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Short Communication

Increased antimicrobial resistance in *Klebsiella pneumoniae* from a University Hospital in Rio Grande do Sul, Brazil

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Abstract

Introduction: The spread of multidrug-resistant Gram-negative bacilli is a health threat, limiting therapeutic options and increasing morbimortality rates. **Methods:** This study aimed to evaluate the antimicrobial susceptibility profile of 1805 *Klebsiella pneumoniae* isolates collected from *Hospital Universitário de Santa Maria* between January 2015 and December 2016. **Results:** Resistance to colistin (239.3%), meropenem (74.2%), ciprofloxacin (68%), gentamicin (35.1%), tigecycline (33.9%), imipenem (29.7%), ertapenem (26.8%), and amikacin (21.4%) was found increased. **Conclusions:** Infection control measures in the hospitals are necessary for reducing the spread of multidrug-resistant microorganisms and preventing efficacy loss of these drugs.

Keywords: Colistin. *Klebsiella pneumoniae*. Multi-drug resistance.

In recent years, antimicrobial resistance has become a global threat to public health. The extensive and indiscriminate use of antimicrobial agents in human and veterinary medicine has led to the dissemination of high-risk clones, capable of accumulating mutations and resistance genes in mobile genetic elements[1]. With the emergence of carbapenem-resistant Gram-negative bacilli, polymyxins have become the drug of choice for the treatment of these pathogens[2]. However, recent studies have shown increased resistance rates due to changes in outer membrane, involving component systems (e.g. *mgrB* gene) and lipid A (e.g. *mcr-1* gene)[3,4].

Increased infections by multidrug-resistant bacteria, associated with limited therapeutic options, imply the failure of empirical treatments, reinforcing the need for antibacterial therapies based on antimicrobial-susceptibility tests[5]. Therefore, this study aimed to evaluate the antimicrobial susceptibility profile of *Klebsiella pneumoniae* isolates collected during the period of January 2015 till December 2016 at *Hospital Universitário de Santa Maria* (HUSM), Santa Maria-RS, Brazil.

This was a retrospective observational study, developed in a university hospital in the Central-West region of Rio Grande do Sul. HUSM is a reference institution for emergency care that has 403 hospital beds, and serves approximately 1.2 million inhabitants from 45 municipalities. The isolates were collected from several clinical specimens of hospitalized patients, and identified according to the automated system VITEK®2 (bioMérieux, Marcy-l'Étoile, France). Besides gender, age, and hospital unit of the patient, susceptibility profile against the antimicrobial agents amikacin, ciprofloxacin, colistin, ertapenem, gentamicin, imipenem, meropenem, and tigecycline were evaluated. The Minimum Inhibitory Concentration (MIC) was analyzed using the Advanced Expert System (AES) program, included in VITEK®2. For the interpretation of susceptibility to colistin and tigecycline, the criteria established by the European Committee on Antimicrobial Susceptibility Testing (EUCAST) were used[6]. For the other antimicrobials, the Clinical and Laboratory Standards Institute (CLSI) guidelines were used[7].

During the study period, 1805 isolates were collected from patients infected or colonized by *K. pneumoniae*, with 784 (43.4%) isolated in 2015 and 1021 (56.6%) isolated in 2016. The specimen from which isolation was maximum was urine (685; 38%), followed by rectal swab (322; 18%), tracheal secretion (230; 12.7%), sputum (137; 7.6%), blood (97; 5.3%), feces (65; 3.6%), surgical wound fluid (25; 1.4%), abdominal fluid (19; 1.1%), peritoneal fluid (17; 0.9%), bronchoalveolar lavage (14; 0.8%), lesion swab (11; 0.6%), catheter tip (11; 0.6%), pleural fluid (9; 0.5%), and others (163; 9%).

