/CMR.00019-11

CALDERÓN, G; GARCÍA, E; ROJAS, P; GARCÍA, E; ROSSO, M; LOSADA, A. (2011). *Chryseobacterium* indologenes infection in a newborn: a case report. *J. Med. Case. Reports.* 5**:**10. doi:10.1186/1752-1947-5-10.

CARVALHO, R.H; GONTIJO FILHO, P.P. (2008). Epidemiologically relevant antimicrobial resistance phenotypes in Pathogens isolated from critically ill patients in a brazilian universitary hospital. *Braz. J. Microbiol.* 39:623-630

Clinical and Laboratory Standads Institute - CLSI (2006). Performance standards for antimicrobial susceptibility testing. Sixteenth informational supplement. CLSI document M100-S16. Wayne, PA, USA.

DELIBERALI, B; MYIAMOTO, K.N; WINCKLER NETO, C.H.D.P; PULCINELLI, R.S.R; AQUINO, A.R.C; VIZZOTTO, B. S; Santos, R. C. V.(2011). Prevalência de bacilos Gram-negativos não fermentadores de pacientes internados em Porto Alegre-RS. *J. Bras. Patol. Med. Lab.* 47 (5), 529-534.

FIGUEIREDO, D.Q; CASTRO, L.F.S; SANTOS, K. R. N; TEIXEITA, L. M; MONDINO, S. S. B Detecção de metalo-beta-lactamases em amostras hospitalares de *Pseudomonas aeruginosa* e *Acinetobacter baumannii.* (2009.) *J. Bras.Patol. Med. Lab.* 45(3), 177-184.

GALES, A.C; MENDES, R. E; RODRIGUES, J; SADER, H.S. (2002). Comparação das atividades antimicrobianas de meropenem e imipenem/celastatina: o laboratório necessita testar os dois antimicrobianos? *J. Bras. Patol. Med. Labor*. 38 (1), 13-20.

HARINGER, D.M.C.(2009). Pneumonia associada a ventilação mecânica. *Pulmão.* Supl 2:S37-S45

LEISER,J.J; TOGNIM, M.C.B; BEDENDO,J.(2007).Infecções hospitalares em um centro de terapia intensiva de um hospital de ensino no norte do Paraná. *Ciênc. Cuid. Saúde.* 6(2),181-186.

LÓPEZ-ROJAS, R; JIMENÉZ-MEIFAS, M.E; LEPE, J.A; PACHÓN J. (2011). *J. Infect. Dis.* 204 (7); 1147-1148. doi: 10.1093/infdis/jir476v

LISBOA, T; FARIA, M; HOHER, J.A; BORGES, L A; GÓMEZ, J; SCHIFELBAIN, L; DIAS, F. S; LISBOA, J; FRIEDMAN, G. (2007). Prevalência de infecção nosocomial em Unidades de Terapia Intensiva do Rio Grande do Sul. *Rev. Bras. Ter. Intensiva*.19(4),414-20.

KALAIL, S; JOiauhai, W; MAHJOUBI, F; GHozzi, R; THABET, L; BEM REJEB, S; HAMMAMI, A; KECHRID, A; BEM HASSEM, A. (2004). *Pseudomonas aeruginosa*: a

Journal of Biology & Pharmacy and Agricultural Management, v. 14., n. 2, abr/jun 2018
ISSN 1983-4209 revista.uepb.edu.br/index.php/biofarm

multicentric study of antibiotic resistence (1999-2000). *Tunis Med.* 82 (12), 1070-1074**.**

MARCONI,C; CUNHA, M. L.R.S; LYRA,J.C; BENTILIN, M.R; BATALHA, J.E.N; SUGIZAKI,M.F; RUGOLO, L.M.S.S. (2008). Comparison between qualitative and semiquantitative catheter tip-cultures: laboratory diagnosis of catheter-related infection in newborns. *Braz. J. Microbiol.* 39:262-267.

MARQUES, P.B; VIEIRA, A.B.R; FARIAS, M.G.F.; SILVA, R.O.F; VIEIRA, J.M.S. (2007). Perfil de suscetibilidade à antibióticos de amostras de *Pseudomonas aeruginosa* isoladas no centro de diagnóstico da Unimed Belém-PA. *Rev. Bras. Anal. Clin.* 39(3), 175-177.

MENEZES, E.A; MACEDO, F.V.V; CUNHA, F.A; ANDRADE, M.S.S; ROCHA, M.V.P. (2004). Perfil de infecção e resistência aos antimicrobianos de bacilos Gram-negativos não fermentadores isolados no Laboratório de Patologia Clínica Dr. Edilson Gurgel, Santa Casa de Misericórdia de Fortaleza-CE. R*ev. Bras. Anal .Clin.* 36(4), 209-212.

MURRAY, P.R; BARON, E.J; JORGENSE, J.H; PFARLLER, M. A; YOLKER R.H. (2003). *Manual of Clinical Microbiology.* 8th ASM Presss,Washington,DC .

