Gram negative bacterial bloodstream infections: resistance patterns and risk factors

Infecţii ale torentului circulator cu bacili Gram negativi: tipuri de rezistenţă şi factori de risc

Adriana Hristea*[1*], Ioana D. Olaru[1], Olga Dorobat[1], Maria Nica[2], Mihaela Ion[1], Ruxandra Moroti[1], Victoria Arama[1], Alexandru Rafila[1], Emanoil Ceausu[2], Lee W. Riley[3]

1. Institutul National de Boli Infectioase "Prof Dr Matei Bals"
2. Spitalul "Dr. Victor Babes"
3. University of California, Berkeley

Abstract

Objectives: *(1) To describe the resistance patterns of Gram negative bacilli (GNB) isolated from blood cultures and (2) identify the epidemiological characteristics associated with multi-drug resistance (MDR).* ***Materials and methods:*** *We performed a retrospective study of the susceptibility profile of GNB strains isolated from blood stream infections (BSI) from patients admitted to two tertiary infectious diseases facilities between January 2009 and January 2011. Bacteria were isolated in BacT/ALERT and identified by classic techniques or automated methods. Susceptibility testing was done by disk-diffusion and automated methods according to CLSI guidelines. Clavulanic acid synergy test was performed to identify extended spectrum beta-lactamase (ESBL) production. Multidrug-resistance (MDR) was defined as resistance to 3 or more classes of antimicrobial agents.* ***Results:*** *From 116 patients, 222 non-duplicate isolates were identified. The isolates included: 120 E. coli (54%), 38 Klebsiella spp. (17%), 16 P. aeruginosa (7%), 11 Acinetobacter spp. (5%), 30 other Enterobacteriaceae (14%) and, 7 other non-fermenters (3%). The prevalence of resistance to one, two, and three classes of drugs were 17.6%, 18.3% and 31.7%, respectively. ESBL production was detected in 12 (10%) of E. coli and 10 (26%) of Klebsiella spp. Carbapenem resistance was identified in 8 (73%) of Acinetobacter spp. and 10 (63%) of P. aeruginosa isolates as compared with 4 (2.1%) in Enterobacteriaceae isolates. Clinical and epidemiological data were available for 142 (64%) patients. The primary source of bacteraemia were suspected to include: urinary tract (43%), gastrointestinal (13%), pulmonary (11%), others (6%), and unknown sites (27%). BSI were hospital associated (HA), health care associated (HCA) and community acquired (CA) in 50.7%, 17.6% and 31.7%, respectively. There was no statistical difference regarding the origin, underlying conditions and epidemiology of the three different epidemiological types of BSI. MDR was more frequently seen in HA (37.5%) and HCA infections (44%) than in CA (15.6%). Prior hospitalization, recent urinary tract infection, antimicrobials use, recent surgery and death were each associated with MDR BSI. In multivariate analysis, only prior use of antimicrobials was associated with MDR BSI (p=0.001).* ***Conclusions:*** *High MDR prevalence (31.7%) was observed in patients with BSI in our study. Combined resistance to third-generation cephalosporins, aminoglycosides, and quinolone was considerably higher than that reported from other European countries, especially in patients with hospital associated infections. Antimicrobial use within the past 3 months was associated with MDR BSI in our patient population.*

Keywords: *Bloodstream infections, Gram negative bacilli, multi-drug resistance*

[*]**Corresponding author:** Adriana Hristea adriana_hristea@yahoo.com, Str. Calistrat Grozovici, nr.1, sector 2 021105 Bucharest, Romania. Phone: +40213186100 ext 5204 or 0723562370, Fax: +40213186090

