Prevalence of Antibiotic Resistance of ESKAPE Pathogens Over Five Years in an Infectious Diseases Hospital from South-East of Romania

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Purpose: This study aimed at identifying the main antimicrobial resistance of ESKAPE (Enterococcus faecium, Staphylococcus aureus, Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa, Escherichia coli) pathogens in a Romanian infectious diseases hospital. This antimicrobial resistance is a global threat, having high rates of multidrug resistance and limited treatment options.

Patients and Methods: This retrospective study (2016–2020) assessed the antimicrobial resistance of ESKAPE pathogens isolated from the patient’s biological samples. The microbiological diagnosis was performed by classical culture methods. The antimicrobial susceptibility analysis used the Kirby–Bauer disk-diffusion method and the method of minimum inhibiting concentration with the automated Vitek, according to the CLSI (Clinical and Laboratory Standards Institute) standards.

Results: Included in this study were 4293 bacterial isolates: 67% Gram-negative bacilli, 31% Gram-positive cocci and 2% other morphotrientorial bacteria. ESKAPE pathogens were found in 97% of the bacterial isolates strains; E. coli (38.26%) and Staphylococcus aureus (26%) were the most prevalent. Most bacterial strains were isolated from urine cultures (45.6%), skin and soft tissue secretions/collections (35.9%) and also blood cultures (4.2%). Increased antimicrobial resistance was observed for methicillin-resistant Staphylococcus aureus (MRSA), extended spectrum beta-lactamase producing (ESBL) Enterobacteriales, carbapenem-resistant (CR) Pseudomonas aeruginosa, Acinetobacter baumannii and Klebsiella pneumoniae. No vancomycin resistance was found for Enterococcus faecium. The highest prevalence rates of multidrug resistance were found in methicillin-resistant Staphylococcus aureus (86.6%), Acinetobacter baumannii (36.8%), Pseudomonas aeruginosa (29.1%) and Klebsiella pneumoniae (24.4%).

Conclusion: ESKAPE pathogens are frequently isolated in the infectious diseases hospital, with main antimicrobial resistance: ESBL, MRSA and CR. The local antimicrobial resistance pattern is essential in updating the local protocols and for appropriately prescribing antibiotics. Streamlining microbiological diagnosis and aligning with the European standards for antimicrobial susceptibility testing are necessary steps in harmonizing the regional network for good antimicrobial resistance control practices.

Keywords: antimicrobial drug resistance, hospital, Romania, ESKAPE

Introduction

Antimicrobial resistance (AMR) is one of the most concerning public health problems globally, a fact that is due to its increasing impact on morbidity, mortality and care costs. Every year, over 700,000 deaths are associated with AMR.1,2 It is...
estimated that in the European Union (EU), in the year 2050, over 33,000 deaths will be caused by an antibiotic-resistant bacteria. AMR is a consequence of the natural evolution and adapting processes of bacteria, accelerated by the selection pressure which stems from the inadequate or excessive use of antibiotics. Multidrug resistance (MDR) is extending continuously across the globe and it is a challenge in treating infections, making it necessary to use the reserve antibiotics which can have higher cost-to-benefit ratios and a decreased security profile. Among MDR germs, “the ESKAPE pathogens” had the biggest impact on health care-associated infections, a group of six germs which have the capacity to elude the biocidal activity of antibiotics: Enterococcus faecium, Staphylococcus aureus, Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa, Escherichia coli. The ESKAPE group is distinguished by pathogenic, transmission and resistance traits, which are represented by enzyme inactivation, target change, cell permeability alteration through porin loss or by increasing the expression of efflux pumps, and mechanical protection through biofilm synthesis. The antiseptic activity of antibiotics can sometimes also contribute to disturbances in the skin microbiome of the face, even that which is linked to endosymbiotes; on the other hand, dermatology is a field which frequently makes use of topical glucocorticoids (as in psoriasis) which register many side effects and have the downside of predisposing the skin to possibly severe infections, some with MDR agents. Likewise, this antibacterial role can also be exerted on germs that are isolated from various isolated skin and mucous membrane diseases, or from systemic ones, either in vivo or in vitro. In the continuous fight against such Gram-negative pathogens, interleukin-6 (IL-6) seems to have an inhibititing effect, expressing antibacterial properties, an intriguing subject which summons further research. The development of new antibiotics is far from keeping the pace with MDR progression rate. Thus, preserving the available antibiotics’ actions through the rational use of antibiotics is a measure adopted as an objective within the programs of antibiotic stewardship and it is mandatory concerning the quality standards of hospital activity, in accordance with Global Action Plan on Antimicrobial Resistance developed by the World Health Organization (WHO) and also with the regulations of the European Antimicrobial Resistance Surveillance Network (EARS-Net), which has Romania as a member. The current COVID-19 (Coronavirus disease of 2019) pandemic context has changed the priorities of the medical activity by influencing antibiotic use with unknown consequences in the long run.

