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Peritonitis Associated with a CDC Group EO-3 Organism

DENISE DALEY,* STEPHEN NEVILLE, AND KATHERINE KOCIUBA

Department of Microbiology and Infectious Diseases, South Western Area Pathology Service, Liverpool, New South Wales 2170, Australia

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A 63-year-old female with chronic renal failure on continuous ambulatory peritoneal dialysis developed chronic peritonitis. A CDC group EO-3 organism was isolated from the peritoneal dialysis fluid on five occasions over a period of 4 months. This is the first reported isolation of this organism in which it is associated with a patient on continuous ambulatory peritoneal dialysis.

Peritonitis remains the major complication of continuous ambulatory peritoneal dialysis (CAPD) (8). Recurrent peritonitis occurs in 20 to 30% of patients and is one of the most common reasons for discontinuation of CAPD. The origin of infection in most cases appears to be contamination of the catheter with common skin organisms and enteric bacteria. Subcutaneous tunnel infections, transient bacteremia, and contamination of the dialysate delivery system during bag exchange may also occur (4, 10). Gram-positive organisms comprise 60 to 80% of clinical isolates, while gram-negative organisms are isolated in 15 to 30% of cases. The recommended empiric regimen is vancomycin in combination with an aminoglycoside, administered via the intraperitoneal (i.p.) catheter; however, removal of the catheter may be necessary in 10 to 20% of patients (3, 4). We report the isolation of a CDC group EO-3 organism on five occasions from the CAPD fluid from a patient with chronic peritonitis. The source and route of infection in our patient's case are not known. To date, the CDC group EO-3 organism has not been described as a causative agent of infection, and its environmental niche remains unknown. To our knowledge, this is the first reported case of chronic peritonitis associated with a CDC group EO-3 organism.

A 63-year-old female with chronic renal failure, due to diffuse vascular disease, commenced CAPD in 1991. In 1993, she was further diagnosed with carcinoma of the breast, which was treated with mastectomy, deep X-ray therapy, and tamoxifen. In January 1994, she presented with a 2-day history of abdominal pain and cloudy CAPD bags. Empiric i.p. vancomycin and gentamicin were commenced, with clinical improvement. A gram-negative coccobacillus was isolated from the CAPD fluid on two separate occasions. The same organism was reisolated from CAPD fluid in early February. In late February, the patient presented with another episode of peritonitis. *Escherichia coli* was isolated on this occasion. The coccobacillus was isolated again in March 1994, with clinical response to empiric i.p. vancomycin and gentamicin.

In April 1994, the patient represented with cloudy CAPD bags, right abdominal pain, fever, nausea, and vomiting. She had lost 4 kg in weight since January and had worsening anemia (hemoglobin, 7.5 g/liter), and the gram-negative coccobacillus was isolated again from CAPD fluid. She was transfused, and her chronic CAPD peritonitis responded to a 4-week course of i.p. vancomycin, amikacin, and oral erythromycin. When symptoms recurred in June, the Tenckhoff catheter was removed, with cure of her chronic peritonitis. Following this, no further episodes of peritonitis associated with this organism occurred. Neither catheter nor exit site swabs were sent to the laboratory. During five separate episodes of peritonitis over a 4-month period, seven peritoneal dialysate fluid samples were submitted to the microbiology laboratory. Total and differential leukocyte counts were performed, and a Gram stain was prepared from a centrifuged deposit. Quantitative cultures were performed with 9% blood agar plates lysed with 0.05% saponin (LBA) (8). The fluid was also cultured onto two plates of 5% horse blood agar (HBA), a MacConkey agar (MAC) plate (Becton Dickinson Pty Ltd., Underwood, Queensland, Australia), and a chocolatized HBA plate (Oxoid, Medvet Science Pty Ltd., West Heidelberg, Victoria, Australia). One of the HBA plates, the LBA plate, and the chocolatized HBA plate were incubated in 5% carbon dioxide at 35°C. The remaining HBA plate was incubated anaerobically at 35°C. The MAC plate was incubated aerobically at 35°C. All plates were incubated for up to 5 days. In addition to the above direct cultures, 10 ml of the fluid was aseptically inoculated into a Bactec Plus 26A blood culture vial (Becton Dickinson, Sparks, Md.), supplemented with 3 ml of sterile horse blood. The bottle was incubated in a Bactec NR-860 Automated Blood Culture System. When flagged as positive by the instrument, the bottle was subcultured onto the agars mentioned above.

The biochemical profile of the isolate was determined by conventional biochemical tests (1) (Table 1). Carbohydrate assimilation was determined by the ATB ID32GN identification strip (bioMerieux-Vitek, Sydney, Australia).

Antimicrobial susceptibility testing was performed by disk diffusion according to National Committee for Clinical Laboratory Standards guidelines (6). MICs were determined by Etest (AB Biodisk, Solna, Sweden) according to the manufacturer's instructions. The organism was referred to the Centers for Disease Control and Prevention (CDC), Ga., for confirmation of the identification.