Rezumat

Obiective: *(1) descrierea rezistenței bacililor Gram negativi (BGN) izolați din hemoculturi şi (2) identificarea caracteristicilor asociate cu multirezistența.* ***Material şi metode:*** *Am realizat un studiu retrospectiv privind profilul de susceptibilitate a BGN izolați din hemoculturi de la pacienții internați în doua spitale de boli infecțioase, unități terțiare de îngrijire, în perioada ianuarie 2009-ianuarie 2011. Bacteriile au fost izolate în BacT/Alert şi identificate prin tehnici clasice sau metode automatizate. Testarea susceptibilității la antimicrobiene s-a realizat prin metoda difuzimetrică şi metode automatizate conform recomandărilor CLSI. Pentru identificarea betalactamazelor cu spectru extins (ESBL) s-a folosit testul sinergiei cu acid clavulanic. Multirezistența a fost definită ca rezistență la 3 sau mai multe clase de antimicrobiene.* ***Rezultate:*** *De la 116 de pacienți s-au obținut 222 de tulpini. Acestea includeau 120 E.coli (54%), 38 Klebsiella spp (17%), 16 P. aeruginosa (7%), 11 Acinetobacter (5%), 30 de alte Enterobacteriaceae (13%) şi 7 alți germeni non-fermentativi (3%). Prevalența rezistenței la una, două sau trei clase de antimicrobiene a fost 17,6%, 18,3% şi respectiv 31,7%. Producția de ESBL a fost detectată la 12 (10%) E.coli şi 10 (26%) Klebsiella spp. Rezistența la carbapeneme a fost identificată la 8 (73%) Acinetobacter şi 10 (63%) din tulpinile de piocianic, în comparație cu 4 (2,1%) din tulpinile de Enterobacterii. Pentru 142 de pacienți (64%) am putut colecta date clinice şi epidemiologice. Sursa primară a bacteriemiei a fost: tractul urinar (43%), gastrointestinal (13%), pulmonar (11%) şi altele (6%), iar in 27 % a fost neprecizată. Bacteriemiile au fost dobândite în spital (50,7%), asociate ingrijirilor de sănătate (17,6%) şi dobândite în comunitate (31,7%). Nu a existat nici o diferență statistică privind originea, comorbiditatile şi epidemiologia celor trei tipuri de bacteriemii. Multirezistența a fost mai frecvent semnalată în infecții nozocomiale (37,5%) şi infecții asociate îngrijirilor de sănătate (44%) decât în infecții comunitare (15,6%). Spitalizarea anterioară, infecțiile urinare recente, administrarea de antimicrobiene, chirurgia recentă şi mortalitatea mai mare au fost asociate cu bacteriemiile cu germeni multirezistenți. In analiza multivariată numai utilizarea de antimicrobiene în antecedente a fost asociată cu bacteriemiile cu germeni multirezistenți (p=0,001).* ***Concluzii:*** *La pacienții cu bacteriemie din studiul nostru s-a observat o prevalență ridicată (31,7%) a multirezistenței. Rezistența combinată la cefalosporine de generația a treia, aminoglicozide şi chinolone a fost mai mare decat cea raportată de alte țări Europene, mai ales la pacienții cu infecții nozocomiale. Bacteriemiile cu germeni multirezistenți s-au asociat cu utilizarea de antimicrobiene în ultimele 3 luni.*

Cuvinte cheie: *bacteriemii, Bacili Gram negativi, multirezistența*

Introduction

Bloodstream infections (BSI) are an important cause of morbidity and mortality not only in patients with health care associated infections (1, 2), but also in patients with community acquired infections. BSI have been classified into community acquired (CA), healthcare-associated (HCA) and hospital associated (HA) infections (3, 4). HCA infections include those with significant recent healthcare contact and procedures (5-7). Gram-negative organisms are major causes of community-acquired bacteraemia. In hospitals in the USA, infections due to Gram-positive bacteria are more commonly reported than those due to Gram-negative bacteria (8). However, a recent meta-analysis reported that Gram-negative bacilli (GNB) are the most common nosocomial isolates in developing countries (9).

The choice of an effective antimicrobial treatment should be guided by local epidemiologic data on resistance, based on continuous surveillance, since in both community- and hospital-acquired BSI, the prevalence of antimicrobial resistance has continued to increase over the last decades (10). In Europe the reported data for 2009 showed a significant increase of antimicrobial resistance in E. coli invasive isolates and an overall increase in BSI caused by this microorganism (11).

