

ANTIMICROBIAL RESISTANCE TO *KLEBSIELLA PNEUMONIAE* IN A NORTH EASTERN ROMANIAN HOSPITAL: a descriptive study

Nicoleta Luchian^{1†}, Marius Gabriel Dabija^{2†}, Lucian Eva^{1†}, Constantin Marcu^{1†},
Mihaela Roxana Popescu^{2†}, Mihaela Monica Scutariu^{2†}, Elena Roxana Bogdan
Goroftei^{1†*}, Cristian Guțu^{1†*}, Oana Cristina Cretu^{3†}, Florina Popa^{1†}, Letiția Doina
Duceac^{1†}

1., „Dunărea de Jos” University of Galați, Faculty of Medicine and Pharmacy, Galați, 47 Domnească Street, RO-800008, Galați, România

2. University of Medicine and Pharmacy "Grigore T. Popa", Iași

3. Md. Phd. Socola Psychiatric Institute, Iași, Romania

*Corresponding Author: Cristian Guțu, dr.c.gutu@gmail.com

Elena Roxana Bogdan Goroftei, elenamed84@yahoo.com

†- all authors have the same contribution

Abstract

Microbial resistance is a rapidly escalating phenomenon worldwide, with complex mechanisms and important involvement in the determinants of healthcare-associated and in-hospital infections. At European level, it is estimated that 80.000 hospitalised patients have at least one HAI every day with multi-antimicrobial resistant germs, leading to an extension of hospital stay by 16 million days every year. In high-income countries, the prevalence is around 7.5%, and in low- and middle-income countries, the prevalence ranges from 5.7% to 19.2%.

Material and method. Through a retrospective, descriptive study carried out in the Cline Emergency Hospital „Prof. Dr. Nicolae Oblu” in Iași, Romania in the period 2020-2021, the 602 identified and reported intra-hospital infections were investigated, focusing on the etiology and microbial resistance of the pathogens involved. **Results.** The most frequent etiological agents of HAI identified in our study were *K. pneumoniae* (13.45%), *A. calcoaceticus* (10.11%), *S. aureus* (8.45%), *E. coli* (7.62%), *P. aeruginosa* (5.47%), noting the predominance of Gram-negative bacteria (GN) and the fact that they are part of the ESKAPE group of pathogens, the percentages obtained being similar to the European average. *Klebsiella pneumoniae* (13.45 %) was the most common in the aetiological palette of HAIs, allowing us to highlight certain peculiarities regarding its antimicrobial resistance. **Conclusions.** The present study provides useful data for monitoring the resistance of pathogens involved in HAIs at hospital level, for guiding antibiotic therapy in healthcare-associated infections and for substantiating the implementation of a set of measures related to antibiotic therapy and antibiotic prophylaxis.

Keywords: multidrug-resistant bacteria, *Klebsiella pneumoniae*, neurosurgery, healthcare-associated infections.

INTRODUCTION

Healthcare-associated infections (HAIs), synonymously known as nosocomial infections or intra-hospital infections, are a major public health problem worldwide with their considerable impact on morbidity, mortality and costs. In 2016 and 2017, ECDC reports approximately 8.9 million HAIs per year across EU/EEA countries, of which 4.5 million HAIs in hospitals[1].

In Romania, the incidence and prevalence of healthcare associated infections (HAIs) are grossly underestimated due to underreporting, with only 0.2-0.25% in official reporting. Data with higher accuracy

were obtained from a 2018 European study that revealed a national prevalence of 2.6%, equivalent to 100,000 cases registered annually.

With the entry into force of the Ministry of Health Order 1101 of 2016, the reporting rate of HAI has increased, but the prevalence value is still far from the European average of 7.1%, and strategies need to be implemented to improve surveillance, prevention and containment of nosocomial infections and to address the causes leading to their underreporting [2,3]

Klebsiella are significant human pathogens, frequently resistant to multiple antibiotics, and recently, ectopic colonization of the gut

by oral *Klebsiella* was documented to induce dysbiosis and inflammation [4].

Oral *K. pneumoniae* was recently shown to induce inflammation and dysbiosis in the gut after ectopic colonization, and it was hypothesized that the oral cavity provides a reservoir for would-be intestinal pathogens, such as *Klebsiella* [5].

One of the causes identified for the increased number of HAIs in Romania would be multidrug resistant (MDR) bacteria, which have a high risk of transmission due to high pathogenicity, undersized accommodation and patient care facilities and poor isolation of those colonised with MDR. Highly pathogenic MDR bacteria are carbapenem-resistant Enterobacteriaceae [2]

Multidrug-resistant bacteria are frequently encountered as the etiology of healthcare-associated infections (HAIs) or nosocomial infections (approximately 20%), associated with increased mortality, the notorious ones being: Methicillin-resistant *Staphylococcus aureus* (MRSA); Vancomycin-resistant/Vancomycin-intermediate *Staphylococcus aureus* (VRSA/VISA); Enterobacteriaceae producing extended-spectrum beta (β) lactamase (ESBL); Enterobacteriaceae producing carbapenemases (CRE); Vancomycin-resistant Enterococcus (VRE); *Pseudomonas* MDR [3]

Antimicrobial resistance is another important global public health problem, closely linked to that of HAIs as these infections are often caused by antibiotic-resistant germs [6], and the emergence of HAIs increases the risk of developing resistant strains[7].

Frequent use of antibiotics over a long period of time has caused some bacteria to develop antibiotic resistance [8]. In the absence of a global intervention to curb untimely and long-lasting antimicrobial therapies, infections with antibiotic-resistant microorganisms are expected to become the leading cause of death by 2050, surpassing cancer mortality in developed countries [9]. Furthermore, aside from being a substantial pathogen in its own right, evidence is accumulating that *K. pneumoniae* serves as a key trafficker of drug resistance loci from the environment to human pathogens [10,11]

Contaminated hospital surfaces, particularly sinks, have been the source of outbreaks of multidrug-resistant *Klebsiella* [12].