This study aimed to evaluate the evolution of the main antimicrobial resistance issues, as part of the stewardship antibiotic program in an infectious diseases hospital which is representative for the south-east region of Romania.

**Patients and Methods**

The current study was conducted in accordance with the Declaration of Helsinki. It is a retrospective type of study, extending from 2016 to 2020, that was done by analyzing the annual reports of the microbiology laboratory, within the antibiotic stewardship activities controlled by law in Romania in a specialized infectious disease and dermatovenereology hospital with 160 beds in the south-east region of this country. Community infections, along with medical assistance associated infections (as transfers from other multidisciplinary hospitals), were admitted.

This study evaluated the antibiotic sensibility of bacteria isolated from biological products taken for diagnostic purposes from hospitalized patients. The bacterial isolates were identified by using classical bacteriology methods for microbial culture, on solid media agar-based, followed by biochemical (multi-testing and automated ones – Vitek 2 Compact) and specific antigen testing of the obtained colonies. Duplicate samples were excluded. Antibiotic sensibility was tested by using the Kirby–Bauer disk diffusion-method and the minimal inhibiting concentration method – Vitek 2 Compact, the resistance to a certain antibiotic being interpreted according to CLSI (Clinical and Laboratory Standards Institute) standards which are renewed annually. ESBL (extended spectrum beta-lactamase) producing Enterobacteriaceae strains were identified by using the test of double diffusion, with the help of ceftoxime in combination with inhibitory clavulanic acid and the Vitek 2 Compact Software. MRSA (methicillin-resistant Staphylococcus aureus) was taken into consideration when the cefoxitin disk diffusion test was over 22 mm.

MDR was defined by being non-susceptible to at least 1 agent from at least 3 antimicrobial categories. The following antibiotic classes were used: penicillins (PEN – penicillin, AMC – amoxicillin clavulanate), cephalosporins (FEP – cefepime, CTX – cefotaxime, CAZ – ceftazidime, CXM – cefuroxime, TZP – piperacillin tazobactam), carbapenems (ERT – ertapenem, IPN – imipenem, MEM – meropenem), fluoroquinolones (CIP – ciprofloxacin, LVX – levofloxacin), aminoglycosides (AMK – amikacin, GM – gentamicin, TOB – tobramycin).
and tetracycline (TET – tetracycline, DOX – doxycycline), sulfonamide (SXT – sulfamethoxazole-trimethoprim).

The resistance analysis was focused on the ESKAPE germs which are monitored globally and regionally within the surveillance networks, and which are in the spotlight for stewardship antibiotic programs: Enterococcus spp., Staphylococcus aureus, Klebsiella spp., Acinetobacter spp., Pseudomonas spp., Enterobacter spp.

Results

The Evolution of Hospital Indicators

During 2016 and 2020, 24,328 patients were hospitalized from which 4293 bacterial strains were isolated. Presented in dynamic, the number of isolated strains diminished progressively from 2016 to 2019, having an even more abrupt decrease in the pandemic year of 2020, when 90% of the hospitalized patients had COVID-19 infection. In comparison to 2016, the number of admissions reduced by 33.4% in 2020, while the number of bacterial strains isolated decreased by 65%, meaning an isolation rate of almost 50% lower, in the context of a reduced diversity of analyzed biological products in viral-infected COVID-19 admitted patients, either as a suspected or confirmed infection (Figure 1).