In spite of the participation of Romania in the European Antimicrobial Resistance Surveillance System (EARSS) network during the last years, the data regarding resistance in Romania are based on a small number of isolates. Hence, we conducted a retrospective study in two tertiary care hospitals in Bucharest to de-

scribe the resistance patterns of GNB isolated from BSI. This study is designed to complement the existing data and guide clinicians in choosing an effective empiric antimicrobial therapy.

Objective

The study aimed to describe the susceptibility profile of GNB strains isolated from BSI from patients admitted to two tertiary infectious diseases facilities between January 2009 and January 2011. Demographic, clinical, and epidemiologic characteristics associated with multidrug resistance (MDR) were also assessed.

Materials and Methods

Blood cultures were obtained from peripheral veins, as part of routine care for admitted patients. When different episodes of bacteraemia occurred in the same patient, we included only those that yielded different bacterial species by culture.

Bacteria were isolated in BacT/ALERT and identified by classic techniques or automated methods: miniAPI, VITEK2C. The susceptibility tests were done by disk-diffusion and automated methods (CMI-ATB Expression/BioMerieux, VITEC2C, E-test) according to CLSI guidelines (12,13). Clavulanic acid synergy test was done to identify extended spectrum beta-lactamase (ESBL) production. Twenty-four antimicrobial agents were tested, including aminoglycosides, beta-lactams +/- beta-lactamase inhibitors, carbapenems, cephems, monobactams, quinolones, cyclines, trimetoprim-sulfametoxazol and in selected cases colistin. For internal quality control we used E. coli ATCC 25922, P. aeruginosa ATCC 27853. MDR was defined as resistance to 3 or more classes of antimicrobial agents.

The data collected included: setting of infection; demographic characteristics; underlying diseases; source of BSI; prior antimicrobial use; major surgery and history of urinary tract infection in the preceding 3 months of the BSI episode; outcome (cured, deceased, discharged).

The BSI episode was defined as HA if the first positive blood culture occurred more than 48h after the current admission; all other episodes were considered community-onset and subsequently classified as HCA or CA. The infections were considered as HCA when any of the following was present: hospitalization for > 2 days in an acute care hospital in the 90 days preceding the BSI; or haemodialysis in the 30 days before the BSI. The remaining episodes were classified as community acquired. The BSI source was established based on clinical and laboratory data.

Statistical analysis

In order to identify the epidemiological characteristics associated with MDR in our study population, we tested for differences between patients with and without MDR using the chi-square test for categorical variables and the Mann-Whitney U test for continuous variables. Statistical analysis was performed with SPSS v17.0.

Results

Predisposing features, sources and outcome according to setting of infection

From 116 patients, 222 non-duplicate isolates were identified and tested for antimicrobial susceptibility. The most common gram-negative bacilli isolated from blood specimens included *E. coli* (120 isolates) and *K. pneumoniae* (38 isolates) which made up 71.2% of all isolates.

For 142 patients clinical and epidemiological data in addition to antimicrobial susceptibility test results were available. The demographic and clinical data of patients with BSI are summarized in *Table 1*. The ratio of male to female patients was highest for individuals with CA and HCA episodes.

There were 31.7% CA BSI, 17.6% HCA BSI and 50.7% HA BSI; the strains isolated are shown in *Table 2*. Bacteraemias were polymicrobial in 3 HCA and 2 CA infections. As expected, MDR was more frequently seen in HA (37.5%) and HCA infections (44%) than in CA (15.6%) *(Table 3)*.

Table 1. Patient characteristics, clinical features and mortality of BSI by epidemiological type of infection

