Actinomyces meyeri sp. nov., Specific Epithet rev.

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We propose that "*Actinobacterium meyeri*" be placed in the genus *Actinomyces* as *Actinomyces meyeri*. A description of the species is presented. The proposed type strain is strain ATCC 35568 (= Prévot 2477B = VPI 8617).

Actinobacterium israelii, the type species of the genus *Actinobacterium* Sampietro 1908, was transferred to the genus *Actinomyces* Harz 1877 by Breed and Conn in 1919 (2). This effectively invalidated the name *Actinobacterium* according to Rule 37a of the *International Code of Nomenclature of Bacteria* (6). The genus name *Actinobacterium* was not included on the Approved Lists of Bacterial Names in 1980 (13) and thus has no standing in nomenclature. In the system of classification of Prévot, the genus *Actinobacterium* was included as late as 1968 (1) and comprised those species of the family *Actinomycetaceae* that were obligately anaerobic, at least on isolation. "*Actinobacterium israelii*" was retained as the type species, and "*Actinobacterium meyeri*," which was proposed by Prévot in 1938 (10), was included in the genus. Strains with the characteristics of "*Actinobacterium meyeri*" (11) have been isolated frequently from brain abscesses, from pleural cavities, and from both healthy and diseased subgingival pockets of humans (8, 9, 12). Because this organism may be a significant human pathogen, we propose the new species *Actinomyces meyeri* for these strains, with strain ATCC 35568 as the type strain.

The results of a study of 16 strains, 5 of which were identified by A.-R. Prévot as "*Actinobacterium meyeri*," are presented below. The sources of these strains are shown in Table 1. The anaerobic methods that we used are described in the *Anaerobe Laboratory Manual* (4). The strains were tested for fluorescent antibody staining as described by Slack and Gerencser (14) with monovalent conjugate prepared against Prévot strain 2801 (= ATCC 33972). Antibiotic susceptibilities were determined by the broth disk method of Wilkins and Thiel (15).

Actinomyces meyeri sp. nov. (basonym, "*Actinobacterium meyeri*" A.-R. Prévot 1938,303; meyeri. n.l. gen. n. *meyeri* named for K. F. Meyer, German bacteriologist who first isolated the species). Cells in peptone-yeast extract-glucose broth cultures supplemented with vitamin K₁, hemin, and Tween 80 (4) are short, gram-positive, nonspore-forming, nonmotile rods 0.5 to 0.9 by 0.9 to 3.0 µm and occur in pairs, short chains, or clumps. Terminal swellings are present occasionally.

Surface colonies on supplemented brain heart infusion blood agar plates are pinpoint to 1 mm in diameter, circular, flat to convex, translucent to opaque, and white, with shiny, smooth surfaces and entire margins. They may be alpha- or nonhemolytic. Growth is abundant on plates incubated anaerobically; very slight growth may occur on plates incubated in air enriched with 5% CO₂; there is no growth on plates incubated aerobically.

Cultures in peptone-yeast extract-glucose broth are only slightly turbid with a smooth or stringy sediment and have a pH of 4.6 to 5.0 after incubation for 5 days.

Strains grow equally well at 30 or 37°C; they grow nearly as well at 25°C but not at all at 45°C. They have an absolute requirement for vitamin K₁, and growth is greatly stimulated by 0.02% Tween 80 and by fermentable carbohydrate. There is no growth in 20% bile or 6.5% NaCl.