The Types of Isolated Bacterial Species (Isolates)

During 2016 and 2020, 4293 bacterial strains were isolated, 67% being Gram-negative bacilli, 31% Gram-positive cocci and 2% other bacteria.

The ESKAPE group made up 97% (4166/4293) of the bacterial strains isolated from 2016 to 2020 (Table 1). On the order of frequency, E. coli strains were most prevalent (38.26%), along with S. aureus (26%), followed by Klebsiella spp. (9.55%), Pseudomonas spp. (8.78%), Proteus spp. (5.71%), Salmonella spp. (3.5%), Enterococcus spp. (3.55%). Enterobacter spp., other Enterobacteriaceae (Serratia spp., Citrobacter spp., Morganella spp., Providencia spp.) and Acinetobacter spp., played a limited part in the ESKAPE group. The annual frequency of the isolated strains was a relatively constant one in those 5 years (Table 2). Thus being said, in the pandemic year of 2020 there was a slight decreasing rate in the numbers of staphylococci and enterococci, while Acinetobacter spp. and Pseudomonas spp. registered a slight increase.

Most isolated bacterial strains were obtained from urine cultures (45.6%), from cutaneous secretions/collections (35.89%) and blood cultures (4.2%), and rarely from other biological products, such as sputum, otic, ocular, genital, pharyngeal or nasal secretions, vomiting, fecal matter, cerebrospinal fluid (Table 2). Strains isolated (isolates) from catheter cultures registered a frequency of 0.9%, and represented either contaminations or health care associated infections.

AMR of ESKAPE Germs

Enterococcus faecium

One hundred and forty-eight strains of Enterococcus spp. were isolated, but species identification was done from 2019, only for 26 cases, from which 2 were E. faecium and 24 E. faecalis. Ciprofloxacin (CIP) sensitivity was low in all species, ranging between 44.4% and 55.2%. E. faecalis’s sensitivity for ampicillin (AMP) was of 95.7%. VAN (vancomycin) and LNZ (linezolid) sensitivity was proven both for E. faecis and E. faecalis that were identified from 2019 to 2020, but during 2017 to 2018, 5
unspecified strains of Enterococcus spp. VAN-R (vancomycin-resistant) were isolated from urine cultures (Table 3).

**Staphylococcus aureus**

*Staphylococcus aureus* makes up 89.0% from staphylococcal isolates, most of them being obtained from infections located in the skin or soft tissues. The rate of MRSA had a decreasing tendency from 2016 to 2018, followed by an increase from 201 to 2020, when it reached its peak level of 46.2% (Figure 2). Methicillin resistance was correlated with a significantly increased rate of resistance as compared to other antibiotics, including VAN-R and MDR (Table 3).

### Table 1 Annual Frequency of ESKAPE Group Species

| Genera                | No-2016 | No-2017 | No-2018 | No-2019 | No-2020 | Total | %   |
|-----------------------|---------|---------|---------|---------|---------|-------|-----|
| Enterococcus spp.     | 62      | 27      | 39      | 12      | 8       | 148   | 3.55%|
| Staphylococcus spp.   | 282     | 229     | 249     | 236     | 91      | 1087  | 26%  |
| Klebsiella spp.       | 96      | 94      | 92      | 75      | 41      | 398   | 9.55%|
| Acinetobacter spp.    | 5       | 5       | 5       | 9       | 4       | 28    | 0.67%|
| Pseudomonas spp.      | 88      | 83      | 67      | 82      | 46      | 366   | 8.78%|

**Enterobacteriaceae**

| Genera                     | No-2016 | No-2017 | No-2018 | No-2019 | No-2020 | Total | %       |
|----------------------------|---------|---------|---------|---------|---------|-------|---------|
| Escherichia coli           | 372     | 414     | 383     | 313     | 112     | 1594  | 38.26%  |
| Proteus spp.               | 65      | 68      | 54      | 29      | 22      | 238   | 5.71%   |
| Enterobacter spp.          | 25      | 22      | 22      | 18      | 13      | 100   | 2.40%   |
| Salmonella spp.            | 35      | 42      | 31      | 25      | 13      | 146   | 3.50%   |
| Other Enterobacteriaceae   | 16      | 21      | 9       | 12      | 3       | 61    | 1.46%   |

| Genera                     | No-2016 | No-2017 | No-2018 | No-2019 | No-2020 | Total | %       |
|----------------------------|---------|---------|---------|---------|---------|-------|---------|
| Enterobacteriaceae         | 1089    | 1022    | 967     | 839     | 376     | 4293  | 100%    |

