Therapeutics, Pharmacology and Clinical Toxicology
Vol XVI, Number 2, June 2012
Pages 95-100

ORIGINAL PAPER

ACINETOBACTER SPP. SKIN AND SOFT TISSUE INFECTIONS IN THE CLINICAL INFECTIOUS DISEASES HOSPITAL OF IAŞI CITY, BETWEEN 2006 AND 2010

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Abstract. Acinetobacter (ACB) species have often revealed themselves as highly resistant to antibiotics, occurring preponderantly in vulnerable patients and sometimes causing severe infections, which are difficult to treat. Material and method: retrospective study on patients diagnosed in the Clinical Infectious Diseases Hospital of Iaşi City, between January 2006 and December 2010; the demographic, clinical and microbiological characteristics of patients suffering from skin and soft tissue (SST) infections caused by ACB were compared to those of other patients with different infection locations. Results: 24.4% of the whole 45 ACB strains isolated throughout the study were involved in SST infections. Most of them (10/11) had a significant number of associated conditions; average age - 59.4 years, preponderantly males (63.6%). The most frequent isolate was Acinetobacter baumanni (8/11 cases, 4 of whom also had bacteremia). The isolated strains were resistant to multiple classes of antibiotics (anti-pseudomonas cephalosporins – 8/11, imipenem 6/11, amikacin 6/9, and ciprofloxacin 8/10); colistin was the only antibiotic that revealed good anti-ACB activity, as only one of its strains was antibiotic resistant. Multidrug-resistance was noticed in 4/11 strains. ACB SST infections were lethal in 9.1% of the cases. There were no statistically significant differences between the demographic, epidemiological or antibiotic-resistance variables of patients with SST infections and those of other patients with different ACB infection locations. Conclusions: the ACB infection located on SST distinguishes itself by its severity and serious therapeutic challenges, whereas its antibiotic-resistance behavior is similar to that of infections with different locations.
Keywords: antibiotic susceptibility, sepsis, nosocomial

Acinetobacter (ACB) microorganisms are short, thick, not motile (*akinetos*), frequently encapsulated, strictly aerobic, non-pigmentogenic, catalase-positive and oxidase-negative Gram-negative bacilli. They are ubiquitous water and soil saprophytes. The genus has suffered major taxonomic changes over the last 30 years, its members having distinguished themselves as some of the most problematic pathogens in hospitalized patients, in patients with associated conditions or in intensive care wards.

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Acinetobacter species may be frequently isolated on normal skin and mucosa, this asymptomatic occurrence having been originally described in 25% of the investigated subjects [1]. More recent studies [2,3] reveal a colonization rate of 43% in outpatients and of up to 75% in hospitalized patients. Notwithstanding, in 99% of the cases, the species colonizing the skin are not among those usually associated with the infection [4].

Acinetobacter spp. are not common pathogens in the etiology of skin and soft tissue (SST) infections, as shown by a comprehensive study [5] on over 1,700 Latin American patients, according to which, their involvement in such infections did not exceed 4.1%.

A wide range of antibiotic-resistance mecha-

nisms has been described for the ACB group members. The fast emergence of *A. baumanii* strains resistant to all the beta-lactam antibiotics, including carbapenems, reveals the incredible capacity of this microorganism to adjust to the changes occurring in its living environment. Most authors indicate the percent of strains resistant to one particular antibiotic, yet they do not refer to those that are multi-resistant. Among other, we chose the definition for multidrug-resistant strains introduced by Peleg et al, in 2008: resistance to more than 2 classes of antibiotics of the following 5: antipseudomonal cephalosporins (ceftazidime, cefepime), antipseudomonal carbapenems (imipenem or meropenem), ampicillin-sulbactam, fluoroquinolones (ciprofloxacin or levofloxacin) and aminoglycosides (gentamicin, tobramycin, amikacin).