MATERIAL AND METHOD. A retrospective descriptive study was conducted in which the 602 healthcare-associated infections identified and reported in the Hospital of Neurosurgery "Prof. Dr. N. Oblu" in the period 2020 - 2021 were analyzed, aiming to identify the top 5 etiological agents and their antibiotic resistance profile.

The case definitions for HAIs were taken from the "Commission Implementing Decision (EU) 2018/945 of 22 June 2018 on communicable diseases and related special health issues to be covered by epidemiological surveillance, as well as relevant case definitions" and ECDC. Antibiotic susceptibility testing was performed using the Kirby- Bauer diffusimetric method on Mueller-Hinton culture medium and the microdilution method for MIC determination of Vancomycin and Colistin according to EUCAST 2019 standards.

RESULTS. In the etiological palette of intra-hospital infections in our study, *Klebsiella pneumoniae* (13.45%) was ranked first, followed by *Acinetobacter calcoaceticus* (10.11%), *Staphylococcus aureus* (8.45%), *E. coli* (7.62%), *Pseudomonas aeruginosa* (5.47%), with a predominance of Gram negative (GN) bacteria, with percentages similar to the European average.

1. Data related to distribution by reporting year, sex, age, ward, detection interval and type of HAI

Of the 81 cases of HAIs with *K. pneumoniae*, 19 (23.45%) were reported in 2020 and 62 cases (76.54%) were reported in 2021. Patients presenting with HAIs with *K. pneumoniae* were aged between 20 and 86 years, predominantly those in the age range 70 - 80 years (24.69%). In terms of gender distribution, they were more frequently diagnosed in males (48 cases, 59.25%) compared to females (33 cases, 40.74%).

The distribution of nosocomial infections with *K. pneumoniae* by hospital wards, in order of their frequency was as follows: the ATI ward reported the most cases (53 cases;

65.43%), followed by the Neurosurgery ward (NC)- 17 cases; 20.98% and the Neurology ward (N) -11 cases; 13.58%.(Fig.1)

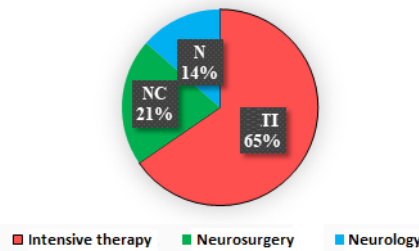


Fig.1. Distribution of HAIs caused by *K. pneumoniae* in hospital wards

The time interval from admission to detection of nosocomial infection with *K. pneumoniae* ranged from 4 to 64 days, with the majority being identified between day 5 and day 20 of admission (65.43%) (Fig. 2).

The type of HAI with *K. pneumoniae* most common was urinary tract infection (UTI), evidenced in 55 cases (67.9%), followed by

lower respiratory infection (LRRI)- 12 cases (14,81%), sepsis - 5 cases (6,17%), Intubation-associated pneumonia (IAP) - 4 cases (4,93%), post-operative meningitis (POM) - 2 cases (2,46%), Peripheral venous catheter infection (PVC) - 2 cases (2,46%) and surgical incision infection (SII) detected in 1 case (1,23%). (Fig.3)

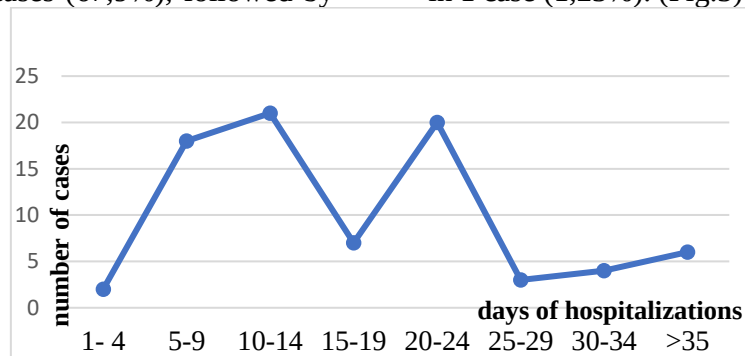


Fig.2. Distribution of cases of HAIs with *K. pneumoniae* according to days of detection/reporting

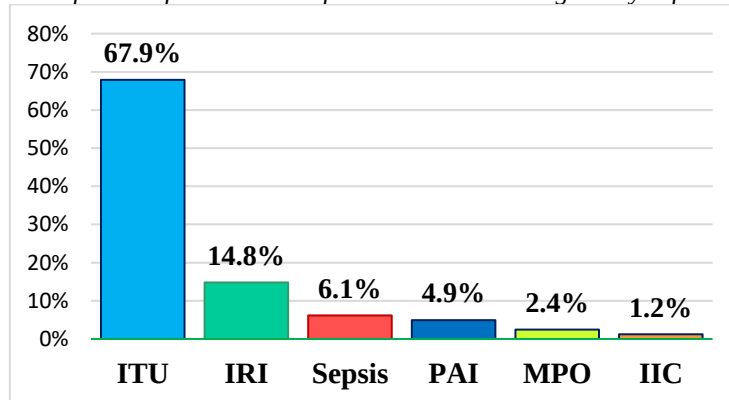


Fig.3. Distribution of *K. pneumoniae* isolates according to the type of HAI determined

1.2. Antibiotic resistance of *K. pneumoniae* isolates involved in HAIs.

Of the 81 *K. pneumoniae* isolates: 28 (34.56%) were MDR (multidrug-resistant), 66 (81.48%) were ESBL-producing and 59

(72.83%) were CR (carbapenem-resistant). According to annual reporting by HAIs, in 2020 of the 19 isolates: 12 (63.15%) were considered MDR, 16 (84.21%) were ESBL/BLSE-producing and 68.42% were