	Community acquired N=45 (31%)	Health care associated N=25 (18%)	Hospital associated N=72 (51%)	Total N=142
Median age, (range)	62 (20-90)	70 (1-89)	58 (1-90)	61 (1-90)
Male gender	25 (55.5%)	19 (76%)	32 (44.4%)	76 (53.5)
Underlying disease	33 (73.3%)	23 (92%)	55 (76.4%)	111/142 (78.1%)
Chronic renal disease	6 (13.3%)	8 (32%)	1 (1.4%)	15/142 (10.5%)
Chronic liver disease	13 (28.8%)	2 (8%)	15 (20.8%)	30/142 (21.1%)
Solid cancer	3 (6.6%)	4 (16%)	6 (8.3%)	13/142 (9.1%)
Hematological cancer	3 (6.6%)	4 (16%)	8 (11.1%)	15/142 (10.5%)
HIV infection	7(15.5%)	0	3 (4.1%)	10/95 (10.5%)
Alcohol abuser	7 (15.5%)	1 (4%)	7 (9.7%)	15/134 (11.1%)
Diabetes	10 (22.2%)	10 (22.2%)	16 (22.2%)	36/142 (25.3%)
Vascular disease*	9 (20%)	6 (24%)	15 (20.8)	30/142 (21.1%)
Recent surgery (the preceding 3 months) #	8 (17.7%)	12 (48%)	17 (23.6)	37/142 (26%)
Recent antimicrobial use	7 (15.5%)	12 (48%)	46 (63.8)	65/107 (60.7%)
Recent history of urinary tract infection	2 (4.4%)	6 (24%)	20 (27.7)	28/91 (30.7%)
Mortality	7 (15.9%)	2 (8%)	13 (18%)	22/142 (15.5%)
Source				
Urinary	22 (48.8%)	14 (56%)	25 (34.7)	61 (43)
Respiratory/Pulmonary	5 (11.1%)	2 (8%)	9 (12.5%)	16 (11)
Intra-abdominal	2 (4.4%)	2 (8%)	14 (19.4%)	18 (12.6)
Others	2 (4.4%)	1 (4%)	6 (8.3%)	9 (6.3)
Unknown	14 (31.1%)	6 (24%)	18 (25%)	38 (26.7)

*coronary heart disease, stroke, peripheral arterial disease; # $p<0.05$

Most BSI were in older patients (median age of 61 years), with associated co-morbid conditions (78.1%), including diabetes (25%), solid or hematological cancers (19%), or with recent surgery in the preceding 3 months (26%). The most common identified source of BSI was urinary tract infection (43%). Corticotherapy was given in 7 (4.9%) patients, and neutropenia and COPD were each reported in 4 (2.8%), travel abroad occurred in 6 (8.9%) and the use of illegal drugs in one patient. With the exception of recent surgery, which was more frequently associated with HCA, there were no statistically significant differences regarding bacteraemia source, underlying conditions between the three different epidemiological types of BSI. Nevertheless, intra-abdominal source of the BSI, recent antimicrobial use and recent history of urinary tract infections were more commonly found in HA and HCA (*Table 1)*.

Antimicrobial susceptibility testing

The resistance patterns of the 222 GNB isolated from BSI is shown in *Table 4*. In *E. coli* and *K. pneumoniae*, resistance prevalence was high except for carbapenems. *E. coli* isolates were more resistant to one and two antimicrobial drug classes (75% and 58%) than *K. pneumoniae* strains, which were more frequently multidrug resistant (39% vs 29%). ESBL producing strains were more frequent among *K. pneumoniae* than among *E. coli* (26.3 vs 10%) (*Table 5 and 6*). The presence of ESBL was suspected in 32 (20.3%) isolates and confirmed in 12 (10%) of *E. coli* (10%) and 10 (26%) of *Klebisella spp.* (26.3%) isolates. Combined resistance to two, and three classes of antimicro-

Table 2. The most common microorganisms isolated from BSI

	Community acquired N=45 (31%)	Health care associated N=25 (18%)	Hospital associated N=72(51%)	Total N=142	p-value
E. coli	29 (64.4%)	15 (60%)	36 (50%)	80 (56.3%)	0.186
Klebsiella spp	5 (11.1%)	7 (28%)	14 (19.4%)	26 (18.3%)	0.002
P. aeruginosa	3 (6.6%)	3 (12%)	5 (6.9%)	11 (7.7%)	0.227
Acinetobacter spp	1 (2.2%)	0	5 (6.9%)	6 (4.2%)	0.219
Other *Enterobacteriaceae*				15 (10.5%)	0.607
Salmonella spp	2 (4.4%)	0	2 (2.7%)	4 (2.8%)	
Enterobacter spp	2 (4.4%)	0	4 (5.5%)	6 (4.2%)	
Proteus mirabilis	1 (2.2%)	0	2 (2.7%)	3 (2.1%)	
Morganella spp	1 (2.2%)	0	0	1 (0.7%)	
Kluyvera	0	0	1 (1.3%)	1 (0.75)	
Other non-fermenters				3 (2.1%)	NA
Stenotrophomonas	0	0	3 (4.1%)		

