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Rahnella aquatilis, an Unusual Gram-Negative Rod Isolated from the Bronchial Washing of a Patient with Acquired Immunodeficiency Syndrome

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***Rahnella aquatilis*, a rare enteric gram-negative rod which is usually found in fresh water, was isolated from the bronchial washing of a patient with acquired immunodeficiency syndrome. Although few clinical isolates have been reported, this is the second isolation of *R. aquatilis* from a human in North Carolina. A case report and discussion of *R. aquatilis* is presented.**

Rahnella aquatilis, a rare gram-negative rod, is a member of the family *Enterobacteriaceae*. In 1976 Gavini et al., using numerical taxonomy and phenotypic characteristics, identified a group of *Enterobacteriaceae* that was designated H2 (3). In 1979, by means of DNA-DNA hybridization techniques, the H2 group was shown not to have a close genetic relationship to any known species or genera of *Enterobacteriaceae*. Thus, the new name *R. aquatilis* was proposed (4): *Rahnella* to honor the German-American microbiologist Otto Rahn, and *aquatilis* because the natural habitat of the species is water. In the culture collection at the Centers for Disease Control (CDC), there are 21 isolates, 11 of which are the original French water isolates (2). There are two isolates in the CDC collection from humans; both were isolated from patients in North Carolina. One isolate is from a burn wound, and the other is from the bronchial washing of a patient with acquired immunodeficiency syndrome. The additional eight isolates in the CDC collection are from four states (Virginia, Georgia, Oregon, and West Virginia); seven are from water, and one is from wood dust of a pine tree. In this paper we present the case report for the acquired immunodeficiency syndrome patient and review the biochemical characteristics of *R. aquatilis*.

Case report. The patient was a 37-year-old male homosexual who was in good health until November 1982, when he presented to his physician with right unilateral cranial nerve V_1 herpes zoster with nerve VI palsy and pupillary motor fiber involvement which resolved after treatment with acyclovir and prednisone. He remained well until July 1984, when he was admitted with a 2-month history of lymphadenopathy, diarrhea, and a 30-lb (1 lb = 453.592 g) weight loss. As an outpatient at another hospital, he had been treated with trimethoprim-sulfamethoxazole for proctitis during the preceding month and then developed an allergic pneumonitis and dermatitis. During his hospitalization he had negative serological results for cytomegalovirus, toxoplasmosis, human immunodeficiency virus, syphilis, and hepatitis B surface antigen. His serology was positive only for hepatitis B surface antibody. He also underwent a lumbar puncture and a bronchoscopy with transbronchial biopsy; both of these procedures yielded normal results, including negative cultures for fungi and mycobacteria. Workup of his diarrhea revealed *Cryptosporidium* species, which disappeared completely with outpatient spiramycin therapy.

In August 1985 he presented to his physician with generalized pruritus secondary to biopsy-proven lichen simplex chronicus, which was treated with topical fluocinonide. He also had decreased and blurred vision in his right eye. On examination, he was found to have diplopia on right lateral gaze, right-hand dysesthesia, and right-arm hypoaesthesia. He was tested by computerized cranial tomography with contrast, and the results were normal. He had converted to seropositive for HIV by enzyme-linked immunosorbent assay. No confirmation by Western blot (immunoblot) was done.

He presented to the hospital in February 1986 after 3 weeks of productive cough, fever, night sweats, diarrhea, weight loss, and headache. Upon physical examination, he had an oral temperature of 38°C, blood pressure of 90/54 mm Hg, pulse of 84/min supine which changed to 100/min standing, and a respiratory rate of 18/min. He appeared to be chronically ill but not in acute distress. His skin showed papular hyperpigmented areas with central scaling. He had a swollen anterior cervical lymph node (1 by 1 cm) and swollen bilateral 0.5-cm inguinal nodes with no axillary or supraclavicular adenopathy. His head, eyes, ears, nose, and throat examination was normal except that he had white adherent patches on the buccal mucosa. His lungs were clear to auscultation and percussion. His cardiovascular examination was normal, with no murmurs, rubs, or gallops. His abdominal, rectal, and genital examinations were normal. His neurologic examination was also normal.

Significant laboratory results were as follows: leukocyte count, 1,800/mm^3; hemoglobin, 9.5 mg/dl; and hematocrit, 31.1%, with 28% neutrophils, 14% lymphocytes, 40% monocytes, and 14% bands with 243,000 platelets per mm^3. He had normal electrolytes, renal function, urinalysis, and electrocardiograph. His arterial blood gas on room air had a pH of 7.55, partial O_2 pressure of 109 mm Hg, and partial CO_2 pressure of 24 mm Hg.