RC. In 2021 there is a decrease in MDR variants (only 25.8%), the number of ESBL-producing strains is stationary and there is also a slight increase in the number of RC strains (75.8%) (Table 3) and (Fig. 4)

Table 3. Number of strains producing ESBL, RC and MDR

	Isolated number <i>K. pneumoniae</i> 2020 – 2021 Total= 81	Percent
MDR	28	34.56%
ESBL	66	81.48%
RC	59	72.83%
	Isolated number <i>K. pneumoniae</i> 2020 Total= 19	Percent
MDR	12	63.15%
ESBL	16	84.21%
RC	13	68.42%
	Isolated number <i>K. pneumoniae</i> 2021 Total= 62	Percent
MDR	16	25.8% ↓
ESBL	50	80.64%
RC	47	75.8% ↑

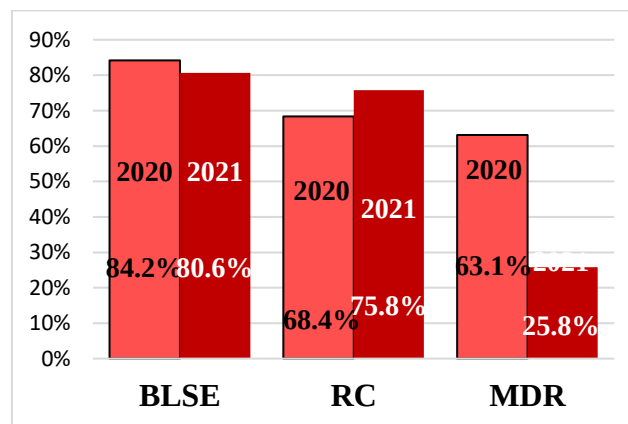


Fig.4. Antibiotic resistance profile (BLSE, RC, MDR) of K. pneumoniae strains compared between 2020 and 2021

K. pneumoniae spp. producing ESBL strains retained their susceptibility to the following antibiotics: Most were sensitive to Ceftazidime (CAZ) (94.11%), more than half to Amikacin (AK) (59.37%), followed by Imipenem (IPM) (26.66%), Gentamicin (G)-

26, 56%, Meronem (MEM)-23.8%, Tazobactam (TZP)-12.69%, Ciprofloxacin (CIP)-12.5%, Trimetoprim (TMP-SXT)-10.16%, Levofloxacin (LEV)-9.52%, Ertapenem (ETP)-3.12%. (Fig 5).

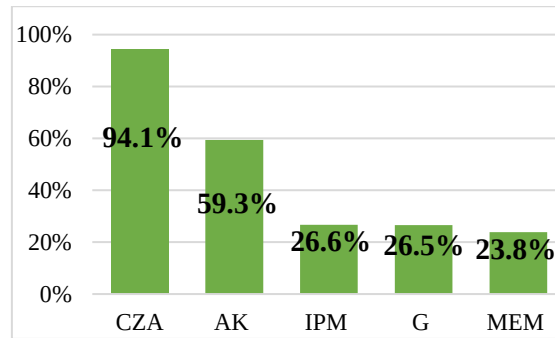


Fig.5. Antibiotic susceptibility of ESBL-positive *K. pneumoniae* strains involved in the aetiology of HAIs in the period 2020-2021

In 2020, of the 19 strains reported, 16 (84.21%) were ESBL positive, and in 2021, 46 of 58 *K. pneumoniae* strains (79.31%) were ESBL/BLSE producing.

Carbapenem resistance was recorded in 72.83% of the strains. Carbapenem-resistant

Klebsiella pneumoniae (KPRC) showed antimicrobial susceptibility to: CZA (46/48; 95.83%), G (12/57; 21.05%), AK (31/57; 54.38%), TMP-SXT (6/53; 11.32%) (Fig 6).

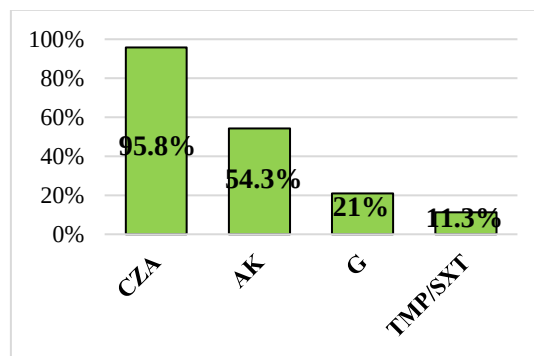


Fig.6. Antibiotic susceptibility of KPRC strains involved in the aetiology of HAIs from 2020-2021

In 2020, of the 19 strains reported, 13 (6.42%) were RC, and in 2021, 43 of 58 strains of *K. pneumoniae* (74.13%) were RC.

MDR *K. pneumoniae* strains were present in 28 cases (34.56%) of HAIs. The

mean age of patients infected with these strains was 58 years, and the mean time interval from admission to HAIs detection was 19 days (most infections reported between day 20-24 of hospitalization) (Fig 7).

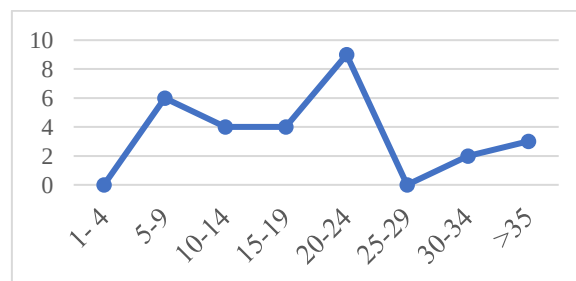


Fig. 7. Distribution of HAIs cases with *K. pneumoniae* MDR according to days of detection/reporting

MDR strains were isolated most frequently from the wards: ATI -78.57%, followed by

Neurology and Neurosurgery with 10.71%. They remain most frequently involved in UTI

(60.71%), followed by IRI (21.42%), sepsis (10.21%), IIC and MPO (3.57%). Most strains retain sensitivity to CZA (88.88%) and a few MDR strains to aminoglycosides (AK 14.28%). In 2020, of the 19 strains reported, 12 (63.15%) were MDR, and in 2021, 16 of 62 *K. pneumoniae* strains (25.8%) were MDR.