**Note:** % annual frequency.

### Table 2 ESKAPE Distribution According to the Biological Product Analyzed (2016–2020)

| Urine Culture                | N   | %    | Wound or Abscess   | N   | %    | Blood Culture   | N   | %    | Catheter Culture | Others | TOTAL |
|-------------------------------|-----|------|--------------------|-----|------|-----------------|-----|------|------------------|--------|-------|
| Enterococcus spp.             | 125 | 84.4%| 16                 | 10.8%| 4    | 2.7%           | 3   | 2.0%| 0                | 0      | 148   |
| Enterococcus spp. untyped     | 106 | 86.8%| 12                 | 9.8% | 2    | 1.6%           | 2   | 1.6%| –                | –      | 122   |
| Enterococcus faecium          | 0   | –    | 1                  | 9.8% | 1    | –              | 0   | –    | –                | –      | 2     |
| Enterococcus faecalis         | 19  | 79.1%| 3                  | 12.5%| 1    | –              | 1   | –    | –                | –      | 2     |
| Staphylococcus spp.           | 6   | 0.5% | 741               | 68.1%| 88   | 8.1%           | 35  | 3.2%| 217              | 19.9%  | 1087  |
| Staphylococcus aureus         | 5   | 0.5% | 724               | 74.8%| 24   | 2.4%           | 23  | 2.3%| 197              | 20.3%  | 968   |
| Klebsiella spp.               | 244 | 61.3%| 80                | 20.1%| 18   | 4.5%           | 0   | 0    | 56               | 14.4%  | 398   |
| Klebsiella pneumoniae         | 159 | 77.9%| 52                | 25.4%| 13   | 6.3%           | 0   | 0    | 34               | 16.6%  | 258   |
| Acinetobacter spp.            | 3   | 10.7%| 11                | 39.2%| 2    | 7.1%           | 2   | 7.1%| 10               | 35.7%  | 28    |
| Acinetobacter baumannii       | 2   | 10.5%| 10                | 52.6%| 2    | 10.5%          | 2   | 10.5%| 3                | 15.7%  | 19    |
| Pseudomonas spp.              | 44  | 12%  | 299               | 81.7%| 0    | 0              | 0   | 0    | 23               | 6.2%   | 366   |
| Pseudomonas aeruginosa        | 39  | 19.1%| 151               | 74.0%| 0    | 0              | 0   | 0    | 14               | 6.8%   | 204   |

**Enterobacteriaceae**

| Genera                     | N   | %    | Urine Culture | Wound or Abscess | Blood Culture | Catheter Culture | Others | TOTAL |
|----------------------------|-----|------|---------------|------------------|---------------|-------------------|--------|-------|
| Escherichia coli           | 1346| 84.4%| 98            | 6.14%            | 53            | 3.3%              | 0      | 97    | 1594 |
| Proteus spp.               | 82  | 34.4%| 145           | 60.9%            | 3             | 1.2%              | 1      | 0.4% | 7    | 238 |
| Enterobacter spp.          | 38  | 38.0%| 50            | 50.0%            | 2             | 2%                | 0      | 0    | 10   | 100 |
| Salmonella spp.            | 0   | 0    | 0             | 0                 | 3             | 2%                | 0      | 0    | 143  | 97.9%| 146 |
| Other Enterobacteriaceae   | 12  | 19.6%| 26            | 42.6%            | 2             | 3.2%              | 0      | 0    | 21   | 34.4%| 61  |

| Total                      | 1900| 45.6%| 1466          | 35.89%           | 175           | 4.2%              | 41     | 0.9% | 561  | 13.4%| 4166|

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Klebsiella pneumoniae

*Klebsiella pneumoniae* sensitivity is reduced in more than one strains for SXT, CIP, GM, MEM, but especially for beta-lactamines, registering a 7.6% resistance to carbapenems and a 24.4% MDR. ESBL frequency varied between 33% (2018) and 20% (2019), with an increasing trend in the pandemic year of 2020 (Table 4).