Six of the seven peritoneal dialysate fluid samples submitted produced total leukocyte counts of $>100/mm^3$. Eighty percent of the leukocytes were polymorphonuclear cells. Only one fluid sample yielded a leukocyte count of $<10/mm^3$. On this occasion, the fluid sample was sterile following successful treatment of the *E. coli* peritonitis. From the seven specimens submitted, a yellow-pigmented gram-negative coccobacillus was isolated on a total of five occasions. From the first three specimens (episodes 1 and 2), the organism was isolated from the broth cultures only. *E. coli* was isolated from the next fluid sample collected (episode 3), with a posttreatment specimen produc-

* Corresponding author. Mailing address: Department of Microbiology and Infectious Diseases, S.W.A.P.S., Locked Mail Bag 90, Liverpool, N.S.W. 2170, Australia. Phone: (02) 9828 5144. Fax: (02) 9828 5129. E-mail: Denise.Daley@swsahs.nsw.gov.au.

TABLE 1. Morphological and biochemical characteristics of case isolate

Feature or test	Result
Growth on MAC	+
Hemolysis	−
Colony size (mm)	<0.5
Gram stain reaction	−
Morphology	Coccobacilli, straight rods
Motility	−
Catalase	+
Oxidase	+
Growth on cetrimide agar	−
Simmons citrate (days)	− (5)
Esculin hydrolysis	−
Indole	−
Nitrate to nitrite	−
Nitrite to gas	−
Urea	+
KIA slant[a]	Alkaline
KIA butt	No change
H_2S KIA	−
Gelatin	−
Nonsoluble pigment	Yellow
Acetimide	−
Serine	−
Acidification (OF medium)[b]	
Glucose (days)	Acid (1–2)
Xylose (days)	Acid (1–2)
Mannitol	Weak acid
Lactose	Neutral[c]
Sucrose	Neutral
Maltose	Neutral
Control	Alkaline
Arginine	+
Lysine	−
Ornithine	−
Growth at:	
25°C	+
35°C	+
42°C	−

[a] KIA, Kligler's iron agar.
[b] OF, oxidation-fermentation. See reference 1.
[c] Neutral, weak acid if control is alkaline.

ing no growth. A further two specimens (episodes 4 and 5) were collected, and both grew the yellow-pigmented gram-negative coccobacillus on direct (>50 CFU/ml) and broth cultures. On each occasion, the organism grew aerobically on MAC and on HBA and LBA in carbon dioxide but not on HBA anaerobically. Colonies on HBA at 24 h were nonhemolytic and pinpoint (<0.5 mm in diameter). After 48 to 72 h, colonies were glossy and a yellow pigment was apparent. On Gram staining, the organism was a pleomorphic gram-negative coccobacillus. There were some gram-positive elements retained in occasional organisms. The biochemical reactions reported in Table 1 were confirmed by the CDC, with the exception of citrate utilization.

The ATB ID32GN produced an identification profile number (06102200100) without a satisfactory identification, although the isolate was able to assimilate glucose, arabinose, maltose, gluconate, mannose, and malate. The results of the cellular fatty acid analysis of the case isolate (79% of 18:1; 10.4% of 16:0; 5% of 18:0; 3% of 19:0cyc [11, 12]; and 1% each of 20:1, 16:1*w*7c, 18:2, and 18:1*w*9c) were consistent with the profile of known strains of CDC group EO-3 (99.14% agreement [11a]).

The organism was found to be susceptible by disk diffusion to ampicillin, cefotaxime, ceftazidime, gentamicin, tobramycin, imipenem, amikacin, ticarcillin, and chloramphenicol and resistant to ciprofloxacin and trimethoprim. The MICs of the following antibiotics were determined at the request of the consulting physician: ampicillin (MIC, 0.25 μg/ml), cefotaxime (MIC, 0.064 μg/ml), ceftazidime (MIC, 1.0 μg/ml), gentamicin (MIC, 1.0 μg/ml), tobramycin (MIC, 1.0 μg/ml), imipenem (MIC, 0.032 μg/ml), and ciprofloxacin (MIC, 16 μg/ml).

The CDC group EO-3 organism (5, 7, 9, 12) is an aerobic, gram-negative coccoid to short thick rod. It is strongly oxidase positive, nonmotile, and indole negative and utilizes glucose, lactose (usually weak or late), xylose, mannitol (weak), and sometimes maltose. The EO-3 organism has a yellow nondiffusible pigment, lacks the distinctive O-shaped cellular morphology found in the EO-2 organism, and is unreactive in the *Psychrobacter immobilis* transformation assay (2).

The isolation of CDC group EO-3 organisms from clinical specimens appears to be rare. The sources of seven strains were as follows: three from blood and one each from eye, forearm, stitch abscess, and vagina (12). The clinical significance of this group of organisms is not known; however, this case report is of value to the clinical laboratory in that it is the first to associate the CDC group EO-3 organism with peritonitis.

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