NEVES, P.R; MAMIZUKA, E.M; LEVY, C.E; LINCOPAN, N. (2011). Pseudomonas multirresistente: um problema endêmico no Brasil. *J. Bras. Patol. Med. Lab*.,47(4),409-420.

PAVIANE, E.R; STADNIK, C.B; HEINEK, I. (2004). Estudo da epidemiologia e perfil de sensibilidade da *Pseudomonas aeruginosa*. *Infarma*. 15(11-12),66-70.

PELLEGRINO, F.L.P.C; TEIXEIRA, L.M.; CARVALHO, M.G.S; NOGUEIRA, S.A; OLIVEIRA, M.P; SAMPAIO, J.L.M.; FREITAS, A.D; FERREIRA, A.L.P; AMORIM, E.L.T; RILEY, L.W; MOREIRA B.M. (2002). Occurrence of a multidrug-resistant *Pseudomonas aeruginosa* clone in different hospitals in Rio de Janeiro, Brazil. *J. Clin. Microbiol.* 40 (7), 2420-2424.

PEÑA, C. et al. Prospective Multicenter Study of the Impact of Carbapenem Resistance on Mortality in *Pseudomonas aeruginosa* Bloodstream Infections. Antimicrob. Agents Chemother. 56(3), 1265–1272. Available in: http://aac.asm.org/content/56/3/1265.full. Accessed: May 6, 2012.

PIRES, E.J.V.C; JÚNIOR, V.V.S; LOPES, A.C.S; VERAS, D.L; LEITE, L. E: MACIEL, M.A.V. (2009). Análise epidemiológica de isolados clínicos de *Pseudomonas aeruginosa* provenientes de hospital universitário. *Rev. Bras. Terap. Intens.* 21(4), 384-390.

PITTET, D; DHARAN, S; TOUVENEAU, S; SAUVAN,V; PERNEGER, T.V. (1999). Bacterial contamination on hands of hospital staff during roitine patiente care. *Arch. Intern. Med.* 159(8), 821- 826.

Journal of Biology & Pharmacy and Agricultural Management, v. 14., n. 2, abr/jun 2018
ISSN 1983-4209 revista.uepb.edu.br/index.php/biofarm

Probac do Brasil. (2012). *Kit não fermentador***.** Available in: http://www.probac.com.br/bulas/kit_nf.pdf. Accessed: April 11, 2012.

SADER, H.S. et al. Perfil de sensibilidade a antimicrobianos de bactérias isoladas do trato respiratório baixo de pacientes com pneumonia internados em hospitais brasileiros: resultados do Programa SENTRY, 1997 e 1998. *J. Pneumol*. 27(2),59-67.

SANTOS, L.F. (2006). Identificação de bastonetes gram negativos não fermentadores. *Manual de Microbiologia Cínica*, 4°ed. João Pessoa-PB.

SILVA FILHO, L.V.F; TATENO, A.F; MARTINS, K.M; AZZUZ CHERNISHEV, A.C; OLIVEIRA GARCIA, D; HAUG, M; MEISNER, C; RODRIGUES, J. C; DÖRING, G. (2007). The combination of PCR and serology increases the diagnosis of *Pseudomonas aeruginosa* colonization/infection in cystic fibrosis. *Pediatr. Pulmonol*. 42: 938–944.

TAKAGI, E.H; LINCOPAN, N; CASSETTAR, V.C; PASSADORE, L.F; MAMIZUKA, E.M; MARTINEZ, M.B. (2009). Carbapenem-resistant A*cinetobacter baumannii* outbreak at University Hospital. *Braz. J. Microbiol.* 40:339-341

TUMITAN, A.R; PIZZOLITTO, A.C. (2003). Padrão de sensibilidade a antimicrobianos de bacilos gram negativos não fermentadores isolados de materiais clínicos de pacientes em Presidente Prudente – SP. *Rev. Ciênc. F*arm, 24 (2),131-139.

UMADEVI, S; JOSEPH, N.M; KUMARI,K; EASON,J.M; KUMAR, S; STEPHEN, S; SRIRANGARAJI, S; RAJ, S. (2011). Detection of extended spectrum beta lactamases, AMPC beta lactamases and metallo betalactamases in clinical isolates of ceftazidime resistant *Pseudomonas aeruginosa*. *Braz. J. Microbiol*. 42(4), 1284-1288.

VOLK, W; BENJAMIN, D; KADNER,R; PARSONS, J.T. (1999). *Essentials of Medical Microbiology*. Grand Rapids, J. B. Lippincott.

ZANOL.F.M; PICOLI, S.U; MORSCH,F. (2010). Detecção fenotípica de metalobetalactamases em isolados clínicos de *Pesudomonas aeruginosa* em hospitais de Caxias do Sul. *J. Bras. Patol. Lab*. 46(4), 309-314.

WOLSKA, K; KOT, B; JAKUBCZAK, A. (2012). Phenotypic on genotypic diversity of *Pseudomonas aeruginosa* strains isolated from hospitals in Siedle (Poland). *Braz. J. Microbiol*. 43(1), 274-282.

Received: 16 November 2015
Accepted: 17 March 2018
Published: 30 June 2018