No gas is formed in peptone-yeast extract-glucose agar deep cultures; neither hydrogen nor H₂S is produced; ammonia may be produced in chopped meat cultures. Acid is produced from glucose, fructose, maltose, starch, sucrose, ribose, and xylose; fermentation of amygdalin, arabinose, galactose, glycogen, glycerol, and lactose is variable (Table 2); neither adonitol, dulcitol, erythritol, inositol, mannitol, sorbitol, cellobiose, esculin, inulin, mannose, melezitose, melibiose, pectin, raffinose, rhamnose, salicin, sorbose, nor trehalose is fermented. Neither indole, acetylmethylcarbinol, catalase, lecithinase, nor lipase is produced. Production of urease is variable when organisms are tested in peptone-yeast extract-urea broth (4). Proline deaminase and α-glucosidase are produced when organisms are tested by the API-ZYM system (Analytab Products, Plainview, N.Y.). Milk is curdled by those strains that ferment lactose. Esculin, starch, and hippurate are not hydrolyzed. Nitrate is not reduced; neutral red is reduced, but the reduction of resazurin is variable. There is no digestion of gelatin or chopped meat.

The products of fermentation in peptone-yeast extract-glucose broth are formic, acetic, and succinic acids, with small amounts of lactic or pyruvic acid occasionally detected. Neither lactate, threonine, nor gluconate is utilized; pyruvate is converted to formate and acetate.

TABLE 1. Sources of strains of *Actinomyces meyeri*

VPI number	Source
8617	purulent pleurisy, Prévot 2477B, ATCC 35568, type strain
8611	cervico-facial actinomycosis, Prévot 3732
8620B	brain abscess, Prévot 2169
8622	purulent meningitis, Prévot 2463
10648	unknown, Prévot 2801, antigen strain, ATCC 33972
10757	chest wall, Maryland
11313	brain abscess, Massachusetts
11346	facial actinomycosis, Delaware
11706	brain abscess, Texas
11857B	bartholin cyst, Tennessee
11858A	subdural abscess, Tennessee
11983	mandible, Texas
12065	empyema, Delaware
12313	thoracentesis fluid, California
D27F26	subgingival-crevice, severe periodontitis, Virginia
D39B11	subgingival crevice, healthy, Virginia

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TABLE 2. Variable reactions of strains of *Actinomyces meyeri*

	ATCC 35568 type strain	reaction positive (no. of strains)
Acid from		
Amygdalin	+	20 (50)
Arabinose	—	19 (50)
Galactose	—	9 (15)
Glycerol	—	16 (33)
Glycogen	+	24 (50)
Lactose	+	22 (50)
Milk curdled	+	37 (50)
Urease produced	+	10 (16)

The patterns of soluble cellular proteins as determined by polyacrylamide gel electrophoresis (7) illustrate the homogeneity of the strains examined (Fig. 1).

In a numerical taxonomic study, Holmberg and Nord (5) reported that four of these strains form a cluster related to but distinct from other species in the genus *Actinomyces*.

All of the strains listed in Table 1 give a positive fluorescent antibody reaction with a monovalent conjugate prepared against strain ATCC 33972. No cross-reactions have been found with conjugates to other species of oral isolates, including two serotypes of *Actinomyces israelii*, two serotypes of *Actinomyces odontolyticus*, three serotypes of *Actinomyces naeslundii*, *Actinomyces viscosus*, and a common but unnamed oral isolate, *Actinomyces* sp. strain WVU 963. Several strains that are otherwise phenotypically identi-

cal to *Actinomyces meyeri* do not react with this conjugate although their protein patterns are similar, which indicates that there are other serotypes of the species.

All of the strains tested are susceptible to achievable blood levels of chloramphenicol, clindamycin, erythromycin, penicillin G, and tetracycline.

Actinomyces meyeri can be distinguished from *Actinomyces israelii* and *Actinomyces naeslundii* by not fermenting raffinose, from *Actinomyces viscosus* by not producing catalase, from *Actinomyces odontolyticus* by not producing pink colonies on blood agar and by not reducing nitrate, from *Actinomyces bovis* by fermenting ribose and xylose, and from *Actinomyces* sp. strain WVU 963 by not hydrolyzing esculin.

The principal habitat of *Actinomyces meyeri* is human periodontal sulci. It has been isolated frequently from brain abscesses and pleural fluid and less often from abscesses of hips, hands, feet, and spleens and from bite wounds.