Acinetobacter baumannii

Nineteen strains of *Acinetobacter baumannii* were identified, being isolated from cutaneous infections, urine cultures, blood cultures, other airway secretions, and also two from catheter cultures. Significant resistant strains were found for cephalosporins, quinolones, and aminoglycosides, with a rate of 36.8% MDR. From 2016 to 2019, almost half of *Acinetobacter baumannii* strains were found to be resistant to carbapenems (8/17), but in 2020, only two strains were isolated, both sensitive to meropenem.

Pseudomonas aeruginosa

Two hundred and four strains of *Pseudomonas aeruginosa* were identified, most of them from skin infections, with significant resistance to all tested antibiotics, with a carbapenems-resistance of 44.8% and an MDR of 29.1%. Annual variations in carbapenems resistance were registered, with an increasing rate from 9% in 2016, to 60.8% in 2019, and again a decrease to 31.1% in the pandemic year of 2020.

Enterobacteriaceae

Each year, *E. coli* was the most frequent germ isolated in the microbiology laboratory, 84% of cases being found in urine cultures. *E. coli* frequency was of 9.8%, while ESBL was maintained in between 6% and 8% during the timeframe between 2016 and 2020, increasing in the pandemic year of 2020 (Figure 3).

From 2016 to 2020, MDR prevalence for Proteus spp. was of 15.1%, similar to that of ESBL, of 15.7%. Observations were made and it was found that ESBL prevalence is more elevated in comparison to MDR for *Enterobacter spp.*, 14.7%

### Table 3 Susceptibility and MDR of Gram-Positive Cocci from ESKAPE Group

|                       | OXA | AMP | CC  | CIP | ERY | GM* | LNZ | TEC | VAN | SXT | MDR |
|-----------------------|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| *Enterococcus spp. untyped* | IR  | 80  | 55.2| –   | –   | 100 | –   | 93.3| –   | 80  | 0   |
| *Enterococcus faecium*  | IR  | IR  | 50  | IR  | IR  | 100 | –   | 100 | IR  | 0   | 0   |
| *Enterococcus faecalis* | IR  | 95.7| 44.4| IR  | IR  | 100 | –   | 100 | IR  | 0   | 0   |
| *Staphylococcus aureus* | 64.3| IR  | 72.6| 32.9| 77.6| 99.7| 87.8| 85.7| 86  | 41.8| 0   |
| MSSA                   | IR  | 62.6| 86.8| 47.8| 89.2| 99.7| 88.9| 77.8| 90.4| 16.5| 0   |
| MRSA                   | IR  | 21.1| 46.9| 5.7 | 56  | 99.6| 86.4| 100 | 78.3| 86.6| 0   |

*Note:* *OXA*, oxacillin; *AMP*, ampicillin; *CC*, clindamycin; *CIP*, ciprofloxacin; *ERY*, erythromycin; *GM*, gentamicin; *LNZ*, linezolid; *TEC*, teicoplanin; *VAN*, vancomycin; *SXT*, sulfamethoxazole-trimethoprim; *MDR*, multidrug resistance; *MSSA*, methicillin sensible *Staphylococcus aureus*; *MRSA*, methicillin resistant *Staphylococcus aureus*.

### Figure 2 MRSA rate progression (2016–2020).
versus 21.3% and for other Enterobacteriaceae (Citrobacter spp., Morganella spp., Serratia spp.), 22.2% versus 28.8%, with an increasing rate in 2020.

Salmonellas maintain their sensitivity to cephalosporins, carbapenems; 95% of cases are sensitive to SXT, while AMP resistance develops in one in four Enterococcus strains, and CIP and GM resistance develops in 22% of isolates.

