## **Short Communinication**

## Nosocomial Outbreak of *Sphingomonas paucimobilis* Bacteremia in a Hemato/Oncology Unit

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(Received April 2, 2007. Accepted July 31, 2007)

**SUMMARY**: Nosocomial *Sphingomonas paucimobilis* infections can arise from contaminated water and the contaminated hands of hospital staff. Within a 1-month period, we isolated six *S. paucimobilis* strains, including four from blood cultures of four patients and two from hospital environment specimens including tap water and a bathtub in a hemato/oncology unit. We described here these strains' molecular epidemiological analyses by pulsed-field gel electrophoresis (PFGE) and antibiotic susceptibilities by E-test. Although clinical and environmental isolates yielded three different antibiotic resistances and PFGE patterns, all four clinical strains had an identical pattern by both methods. Thus, the isolated clinical strain clone could be traced neither to health care workers nor to environmental samples. It was concluded that *S. paucimobilis* strains can cause outbreaks in hemato/oncology units. We did not demonstrate genetic relatedness between clinical and environmental isolates by PFGE, but did find PFGE a useful identification technique for epidemiological investigation.

Sphingomonas paucimobilis (group IIK, biotype 1) is a yellow-pigmented, aerobic, motile with polar flagellum, nonfermentative, Gram-negative bacteria (1). The natural habitat of this organism has not been totally defined, but it is widely distributed in the natural environment especially in water and soil (2), and has also been recovered from hospital environments including tap water, distilled water, nebulizers, respirators, dialysis fluid, and other equipment (3). *S. paucimobilis* is an opportunistic pathogen, which has been isolated from hospital infections, and is considered to originate from contaminated hospital equipment or manipulation of some medical devices (4,5).

The aims of this study were to investigate the genotypic relatedness of four clinical and two environmental strains of S. paucimobilis, which caused an outbreak in the hemato/ oncology unit of our hospital, by pulsed-field gel electrophoresis (PFGE) and to determine the antibiotic resistance patterns by the E-test method. Gulhane Military Medical Academy Hospital is a teaching hospital with more than 1,500 beds in Ankara, the capital of Turkey. Between 27 March and 12 April 2006, S. paucimobilis bacteremia developed in four patients during a 2-week period in the hemato/oncology unit, which has 11 beds. Patient characteristics are shown in Table 1. Infection was suspected when patients' fever rose above 38°C. Patients' blood samples were processed from two different venous vessels with the Bactec 9240 non-radiometric blood culture system (Becton Dickinson, Cockeysville, Md., USA). If the two blood cultures become positive, they were sub-cultured onto trypticase soy agar supplemented with 5% sheep blood (Merck, Darmstadt, Germany) (2). Four isolates of S. paucimobilis were recovered from the blood cultures of four patients.

Medical devices, tap water, bathtub, aerators, intravenous

fluids, and the hands of health care workers were investigated to evaluate potential risk factors and sources of bacteremia caused by *S. paucimobilis*. Tap water samples (100 mL) were filtered through sterile filters and the filters were cultured on suitable agar plates that were incubated for 3 days at 35 to  $37^{\circ}$ C. Twenty-eight environmental samples were obtained with reliable methods from medical devices, bathtub, aerators, and intravenous fluids and were streaked onto trypticase soy agar supplemented with 5% sheep blood. All plates were incubated for up to 3 days at 35 to  $37^{\circ}$ C. After incubation, two organisms were identified on the basis of biochemical tests (2) and then confirmed as *S. paucimobilis* by the API ID32GN system (bioMerieux, Marcy L'Etoile, France). All isolates were stored at  $-70^{\circ}$ C in trypticase soy broth (Merck) supplemented with 15% glycerol.

The susceptibility of the isolates to eight antimicrobial agents was tested by E-test (AB BIODISK, Solna, Sweeden) according to the manufacturer's directions. The antibiotics tested were ciprofloxacin, trimethoprim-sulfamethoxazole, imipenem, meropenem, amikacin, cefotaxime, gentamicin, and piperacillin-tazobactam. The susceptibility breakpoint for the isolates was applied as described for *Pseudomonas aeruginosa*. Results were expressed according to the criteria recommended by the Clinical Laboratory Standards Institute (6). *Escherichia coli* ATCC 25922, *P. aeruginosa* ATCC 27853 were used as control strains.

PFGE typing of *Xba*I-digested DNA was performed by modification of a previously described method (7). To achieve optimal separation of the fragments, *Xba*I-digested DNA was electrophoresed with a run time of 19.5 h under 5.3- to 34.9sec linear ramped pulse times. The gels were stained with ethidium bromide (0.5  $\mu$ g/ml) for 40 to 60 min. Banding patterns were photographed under UV transillumination (BioRad, Hercules, Calif., USA).

