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Bacteremia by Multidrug-Resistant *Capnocytophaga sputigena*

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A case of bacteremia caused by a multiresistant strain of *Capnocytophaga sputigena* in a patient with hematological malignancy is described. The strain presented with a pattern of marked resistance to β-lactams, with MICs of >256 mg/liter for ampicillin, ticarcillin, piperacillin, cefazolin, and cefuroxime, 64 mg/liter for cefotaxime, and 32 mg/liter for ceftazidime. In addition, the MIC of ciprofloxacin was 16 mg/liter. Both of these groups of antimicrobial agents are frequently used in the empiric treatment of infections in immunocompromised patients. The appearance of resistant strains suggests the need for antimicrobial susceptibility studies in all patients with severe infections caused by *Capnocytophaga* spp. or other capnophilic organisms present in the oral microflora of these patients.

The genus *Capnocytophaga* is made up of a group of slowly growing, capnophilic, fusiform, and filamentous gram-negative rods. In 1979 the genus was redefined to include the organisms previously classified as CDC biogroup DF-1, *Capnocytophaga* spp. and *Bacteroides ochraceus* (7, 11). These microorganisms form part of the normal gingival flora and have been implicated in cases of juvenile gingivitis and periodontitis (6). In addition, *Capnocytophaga* spp. have repeatedly been recognized as the causative agent of sepsis in immunocompromised patients (3, 8). In immunocompromised patients, it is frequently necessary to provide empiric antibiotic treatment covering a broad spectrum of potential pathogens, among which must be included *Capnocytophaga* spp. In general, *Capnocytophaga* spp. are found to be susceptible to a large number of antibiotics, of which the penicillins, clindamycin, the macrolides, broad-spectrum cephalosporins, and the quinolones are outstanding examples (5, 9, 10). There have been occasional reports of β-lactamase-producing strains (1, 10) and, very rarely, descriptions of resistance to the fluoroquinolone group (2).

We describe here a case of sepsis caused by a strain of *Capnocytophaga sputigena*, which proved to be a β-lactamase producer and resistant to the quinolones, in a neutropenic patient. These characteristics have very rarely been reported in the literature to date.

A 55-year-old woman, who had been diagnosed with chronic B lymphatic leukemia 3 years previously was admitted to the hospital with a clinical picture of acute appendicitis, for which an appendectomy was performed. She had been asymptomatic until 2 years previously, when the progression of her disease manifested itself in fatigue, cervical adenopathy, increase in the peripheral blood leukocyte count, anemia, raised lactate dehydrogenase levels, and diffuse infiltration of the bone marrow, compromising residual hemopoiesis. A lymph node biopsy performed at that time showed the presence of a well-defined lymphocytic lymphoma. During the following year she received treatment with chlorambucil and steroids, but with a poor response. Over this period she developed symptomatic anemia which required repeated transfusions and thrombopenia together with the following various infectious complications: an episode of staphylococcal folliculitis, various urinary tract infections caused by *Escherichia coli*, lobar pneumonia of unknown etiology, various episodes of dacriocystitis caused by *Staphylococcus aureus*, and recurrent orolabial herpes.

Histologic examination of the appendix showed appendicular inflammation and tumor infiltration. In the postoperative period, she developed sepsis associated with a surgical wound infection. Cultures of blood in commercial blood culture bottles grew *Bacteroides fragilis*, and cultures of the wound exudate grew *E. coli*. Ceftriaxone and metronidazole were used successfully to treat the infection. However, the patient developed a large adenopathic mass on the lateral surface of the neck which failed to respond to a cycle of fludarabine. Local radiation therapy was administered (3,400 rads). A severe radiation-associated mucositis developed; this was accompanied by a high fever and neutropenia (400/mm^3). Empiric treatment with ceftazidime and amikacin at standard doses was begun. The patient's temperature did not abate, and the patient continued to complain of pharyngeal pain and dysphagia, prompting the addition of oral ciprofloxacin (750 mg twice daily) to the treatment regimen. *C. sputigena* was isolated in the two cultures of blood (Hemoline; BioMérieux). Taken at the onset of this episode, and once the antibiotic susceptibilities of the isolate were ascertained, antimicrobial treatment was changed to amoxicillin-clavulanic acid (1 g given intravenously every 8 h). Nevertheless, the patient died 2 days later as a consequence of progressive refractory disease, severe pancytopenia, mucositis, malnutrition, and systemic infection.

C. sputigena was isolated from both ventilated and nonventilated bottles after 3 days of incubation. Subculture of the blood culture to blood agar plates incubated in a CO_2 atmosphere showed, after 48 h, the growth of gliding, yellowish orange colonies which were catalase and oxidase negative, fusiform and occasionally branching, gram-negative rods. Indole, urease, and H_2S tests, as well as growth in MacConkey agar, were negative. Gelatin hydrolysis and nitrate reduction were positive. The bacteria fermented glucose, maltose, and sucrose but were inactive against lactose, galactose, and raffinose. Differentiation from other species belonging to the genus *Capnocytophaga* was based on the lack of oxidase and catalase reaction of *C. sputigena*, while *Capnocytophaga canimorsus* and *Capnocytophaga cynodegmi* (former CDC DF-2 group) gave positive reactions. Gelatin hydrolysis, nitrate