Discussion
The pandemic year 2020 had a particular profile of bacterial reporting and antibiotic resistance in the infectious diseases hospital by reducing the number of isolates analyzed. Enterococcus faecium and Acinetobacter baumannii did not raise any resistance problems during this year, and Pseudomonas aeruginosa strains with CR decreased. In the same timeframe, the frequency of MRSA, ESBL and MDR resistance increased for Enterobacter spp. and Klebsiella pneumoniae.

Each year, between 2016 and 2020, bacteria genre from the ESKAPE pathogens group made up the majority of the hospital's isolated germ distribution. The laboratory’s difficulties concerning microbiological diagnosis by classical methods and changes in CLSI antibiotic sensitivity testing standards by annual update have a limiting effect in the multi-annual analysis of hospital data. The low number of blood cultured isolated strains in the course of invasive infections and the different standards used for

| Table 4 Susceptibility and MDR of Gram-Negative Bacilli from the ESKAPE Group |
|-------------------|---|---|---|---|---|---|---|---|---|---|
|                  | AMC | CXM | CTX | CAZ | FEP | TZP | ERT | MEM | CIP | GM | SXT | MDR |
| Acinetobacter baumannii | *IR* | NT# | NT | 27.3 | 38.5 | 66.7 | *IR* | 57.1 | 47.4 | 55.6 | NT | 36.8 |
| Pseudomonas aeruginosa | *IR* | NT/IR* | NT/IR* | 66.5 | 60.3 | 73.7 | *IR* | 55.2 | 64.8 | 61.6 | NT/IR* | 29.1 |
| Klebsiella pneumoniae | 50.4 | 63.7 | 66.1 | 22.2 | 38.7 | 72.5 | 43.3 | 73.2 | 63 | 74.1 | 60.4 | 24.4 |

Enterobacteriaceae

| Species                  | AMC | CXM | CTX | CAZ | FEP | TZP | ERT | MEM | CIP | GM | SXT | MDR |
|--------------------------|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Escherichia coli         | 60.2 | 85.2 | 87.3 | NT  | 77.8 | 91.6 | 95.7 | 96  | 75.3 | 91.5 | 64.3 | 9.8  |
| Proteus spp.             | 57  | 68.4 | 82.3 | NT  | 66.3 | 93.8 | 98  | 100 | 73.8 | 68.5 | 44.3 | 15.1 |
| Enterobacter spp.        | 15.7 | 34.9 | 70  | NT  | 81.8 | 87  | 80  | 80  | 33.3 | 82.2 | 78.5 | 70   |
| Salmonella spp.          | 75  | NT  | 100** | NT  | NT  | NT  | 100 | 100 | 77.2 | 77.8 | 95   | 4    |
| Other Enterobacteriaceae | 14.9 | 43.5 | 63.3 | NT  | 50  | 89.3 | 87.5 | 73.3 | 72.5 | 75.6 | 62   | 22.2 |

Notes: **For Salmonella spp. 1st and 2nd generation cephalosporins may appear to be as such in vitro, but are not effective clinically and should not be reported as susceptible.

Abbreviations: *IR, intrinsic resistance; #NT, not tested; AMC, amoxicillin clavulanate; CXM, cefuroxime; CTX, cefotaxime; CAZ, ceftazidime; FEP, cefepime; TZP, piperacillin- tazobactam; ERT, ertapenem; MEM, meropenem; CIP, ciprofloxacin; GM, gentamicin; SXT, sulfamethoxazole-trimethoprim; MDR, multidrug resistance.

Figure 3 Escherichia coli ESBL+ progression rate (2016–2020).
reporting in the European space – the European Committee on Antimicrobial Susceptibility Testing (EUCAST) – also limit the possibility of comparing the obtained data with the level of ESKAPE resistance within the surveillance networks from the EU, part of which many multi-disciplinary hospitals from Romania are.\textsuperscript{3,33–35}

The resistance profile for the ESKAPE germs isolated in the infectious diseases hospital from various biological products did not register significant statistical annual variations during the analyzed timeframe, although increasing tendencies in MRSA, ESBL and MDR rates were observed. VAN-R and CR prevalence is still low, but the inconsistent identification of species from these bacterial genes and the low number of strains analyzed annually do not support an adequate statistical analysis.