Given its high antibiotic resistance and targeting of vulnerable patients, who have many associated conditions, most of the time, this type of infection poses special therapeutic challenges. While waiting for the results of the susceptibility tests, such infections should be treated by combining a broad-spectrum beta-lactam agent and an aminoglycoside, any foreign body should be removed and the necrotic tissue, if any, should be cleansed by debridement.

Material and method

A retrospective study was conducted on 45 patients suffering from infections caused by ACB strains, who were hospitalized in the „Sfânta Parascheva" Clinical Infectious Diseases Hospital of Iaşi City, between 2006 and 2010. Eleven of them had skin and soft tissue (SST) infections, while in the others, the strains were isolated from the cerebrospinal fluid, hemocultures, bronchial aspirates or urine cultures.

Demographic variables, associated diseases, and clinical, bacteriological and therapeutic aspects were analyzed.

These characteristics of the SST infection patients (included in the 1st group – 11 cases) were compared to those of other patients with different infection locations (2nd group - 34 cases).

The bacteriologic diagnosis consisted of:

- the microscopic examination of pathological product smears or of Gram- and methylene blue-stained culture smears, in order to reveal bacteria morphology and their relation to PMNs.
- culture was done on nutritive, differential or poorly selective agar media (5% sheep blood agar, chocolate agar, CLED medium, Mac Conkey agar). Blood cultures were done in flasks containing heart brain infusion broth or in special flasks (PF,SA,FA with BacT/Alert incubation). Solid media cultures underwent 35-37 degree incubation for 24-48 hours, whereas the incubation time for liquid media cultures was 7 days.
- species identification was done considering the biochemical characteristics, by means of multitest media and modern methods: API 20 NE systems with automated reading capability.
- antibiotic sensitivity was tested by the Kirby-Bauer method (according to the NCCLS 2005 standard), using Mueller-Hinton media and micro tablets (Oxoid) or on API systems designed for ATB PSE5 antibiotic susceptibility testing.

The distinction between colonization and the actual infection was made relying on clinical grounds, on isolates sampled simultaneously from several biological products and on their response to antibacterial therapy.

The data was analyzed using computerized programs (Microsaoft Excel + Analyze-it) to calculate means, 95% confidence intervals, t Student test, chi square. A $p<0.05$ was considered relevant.

Results

Eleven patients were diagnosed with Acinetobacter skin and soft tissue infections throughout the period under survey. They made up 24.4% of the total number of infections caused by this type of pathogen in our clinic.

The age of SST infection patients varied between 32 and 80 years, with an average age of 59.4 years (95CI: 48.0 - 70.8 years), which is comparable to that of the patients included in the group with different ACB infection locations (49.7 years, $t=1.38$, $p=0.17$)

Men were more frequently concerned, both among the SST infection patients – 63.6%, and among the patients with different infection locations – 70.6% of the cases (no significant differences between the two groups - $\chi2=0.003$, $p=0.95$).

About half of the cases (45.4%) had previously been hospitalized in other medical units for skin conditions or other disorders, and then transferred to the infectious diseases clinic.

Having analyzed the history of the disease and the patients' previous contacts with different hospitals, we concluded that in the first group (SSTI) 6/11 of the isolated ACB strains had been community-acquired, whereas the other 5 most likely had nosocomial origins. 4/11 patients had been hospitalized in intensive care wards before coming to our infectious diseases clinic. The second group of patients (other infection locations) enjoyed a slightly different situation: a somewhat lower transfer rate (38.2%), a sub unitary community-acquired/hospital-acquired strain ratio (0.78) and a

29.9 percent of cases with a previous contact with an ICU. The comparative statistical analysis of these variables revealed no significant differences.