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TABLE 1. In vitro activities of 20 antimicrobial agents against a multidrug-resistant strain of *C. sputigena*

Antimicrobial agent	MIC (mg/liter)
Ampicillin	>256
Ticarcillin	>256
Piperacillin	>256
Amoxicillin-clavulanic acid	1
Ampicillin-sulbactam	1
Piperacillin-tazobactam	0.5
Cefazolin	>256
Cefoxitin	4
Cefuroxime	>256
Cefotaxime	64
Ceftazidime	32
Aztreonam	1
Imipenem	0.5
Clindamycin	≤0.06
Erythromycin	≤0.25
Gentamicin	>32
Amikacin	>32
Trimethoprim	>128
Ciprofloxacin	16
Tetracycline	1

reduction, and an inability to produce acid from galactose and raffinose are characteristics that help to differentiate these microorganisms from the remaining species of the former CDC DF-1 group (*Capnocytophaga ochracea* and *Capnocytophaga gingivalis*).

The β-lactamase reaction was positive by the nitrocefin test. The antibiotic susceptibilities of the microorganism (Table 1) were determined by an agar dilution method. Columbia agar base (Difco) supplemented with 1% PolyVitex (bioMérieux) and 1% hemoglobin (Difco) was used, because Rummens et al. (10) have demonstrated that the best growth of *Capnocytophaga* spp. is obtained on this medium. A fresh culture that had been growing for 48 h was diluted in Mueller-Hinton broth (Oxoid) and was inoculated with a Steers replicating device to give a final inoculation spot of ca. 10^4 CFU. The plates were incubated for 2 days at 35°C in 10% CO_2. The MIC was defined as the lowest concentration of antibiotic that completely inhibited growth. The presence of a barely visible haze at the inoculation site was disregarded. *E. coli* ATCC 25922 and *Pseudomonas aeruginosa* ATCC 27853 were used as control strains.

Bacteria belonging to the genus *Capnocytophaga*, particularly the three species previously known as CDC group DF-1, have been implicated as causative agents in severe infections in immunocompromised patients (3). In these patients, antibiotic treatment is usually commenced on an empiric basis with broad-spectrum β-lactam drugs active against *P. aeruginosa* in combination with aminoglycosides and, in a large number of patients, vancomycin. Many of these patients develop severe episodes of mucositis in the course of their underlying disease.

Until a few years ago the oral capnophilic bacteria were not included among the causative agents of sepsis in these patients. Nevertheless, in recent years we have witnessed the substitution of the classic intestinal flora by these other agents from the microflora of the oral cavity, where, in addition, they enjoy a densely populated microenvironment which permits easy interchange among its components (2).

Five noteworthy series concerning the antibiotic susceptibilities of *Capnocytophaga* spp. have recently been published (9). These microorganisms were reported to be almost uniformly susceptible to penicillin, clindamycin, chloramphenicol, the quinolones, and broad-spectrum cephalosporins, while susceptibilities to other cephalosporins were found to vary. Only 4 of the 287 strains described in the series described above produced β-lactamase, and until 1987 there had been no reports of cases of sepsis related to these β-lactamase-producing strains (1, 10). On the other hand, in these published series one may observe important discrepancies in connection with the techniques used in the susceptibility determinations. We opted to use hemoglobin-supplemented Columbia agar, as described by Rummens et al. (10). We avoided broth dilution techniques because *Capnocytophaga* spp. tend to form a flocculent suspension when transferred into liquid media. Blood agar media were excluded because of the variability that different sources of blood might introduce (10). Our strain was found to be highly resistant to ampicillin, ticarcillin, and cephalosporins as a result of the production of β-lactamase. This may be inactivated by the association of β-lactamase inhibitors such as clavulanic acid, sulbactam, or tazobactam. The characterization of the enzyme is still the subject of study, and the possibility that it belongs to a recently described group of β-lactamases associated with the membrane in a way similar to that found in other gram-negative bacteria is suggestive from a clinical point of view. This would provide the bacteria with resistance to the broad-spectrum cephalosporins habitually used in the empiric treatment of patients such as ours (4, 9).

The other interesting fact concerning the susceptibility of the microorganism was the high MIC of ciprofloxacin for the organism; ciprofloxacin is usually very active against this group of bacteria, however. In the series published by Rummens et al. (10), MICs for none of the 120 strains studied were greater than 0.5 mg/liter, giving this compound the highest activity among the quinolone group. Similar results were reported by Hawkey et al. (5), who found no ciprofloxacin MIC greater than 0.24 mg/liter for a series of 33 clinical isolates. Similarly, Roscoe et al. (9) recently reported the susceptibilities of 19 isolates, 6 of which produced β-lactamases, to ciprofloxacin, which varied between ≤0.008 and 1 mg/liter. We found only one report in the literature, published by Baquero et al. (2), but that report did not specify the MICs.

Finally, our isolate retained good susceptibilities to clindamycin and erythromycin, as is usual, and displayed susceptibilities to aztreonam, imipenem, and, to a lesser degree, cefoxitin.

We conclude, in view of the results of this and other reports, that physicians should bear in mind the possibility that the susceptibilities of microorganisms may not follow the expected pattern, particularly in immunocompromised patients who do not respond to empiric treatment. We recommend that clinical laboratories perform antibiotic susceptibility testing of these microorganisms by using rapid techniques such as the E test until the results of an agar dilution technique are available.

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