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Third Human Isolate of a *Desulfovibrio* sp. Identical to the Provisionally Named *Desulfovibrio fairfieldensis*

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***Desulfovibrio fairfieldensis* was isolated from the urine sample of a patient with a urinary tract infection and meningoencephalitis. It was identified by 16S rRNA gene amplification and sequencing.**

CASE REPORT

A 46-year-old woman was admitted to a hospital for exploration of febrile meningoencephalitis. A *Desulfovibrio fairfieldensis* strain was isolated from a urine specimen of this patient. All microbiological investigations, including blood and cerebrospinal fluid cultures, remained negative. The patient was treated with rifampin, ethambutol, isoniazid, acyclovir, amoxicillin, and, several days later, corticosteroids. On admission, a microscopic examination of urine yielded 10^6 polymorphonuclear leukocytes per ml and long motile bacteria with spirochetal morphology. Treatment with antimycobacterial drugs was continued, and clinical symptomatology improved slowly. Two weeks later neither pyuria nor bacteriuria was detected. Six months later, the patient's condition had deteriorated. No pyuria or bacteriuria was detected, and a urine sample showed no growth aerobically or anaerobically. The patient's condition fluctuated between periods of improvement and relapse over the following months. After another 6 months the patient died. The family of the patient refused permission for an autopsy to be carried out.

Two weeks after inoculation of urine into antibiotic-free Ellinghausen and McCullough medium modified by Johnson and Harris (EMJH medium; Difco, Detroit, Mich.) incubated at 30°C, bacilli in a spirochete form were detected. As growth occurred mostly at the bottom of the tube, EMJH culture was inoculated onto 5% sheep blood agar and incubated under strict anaerobic conditions. After 5 days, numerous pinpoint nonhemolytic brown colonies were detected. In the API 20 A system (BioMerieux, Marcy l'Etoile, France) the organism remained inactive, with the exception of esculin, which was positive. Identification was performed by using 16S rRNA gene amplification and sequencing (6). Briefly, DNA extracts were obtained from 200 μl of bacterial suspension by using the QIA ampBlood kit (Qiagen, Hilden, Germany). These were amplified by a PCR incorporating universal primers fD1 (5′-AGA GTTTGATCCTGGCTCAG-3′) and rP2 (5′-ACGGCTACCT TGTTACGACTT-3′). After purification of the amplification products, sequencing reactions were prepared with the Amplicycle sequencing kit (Perkin-Elmer, Norwalk, Conn.). Reaction products were resolved by electrophoresis on a 6% polyacrylamide gel incorporated onto an ALF automatic sequencer (Pharmacia Biotech, Uppsala, Sweden). The complete sequence was compared to all bacterial sequences available from the GenBank database by using the Blast 2.0 program (National Center for Biotechnology Institute) and showed 100% similarity to that of *D. fairfieldensis* (GenBank accession no. U42221). Antibiotic susceptibility was determined by using the Epsilometer test (E test; AB Biodisk, Solna, Sweden) on Wilkens-Chalgren agar, incorporating 5% defibrinated sheep blood (1). An inoculum with a turbidity equivalent to that of McFarland standard no. 1 was used. Because of the slow growth of the organisms MICs were determined after 72 h of anaerobic incubation at 37°C. The MICs of the following antibiotics were determined: penicillin (>256 μg/ml), amoxicillin (>256 μg/ml), amoxicillin-clavulanate (3 μg/ml), imipenem (0.75 μg/ml), ciprofloxacin (0.38 μg/ml), rifampin (0.5 μg/ml), clindamycin (0.75 μg/ml), metronidazole (0.06 μg/ml), and chloramphenicol (0.125 μg/ml). An assay for the detection of beta-lactamase by using a nitrocefin disk (7) was negative.

Desulfovibrio species are members of the delta subgroup of *Proteobacteria*. They are phylogenetically closely related to several other anaerobic sulfate-reducing bacteria, such as *Desulfomonas* and *Desulfomicrobium*, and especially several animal enteric pathogens including *Lawsonia intracellularis* (3). Some species have been proposed as possible agents of ulcerative colitis in humans (4). Spirillar forms occur in late cultures, in cultures stressed with antibiotics, in Mg^{2+} deprivation, and in very sulfide-rich cultures. To date, six clinical isolates of *Desulfovibrio* species have been reported, including one from a patient with septicemia (8), two from cases of acute appendicitis (2), and a fourth from a patient with an abdominal abscess (5). The fifth and sixth cases, both described in Australia, involved a patient with a liver abscess (9) and another with bleeding colonic polyps (7). In our leukocyturia patient, it is possible that *D. fairfieldensis* was responsible for a urinary tract infection, since it was the sole microorganism isolated from urine and it was observed by direct examination. With the exception of esculin, phenotypic characteristics were identical to those of previous reports. Bacteria with internal bulbous swelling as previously described were not observed (7).

The antibiotic susceptibility pattern of our isolate was comparable to that observed in the second described isolate of *D. fairfieldensis* (9). Even though the MIC of amoxicillin was lowered by the addition of a beta-lactamase inhibitor, beta-lactamase activity was not detected.

This case highlights the value of PCR amplification, sequencing, and comparison to sequence databases of 16S rRNA genes for the identification of uncommon or new pathogens.

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