Only one of the 11 SST infection patients did not have significant associated conditions. He was a 35 year-old man having suffered open tibia fracture, for which he underwent surgical treatment, followed by severe *A. baumanni* surgical wound infection, the treatment of which required 14 days of hospitalization in the infectious diseases clinic. The immune system of the other 10 patients was already weakened by their associated conditions: 3 patients suffered from advanced chronic liver conditions, 2 patients had diabetes mellitus, 2 patients had lung and liver neoplasm, respectively, 2 suffered from disabling neurological and mental conditions (Alzheimer's disease, hemiparesis) and one patient had 4th degree obesity. The percent of patients without associated conditions was higher in the group of patients with other ACB infection locations, namely 38.2%, yet the difference was not statistically significant ($\chi 2=2.07$, $p=0.14$).

The clinical aspects varied: in 4 of the patients the pathogen was isolated in the purulent secretions of their bed sores (3 in the sacrum area and 1 in the calcaneus area), 4 patients developed inflammatory processes materialized in violaceous erythematous plaques around surgical wounds, which triggered/enhanced purulent secretions in the wound or drain tubes; a 40 year-old obese patient developed a cellulitis on the right thigh, complicated with thigh and buttock phlegmon, subsequent to a wound caused by a pointed object, which had been neglected at the time, and other 2 diabetic patients suffered from erysipelas-like calf cellulitis, which later turned into bullous cellulitis and hypodermic necrosis.

In the first group, 8/11 patients the infection was accompanied by fever, which sometimes reached 39.5°C and lasted 2 to 7 days. In 7/11 cases the clinical evolution of the infection was severe and fulfilled the sepsis criteria, whereas the other 4/11 cases required medical care in the hospital's intensive care ward. The 2nd group included 12 respiratory infections, 8 meningitis cases, 5 urinary tract infections and 9 septicemia cases with multiple septic determinations; fever was present here in a higher number of patients – 85.7% of the cases (statistically insignificant difference: $\chi 2=0.57$, $p=0.45$), and the sepsis cases more numerous - 91.2%, the percent being close to the significance threshold ($p=0.08$).

Most of the patients (6/11) had leukocytosis on their hospitalization day, with values between 11.200 and 23.000/mm^3; 3 patients had leukopenia, and the other 2 had a normal number of white blood cells. All the patients had a high PMNs percent in their white blood cell count, which varied between 71 and 89%. The ESR was high (between 32 and 110 mm/h) in all the 9 patients in whom this test was done. Fibrinogen was high in 4/5 of the patients in whom it was measured.

Acinetobacter baumanni was isolated in 8/11 patients of the 1st group, whereas in the other 3 cases the species remained unknown. As concerns the 2nd group, *A. baumanni* was identified in 23 patients, *A. lwolffi* in 4, and in the other 7 cases the species could not be identified.

Four of the patients in the SST infection group also had positive blood cultures at the same time, and 2 of them also had secondary septic determinations (bronchopneumonia and meningitis, respectively).

Three of the patients came to the infectious diseases clinic after the microorganism had been isolated in other medical units, and the etiological diagnosis setting for the remaining 8 took 3 to 7 days.

Ten of the 11 patients had received antibiotic therapy (either one or several associated antibiotics) before the setting of their etiological diagnosis. 5 had been given 3rd generation cephalosporins, 6 fluoroquinolones, 3 aminoglycosides, 2 carbapen-

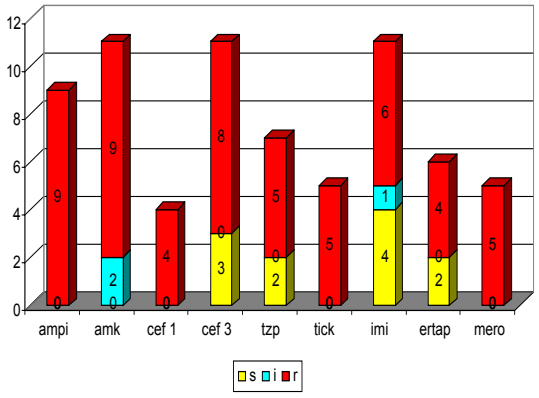

Figure 1. Beta-lactam susceptibility of Acinetobacter strains involved in SST infections