1.2.1. Antibiotic resistance to beta-lactams

The antimicrobial resistance to Betalactams of *K. pneumoniae* strains responsible for AMR from 2020-2021 was 88.9%.

Antibiotic resistance to broad-spectrum penicillins: Piperacillin with tazobactam (TZP), a ureidopenicillin combined with a beta-lactamase inhibitor, was tested for 78 out of 81 samples. It showed a 73.07% TZP resistance rate. In 2020 the TZP resistance rate of the strains is 70.58% and in 2021 it is 73.68%.

b) *Cephalosporin class*. Antibiotic resistance to 2nd generation cephalosporins. Cefuroxime (CXM) was used as a representative of this class. Sensitivity to CXM was tested for 72 out of 81 cases. Sixty-three of the *K. pneumoniae* strains (87.5%) were found to be resistant. In 2020 the percentage of resistant strains is 88.88% and in 2021 it is 86%.

Antibiotic resistance to Cephalosporin III and cephalosporin III associated with beta-lactamase inhibitor. Susceptibility testing of these showed the following antimicrobial resistance percentages: 87.34% of strains were resistant to CAZ (in 2020 the prevalence of CAZ-resistant strains is 83.33% and in 2021 it is 87.71%), 90.9% were resistant to CFP, and 87.5% of *K. pneumoniae* tested were resistant to ceftibuten (CBT); 84.81% of *K. pneumoniae* strains are resistant to CRO (in 2020 a percentage of 77.7% of strains were resistant to CRO and 85.96% in 2021). Ceftazidime susceptibility testing with avibactam (CZA), a third-generation cephalosporin combined with a beta-lactamase inhibitor, was performed on 58 cases, revealing a percentage of CZA-resistant strains of 5.17% (3 strains). All 3 CZA-resistant strains were reported in 2020, 2 of which were resistant to all classes of

a) *Penicillin class*. Antimicrobial resistance to Aminopenicillins and aminopenicillins associated with a beta-lactamase inhibitor: AMP testing identified 100% resistance. From the class of aminopenicillins associated with beta-lactamase inhibitor, AMC and AMS were tested in 78 out of 81 samples.

Resistance to AMC was observed in 68 cases (87.17%) and resistance to AMS was present in 66 cases (84.61%). In 2020 the prevalence of resistance to both AMC and AMS was 82.35% and in 2021 it was 87.71% and 84.21% respectively.

antibiotics tested (PDR). In 2021, there were no cases of IAAM with CZA-resistant *K. pneumoniae*.

c) *Carbapenems class*. Carbapenem sensitivity testing of *K. pneumoniae* strains revealed the following antibiotic resistance rates: 56.41% of strains were resistant to MPI, 59.25% to MEM and 89.47% of strains to ETP. In 2020 there was a 62.5% resistance rate to MPI and in 2021 a 54.54% resistance rate.

d) *Monobactam class*. Resistance to Aztreonam (AZT) was identified in 85.29% of *K. pneumoniae* strains. In 2020, AZT resistance was 85.71% and in 2021 it was 86.66%.

1.2.2. Antibiotic resistance to Chinolone

Of the 81 *K. pneumoniae* isolates tested, 69.1% were resistant to quinolone, similar to resistance to Betalactam.

Ciprofloxacin susceptibility testing (CIP) revealed that 81.01% of *K. pneumoniae* strains were resistant. Antimicrobial resistance to Levofloxacin (LEV) was similar to CIP resistance at 79.22%. In 2020, quinolone resistance was present in 82.35% of strains for CIP and 88.23% for LEV, and in 2021, 82.25% for CIP and 78.83% for LEV.

1.2.3. Aminoglycoside resistance

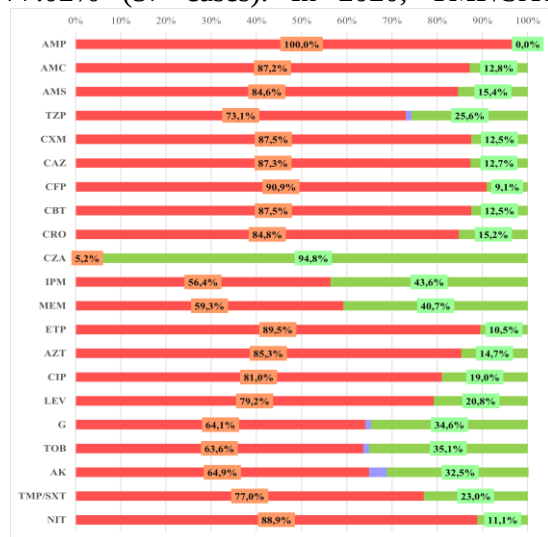
Sensitivity to Gentamycin (G) was shown in 50 of the *K. pneumoniae* strains (64.1%). Similar results were observed for Tobramycin (TOB)- 63.63%. Amikacin (AK) susceptibility testing showed that the majority of *K. pneumoniae* strains (64.94% representing 50 cases) are susceptible to AK, only 32.46% (25 cases) of strains show resistance, and 3 strains (3.89%) show

intermediate resistance. In 2020, the resistance of *K. pneumoniae* strains to aminoglycosides was: 88.23% for G, 64.7% to AK, and in 2021 a value of 59.01% for G and 24.59% to AK is reported.