The type strain of *Actinomyces meyeri* is strain ATCC 35568 (= Prévot 2477B = VPI 8617). The guanine-plus-cytosine content of this strain, as determined by chromatographic analysis, is 67 mol% (1).

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LITERATURE CITED

1. Bouisset, L., J. Brouillard, G. Michel, and G. Larrouy. 1968. Bases nucléiques des bactéries. Application au genre *Actinobacterium*. Ann. Inst. Pasteur Paris 115:1063-1081.
2. Breed, R. S., and H. J. Conn. 1919. The nomenclature of the *Actinomycetaceae*. J. Bacteriol. 4:585-602.
3. Buchanan, R. E., J. G. Holt, and E. F. Lessel, Jr. (ed.). 1966. Index bergyana. The Williams & Wilkins Co. Baltimore.
4. Holdeman, L. V., E. P. Cato, and W. E. C. Moore (ed.). 1977. Anaerobe laboratory manual, 4th ed. Virginia Polytechnic Institute and State University, Blacksburg.
5. Holmberg, K., and C.-E. Nord. 1975. Numerical taxonomy and laboratory identification of *Actinomyces* and *Arachnia* and some related bacteria. J. Gen. Microbiol. 91:17-44.
6. Lapage, S. P., P. H. A. Sneath, E. F. Lessel, V. B. D. Skerman, H. P. R. Seeliger, and W. A. Clark (ed.). 1975. International code of nomenclature of bacteria. 1975 Revision. American Society for Microbiology, Washington, D.C.
7. Moore, W. E. C., D. E. Hash, L. V. Holdeman, and E. P. Cato. 1980. Polyacrylamide slab gel electrophoresis of soluble proteins for studies of bacterial floras. Appl. Environ. Microbiol. 39:900-907.
8. Moore, W. E. C., L. V. Holdeman, R. M. Smibert, D. E. Hash, J. A. Burmeister, and R. R. Ranney. 1982. Bacteriology of severe periodontitis in young adult humans. Infect. Immun. 38:1137-1148.
9. Moore, W. E. C., R. R. Ranney, and L. V. Holdeman. 1982. Subgingival microflora in periodontal disease: cultural studies, p. 13-26. In R. J. Genco and S. E. Mergenhagen (ed.), Host-parasite infections in periodontal diseases. American Society for Microbiology, Washington, D.C.
10. Prévot, A.-R. 1938. Études de systématique bactérienne. Ann. Inst. Pasteur Paris 60:285-307.
11. Prévot, A.-R. 1966. Manual for the classification and determination of the anaerobic bacteria. Translated by V. Fredette. Lea & Febiger, Philadelphia.
12. Schaal, K. P., and G. Pulverer. 1981. The genera *Actinomyces*, *Agromyces*, *Arachnia*, *Bacterionema*, and *Rothia*, p. 1923-

FIG. 1. PAGE protein patterns of strains of *A. meyeri*. Lane 1. *Streptococcus faecalis* U4-20, reference strain; lanes 2-11, *Actinomyces meyeri*: 2 = ATCC 33972 (antigen strain); 3 = 8611; 4 = ATCC 35568 (type strain); 5 = 8620B; 6 = 8622; 7 = 11346; 8 = 12065; 9 = D39B11; 10 = D27F26; 11 = 10757.

1950. In M. P. Starr, H. Stolp, H. G. Trüper, A. Balows, and H. G. Schlegel (ed.), *The prokaryotes*, vol. 2. Springer-Verlag, New York.
13. Skerman, V. B. D., V. McGowan, and P. H. A. Sneath (ed.). 1980. Approved lists of bacterial names. *Int. J. Syst. Bacteriol.* **30**:225–420.
14. Slack, J. M., and M. A. Gerencser. 1975. *Actinomyces*, filamentous bacteria: biology and pathogenicity. Burgess Publishing Co., Minneapolis.
15. Wilkins, T. D., and T. Thiel. 1973. A modified broth-disk method for testing the antibiotic susceptibility of anaerobic bacteria. *Antimicrob. Agents Chemother.* **3**:350–356.