Regarding gender and age, there was a prevalence among males (940; 52.1%) over 60 years of age (925; 51.3%), followed

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by those between 21 to 59 years (717; 39.7%), and zero to 20 years (163; 9%). The Intensive Care Unit (319; 17.7%) was the hospital sector with the highest isolation rates, followed by the Semi-Intensive Care Unit (295; 16.4%), clinic (290; 16.1%), General Surgery Unit (253; 14.1%), Adult Emergency Room (245; 13.6%), Cardiac Intensive Care Unit (93; 5.2%), Nephrology Unit (60; 3.4%), Surgical Block (50; 2.8%), Medical Clinic Unit (34; 1.9%), Recovery Room (31; 1.8%), Obstetric Center (30; 1.7%), Treatment Center for Children with Cancer (29; 1.6%), Child and Adolescent Health Care, Child Intensive and Semi-Intensive Care (28; 1.5%), Maternal-Infant and Women's Health Unit (14; 0.8%), and others (25; 1.4%).

The antimicrobial susceptibility profile is described in **Table 1**. We observed that *K. pneumoniae* showed increased resistance to colistin (239.3%), meropenem (74.2%), ciprofloxacin (68%), gentamicin (35.1%), tigecycline (33.9%), imipenem (29.7%), ertapenem (26.8%), and amikacin (21.4%). **Figure 1** represents the percentage of antimicrobial resistance per quarter.

The phenotype of multi-resistance can be explained by the spread of carbapenemases, especially *Klebsiella pneumoniae* carbapenemase (KPC), as well as the extensive use of monotherapy in this hospital[8]. A study by Santos, La Rocca and Hörner[9] had also described low susceptibility to carbapenem and ciprofloxacin in colistin-resistant *Pseudomonas aeruginosa* isolates during the same period. Rossi et al.[10] have observed an increase in colistin-resistant *K. pneumoniae* between 2010 and 2014 at *Hospital de Clínicas in São Paulo* (Brazil).

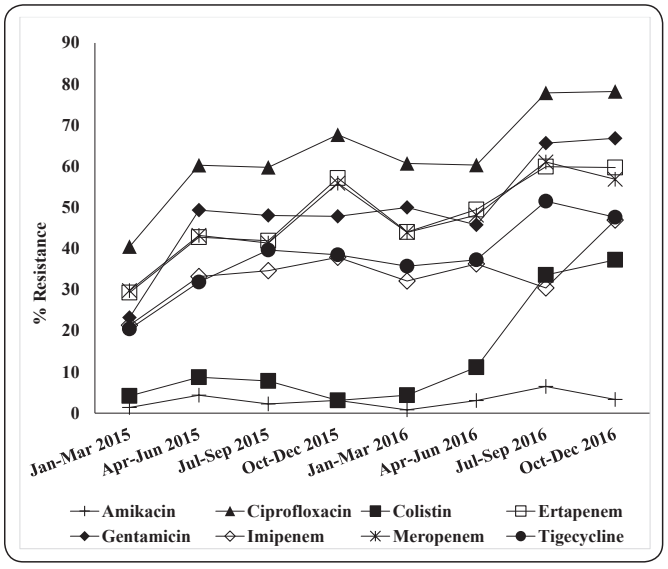

FIGURE 1: Increased antimicrobial resistance of *Klebsiella pneumoniae* isolates between 2015 and 2016.

In our study, all colistin-resistant *K. pneumoniae* isolates showed high resistance to carbapenems and tigecycline (**Table 2**), which is alarming, considering that these antimicrobials are used for the treatment of carbapenem-resistant *Enterobacteriaceae* in combination with amikacin, which has demonstrated good activity against the isolates, with susceptibility greater than 80%[2].

TABLE 1: Antimicrobial susceptibility profile of *Klebsiella pneumoniae* isolates between 2015 and 2016.