1.2.4 Antibiotic resistance to Sulfamides

Trimethoprim/Sulfamethoxazole

(TMP/SXT) susceptibility testing showed a resistance of *K. pneumoniae* strains of 77.02% (57 cases). In 2020, TMP/SXT



DISCUSSION. *K. pneumoniae* is a major cause of in-hospital infections, accounting for approximately 10% of them [8-11]. It is also part of the top 3 pathogens of international concern, documented in 2017 in the WHO Global Priority List of Antibiotic-Resistant Bacteria for Guidance, Research, Discovery and Development of New Antibiotics. [12] In Eastern, Central and Southern Europe, an increase in the frequency of *K. pneumoniae* infections resistant to third-generation cephalosporins, fluoroquinolones and aminoglycosides is evident.

ESBL-producing *Klebsiella pneumoniae*. The management of infections caused by antibiotic-resistant *K. pneumoniae* is difficult because of intrinsic and acquired resistance to broad-spectrum antibiotics (e.g. betalactams)[10]. ESBLs are enzymes that confer resistance against betalactams, a class of antibiotics commonly used in clinical practice. [12,13]

K. pneumoniae is considered naturally resistant to ampicillin and carbenicillin. [14,15]. To date, more than 400 types of ESBLs are known [16,17].

resistance of *K. pneumoniae* strains isolated from HAIs patients was 78.57% and in 2021 80%.

1.2.5. Antibiotic resistance to Nitrofurans

Nitrofurantoin (NIT) susceptibility was tested in 45 out of 81 cases (UTI-HAIs cases only). It was found that 88.88% of *K. pneumoniae* strains (40 cases) were resistant, and only 11.11% (5 cases) were susceptible.

Fig.8. Resistance to different classes of antibiotics of *K. pneumoniae* isolates involved in HAIs, period 2020-2021

A meta-analysis conducted in 2021, which included 47 studies, found 91.49% betalactam resistance in *K. pneumoniae* implicated in healthcare-associated infections[20].

Another retrospective study conducted from 2016 to 2020 in a tertiary center in China, aiming to observe the trend of antibiotic resistance profile of *K pneumoniae* isolated from 396 blood cultures from patients diagnosed with healthcare-associated sepsis, mentions 61.4% resistance to Ampicillin with sulbactam, 42.1% to Ceftazidim, 59.64% to Cefotaxime, 59.64% to Ceftriaxone, 50.87% to Aztreonam. [18].

A study in Portugal, conducted on 81 isolates of *K. pneumoniae* from urocultures collected between January-March 2010, aiming to compare the antibiotic resistance profile of strains involved in community UTIs and those involved in healthcare-associated UTIs, showed the following percentages of beta-lactam resistance of nosocomial strains (31 samples): 93.5% for Ceftazidim, 87.1% for Cefotaxime, and 64.5% for Amoxicillin clavulanate[12].

Carbapenem-resistant *Klebsiella pneumoniae*. Carbapenems are the most commonly used class of antibiotics for the treatment of ESBL-producing *K. pneumoniae* infections and are a last choice of antibiotic therapy for infections with this pathogen [9,10]. In 2018, at European level, *K. pneumoniae* resistance to carbapenems (backup antibiotic) ranged from 0% to 63.9%, with the highest prevalence in Greece, followed by Romania (29.5%) and Italy (26.8%) [9].

A retrospective study from China conducted from 2016 -2020 shows a prevalence of carbapenem resistance of *K. pneumoniae* involved in healthcare-associated sepsis of: 36.84% to imipenem and 35.08% to meropenem. [18,21].

At the national level, a study conducted in Romania, Craiova County Hospital between 2017 and 2019 on 251 strains of *Klebsiella* spp. isolated from patients with in-hospital infections found a resistance to meropenem of 27.78% [8]

Fluoroquinolone-resistant *Klebsiella pneumoniae*. Fluoroquinolones, antibiotics of synthetic origin, are extensively used in clinical practice due to their broad-spectrum activity against a wide range of important pathogens and excellent tissue penetration. *Klebsiella pneumoniae* is one of the main fluoroquinolone-resistant Enterobacteriaceae [22,23]

In the study by Cao Z et al. On 396 positive blood cultures from patients with healthcare-associated sepsis for *K. pneumoniae*, it reveals a strain resistance to ciprofloxacin of 54.38% and 40.35% to levofloxacin. [18].

At national level, the same study conducted in the County Hospital of Craiova on 251 strains of *K. pneumoniae* from patients with healthcare-associated infection revealed a resistance to Ciprofloxacin of 30.57%. [8].

A study of patients diagnosed with HAIs admitted to the ICU of a hospital in Saudi Arabia reports a 64.2% resistance to Ciprofloxacin, 63.2% to Levofloxacin, 68.9% to Moxifloxacin and 40% to Norfloxacin of isolated *K. pneumoniae* strains [24].

Aminoglycoside-resistant *Klebsiella pneumoniae*. At European level, an average

In 2021 ECDC publishes a report based on data from EARS-Net, in which it differentiates between the North-West European region (14 out of 45 countries, 31%) where carbapenem resistance of *K. pneumoniae* is estimated to be less than 1%, and the other countries: of which 15 (33%) report a prevalence of 25% or higher, of carbapenem-resistant strains of *K. pneumoniae* and 8 countries (18%) record a percentage greater than or equal to 50%. [19,20]

prevalence of aminoglycoside (gentamicin/netilmicin/tobramycin)-resistant *K. pneumoniae* strains of 23.7% (between 0 and 67%) is reported for the year 2020 [19,25].

At national level, in the Craiova County Hospital, the same study reveals an Amikacin resistance of 13.5% of *Klebsiella* spp. strains [8]

As an aside, our study retrospectively spanned the pandemic period 2020-2021 and thus various HAIs with MDR type K germs were reported. *pneumoniae*, in patients infected with SARS-CoV-2, who presented a high degree of immunosuppression, leading to a thrash evolution of HAIs and implicitly more days of hospitalization [26-28].