His chest X ray showed bilateral interstitial infiltrates which were most marked in the middle and lower lobes. Because his clinical presentation was consistent with *Pneumocystis carinii* pneumonia, he was started on intravenous (i.v.) pentamadine at 4 mg/kg of body weight per day. He was also given ampicillin and gentamicin, pending sputum

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culture results. The day after admission he underwent fiberoptic bronchoscopy with transbronchial biopsy (1). All special stains of the bronchial washings and biopsy specimens were negative, but routine culture revealed normal flora, moderate amounts of yeast cells, *Klebsiella pneumoniae*, and a lactose-fermenting gram-negative rod that was later identified as *R. aquatilis* by the CDC. The patient also underwent a lumbar puncture. Results for cerebrospinal fluid were as follows: protein, 19 mg/dl; glucose, 56 mg/dl (serum glucose, 77 mg/dl); leukocyte count, 1/mm^3 (70% lymphocytes, 30% monocytes); erythrocyte count, 3/mm^3; and a negative test for syphilis. The India ink preparation was negative, but the cerebrospinal fluid cryptococcal antigen was 1:2 (Crypto-LA kit; International Biological Laboratories, Inc., Cranbury, N.J.) and the culture grew *Cryptococcus neoformans*. He was started on amphotericin B (0.3 mg/kg per day i.v.) on day 2 of hospitalization. He became afebrile on day 3 of hospitalization, and the pentamadine, ampicillin, and gentamicin were discontinued when the bronchoscopy results were known.

During the second week of hospitalization he had recurrent fever. Workup of his fever included a cranial computerized tomography with contrast, which revealed sphenoid sinusitis and early communicating hydrocephalus. His sinusitis was treated with resumption of ampicillin and gentamicin i.v. for 10 days; rapid defervescence resulted. He was discharged after a 1-month hospitalization and was put on home i.v. amphotericin B therapy (total dose, 606 mg, and then a maintenance dose of 72 mg i.v. three times per week) with a Karnofsky status of 60 to 70%.

His subsequent course was significant for a Hickman catheter exit site infection with methicillin-resistant *Staphylococcus aureus* which was successfully treated with vancomycin and gentamicin for 2 weeks. He remained on amphotericin B treatment at a reduced dose (maintenance dose, 72 mg/week) for his cryptococcal meningitis. *R. aquatilis* was not isolated from any further clinical specimens. He had an inexorable downhill course and died from complications of acquired immunodeficiency syndrome in July 1986. No autopsy was done.

Bacteriology. *R. aquatilis* was isolated from the bronchial washing on MacConkey agar after 48 h of incubation at 35°C. It was lactose fermenting, and triple sugar iron agar results showed an acid-acid reaction with gas. The API 20E (Analytab Products, Inc., Plainview, N.Y.) profile number 1105523 gave an excellent identification for CDC enteric group 17. Biochemical results from the MicroScan Combo Plus broth dilution panel (Baxter MicroScan, West Sacramento, Calif.) did not render an identification. The isolate was then referred to the North Carolina State Laboratory of Public Health and thence to CDC, where, on the basis of biochemical reactions listed in Table 1, it was identified as *R. aquatilis*. Key sugars which were fermented included D-glucose, lactose, maltose, L-rhamnose, raffinose, and salicin. The isolate was weakly positive for phenylalanine deaminase, but tests for lysine and ornithine decarboxylases and arginine dihydrolase were negative. The Voges-Proskauer test was positive, motility was noted at 22°C, and yellow pigment was absent.

Antimicrobial susceptibility tests were performed with the MicroScan Combo Plus broth dilution panel. The *R. aquatilis* and *K. pneumoniae* isolates were susceptible to gentamicin, tetracycline, mezlocillin, chloramphenicol, and trimethoprim-sulfamethoxazole. Both microorganisms were resistant to ampicillin, but only *R. aquatilis* was resistant to cephalothin and cefazolin.

TABLE 1. Biochemical characteristics of *R. aquatilis*

Test	Reaction[a]
Motility (22°C)	+
Nitrate reduction to nitrite	+
Lysine decarboxylase	−
Ornithine decarboxylase	−
Arginine dihydrolase	−
Phenylalanine deaminase	+
Methyl red	+
Voges-Proskauer	+
Carbohydrate fermentation	
D-Glucose	+
Lactose	+
Maltose	+
L-Rhamnose	+
Raffinose	+
Salicin	+
Yellow pigment (25°C)	−

[a] −, Negative at the end of the appropriate incubation period; +, positive at 24 h or at time of test. Tests were incubated at 35 ± 1°C for 48 h except as noted.

Discussion. *R. aquatilis* is difficult to distinguish from other *Enterobacteriaceae* because it has no single distinguishing characteristic. Often the first clues to its identification are a weakly positive phenylalanine deaminase reaction and the absence of yellow pigment. Otherwise, it resembles *Enterobacter agglomerans*. The ability to distinguish the organism is lacking in the data bases of all commercial identification systems currently on the market.

Because this isolate was found in only one specimen from the patient discussed here and was present along with another potential pathogen, *K. pneumoniae*, its clinical significance is unclear. However, the patient had an apparent clinical improvement with defervescence after resumption of therapy with gentamicin, to which the organism is susceptible, and presumably all solutions used during bronchoscopy were sterile. Hence, the clinical picture is not inconsistent with a pulmonary infection caused by *R. aquatilis* or *K. pneumoniae* or both.

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