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Endocarditis Associated with Cardiac Catheterization Due to a Gram-Positive Coccus Designated *Micrococcus mucilaginosus incertae sedis*

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A gram-positive coccus, presently named *Micrococcus mucilaginosus incertae sedis,* was isolated from 14 blood cultures from a patient with endocarditis. The first positive blood culture was drawn 5 days after the patient underwent cardiac catheterization.

Although many genera of bacteria have been incriminated as the etiological agents of endocarditis, close to 90% of infections of the endocardium are due to either streptococci or staphylococci (8). The most common agents are the viridans streptococci, which are part of the normal oral flora. Other members of the oral flora have been reported as infrequent causes of endocarditis. These include *Streptococcus pneumoniae* (8), *Corynebacterium* (13), *Haemophilus influenzae* and *H. parainfluenzae* (6), *Neisseria catarrhalis* (11), and *Micrococcus* (9). Recently, we observed a case of endocarditis due to a gram-positive coccus presently named *Micrococcus mucilaginosus incertae sedis* (strain SFH). This appears to be the first documented infection due to this bacterium.

CASE REPORT

A 63-year-old white man was admitted to Saint Francis Hospital and Medical Center, Hartford, Conn., on 4 March 1977 because of increasing shortness of breath. The patient is known to have arteriosclerotic and rheumatic heart disease, with moderate mitral stenosis and regurgitation.

On examination, the patient was found to have a temperature of 99°F (ca. 37.2°C) and signs of minimal congestive heart failure. Fundoscopic examination was normal. The mouth was adentulous. The heart sounds were irregular, with a grade II pansystolic murmur at the apex with radiation to the axilla and a low-pitched opening snap. Laboratory data included a leukocyte count of 6,400 cm^3, with 54% polymorphonuclear leukocytes, 10% band forms, 25% lymphocytes, 6% monocytes, 5% eosinophils; 40% hematocrit; hemoglobin, 14.6 g/100 ml; and a sedimentation rate of 59 mm/h.

The patient had temperatures between 99 and 100°F during the initial hospital course. On 25 March 1977, he underwent cardiac catheterization, which demonstrated the mitral lesion and coronary artery disease. He received prophylactic cephalosporin for the procedure. The day after catheterization, the patient developed inflammation at the catheterization site, accompanied by a temperature to 102°F which persisted for 2 weeks. Cultures from the catheterization site were negative on three different occasions. Blood cultures at that time were negative; a culture drawn 5 days later was positive for gram-positive cocci, as were 13 other blood cultures over the next week. The patient was treated with aqueous penicillin, 2×10^6 U intravenously every 4 h. The serum was bactericidal on 9 April 1977 at a dilution of 1:256. The temperature rapidly returned to normal, and the patient clinically improved. There was no change in his heart murmur. He was discharged after 5 weeks of intravenous antibiotic therapy.

RESULTS AND DISCUSSION

Laboratory findings. The Bactec blood culture system (Johnston Labs, Inc.) with an aerobic and anaerobic bottle (6A and 7B) for each specimen is used in our laboratory. Cultures are read daily on a Bactec 301 for 7 days. Smears of cultures with a positive growth index are examined. These cultures are then subcultured to sheep blood agar and chocolate agar plates. Initial cultures drawn from 25 to 26 March 1977 were negative by growth index. One of the cultures drawn on 30 March 1977 gave a positive growth index after 72 h of incubation. Subsequent blood cultures were positive between 3 and 7 days. The majority of the blood cultures were not positive until day 7. The smear of the initial bottle revealed gram-positive cocci 1 to 1.5 μm in diameter arranged in pairs and packets.