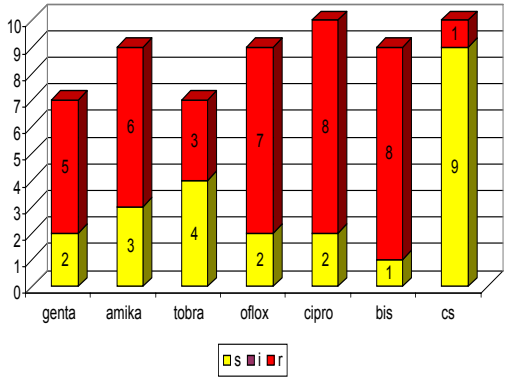

Figure 2. Susceptibility to various classes of antibiotics of Acinetobacter strains involved in SST infections

ems, 1 penicillin and 1 oxacillin.

The antibiotic susceptibility of the isolated strains was tested in all the cases.

As shown in figure 1, the beta-lactam susceptibility of Acinetobacter strains involved in SST infections was extremely low: none of the 9 and 11 strains, respectively, which were ampicillin or amoxicillin-clavulanate tested was susceptible, 8/11 were resistant to third generation cephalosporins, and the rather wide carbapenem resistance, especially to new generation carbapenems (ertapenem), is alarming.

Table I shows the share of strains resistant to antibiotics in the 2nd group:

Antibiotic	% tested strains	Antibiotic	% tested strains
ampi	91.3	**genta**	90.9
amk	81.0	**amika**	48.1
cef3	87.1	**netro**	37.5
cefpirom	100.0	**tobra**	17.6
tzp	42.1	**oflox**	77.8
imi	32.3	**peflo**	75.0
ertap	92.1	**cipro**	89.7
mero	68.0	**bis**	78.6
		cloro	66.7
		colistin	0.0

Table I. Share of antibiotic-resistant strains

According to the definition, 4/11 of the ACB strains isolated in the SST infections were multidrug-resistant and 35.3% of the strains in the 2nd group had the same behavior (Tabel II).

The death rate in our ACB infection group was 9.1%. The only death among our cases was that of a 69 year-old patient, with a serious cardiovascular history, with sacral bed sores, bacteremia and nosocomial pneumonia. The death rate was higher in the 2nd group, amounting to 23.5%, which made no statistically significant difference (p=0.5), possibly due to the small number of cases.

Discussions

Increasingly higher numbers of Acinetobacter spp infections have been reported recently, both worldwide and in Europe. The Clinical Infectious Diseases Hospital of Iasi City is no exception, where 45 cases have been reported over the last 5 years.

Although it may sometimes be difficult to distinguish between a simple ACB colonization and an actual infection, corroborating clinical data with laboratory changes – the inflammatory syndrome and the bacteriological isolation of the same strain from 2 or more pathological products, helped us a great deal.

Acinetobacter spp. skin infections have not been extremely frequently reported in the absence of a traumatic context (shot wounds [4] or tibia fractures [6]), and in our case they made up only 24.4% of the whole number of occurrences and they were associated with surgical wound superinfection (4 cases) or bed sore superinfection (4 cases), and even with primary cellulitis in patients with associated conditions.

Unlike Sebeny's study [4], where the average patient age was 25.5 years, as it concerned a particular population (the military), the average age in our study was 59.4 years, skin infections occurring especially in old people with associated conditions (10/11).

The strains involved may be transmitted from one hospital to another, especially via the infected or colonized patients that are transferred; almost half (5/11) of the SST infection patients had been previously hospitalized in other medical units before coming to the Clinical Infectious Diseases Hospital of Iaşi, 4/11 in other IC wards.

Fever was more frequent in the group of patients with other *Acinetobacter spp*. infection locations, namely 85.7% vs. 72.7% in the SST infection group, this difference approaching the statistical significance threshold (p=0.08).