p values calculated for CA vs. HCA and HA infections

Table 3. Resistance according to setting of infection

	Community acquired N=45 (31%)	Health care associated N=25 (18%)	Hospital associated N=72 (51%)	Total N=142	p-value	OR (95% CI)
Fully susceptible	20 (44.4%)	3 (12%)	23 (31.9)	46 (32.4%)	0.037	0.46 (0.22-0.96)
Resistance to one class	8 (17.8%)	5 (20%)	12 (16.7%)	25(17.6%)	0.971	0.98 (0.39-2.48)
Resistance to two antimicrobials classes	10 (22.2%)	6 (24%)	10(13.9%)	26(18.3 %)	0.412	0.69 (0.29-1.67)
Resistance to at least three antimicrobials classes (MDR)	7 (15.6%)	11 (44%)	27(37.5%)	45 (31.7%)	0.005	3.5 (1.42-8.63)
ESBL	0	5 (20%)	9(12.5)	14(13.1%)	0.002	13.08 (1.68-102.02)

p values and OR calculated for CA vs. HCA and HA infections; * for *E. coli* and *Klebsiella spp*

bial agents (aminopenicillin, fluorquinolones, aminoglycosides) is shown in *Table 7.*

Epidemiological and clinical characteristics associated with multi-drug resistance (MDR)

The in-hospital mortality was higher among MDR BSI patients (25,6%) than among others (12,9%), but the difference was not statistically significant. The following variables were found to be significant in univariate analysis for MDR vs. non-MDR BSI: recent history of urinary tract infection (p-value, OR, 95% CI); antibiotic use within the last 3 months; hospitalization within the last 3 months; and recent surgery (*Table 8*). Multivariate analysis was then performed by stepwise logistic regression including variables that were found to be significantly associated with MDR in univariate analysis. Seventy-nine patients were included in the analysis (the others had missing data or the information was unavailable). Only recent antibiotic use was found to be significantly associated with MDR BSI by multivariate analysis (p = 0.001).

Table 4. Antimicrobial resistance of the 222 GNB isolated from BSI

Isolates	Amino-penicillin N (%)	Third generation cephalosporins N (%)	Carbapenems N (%)	Amino-glycosides N (%)	Quinolones N (%)	Trimetoprim-Sulfametoxazol N (%)
E. coli N=120 (54.1%)	81 (67.5)	17 (14.2)	2 (1.7)	23 (19.2)	23 (19.2)	41 (34.2)
Klebsiella spp N=38 (17.1%)	NA	15 (39)	2 (5.3)	15 (39.5)	14 (36.8)	12 (31.6)
P. aeruginosa N=16 (7.2%)	NA	13 (81)	10 (62)	10 (62)	9 (56)	15 (94)
Acinetobacter spp N= 11 (5%)	NA	10 (91)	8 (73)	8 (73)	8 (73)	5 (45)
Other *Enterobacteri-aceae* N=30 (13.5%)	12 (40)	5 (16.7)	0	3 (10)	2 (6.7)	5 (16.7)
Other non-fermenters N=7 (3.2%)	NA	4 (57.1)	2 (28.6)	4 (57.1)	4 (57.1)	2 (28.6)

Table 5. Combined antimicrobial resistance to two and three antibiotic classes among *E. coli* isolated from BSI

