## Peritonitis Caused by *Pseudomonas mesophilica* in a Patient Undergoing Continuous Ambulatory Peritoneal Dialysis

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Received 13 May 1988/Accepted 10 August 1988

We describe a case of recurrent peritonitis caused by *Pseudomonas mesophilica* in a diabetic man receiving continuous ambulatory peritoneal dialysis. Stagnant water on a bath rail used for support by the patient while showering was implicated as the probable source of these infections. We believe this to be the first report of the isolation of this water-associated bacterium in such infections.

The management of end-stage renal failure by continuous ambulatory peritoneal dialysis (CAPD) is now well established. Peritonitis remains the main complication of CAPD and is indicated when the patient has abdominal pain and produces cloudy dialysates with more than 100 leukocytes per mm<sup>3</sup>. A review of the usual microbial etiology of peritonitis complicating CAPD (2) indicated that coagulasenegative staphylococci cause about half of all episodes, Staphylococcus aureus causes about 15%, and members of the family Enterobacteriaceae and Pseudomonas aeruginosa together are responsible for about 20%. Fungi, usually yeasts, cause about 2% of infections. Streptococci, diphtheroids, miscellaneous microorganisms, and, rarely, anaerobes and mycobacteria account for the remainder of the infections. Almost any microorganism seems to be capable of producing peritonitis in CAPD patients, but this appears to be the first time that *Pseudomonas mesophilica* has been implicated in such an infection.

A 53-year-old man with insulin-dependent diabetes mellitus, associated with multiple complications, suffered repeated episodes of peritonitis. The complications included end-stage renal failure secondary to diabetic nephropathy, hypertension, peripheral vascular disease, peripheral and autonomic neuropathy, and diabetic retinopathy resulting in blindness. Chronic renal failure secondary to diabetic nephropathy was first diagnosed in 1984; the patient's renal function steadily deteriorated, requiring the commencement of peritoneal dialysis in July 1986. After stabilization of his uremic state, he was discharged from the hospital to continue CAPD procedures at home. Because of his blindness, the bag changes and administration of intraperitoneal insulin were performed by his wife. The patient's course on CAPD was complicated by intermittent peritonitis, chronic infection of the catheter tract with abscess and cutaneous sinus infection, and a deterioration in his peripheral vasculature.

The first episode of peritonitis associated with CAPD occurred on 4 August 1986, with *Enterobacter taylorae* implicated as the causative agent. Further episodes of peritonitis, with abdominal pain, cloudy dialysates, and raised total leukocyte counts, occurred on 16 August 1987, 11 September 1987, and 13 October 1987. On each occasion, *P. mesophilica* was isolated.

Although suspected initially of being the cause, the chronic tract infection was not related to the episodes of peritonitis. *S. aureus* was isolated from the swabs of the exit

site of the dialysis catheter and sinus tract, but despite repeated culture, *P. mesophilica* was never isolated from these sites. Treatment with intraperitoneal piperacillin was initiated on 19 August 1987, but although initial clearing of the dialysate was observed, the peritonitis recurred. Treatment with gentamicin was begun, and the patient responded well. There have been no further episodes of peritonitis since that time.

On receipt of the peritoneal dialysates for culture in the laboratory, a sample of 50 ml was withdrawn aseptically from the bag. A volume of 20 ml was delivered into each of 50 ml of tryptone soya broth (Oxoid Ltd., Basingstoke, England) and 80 ml of thioglycolate broth USP (CM173; Oxoid), and the remaining 10 ml was centrifuged in a sterile centrifuge tube at  $1,000 \times g$  for 5 min. A few drops of uncentrifuged dialysate were reserved for a total leukocyte count. The supernatant fluid was discarded, and the centrifuged deposit was used to make a film for Gram staining and to inoculate a 5% (vol/vol) horse blood agar (HBA) plate (Columbia base; Oxoid) containing 0.05% sodium polyanetholesulfonate (Roche Diagnostics, Basel, Switzerland), MacConkey agar no. 2 (CM109; Oxoid), and a Sabouraud dextrose agar (SAB) plate (Oxoid) without antibiotics. The broths were incubated at 35°C for 1 week, the HBA and MacConkey plates were incubated at 35°C for 48 h in 10% CO<sub>2</sub> atmosphere, and the SAB plate was incubated at 30°C aerobically for 1 week.