Acinetobacter baumannii is considered to be the most important hospital-acquired species for humans [7] and it actually was the most frequently isolated species both in the 1st (8/11) and in the 2nd (65.7%) groups. Although it may be present in the colonizing flora, its occurrence on the skin is rare (0.5-3% of the tested subjects [2, 1]).

Antibiotic resistance was a major problem in the overwhelming majority of cases, being enhanced by factors such as: the incredible capacity of this microorganism to adjust to the changes occurring in its living environment, the nosocomial origin of most of the strains and the previous antibiotic therapy.

In most of the cases, beta-lactams do not seem to be the solution for the treatment of this type of infection, since only 3/11 of the tested strains were susceptible to third generation cephalosporins. Despite the fact that in Sebeny's study all the 8 strains were sensitive to imipenem and although there are authors recommending carbapenems for ACB infection treatment, in our study the resistance rate was high, as 6/11 strains were resistant to imipenem, 5/5 to meropenem and 4/6 to ertapenem; the emergence of carbapenem-resistant strains was first reported back in 2006 [8] and their frequency has been increasing ever since. Similar carbapenem resistance figures were revealed in the 2nd group,

with even higher ertapenem resistance percents (p=0.006).

Aminoglycoside resistance was high, especially to gentamicin (5/7 and 90% in the 1st and 2nd group, respectively), fluoroquinolones (8/10 and 89% to ciprofloxacin in the 1st and 2nd group, respectively) or co-trimoxazole (1/9 vs. 78.6%).

The only antibiotic which still had good anti-acinetobacter activity was colimicin, to which only 1 of the 23 tested strains was resistant. It may be useful for topical or injectable SST infection therapy. Its use as parenteral therapy may sometimes be difficult in aged patients, with associated (especially renal) conditions, due to its topical and systemic toxicity.

There were no statistically significant differences between the share of resistant strains found in the SST infection group and those found in the group of patients with other acinetobacter infection locations (except for ertapenem).

There are not many studies in literature providing data on the proportion of MDR ACB strains, and this is due to the use of different definitions or to the lack of clear systematization [7]. In our groups, this share was about 35% (4/11 in the 1st group and 35.3% in the 2nd group), which supports the assumed (nosocomial/community) origin of the strains involved.

The data on the rate of deaths caused by acinetobacter infections are slightly contradictory, some studies placing it at 21.8% [9], while others considering it to be similar to that of other pathogens (frequent in respiratory infections)[10]. In Sebeny's study [4], the rate of deaths due to SST infections was 12.5%, which was higher than from our group (9.1%).

Conclusions

- 24.4% of the whole 45 ACB strains isolated throughout the study were involved in SST infections (4 infected surgical wounds, 4 bed sores, 3 primary cellulites).
- the great majority of SST infection patients (10/11) had significant associated conditions; the average age was 59.4 years, males were more frequently involved (63.6%).
- the most frequently isolated species was *Acinetobacter baumannii* (8/11 cases), 4 of whom also had bacteremia.
- the isolated strains from SST infections were resistant to many classes of antibiotics (anti-pseudomonas cephalosporins – 8/11, imipenem 6/11, amikacin 6/9, ciprofloxacin 8/10); colistin was the only antibiotic with good anti-acinetobacter activity, as only one of the strains was resistant to it. Multidrug-resistance occurred in 4/11 strains.
- the lethality of ACB SST infections was 9.1%
- there were no statistically significant differences between the demographic, epidemiological or antibiotic-resistance variables of patients with SST infections and those of other patients with different ACB infection locations (possibly due to the small number of cases)

Abbreviations:

ACB – acinetobacter, SST – skin and soft tissue, Ampi – ampicillin, Amk – amoxicillin-clavulanate, Cef3 – antipseudomonal cephalosporins (ceftazidime), Tzp – piperacillin-tazobactam, Imi – imipenem, Ertap – ertapenem, Mero – meropenem Genta – gentamicin, Amika – amikacin, Netro – netilmicin, Tobra – tobramycin, Oflox – ofloxacin, Peflox – pefloxacin, Cipro – ciprofloxacin, Bis – co-trimoxazole, Cloro – chloramphenicol, MDR – multidrug-resistance