MDR bacteria, which generate intra-hospital infections and thus microbial antibiotic resistance, can be combated with broad-spectrum antibiotics: third generation cephalosporins, colistin, carbapenems, ceftriaxone, which can be more easily transported in the body with the help of transport nanoparticles, penetrating down to the cellular and molecular level. Their action is potentiated by probiotics, in order not to cause gastrointestinal disorders. [37 - 47]

In limiting and controlling intra-hospital infections, there must also be a concern for decontamination of the hospital environment with appropriate methods and biocides [48-50].

CONCLUSIONS. The most common type of HAI with *K. pneumoniae* was UTI (67.9%), and 65% of infections were detected in the ICU. Of the *K. pneumoniae* isolates: 34.56% were MDR; 81.48% were ESBL-producing; 72.83% were resistant to carbapenems, 69.1% were resistant to fluoroquinolones and

30.9% to aminoglycosides. There is a decrease in MDR strains from 63.15% in 2020 compared to 25.8% in 2021.

The study provides useful data for surveillance of resistance of pathogens involved in HAIs at hospital level, for guiding

antibiotic therapy in healthcare-associated infections, and can form the basis for implementation of antibiotic therapy and antibiotic prophylaxis measures at national, European and global levels.

References

1. European Centre for Disease Prevention and Control. Infecțiile asociate asistenței medicale - o amenințare pentru siguranța pacienților din Europa.
2. Sandor Szabo, Bogdan Feier, Denisa Capatina et al., An Overview of Healthcare Associated Infections and Their Detection Methods Caused by Pathogen Bacteria in Romania and Europe. J. Clin. Med. 2022, 11(11), 3204.
3. Barlean L, Tatarciuc M, Balcos C, Vitelariu AM, Moisei M, Chiscop I, Scutariu MM. Dentists Occupational Exposures to Chemicals During Hands Hygiene in the Dental Offices in Iasi. Rev. Chimie 2015;10(66): 1696-1699.
4. Scutariu MM, Surdu A, Macovei G. Contributions to recognition of some particularities of the lingual mucosa conditions. Rev. Med. Chir. 2015;119(4): 1147-1152.
5. Popescu GA, Șerban R, Pistol A. Consumul de antibiotice, Rezistența microbiană și Infecții Nosocomiale în România – 2016. București, 2018, ISSN 2537 – 1541.
6. McEwen SA, Collignon PJ. Antimicrobial Resistance: a One Health Perspective. Microbiol Spectr. 2018 Mar;6(2).
7. Bryce T Fukunaga, Wesley K Sumida, Deborah A Taira, James W Davis, Todd B Seto; Hospital-Acquired Methicillin-resistant *Staphylococcus aureus* Bacteremia Related to Medicare Antibiotic Prescriptions: A State-Level Analysis; Hawaii J Med Public Health. 2016 Oct; 75(10):303-309.
8. Mulani MS, Kamble EE, Kumkar SN, Tawre MS, Pardesi KR. Emerging Strategies to Combat ESKAPE Pathogens in the Era of Antimicrobial Resistance: A Review. Front Microbiol. 2019 Apr 1; 10:539.
9. Ghenea AE, Cioboată R, Drocaș AI, Țieranu EN, Vasile CM, Moroșanu A, Țieranu CG, Salan AI, Popescu M, Turculeanu A, Padureanu V, Udriștoiu AL, Calina D, Cârțu D, Zlatian OM. Prevalence and Antimicrobial Resistance of *Klebsiella* Strains Isolated from a County Hospital in Romania. Antibiotics (Basel). 2021 Jul 16;10(7):868.
10. Scutariu MM, Matei MN, Macovei G, Surdu A. A Clinical-statistical Study of Oral Mucosa Pathology Induced by the Acrylic Resins from Removable Dentures in Older Patients. 2015;52(3): 402-407.
11. Drugus D, Ciupilan C, Budala DG, Surdu A, Scutariu MM. Oral Health Problems, Healthcare Insurance and Oral Health Care Spending Among the Elderly Patients in Romania. Romanian Journal of Oral Rehab. 2022;14(4):35-40.
12. Scutariu MM, Budala DG, Davidescu A, Romanec C, Ciupilan C, Drugus D. The performance of the Romanian health system in the European context. Romanian Journal of Oral Rehab. 2022;14(4):96-104.
13. Lingyi Zeng, Chengru Yang, Jisheng Zhang, et al. An Outbreak of Carbapenem-Resistant *Klebsiella pneumoniae* in an Intensive Care Unit of a Major Teaching Hospital in Chongqing, China. Front. Cell. Infect. Microbiol., 2021; 11.
14. Mojgan Farhadi, Mohammad Ahanjan, Hamid Reza Goli et al. High frequency of multidrug-resistant (MDR) *Klebsiella pneumoniae* harboring several β -lactamase and integron genes collected from several hospitals in the north of Iran. Annals of Clinical Microbiology and Antimicrobials, 2021; 20 (70).
15. Zhezhe Lin, Jianhua Yu, Mingli Zhu. Prevalence and antibiotic resistance of *Klebsiella pneumoniae* in a tertiary hospital in Hangzhou, China, 2006–2020. Journal of International Medical Research, 2022 Feb.