Bacteria were not detected in the Gram-stained smears. Total leukocyte counts ranged from 220 to  $490/\text{mm}^3$ . After incubation at 30°C for 3 to 4 days, small, pink-pigmented colonies appeared on the SAB plates. The broth cultures were subcultured on day 4 to HBA and MacConkey agar and yielded the same bacterium. The isolate stained as a vacuolated, gram-negative, rod-shaped bacterium. It was referred to the Microbiological Diagnostic Unit, where it was identified as *P. mesophilica*.

All bag changes had taken place in the patient's home. Four samples of his domestic tap water were taken, one from each of the hot and cold water faucets over a washbasin in the bathroom and one from each of the hot and cold water supplies to the shower head. The samples were collected in 200-ml sterile bottles containing 0.2 ml of 3% (wt/vol) sodium thiosulfate to neutralize residual chlorine. Four 10-ml aliquots of tap water from each of the four sites were centrifuged at 1,000  $\times$  g for 20 min. The supernatant fluids were discarded, and the centrifuged deposits were used to inoculate buffered charcoal yeast extract agar (BCYE) plates

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 TABLE 1. Properties of the isolates of P. mesophilica from peritoneal dialysates and environmental waters

Characteristic or test	Result
Microscopic morphology	Vacuolated rods
Flagella	Single polar
Pigmentation	Pink or orange-pink
Growth at 30 and 35°C	+
Growth at 37 and 42°C	
Catalase	+
Oxidase	+
Urease (Christensen)	+
Nitrate reduction	
Gas from nitrate	
Simmons citrate	–
Malonate utilization	+
Indole production	–
Acid from TSI <sup>a</sup> slant	
Acid from TSI butt	–
H <sub>2</sub> S (TSI butt)	
Acid from ASS <sup>b</sup>	
Arabinose, ethanol, glycerol, xylose	+
Glucose, lactose, maltose	
Mannitol, rhamnose, sucrose	
Hydrolysis of:	
Casein, esculin, gelatin	
Tween 80, tyrosine, lecithin	
Starch	+

" TSI, Triple sugar iron agar.

<sup>b</sup> ASS, Ammonium salt-sugar medium.

(medium for Legionella spp.) and blood agar base no. 2 and SAB plates without antibiotics. All plates were incubated at  $30^{\circ}$ C for 1 week. *P. mesophilica* was cultured from the hot water from the faucet over the washbasin, but the other three water samples did not grow *P. mesophilica*. Swabs were also taken from stagnant water observed lying on top of the bath rail on the wall under the shower head and from the mixer tap of the shower, and these samples were similarly cultured on BCYE and SAB. The swab of the stagnant water on the bath rail yielded a profuse growth of *P. mesophilica* on day 3 of incubation. The swab from the mixer tap of the shower gave 3 CFU of *P. mesophilica*.

The growth characteristics and biochemical reactions of the isolates from the peritoneal dialysates and the environmental waters are given in Table 1. All isolates are of the same biotype since they gave the same results for those characteristics for which strain differences have been reported for *P. mesophilica* (3-5), e.g., nitrate reduction, utilization of citrate (Simmons), and acid production from glucose, rhamnose, and xylose.

The antimicrobial susceptibility of the isolates was tested by the disk diffusion method (8). The method was modified by reducing the temperature of incubation to 30°C and extending the time of incubation to 48 h to comply with the growth requirements of the species. Furthermore, the isolate from the water on the bath rail failed to grow on the recommended Mueller-Hinton medium, so the antimicrobial susceptibility of all isolates was also tested on blood agar base no. 2, which did support the growth of the isolate from the bath rail. All isolates were susceptible to piperacillin, cephalothin, and gentamicin and were resistant to co-trimoxazole on both media. They were also susceptible to ampicillin, although one of the isolates from the patient showed reduced susceptibility (intermediate susceptibility) when tested on the blood agar base no. 2.