Antimicrobial agent	MIC Range (µg/mL)	2015 (n = 784)			2016 (n = 1021)		
		MIC_{50}/MIC_{90}	R (n;%)	S (n;%)	MIC_{50}/MIC_{90}	R (n;%)	S (n;%)
AMI	0.5-64	2/16	22; 2.8	762; 97.2	2/16	35; 3.4	986; 96.6
CIP	0.25-16	4/4	321; 40.9	463; 59.1	4/4	701; 68.7	320; 31.3
COL	0.5-16	0.5/0.5	48; 6.1	736; 93.9	0.5/16	211; 20.7	810; 79.3
ERT	0.5-8	0.5/8	328; 41.8	456; 58.2	4/8	541; 53	480; 47
GEN	1-16	1/16	326; 41.6	458; 58.4	16/16	574; 56.2	447; 43.8
IMI	0.25-16	0.25/16	245; 31.3	539; 68.7	0.25/16	414; 40.6	607; 59.4
MER	0.25-16	0.25/16	237; 30.2	547; 69.8	2/16	537; 52.6	484; 47.4
TIG	0.25-8	0.5/8	250; 31.9	534; 68.1	1/8	436; 42.7	585; 57.3

MIC50: minimum inhibitory concentration required to inhibit the growth of 50 % of microorganisms; **MIC90:** minimum inhibitory concentration required to inhibit the growth of 90 % of microorganisms; **R:** resistant and intermediate; **S:** sensitive; **AMI:** amikacin; **CIP:** ciprofloxacin; **COL:** colistin; **ERT:** ertapenem; **GEN:** gentamicin; **IMI:** imipenem; **MER:** meropenem; **TIG:** tigecycline.

TABLE 2: Antimicrobial susceptibility profile of colistin-resistant *Klebsiella pneumoniae* isolates between 2015 and 2016*.

Antimicrobial agent	MIC Range (µg/mL)	2015 (n = 48)			2016 (n = 211)		
		MIC_{50}/ MIC_{90}	R (n;%)	S (n;%)	MIC_{50}/ MIC_{90}	R (n;%)	S (n;%)
AMI	0.5-64	8/16	6; 12.5	42; 87.5	4/16	13; 10.9	198; 89.1
CIP	0.25-16	4/4	46; 95.8	2; 4.2	4/4	208; 98.6	3; 1.4
ERT	0.5-8	8/8	40; 83.3	8; 16.7	8/8	201; 95.3	10; 4.7
GEN	1-16	16/16	32; 66.7	16; 33.3	16/16	195; 92.4	16; 7.6
IMI	0.25-16	16/16	40; 83.3	8; 16.7	8/16	203; 96.2	8; 3.8
MER	0.25-16	16/16	40; 83.3	8; 16.7	16/16	203; 96.2	8; 3.8
TIG	0.25-8	8/8	42; 87.5	6; 12.5	2/8	187; 88.6	24; 11.4

MIC50: minimum inhibitory concentration required to inhibit the growth of 50 % of microorganisms; **MIC90**: minimum inhibitory concentration required to inhibit the growth of 90 % of microorganisms; R: resistant and intermediate; **S:** sensitive; **AMI:** amikacin; **CIP,** ciprofloxacin; **COL**: colistin; **ERT**: ertapenem; **GEN,** gentamicin; **IMI**: imipenem; **MER**: meropenem; **TIG:** tigecycline; *MIC50/ MIC90 16 µg/mL in both years.

The colonization of carbapenemase-producing microorganisms, prolonged antimicrobial therapy, and previous use of polymyxins contributed to the emergence of heteroresistance[11,12]. Giani et al.[13], had observed an outbreak of colistin-resistant KPC-3-producing *K. pneumoniae* due to the deletion of *mgrB* gene. The presence of *mcr-1* gene in carbapenemase-producing isolates had also been reported by some authors[14,15].

The resistance profile observed in this institution, during the period of analysis, raises concern, since *K. pneumoniae* showed increased resistance to the antimicrobial agents tested, prompting the adoption of control and preventive measures against multidrug-resistant microorganisms to reduce their spread in the hospital environment and avoid efficacy loss of these drugs.

Ethical considerations

This research was approved by the Ethical Research Committee, Universidade Federal de Santa Maria, under approval number 38850614.4.0000.5346.

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Conflicts of interest

The authors declare that there is no conflict of interest.

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