Romanian national data concerning AMR progression inside hospitals are limited to reports of blood culture strains in invasive infections, within the ECDC (European Centre for Disease Prevention and Control) network, which is done only by several multidisciplinary university hospitals which are not representative regionally. According to these data, Romania is one of the leading countries in Europe concerning AMR of invasive infections isolated bacteria: \textit{Klebsiella pneumoniae} intermediate resistance to carbapenems 35.9\%, \textit{Pseudomonas aeruginosa} CR 52\%, \textit{Acinetobacter spp.} multi-resistant to carbapenems, aminoglycosides and quinolones 82.9\%, \textit{Staphylococcus aureus} MRSA 50.5\%, \textit{Enterococcus faecium} VAN-R 39\%.\textsuperscript{3,34} AMR is an increasing concern not only in Europe, but in the Middle-East countries as well; studies carried out by Abbasi et al and Mansour et al have reported frequent MDR concerning \textit{E. coli} and coagulase-negative staphylococci in Iran. \textit{E. coli} MDR strains were found in high frequency (96.9\%), mostly being resistant to AMP, cephalosporins and co-trimoxazole; coagulase-negative staphylococci also registered a high MDR frequency, being found resistant to 8 antibiotics, with the highest resistance rate to ERY (84.1\%).\textsuperscript{36,37}

A study done by the European Public Health Alliance identifies Romania to be “in the red zone” in what concerns the excessive use of antibiotics and the prevalence of antibiotic resistant bacteria, to which insufficient funding, insufficient medical equipment and technologies and the lack of qualified medical personnel contribute.\textsuperscript{38} Although these challenges are common to other European states, there are major differences between the resources and capacities of the national health systems, thus explaining “the red zones” within the ECDC network.\textsuperscript{39} Furthermore, these resources have an uneven distribution nationally, a fact reflected in the AMR data between different regions of Romania.\textsuperscript{34} Such challenges are present in the Romanian health system not only concerning MDR but also cancer diagnosis and treatment, associated high-risk diseases and treatment side effect risk, being major stressors not only for treating physicians but also for the patients, whom not rarely are of young age (such being the case of spiradenocarcinoma or AIDS (acquired immunodeficiency syndrome) patients) or can develop side effects even to commonly used drugs (such as beta-blockers); adequate treatment is thus an issue with consequent severe psychological impact.\textsuperscript{40–43}

ESKAPE AMR evaluated in this study is lower than other resistances reported nationally in invasive infections, but it reflects their prevalence and persistence within the community. Pursuit for superbug surveillance in the infectious diseases hospital must develop by increasing the quality and accuracy of the identifying methods used, by adopting the EUCAST standards, increasing the clinical vigilance for infectious diagnosis and justified use of antibiotics.

\textbf{Conclusion}

ESKAPE pathogens are the majority of germs isolated in the infectious diseases hospital, the main AMR problems being ESBL and MRSA. Recognition of AMR is essential in understanding this phenomenon with the aim of updating the local protocols for antibiotic prescription. Streamlining the microbiological diagnosis and aligning with the European antimicrobial resistance testing are necessary steps in harmonizing the regional network for good AMR control practices.

\textbf{Abbreviations}

AMR, antimicrobial resistance; EU, the European Union; MDR, multidrug resistance; ESKAPE pathogens – \textit{Enterococcus faecium} – \textit{Staphylococcus aureus} – \textit{Klebsiella pneumoniae} – \textit{Acinetobacter baumannii} – \textit{Enterobacter spp}; IL-6, interleukin-6; WHO, World Health Organization; EARS-Net, The European Antimicrobial Resistance Surveillance Network; COVID-19, Coronavirus Disease of 2019; CLSI, Clinical and Laboratory Standards Institute; ESBL, Extended-Spectrum Beta-lactamase Producers Enterobacteriaceae; MRSA, Methicillin resistant \textit{Staphylococcus aureus}; PEN, penicillin; AMC, amoxicillin-clavulanate; FEP, cefepime; CTX, cefotaxime; CAZ, ceftazidine; CXM, cefuroxime; TZP, pipercillin-
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