Swabs were also taken of the Tenckhoff catheter exit site, an abdominal sinus, the inside and outside of the tubing connecting the bag to the catheter, and the povidone-iodine sponge which overlaid the connection of the catheter tube and the bag tube. The swab of the abdominal sinus grew S. *aureus*. None of the other swabs yielded any growth.

The chlorhexidine antiseptic skin cleanser (Hibiclens; ICI Australia, Villawood, New South Wales, Australia) used by the patient's wife to disinfect her hands before bag changes was tested by the in-use test of Kelsey and Maurer for disinfectant failure leading to bacterial contamination (7). No growth was obtained, indicating that the antiseptic skin cleanser was not contaminated and not a source of infection.

*P. mesophilica* is a pink-pigmented, gram-negative, rodshaped bacterium originally described as being isolated from leaf surfaces (1). It has been isolated from tobacco leaves, perennial rye grass leaves, methane-oxidizing enrichment cultures from soil, sewage, the rumen of cows, water, nebulizers, and human clinical materials such as throat swab, bronchial washing, and blood (3, 4). A few cases of human infection have been described, including severe skin ulcers in a Puerto Rican woman (6) and bacteremia in a patient with metastatic adenocarcinoma of the lung (9). *P. mesophilica* has also been reported from blood and nasopharyngeal culture from patients in a bone marrow transplant unit who had received oral or perineal irrigations with saline made up from tap water which was contaminated with *P. mesophilica* (5).

*P. mesophilica* has been known by a variety of synonyms, including "Mycoplana rubra," "Vibrio extorquens," "Protaminobacter ruber," and "Pseudomonas methanica" (1, 4). Urakami and Komagata (10) have proposed a new genus, Protomonas, for *P. mesophilica*, with the type species Protomonas extorquens comb. nov. They demonstrated that *P. mesophilica* is a methanol-utilizing bacterium, distinguishable from typical species of *Pseudomonas* by its cellular fatty acid composition, its ubiquinone systems, and the formation of bacteriochlorophyll.

Our investigations indicate that the original source of infection was probably the domestic water supply in the patient's home, but the exact mode of entry into the peritoneal cavity was not demonstrated. Shower water was allowed to remain for long periods, possibly continuously, on the bath rail under the shower, allowing the bacteria to multiply in that site. The patient used this rail to support his bag and line while showering. When in the shower, he also leaned on the bath rail to support himself, as he had difficulty in balancing. Observation of the bag-changing procedure indicated that the wife might not have left the Hibiclens solution on long enough for effective skin disinfection after washing her hands in the contaminated hot water. The patient's wife was instructed on how to disinfect the rail and was advised to keep it dry and not to allow pools of water to accumulate anywhere. A review of bag-changing and handwashing procedures was also undertaken. No further episodes of peritonitis have occurred since that time.

We agree with the views of Gilchrist et al. (5) that many clinical laboratory protocols do not include media, incubation temperature, or times appropriate for the detection of *P. mesophilica*. Like them, we first isolated this bacterium on medium used for the culture of fungi. Apart from SAB, the isolates grew well on BCYE and Oxoid blood agar base no. 2. They also grew on 5% HBA, but pigmentation was not as obvious as it was on other media. The isolates grew at 35°C but not at 37°C. Another problem is that the databases of the Vitek AMS system (Vitek Health Systems, Hazelwood, Mo.) and the 20 NE API system (La Balme Les Grottes, Montalieu-Vercieu, France) do not include *P. mesophilica*. Although skin bacteria cause the majority of infections in patients undergoing CAPD, microorganisms associated with water also represent a hazard. City water supplies contain many different species of bacteria which, under certain conditions, may proliferate and act as sources of infection, resulting in peritonitis in such patients. Improvements in technology and treatment during the past decade have greatly improved the prospects for diabetics with end-stage renal failure who are undergoing CAPD, but the potential for infection by water-associated microorganisms, among other opportunistic pathogens, must be recognized in the management of disease in these patients.

We thank Lorna Barclay for her technical assistance.

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