	***E coli* N (%) N=120**
Fully susceptible	**30 (25)**
Resistance to one class	**20 (16.7)**
Resistance to two antimicrobials classes	**35 (29.2)**
Aminopenicillin+Fluorquinolones	23 (19)
Aminopenicillin+Aminoglycosides	20 (16)
Fluoroquinolones+Aminoglycosides	14 (11.7)
Third cephalosporins+Fluorquinolones	15 (12)
Third cephalosporins+ Aminoglycosides	12 (10)
Resistance to at least three antimicrobials classes/	**35 (29.2)**
Aminopenicillin+Fluoroquinolones+Aminoglicosides	14 (11.7)
Third cephalosporins + Fluoroquinolones+ Aminoglicosides	12 (10)
Aminopenicillin+Fluoroquinolones+ Trimetoprimsulfametoxazol	14 (11.7)
Third cephalosporins + Fluoroquinolones+ Trimetoprimsulfametoxazol	8 (6.7)
Aminopenicillin+Aminoglicosides+ Trimetoprimsulfametoxazol	11 (9)
Third cephalosporins +Aminoglicosides + Trimetoprimsulfametoxazol	7 (5.8)
ESBL	**12 (10)**

Discussion

According to the European Centre for Disease Prevention and Control (ECDC) report from Europe in 2009, the majority of countries (19 of 28) reported that 50% to 66.5% of the E. coli isolated from BSI were resistant to amino-penicillin, while in Romania the reported resistance was 83.3%. However, only 12 isolates from Romania were analyzed in that report (11). In this present study including 116 patients, resistance to amino-penicillin among the most common BSI pathogen *E. coli* was 67.5%. Nine of 28 countries reported third-generation cephalosporin resistance exceeding 10%, with the highest at 19.2% (8). In our study, resistance to the third-generation cephalosporins was 13.4%, less than that reported by the ECDC (17%) for Romania.

Table 6. Combined antimicrobial resistance to two and at least three antimicrobial classes among *Klebsiella spp* isolated from BSI

	Klebsiella spp **N (%) N=38**
Fully susceptible	**17 (44.7)**
Resistance to one antibiotic class	**3 (7.9)**
Resistance to two antibiotic classes	**3 (7.9)**
Resistance to at least three antibiotic classes/	**15 (39.5)**
Third cephalosporins+Fluoroquinolones+Aminoglicosides	11 (29)
Third cephalosporins+Fluoroquinolones+Trimetoprimsulfametoxazol	10 (26)
Third cephalosporins+Aminoglicosides+Trimetoprimsulfametoxazol	8 (21)
ESBL	**10 (26.3)**

Table 7. Resistance combinations among invasive *E coli* isolates in Romania compared with overall resistance combinations in Europe, 2009

Resistance pattern	**Romania N (%) N=120**	**Europe N (%) N=42 898**
Aminopenicillin + Fluorquinolones	23 (19)	3 718 (8.7)
Aminopenicillin + Aminoglycosides	20 (16)	555 (1.3)
Fluoroquinolones + Aminoglycosides	14 (11.7)	40 (0.1)
Aminopenicillin + Fluoroquinolones + Aminoglycosides	14 (11.7)	1 387 (3.2)

Table 8. Epidemiological characteristics and in-hospital mortality associated with multi-drug resistance (MDR)

	MDR		**Non-MDR**		**OR (95% CI)**	**p-value**
	Number	**%**	**Number**	**%**		
History of urinary tract infections	15/28	53.5	13/63	20.6	4.4 (1.7-11.6)	0.002
Antibiotic use within last 3 months	31/34	91.1	34/73	46.5	11.8 (3.3-42.3)	0.001
Hospitalization within last 3 months	30/40	75	37/87	42.5	4.0 (1.8-9.3)	0.001
Recent surgery	20/45	44.4	17/93	18.2	3.5 (1.6-7.9)	0.001
Lethality	10/39	25.6	12/93	12.9	2.3 (0.8-6.5)	0.07
Total	45		97			

Eleven of 28 countries reported the prevalence of resistance to aminoglycosides higher than 10%, ranging up to 21.4%. Our study showed a prevalence of 18.5%, as compared with 12.5% in the ECDC report. Despite the high level of aminoglycoside resistance in Romania, it decreased over the last four years from > 40% in 2006 to < 20% in 2009, while a significant increase was observed for 10 other countries.

The majority of countries (16 of 28) reported fluoroquinolones resistance levels higher than 20%, ranging up to 43.4%. In our study the fluoroquinolones resistance was 18.5%, comparable with the ECDC reported resistance for Romania of 22.6%.