During the outbreak period, all four strains of *S. paucimobilis* isolated from blood culture samples of four patients were confirmed by the API ID32GN system as *S. paucimobilis*. These organisms were susceptible to ciprofloxacin, trimethoprim-

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Table 1. Clinical characteristics of patients with bacteremia caused by Sphingomonas paucimobilis

No.	Gender	Age (y)	Admission date (m/d/y)	Diagnosis at admission	Antibiotics	Procedures	Evaluation	Date of isolation (m/d/y)
1.	Male	11	2/29/2006	Non-Hodgkin's lymphoma	Imipenem and teicoplanin	Chemotherapy	Survived and recovered	3/27/2006
2.	Male	3	3/27/2006	Acute myeloid leukemia	Imipenem and teicoplanin	Bone marrow transplantation	Survived and recovered	4/2/2006
3.	Female	6	3/23/2006	Lymphoma	Imipenem and teicoplanin	Chemotherapy	Survived and recovered	4/2/2006
4.	Female	5	3/16/2006	Neuroblastoma	Imipenem and teicoplanin	Chemotherapy	Survived and recovered	4/12/2006

sulfamethoxazole, imipenem, meropenem, amikacin, cefotaxime, gentamicin, and piperacillin-tazobactam by the E-test method. Two environmental isolates recovered from tap water and a bathtub were identified and confirmed as. *S. paucimobilis*. However, no *S. paucimobilis* was isolated from medical staff. All isolates were susceptible to all antimicrobial agents tested, except ciprofloxacin and gentamicin. The isolates yielded three different banding patterns by PFGE analysis (Fig. 1). All clinical strains had the same genotype, but environmental samples had two distinct PFGE patterns differing from clinical isolates.

In this report, we described an outbreak of bacteremia caused by a rare pathogen, *S. paucimobilis*, during a 2-week period in a hemato/oncology unit. Hemato/oncology units have the highest incidence of nosocomial infection because of the intensified treatments and severe and prolonged immunosuppression. Rates of nosocomial infection among pediatric hemato/oncology patients have been found to range from 1.08 to 13.3%, with the most frequent episodes of nosocomial infection being causes of bacteremia (8). *P. aeruginosa* is one of the most common nosocomial pathogens isolated from this group of patients and represents an important cause of mortality and morbidity (9). *S. paucimobilis* especially causes opportunistic nosocomial infection in debilitated patients, but an established link with the source of infection generally fails (3).

Bacteremia caused by Gram-negative bacteria frequently results in considerable mortality and morbidity rates, especially among patients in hemato/oncology units. However, *S. paucimobilis* strains have not been strongly associated with death. The lack of the typical lipopolysaccharide constituent of the cellular membrane associated with endotoxin activity has been suggested to explain the apparent lack of lethality of these organisms. For treatment of *S. paucimobilis* infec-



tions, it has been suggested that imipenem alone or an aminoglycoside plus a third-generation cephalosporin might be adequate (2). In this outbreak, our isolates were susceptible to these antimicrobials. After intravenous antimicrobial therapy combining imipenem and teicoplanin, all patients survived.

S. paucimobilis isolates have been recovered from diverse sources including hospital water systems and respiratory therapy equipment (1). It also colonizes in the patients' digestive tract and perineal area during hospitalization. Various clones of S. paucimobilis are widely distributed in hospital environments including various indwelling devices and contamination of sterile or non-sterile water, and subsequently cause nosocomial infection (2). We isolated two S. paucimobilis strains from tap water and a bathtub, but they were distinct from the clinical strain by both antibiotic susceptibility and PFGE pattern. Although S. paucimobilis could be transmitted possibly via the hands of health care workers, we did not recover any S. paucimobilis strains in personnel samples during the outbreak period. During the outbreak, although no evidence for a unique source was found, the patient's rooms, beds, bedside equipment, bathroom surfaces, bathtubs, and sinks were cleaned and taps were changed thoroughly in the hemato/oncology unit. Implementation of these measurements successfully stopped the post-outbreak recurrence of S. paucimobilis infections.

In previous studies, molecular epidemiological typing has been used to determine the relatedness of clinical and environmental *S. paucimobilis* strains (2,10). But there are few typing techniques that can reliably determine the clonal relationship of samples of the same strain. The PFGE method is a useful basic tool for epidemiological studies in nosocomial infection. It has high discriminatory power and good reproducibility (11). In this study, *S. paucimobilis* strains yielded three different clones using the PFGE technique. All clinical strains were revealed as identical PFGE clones; however, environmental strains differed from them and from each other. We suggest that PFGE is a useful method for discrimination of *S. paucimobilis* stains causing outbreaks in oncology/ hematology units.

Although our clinical isolates and environmental isolates did not yield identical banding patterns using PFGE, it is clear that tap water should not be used for washing immunosuppressive children. The PFGE method can be effectively used for the epidemiological investigation of *S. paucimobilis* infections.

## ACKNOWLEDGMENTS

Fig. 1. Pulsed-field gel electrophoresis patterns of *Xba*I-digested genomic DNA of *Sphingomonas paucimobilis* isolates. Lanes 1 to 6, isolates from water, bathtub, patient 1's blood, patient 2's blood, patient 3's blood, and patient 4's blood samples. Molecular size (M) is given in kilobases.

We thank to Aylin Üskudar, Gulhane Military Medical Academy Microbiology Laboratory for his technical assistance.

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