16. Cátia Caneiras, Luis Lito, José Melo-Cristino et al. Community- and Hospital-Acquired *Klebsiella pneumoniae* Urinary Tract Infections in Portugal: Virulence and Antibiotic Resistance. *Microorganisms*. 2019 ; 7(5): 138.
17. Mohd Asri NA, Ahmad S, Mohamud R, Mohd Hanafi N, Mohd Zaidi NF, Irekeola AA, Shueb RH, Yee LC, Mohd Noor N, Mustafa FH, Yean CY, Yusof NY. Global Prevalence of Nosocomial Multidrug-Resistant *Klebsiella pneumoniae*: A Systematic Review and Meta-Analysis. *Antibiotics* (Basel). 2021 Dec 8;10(12):1508.
18. Esther Calbo, Javier Garau. The changing epidemiology of hospital outbreaks due to ESBL-producing *Klebsiella pneumoniae*: the CTX-M-15 type consolidation. *Future Microbiology*, 2015; 10:6.
19. Anton Spadar, Jody Phelan, Rita Elias, et al. Genomic epidemiological analysis of *Klebsiella pneumoniae* from Portuguese hospitals reveals insights into circulating antimicrobial resistance. *Scientific Reports*, 2022; 12:13791.
20. Mohammad Latifpour, Abolfazl Gholipour, Mohammad Sadegh Damavandi. Prevalence of Extended-Spectrum Beta-Lactamase-Producing *Klebsiella pneumoniae* Isolates in Nosocomial and Community-Acquired Urinary Tract Infections. *Jundishapur J Microbiol*. 2016; 9(3): e31179
21. Caiqian Liang, Bangrong Xing, Xiaoyan Yang et al. Molecular epidemiology of aminoglycosides resistance on *Klebsiella pneumoniae* in a hospital in China. *Int J Clin Exp Med*. 2015; 8(1): 1381–1385.
22. Cao Z, Yue C, Kong Q, et al. Risk Factors for a Hospital-Acquired Carbapenem-Resistant *Klebsiella pneumoniae* Bloodstream Infection: A Five-Year Retrospective Study. *Infection and Drug Resistance* 2022;15: 641—654
23. Jarlier V, Diaz Högborg L, Heuer OE, Campos J, Eckmanns T, Giske CG, Grundmann H, Johnson AP, Kahlmeter G, Monen J, Pantosti A, Rossolini GM, van de Sande-Bruinsma N, Vatopoulos A, Žabicka D, Žemličková H, Monnet DL, Simonsen GS; EARS-Net participants. Strong correlation between the rates of intrinsically antibiotic-resistant species and the rates of acquired resistance in Gram-negative species causing bacteraemia, EU/EEA, 2016. *Euro Surveill*. 2019 Aug;24(33):1800538.
24. Ricardo Oliveira, Joana Castro, Sónia Silva, et al. Exploring the Antibiotic Resistance Profile of Clinical *Klebsiella pneumoniae* Isolates in Portugal. *Antibiotics* (Basel). 2022;11(11): 1613.
25. <https://chinets.com/Data/GermYear>
26. Núñez-Núñez M, Navarro MD, Palomo V, Rajendran NB, Del Toro MD, Voss A, Sharland M, Sifakis F, Tacconelli E, Rodríguez-Baño J; EPI-Net, Combacte-Magnet and EUCIC Group for SUSPIRE. The methodology of surveillance for antimicrobial resistance and healthcare-associated infections in Europe (SUSPIRE): a systematic review of publicly available information. *Clin Microbiol Infect*. 2018;24(2):105-109.
27. Pacha Venkataramana Geetha, Kayanam Vijaya Lalitha Aishwarya, Shanthi Mariappan et al. Fluoroquinolone Resistance in Clinical Isolates of *Klebsiella Pneumoniae*. *J Lab Physicians*. 2020; 12(2): 121–125.
28. Ali Al Bshabshe, Ahmed Al-Hakami, Basel Alshehri et al. Rising *Klebsiella pneumoniae* Infections and Its Expanding Drug Resistance in the Intensive Care Unit of a Tertiary Healthcare Hospital, Saudi Arabia. *Cureus* 12(8): e10060.
29. Gelareh Nasiri, Amir Peymani, Taghi Naserpour Farivar et al. Molecular epidemiology of aminoglycoside resistance in clinical isolates of *Klebsiella pneumoniae* collected from Qazvin and Tehran provinces, Iran. *Infect Genet Evol*. 2018; 64:219-224.
30. Popa MV, Bogdan Goroftei ER, Guțu C, Duceac M, Marcu C, Popescu MR, Druguș D, Duceac LD: Observational study of post-Covid-19 in Health Care Workers Infected with SARS-CoV-2 Virus: General and oral cavity complications, *Romanian Journal of Oral Rehabi*. 2023;3:198-207.
31. Duceac LD, Eva L, Dabija M, Ciuhodaru T, Guțu C, Romila L, Nazarie Prevention and Limitation of Coronavirus SARS-CoV-2 Cases in Hospitals and Dental Medicine Offices, *IJMD*. 2020; 24(2):149-156.
32. Botezatu C, Duceac LD, Mastalier B, Stafie L, Jităreanu CM, Luca AC, Țarcă E, Mitrea G, Iordache AC, Pătrașcu T. Hepatic cystic echinococcosis studied in family group *IJMD*. 2018;22(4):346-350.