This positive blood culture was subcultured to sheep blood agar and chocolate agar and incubated anaerobically and in the presence of 8% carbon dioxide. No growth was observed on any of the agar plates. Because there was no growth on solid media, the original blood culture was subsequently inoculated into several liquid media. Growth was obtained in Trypticase soy broth and Mueller-Hinton broth. Growth was best in thioglycolate broth. After several subcultures in thioglycolate broth, all of the isolates grew well on sheep blood agar and chocolate agar; however, the organism only grew when the plates were incubated in a CO_2 incubator (8% CO_2). To obtain growth on solid media, each smear positive blood culture required subculture in thioglycolate broth. A total of 14 cultures (26 bottles) was positive for the same gram-positive coccus.

The colonies on sheep blood agar were nonhemolytic, catalase negative, shiny, and very white with an entire edge. They were very adherent to the agar and became even more so after 48 h. They did not emulsify in broth. Because of a negative catalase test, the organism was tentatively identified as a nonhemolytic streptococcus and sent to the Connecticut State Department of Health for identification as to species. The Connecticut State Department of Health laboratory forwarded the organism to the Center for Disease Control where it was identified as *Micrococcus mucilaginosus*. Their results were confirmed in our laboratory and compared with those obtained by Gordon (7) and Bergan et al. (3). Gordon examined 24 strains of gram-positive cocci named *Staphylococcus salivarius* which were isolated from the oral cavity, and Bergan et al. studied 18 isolates named *M. mucilaginosus* isolated from clinical sources. These results are compared with our isolate (SFH) in Table 1. Gordon found the *S. salivarius* strains to be acetoin positive, whereas our strain was negative. The *M. mucilaginosus* strains reported by Bergan et al. produced acid from glycerol, whereas the SFH strain did not. Their strains were also unable to produce acid anaerobically from glucose. Unlike the other reported strains, the SFH strain would only grow on solid media if incubated in the presence of CO_2. Although not identical, our isolate does appear to be a member of the same species as those previously described.

The antimicrobial susceptibilities of strain SFH and the strains of Bergan et al. are listed in Table 2. Because the isolate grew well in Mueller-Hinton broth, the macrodilution technique described in the *Manual of Clinical Microbiology* (14) was used to determine minimum inhibitory concentrations. The Bauer-Kirby test was used for disk susceptibility tests (2) except that blood Mueller-Hinton agar plates were incubated in a CO_2 incubator. The incubation in CO_2 and the use of blood could affect the zone sizes; however, there was good correlation between disk susceptibility and minimal inhibitory concentration.

The taxonomic position of this gram-positive coccus needs clarification. It is certainly a member of the family *Micrococcaceae* because it does yield a positive benzidine test indicating the presence of cytochrome enzymes (5). Attempts have been made to assign it to either the genus *Staphylococcus* or *Micrococcus*. Gordon

TABLE 1. *Characteristics of M. mucilaginosus i. s.*

Test/substrate	*M. mucilaginosus* (3) (% positive)	*S. salivarius* (7) (% positive)	Strain SFH
Benzidene hydrochloride	ND[a]	100	+
Catalase	70	42	−
Deoxyribonuclease	ND	0	−
Esculin	95	100	+
Gelatin hydrolysis	100	100	+
Growth at			
10°C	ND	0	−
45°C	ND	ND	+
Growth in			
6.5% NaCl	ND	0	−
4.0% NaCl	ND	0	−
2.0% NaCl	ND	ND	+
10% Bile	ND	0	−
40% Bile	ND	0	−
Hemolysis	0	0	−
Indole	0	0	−
Litmus milk	ND	ND	−
Oxidase	0	83	−
Sodium hippurate	ND	ND	−
Starch hydrolysis	0	ND	−
Arabinose	0	0	−
Glucose (anaerobic)	0	100	+
Glycerol	94	0	−
Inulin	0	ND	−
Lactose	ND	0	−
Maltose	70	100	+
Mannitol	0	0	−
Melibiose	0	ND	−
Raffinose	0	0	−
Salicin	ND	100	+
Sorbitol	0	ND	−
Sucrose	ND	100	+
Trehalose	79	100	+
Urea	0	0	−
Voges-Proskauer	ND	100	−
G+C content[b]	59	55.4–58.3	60.4

[a] ND, Not done.