Overall, in Romania, high resistance levels to aminopenicillins, third generation cephalosporins and aminoglycosides are encountered; this is not however the case of fluo-

roquinolones, where resistance levels are lower than in other European countries.

It is important to note that in Romania *E. coli* combined resistance to two or three classes was significantly higher in our study than overall in Europe. Twenty eight countries reported 42 898 *E. coli* isolates in 2009 and 57% of these isolates were resistant to one or more of the antibiotic classes tested (aminopenicillins, third-generation cephalosporins, fluoroquinolones and aminoglycosides). Only in nine European countries the proportion of multi-resistant isolates (resistance to third-generation cephalosporins, fluoroquinolones and aminoglycosides) was higher than 5%.

The HCA episodes of BSI have been found by other authors to be more similar to HA than to CA episodes in terms of frequency of various comorbid conditions, source of infection, pathogens and their susceptibility patterns, and mortality (4-7). In the present study the oldest patients with BSI were those with HCA episodes, although the median age of patients with community and hospital associated infections was similar as reported from Spain (14). Also similar to that study, and with other studies (9, 14) chronic renal disease or solid/hematological cancer were more frequently associated with HCA episodes. In CA BSIs, HIV infection, alcohol abuse or chronic liver disease were more frequently found. Recent surgery and previous antimicrobial use have been found to be more frequently associated with HCA and HA in the Spanish study, as well as in our study.

The most common source of bacteraemia was urinary tract infection, similar to the findings in other studies (10, 14). When Gram positive BSIs were included, vascular catheter was identified as the main origin for HA episodes, compared with urinary tract infections (9, 14). In our study, the most common source was urinary tract infections, but there were no differences among between CA, HCA and HA infections. Actually, in BSI the source may vary largely according to the type of hospital (community or tertiary) or to the differences between different healthcare systems.

Our study shows that Enterobacteriaceae MDR resistance problem is not just a problem of HA (37.5%) or HCA BSI (44%) infections, but also of CA (15.6%) infections. The antimicrobial- resistant organisms, like ESBL producing *E. coli* and, *Klebsiella spp.* were either not present or rare in CA episodes, but should be considered in patients with a suspected HCA or HA BSI. Curiously, although ESBL producers among *E. coli* were found at similar frequencies in CA, HCA and HA episodes in other studies, (15) in the present study the confirmed ESBL producers were absent in CA episodes.

In HA infections a slightly higher mortality (18%) was recorded compared with CA infections (15.9%) and the relatively low mortality in HCA episodes is surprising; the number of HCA episodes was low however. Infections due to MDR strains had the highest mortality in our study (45.4%). In a recent study HCA BSI increased mortality while the additional effect of the most common antimicrobial resistance patterns was comparatively low (16). It was suggested that inappropriate initial antimicrobial therapy contributes poor outcome in these cases (17).

One limitation of our study was the low number of cases, especially HCA episodes, compared with that of HA infections and the retrospective design associated with the lack of clinical and epidemiological data in one third of cases.

Conclusions

Combined resistance to third-generation cephaloporins, aminoglycosides and fluoroquinolones was considerably higher than that reported for other European countries, especially in patients with HA episodes. MDR and ESBL production was more frequently seen in *Klebsiella pneumoniae* than in *E. coli*. Antibiotic use within the past 3 months was associated with MDR BSI in our patient population.

Acknowledgements

This paper is partially supported by the Sectoral Operational Programme Human Re-

sources Development, financed from the European Social Fund and by the Romanian Government under the contract number POSDRU/89/1.5/S/64109. The authors declare no conflict of interest. Data from this study were presented at CCMID-ICC 2011, Milan Italy.