33. Duceac M, Eva L, Dabija MG, Stoica MG, Druguş D, Duceac LD, Voinescu DC. Does oral health affect your bone loss? The link between periodontitis disease and osteoporosis – an epidemiological approach to lumbar disc herniation in a neurosurgery hospital in the northeastern region of Romania, *Romanian Journal of Oral Rehab.*2023;2:44-53.
34. Luchian N, Eva L, Dabija MG, Druguş D, Duceac M, Mitrea G, Marcu C, Popescu MR, Duceac LD. Health – associated infections in hospital in the North-East region of Romania – a multidisciplinary approach, *Romanian Journal of Oral Rehab* 2023; 2:219-229.
35. Popescu MR, Ilinca DA, Balcan RM, Novac O, Dobrovat BI, Boev M, Marciuc EA, Duceac LD, Temneanu OR, Haba D. Imaging Principles In Diagnosis Of Nasopharyngeal Lesions Using Computed-Tomography And Magnetic Resonance In Paediatric Patients. *Romanian Journal of Oral Rehab.*2023;15(4): 190-203.
36. Al Namat R, Duceac LD, Chelaru L, Dabija MG, Guţu C, Marcu C, Popa MV, Popa F, Bogdan Goroftei ER, Țarcă E. Post-Coronary Artery Bypass Grafting Outcomes of Patients with/without Type-2 Diabetes Mellitus and Chronic Kidney Disease Treated with SGLT2 Inhibitor Dapagliflozin: A Single-Center Experience Analysis. *Diagnostics (Basel).* 2023.21;14(1):16.
37. Popescu MR, Dobrovăţ BI, Marciuc EA, Istrate AC, Temneanu OR, Pomohaci D, L Duceac LD, Haba D: The role of the MRI Examination in the diagnosis and monitoring of tumoral lesions of the parotid gland. *Romanian Journal of Oral Rehab.*2023;14(2):54-58,
38. Hizanu M, Bogdan Goroftei ER, Popa F, Duceac M, Marcu C, Ciuhodaru MI, Druguş D, Duceac LD. Dental Disease in Children Diagnosed with Life-Limiting Diseases. Retrospective study. *Romanian Journal of Oral Rehab.*2023;3:160-172.
39. Stefanache A, Lungu II, Butnariu IA, Calin G, Gutu C, Marcu C, Grierosu C, Bogdan Goroftei ER, Duceac LD, Dabija MG, Popa F, Damir D. Understanding How Minerals Contribute to Optimal Immune Function. *J Immunol Res.* 2023 Nov 1;2023:3355733.
40. Duceac LD, Țarcă E, Ciuhodaru MI, Țănuţu MM, Bogdan Goroftei RE, Banu EA, Damir D, Glod M, Luca AC. Study of the mechanism of antibiotic resistance. *Rev.Chimie.*2019;70(1):199-201.
41. Duceac LD, Banu EA, Baciuc G, Lupu VV, Ciomaga IM, Țarcă E, Mitrea G, Ichim DL, Luca AC. Assessment of bacteria resistance according to antibiotics chemical structure. *Rev.Chimie.*2019;70(3): 906-908.
42. Duceac LD, Calin G, Eva L, Marcu C, Bogdan Goroftei ER, Dabija MG, Mitrea G, Luca AC, Hanganu E, Gutu C, Stafie L, Banu EA, Grierosu C, Iordache AC. Third-Generation Cephalosporin-Loaded Chitosan Used to Limit Microorganisms Resistance. *Materials (Basel).* 2020 Oct 27;13(21):4792.
43. Ichim DL, Duceac LD, Marcu C, Iordache AC, Ciomaga IC, Luca AC, Mitrea G, Bogdan Goroftei ER, Stafie L: Synthesis and Characterization of Colistin Intercalated Nanoparticles Used to Combat Multi-Drug Resistant Microorganisms, *Rev Chimie.*2019;70(10):3734-3737.
44. Duceac LD, Geta Mitrea, Elena-Ariela Banu, Mădălina- Irina Ciuhodaru, Irina-Mihaela Ciomaga, Daniela-Luminița Ichim, Constantin Marcu, Alina-Costina Luca: Synthesis and characterization of carbapenem based nanohybrids as antimicrobial agents for multidrug resistant bacteria, *Rev Mater Plastice* . 2019;6(2):388-391.
45. Duceac LD, Luca AC, Mitrea G, Banu EA, Ciuhodaru MI, Ciomaga IM, Ichim DL, Baciuc G. Ceftriaxone intercalated nanostructures used to improve medical treatment, *Rev.Mater.Plastice*, 2018;55(4):613-615.
46. Luca AC, Duceac LD, Mitrea G, Ciuhodaru MI, Ichim DL, Baciuc G, Banu EA, Iordache AC: Antibiotic encapsulated nanomaterial with application in medical area, *Rev. Mater.Plastice.*2018;55(4):552-554.
47. Grierosu C, Calin G, Damir D, Marcu C, Cernei R, Zegan G, Anistoroaei D, Moscu M, Carausu EM, Duceac LD, Dabija MG, Mitrea G, Gutu C, Bogdan Goroftei ER, Eva L. Development and Functionalization of a Novel Chitosan-Based Nanosystem for Enhanced Drug Delivery. *J Funct Biomater.* 2023 Nov 1;14(11):538.
48. Forna N, Damir D, Duceac LD, Dabija MG, Calin G, Ichim DL, Gutu C, Grierosu C, Eva L, Ciuhodaru MI, et al. Nano-Architectonics of Antibiotic-Loaded Polymer Particles as Vehicles for Active Molecules. *Applied Sciences.* 2022; 12(4):1998.

49. Maftai NM, Iancu AV, Elisei AM, Gurau TV, Ramos-Villarroel AY, Lisa EL. Functional Characterization of Fermented Beverages Based on Soy Milk and Sea Buckthorn Powder. *Microorganisms*. 2023 Jun 4;11(6):1493.
50. Maftai NM, Iancu AV, Goroftei Bogdan RE, Gurau TV, Ramos-Villarroel A, Pelin AM. A Novel Symbiotic Beverage Based on Sea Buckthorn, Soy Milk and Inulin: Production, Characterization, Probiotic Viability, and Sensory Acceptance. *Microorganisms*. 2023 Mar 13;11(3):736.
51. Duceac LD, Dobre CE, Păvăleanu I, Călin G, Nichituș S, Damir D. Diseases prevention by water defluoridation using hydrotalcites as decontaminant materials, *Rev. Chimie*. 2017;68(1):168-171.
52. Reygaert WC. An overview of the antimicrobial resistance mechanisms of bacteria. *AIMS Microbiol*. 2018 Jun 26;4(3):482-501.
53. Khan A, Miller WR, Arias CA. Mechanisms of antimicrobial resistance among hospital-associated pathogens. *Expert Rev Anti Infect Ther*. 2018 Apr;16(4):269-287.