[b] Percent G+C was determined by thermal denaturation by Alan Coykendall, University of Connecticut Health Center.

TABLE 2. *Antimicrobial susceptibility of M. mucilaginosus i. s.*

Antibiotic	Disk test[a] (Bauer-Kirby) for:		Tube dilution test (MIC,[b] μg/ml) for SFH
	SFH	Strains of Bergan et al. (3)	
Ampicillin	S	S	0.75
Carbenicillin	S	ND	1.5
Cephalothin	S	ND	0.2
Chloramphenicol	S	S	0.75
Gentamicin	S	R	0.6
Methicillin	S	ND	0.1
Penicillin	S	S	0.1[c]
Tetracycline	S	S	1.5

[a] S, Susceptible; R, resistant; ND, not done.
[b] MIC, Minimal inhibitory concentration.
[c] Units per milliliter.

(7) suggested the name *S. salivarius* because the catalase-variable, benzidine-positive strains that he examined were able to produce acid from glucose anaerobically. Our strain also produced acid from glucose when incubated anaerobically. The strains of Bergan et al. (3) were unable to ferment glucose, although the standard medium (5) was used in all cases. Baird-Parker (1) suggests that one may not always obtain clear-cut results with this test because some strains may not grow well anaerobically.

Bergan et al. (3) proposed that the organism be classified as a micrococcus. It is resistant to lysostaphin, and its cell wall composition differs from that of staphylococci (4). They found a guanine plus cytosine (G+C) content for their strains of 59% and a G+C of 60% for one of Gordon's strains. Although they concluded that the G+C content supported the placement of these organisms in the genus *Micrococcus,* the present classification (1) is based on a G+C content for *Micrococcus* of 66 to 75% and one for *Staphylococcus* of 30 to 40%. Gordon (7) obtained G+C contents for his *S. salivarius* strains of 55.4 to 58.3%. Our isolate had a G+C content of 60.4% (Table 1). Thus, the G+C contents reported for this organism are intermediate between *Micrococcus* and *Staphylococcus.* Baird-Parker (1) feels that these gram-positive cocci described by Bergan et al. and Gordon belong to neither genus but, based on G+C content, are intermediate between the two. Thus, based on available data, these gram-positive cocci presently called *M. mucilaginosus i. s.* are probably neither staphylococci nor micrococci. To clarify their taxonomic position further, studies such as deoxyribonucleic acid hybridization with other gram-positive bacteria and cell wall analysis need to be done.

The diagnosis of endocarditis in this patient was based on the persistently positive blood cultures and mitral valve disease without another source of infection.

Because he had a low-grade fever at the time of admission, it is possible that his endocarditis antedated the cardiac catheterization. Endocarditis after cardiac catheterization has been reported, but it is rare (10, 12, 15). In an extensive review of over 12,000 catheterizations, there were only three cases of endocarditis, an incidence of less than 0.03% (12).

M. mucilaginosus i. s. appears to be an important part of the oral flora, comprising about 3.5% of the predominant cultivable microorganisms on the human tongue (7). Our isolate appears to be the first confirmed cause of any kind of infection by the species. The 18 isolates of Bergan et al. (3) were all from clinical sources, but a role in disease is doubtful for the majority because they were from various oral sources (throat, nasopharynx, bronchial secretions). Three of their strains were isolated from blood cultures from febrile patients. In one case, two cultures taken simultaneously both grew *M. mucilaginosus i. s.* as well as a viridans streptococcus. In our patient, there is little question that *M. mucilaginosus i. s.* was the infecting organism.

The role of this organism in infection appears limited, but like a number of other bacteria found in the oral cavity, it is capable of attacking damaged heart valves. Because it is often catalase negative, it may be misidentified as a *Streptococcus* sp. The clue to its identity is its strong adherence to an agar surface. More complete identification of blood isolates may lead to a better understanding of the role of *M. mucilaginosus i. s.* in endocarditis.

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