

Leptotrichia buccalis Bacteremia in Neutropenic Children

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Two new cases of *Leptotrichia buccalis* bacteremia in seriously ill patients were described. The anaerobic, gram-negative microorganism *L. buccalis* was isolated from blood cultures of two children with severe leukopenia. Anaerobic organisms should be taken into account when a standard protective antibiotic chemotherapy is considered in the immunosuppressed host.

Leptotrichia buccalis is the only species of the genus *Leptotrichia* currently included in the family *Bacteroidaceae*. It is an anaerobic gram-negative bacillus, as are the members of the other genera of the family, *Bacteroides* and *Fusobacterium*, but some strains can grow in air in the presence of CO₂. Like other *Bacteroidaceae*, *L. buccalis* forms part of the normal human flora (10). As with other components of the dental plaque, *L. buccalis* may play a role in the etiology of periodontal diseases or in oral-related abscesses. There are only three reports (2, 9, 12) in which *L. buccalis* has been recovered in blood cultures from seriously ill patients. Two new cases of *L. buccalis* bacteremia associated with severe disease in neutropenic patients are described in this paper, and these are the first observations of this type of bacteremia in children.

Case 1. A 14-year-old boy suffering from osteogenic sarcoma was induced (5th cycle) with methotrexate (14 mg per day) and vincristine (1.9 mg per day). A day 6 after induction the patient had a temperature of 38.5°C, nausea, vomiting, diarrhea, mouth ulcerations, thrombocytopenia, and leukopenia (1,000/mm³). Blood cultures were taken by inoculation of four bottles of brain-heart infusion broth, and treatment of the patient was begun with intravenous ampicillin (1.5 g/6h) and tobramycin (50 mg/8 h). After 2 days an anaerobic organism was detected in the blood culture bottles, and metronidazole (160 mg/6 h) was added to the treatment. On the day 6 after the commencement of symptoms, fever decreased to normal (previous maximum temperature, 39°C). The anaerobic organism isolated in pure culture from the bottles was identified as *L. buccalis* (8).

Case 2. A 9-year-old girl (HLA type ABC4a4b) was admitted to the hospital with severe thrombocytopenia and leukopenia (900/mm³) due to a medullary aplasia of uncertain etiology. Under appropriate protective isolation, standard antibiotic prophylaxis was begun with intravenous cefotaxime (2 g/8 h) and tobramycin (40 mg/8 h). This treatment was discontinued after 1 week. Fever then appeared (38.8°C), and *Escherichia coli* was isolated by blood cultures. Tobramycin treatment was recommended, but 3 days later temperature remained high, mouth ulcerations appeared, and a sterile abscess had formed at the site of the medullary biopsy. New blood cultures yielded gram-negative anaerobic organisms and metronidazole treatment (250 mg/6 h) was started. Despite the improvement of the abscess, an acute multifocal interstitial pneumonia developed, and the patient died owing to cardiac arrest 4 days later. The anaerobic organism isolated in pure culture was identified as *L. buccalis*.

In both cases Gram stain of the macroscopically positive blood culture bottles at 48 h of incubation showed large gram-negative rods with gram-positive granules, and the direct gas-liquid chromatographic analysis of the volatile fatty acids revealed only a discrete amount of acetic acid. After 48 h of incubation at 37°C, the supplemented brucella agar subculture plates incubated anaerobically yielded a positive growth. In case 1, colonies were brittle with a flecked appearance, and after two subcultures they grew in air with CO₂. In case 2, butyrous colonies with the typical cerebroform convoluted surface were obtained, and even after several subcultures they did not grow in air with CO₂. Both isolates hydrolyzed esculin and produced acid from glucose, maltose, salicin, sucrose, and trehalose but not from arabinose, mannitol, rhamnose, and xylose. Catalase and indol were not produced, and nitrate reduction was negative. Gas-liquid chromatographic analysis of cultures on peptone-yeast-glucose broth revealed that lactic acid was the only major end product of their metabolism. The identification of the strains as *Leptotrichia buccalis* was subsequently confirmed by the Anaerobe Laboratory of the Virginia Polytechnic Institute and State University, Blacksburg. The results of the susceptibility testing are shown in Table 1.

The taxonomic position of *Leptotrichia buccalis* has been far from clear. Because younger cells (less than 6 h of culture) show gram-positive characteristics, *Leptotrichia* spp. were confused with members of the family *Lactobacteriaceae* (5). Regardless of the early Gram reaction, studies on the fatty acid composition of the organism and particularly on its cell-wall ultrastructure, lipopolysaccharide chemical composition, and immunological properties conclusively determined its taxonomic position as a gram-negative rod (1, 3, 6, 7).

Because of their morphological similarity, *L. buccalis* was also confused with *Fusobacterium nucleatum* in the 7th edition of the *Bergey's Manual*. Nevertheless in both the 8th edition, and in the recent *Bergey's Manual of Systematic Bacteriology* (1984), *Leptotrichia* constitutes a distinct genus, clearly separated from the genus *Fusobacterium* by the very different end products produced by *Leptotrichia* fermentation of glucose and by DNA hybridization criteria (13).

L. buccalis, possessing a lipopolysaccharide with well-documented endotoxic activity (4), probably has pathogenic capabilities similar to the other members of the family *Bacteroidaceae*. Nevertheless, its incidence in serious bacteremic infections remains comparatively very low. This is probably related to the fact that at least two-thirds of adults have high levels of antibodies against this plaque

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TABLE 1. MICs of selected antibiotics for clinical isolates of *L. buccalis*

Antibiotic	MIC ($\mu\text{g/ml}$)	
	<i>L. buccalis</i> RYC29853 (case 1)	<i>L. buccalis</i> RYC30220 (case 2)
Penicillin G	<0.25	<0.25
Cefoxitin	<0.25	<0.25
Clindamycin	<0.25	<0.25
Chloramphenicol	8	4
Metronidazole	4	2
Gentamycin	>32	>32

organism; antibody levels are at their lowest in the 1- to 12-year age group (11). However, a case of infective endocarditis due to *L. buccalis* has been reported in the absence of immunosuppression (2). The two previously described cases of *L. buccalis* bacteremia associated with severe infection had profound leukopenia (800 to 1,000/mm³) as a result of intensive myelosuppressive therapy (9, 12). In both of our pediatric patients, *L. buccalis* bacteremia also occurred in relationship to severe leukopenia (900 to 1000/mm³). In one patient (case 2) normal levels of immunoglobulins G, M, and A antibodies were documented. From the analysis of these cases of bacteremia, it could be suggested that granulocytopenia might be more important than humoral immune dysfunction in predisposition to *L. buccalis* bacteremia. Moreover, in three of the four cases of infection in leukopenic patients, oral mucosal lesions were present and could have served as the portal of entry for *L. buccalis*.

Although anaerobic organisms are infrequently found as a cause of severe bacteremic infections in immunocompromised patients, the pediatric cases described here may suggest that the standard protective antibiotic chemotherapy used in patients suffering from severe granulocytopenia should perhaps include antibiotics with proved efficacy against anaerobic microorganisms.

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