References

1. Wisplinghoff H, Bischoff T, Tallent SM, Seifert H, Wenzel RP, Edmond MB. Nosocomial bloodstream infections in US hospitals: analysis of 24,179 cases from a prospective nationwide surveillance study. Clin Infect Dis 2004; 39: 309–17
2. Rodríguez-Créixems M, Alcalá L, Muñoz P, Cercenado E, Vicente T, Bouza E. Bloodstream infections. Evolution and trends in the microbiology workload, incidence and etiology, 1985–2006. Medicine 2008; 87: 234–49
3. Garner JS, Jarvis WR, Emori TG, Horan TC, Hughes JM. CDC definitions for nosocomial infections. Am J Infect Control 1988; 16:128–40
4. Friedman ND, Kaye KS, Stout JE, McGarry SA, Trivette SL, Briggs JP, et al. Health care-associated bloodstream infections in adults; a reason to change the accepted definition of community-acquired infections. Ann Intern Med 2002; 137:791–7
5. Siegman-Igra Y, Fourer B, Orni-Wasserlauf R, Golan Y, Noy A, Schwartz D, et al. Reappraisal of community-acquired bacteremia: a proposal or a new classification for the spectrum of acquisition of bacteremia. Clin Infect Dis 2002; 34: 1431–9
6. Shorr AF, Tabak YP, Killian AD, Gupta V, Liu LZ, Kollef MH. Healthcare- associated bloodstream infection: a distinct entity? Insights from a large U.S. database. Crit Care Med 2006; 34: 1588–95
7. Vallés J, Calbo E, Anoro E, Fontanals D, Xercavins M, Espejo E, et al. Boodstream infections in adults: importance of healthcare-associated infections. J Infect 2008; 56: 27–34.
8. Hidron AI, Edwards JR, Patel J, Horan TC, Sievert DM, Pollock DA, et al. NHSN annual update: antimicrobial-resistant pathogens associated with healthcare-associated infections: annual summary of data reported to the National Healthcare Safety Network at the Centers for Disease Control and Prevention, 2006-2007. Infect Control Hosp Epidemiol 2008; 29:996-1011
9. Allegranzi B, Nejad SB, Combescure C, Graafmans W, Attar H, Donaldson L, et al. Burden of endemic health-care-associated infection in developing countries: systematic review and meta-analysis. The Lancet 2011; 377:186–8
10. Biedenbach DJ, Moet GJ, Jones RN. Occurrence and antimicrobial resistance pattern comparisons among bloodstream infection isolates from the SENTRY Antimicrobial Surveillance Program (1997–2002). Diagn Microbiol Infect Dis 2004; 50:59–69
11. European Centre for Disease Prevention and Control. Antimicrobial resistance surveillance in Europe 2009. Annual Report of the European Antimicrobial Resistance Surveillance Network (EARS-Net). Stockholm: ECDC; 2010.
12. Clinical and Laboratory Standards Institute 2009. Performance standards for antimicrobial susceptibility testing; Nineteenth Informational Supplement, vol. 29, no.3. M100-S19. Clinical and Laboratory Standards Institute, Wayne, PA.
13. Clinical and Laboratory Standards Institute 2010. Performance standards for antimicrobial susceptibility testing; Twentieth Informational Supplement, vol. 30, no.1. M100-S20. Clinical and Laboratory Standards Institute, Wayne, PA.
14. Rodríguez-Baño J, López-Prieto MD, Portillo MM, Retamar P, Natera C, Nuño E, et al. Epidemiology and clinical features of community-acquired, healthcare-associated and nosocomial bloodstream infections in tertiary-care and community hospitals. Clin Microbiol Infect 2010; 16: 1408-13
15. Rodríguez-Baño J, Navarro MD, Romero L, Muniain MA, de Cueto M, Ríos MJ, et al. Bacteremia due to extended-spectrum beta-lactamase-producing Escherichia coli in the CTX-M era: a new clinical challenge. Clin Infect Dis 2006; 43: 1407–14
16. Lambert ML, Suetens C, Savey A, Palomar M, Hiesmayr M, Morales I, et al. Clinical outcomes of health-care-associated infections and antimicrobial resistance in patients admitted to European intensive-care units: a cohort study. The Lancet Infectious Diseases 2011; 11:30-8
17. Kang CI, Kim SH, Park WB, Lee KD, Kim HB, Kim EC, et al. Bloodstream infections due to extended-spectrum beta-lactamase-producing Escherichia coli and Klebsiella pneumoniae: risk factors for mortality and treatment outcome, with special emphasis on antimicrobial therapy. Antimicrob Agents Chemother 2004; 48:4574–81.