References

1. Taplin D, Zaias N. The human skin as a source of Mimaherellea infections. *JAMA* **1963; 186:952–5.**

2. Seifert, H., L. Dijkshoorn, P. Gerner-Smidt, N. Pelzer, I. Tjernberg, and M. Vaneechoutte. Distribution of Acinetobacter species on human skin: comparison of phenotypic and genotypic identification methods. *J. Clin.Microbiol*. **1997; 35:2819–2825.**

3. Berlau, J., H. Aucken, H. Malnick, and T. Pitt. Distribution of Acinetobacter species on skin of healthy humans. *Eur. J. Clin. Microbiol. Infect. Dis*. **1999;18:179–183.**

4. Sebeny PJ, Riddle MS, Petersen P. Acinetobacter baumannii Skin and Soft-Tissue Infection Associated with War Trauma. *Clinical Infectious Diseases* **2008; 47:444–9.**

5. Sader H, Jones RN, Silva JB;. SENTRY Participants Group (Latin America). Skin and soft tissue infections in Latin American medical centers: four-year assessment of the pathogen frequency and antimicrobial susceptibility patterns. *Diagn Microbiol Infect Dis* **2002; 44:281–8.**

6. Johnson E N, T C Burns, R A Hayda, D R.Hospenthal, C K Murray. Infectious complications of open type III tibial fractures among combat casualties. *Clin. Infect. Dis*. **2007; 45:409–415.**

7. Peleg AI, Seifert H, Paterson DL Acinetobacter baumannii: Emergence of a Successful Pathogen. *Clinical Microbiology Reviews* **2008; 28: 538–582.**

8. Lolans K, Rice TW, Munoz-Price LS, Quinn JP. Multicity outbreak of carbapenem-resistant Acinetobacter baumannii isolates producing the carbapenemase OXA-40. *Antimicrob Agents Chemother* **2006; 50:2941–5.**

9. Lee NY, Lee HC, Ko NY, et al. Clinical and economic impact of multidrug resistance in nosocomial Acinetobacter baumannii bacteremia. *Infect Control Hosp Epidemiol* **2007;28:713-9.**

10. Garnacho-Montero J, Ortiz-Leyba C, Fernández-Hinojosa E, et al. Acinetobacter baumannii ventilator-associated pneumonia: epidemiological and clinical findings. *Intensive Care Med* **2005;31:649-55.**

no	name	sex	age	sepsis	transfer	community/ hospital	hospitalization days	IC	surgery	associated conditions	death	species	source
1	CP	m	71	yes	no	community	18	no	no	stroke, hemiparesis	no	baumannii	bed-sore
2	AL	f	76	yes	no	community	12	no	no	Alzheimer	no	baumannii	bed-sore
3	DV	m	69	yes	yes	hospital	10	yes	no	cardiovascular	yes	baumannii	bed-sore
4	BC	m	32	yes	yes	hospital	90	yes	yes	hepatocellular carcinoma	no	unidentified	surgery wound
5	TC	m	40	no	no	community	23	no	no	obesity	no	baumannii	wound
6	CE	f	68	no	no	community	21	no	no	cirrhosis	no	baumannii	bed-sore
7	PI	m	59	no	yes	hospital	16	yes	yes	lung neoplasm	no	baumannii	surgery wound
8	PM	m	35	yes	yes	hospital	14	no	no	none	no	baumannii	surgery wound
9	FV	m	53	yes	no	community	16	no	no	diabetes	no	unidentified	pus
10	RE	f	80	no	no	community	27	no	no	diabetes	no	unidentified	pus
11	SL	f	71	yes	yes	hospital	8	yes	yes	obesity, chronic hepatitis	no	baumannii	drain tube secretion

Table II. Characteristics of SST infection patients