Isolation of *Actinomyces viscosus* from two patients with clinical infections

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SUMMARY The isolation of *Actinomyces viscosus* from two patients is described. One was a case of multiple myeloma, the organism being found on blood culture; the other was a patient with a submandibular abscess. These are believed to be the first such isolations of *A. viscosus* in this country.

In 1963 a Gram-positive, non-acid-fast, branching, filamentous, catalase-positive organism that had not been previously described was isolated from periodontal plaque in hamsters (Howell, 1963; Howell and Jordan, 1963). It was later considered to be a new genus and species and was named *Odontomyces viscosus* (Howell *et al.*, 1965). After the genus *Actinomyces* had been redefined to include catalase-positive organisms the name was changed from *O. viscosus* to *Actinomyces viscosus* (Georg *et al.*, 1969).

A. viscosus has been found in the oral cavity of hamsters, being associated particularly with periodontal plaque (Howell, 1963). It has also been isolated from the lungs of two pigs with enzootic pneumonia, from a prescapular lymph node in a goat, and from suppurative lesions in two dogs (Georg *et al.*, 1972). It has been found in dental calculus in man (Gerencher and Slack, 1969), and recently two cases of human disease due to *A. viscosus* have been reported in America, one from a mass in the chest wall (Lewis and Gorbach, 1972) and the other from a branchial cyst (Adeniyi-Jones *et al.*, 1973).

The isolation of *A. viscosus* in this laboratory from a submandibular duct orifice in one patient and from a blood culture in another patient with myelomatosis is described.

Case reports

CASE 1

A 62-year-old woman was admitted to hospital in April 1975 as an emergency with a submandibular abscess. She had a hard, tender, mobile lump in the left submandibular region towards the angle of the

jaw. No calculi were seen on radiographic examination. Cultures from the orifice of the left submandibular duct yielded a normal mouth flora and a moderate growth of a branching Gram-positive bacillus. No pus could be expressed from the gland. The submandibular swelling, which was not incised, responded to treatment with flucloxacillin, 500 mg four times a day, and had almost disappeared after 15 days.

CASE 2

A 76-year-old woman, a known case of multiple myeloma, was admitted for transfusion of four units of blood for cytopenia. The next day a temperature developed with crepitations in both lower lobes. Chest infection was diagnosed and a blood culture was taken. A branching Gram-positive bacillus was isolated from one bottle only, the remaining bottle showing no bacterial growth. The patient was treated with ampicillin and cloxacillin for two days. The temperature settled and her condition improved. She was discharged eight days after admission.

Material and methods

Except where otherwise indicated, these were as described by Cowan (1974). Nitrate reduction tests were performed by the nitrate paper strip method of Cook (1950). Gelatin hydrolysis was tested using Frazier's (1926) method. One per cent peptone water sugars containing Andrade's indicator was used for the carbohydrate fermentation tests. The media used for blood culture in this laboratory are nutrient broth with 0.2% glucose and thioglycollate nutrient broth.

Anaerobic cultures were incubated in a McIntosh and Fildes' jar incorporating a cold catalyst.

Sensitivity testing was by paper disc diffusion.

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Microbiological results

ISOLATION

The organism from case 1 was isolated on blood agar after a few days' incubation at 37°C in an anaerobic jar containing 10% CO₂.

The organism from case 2 was isolated in 0.2% glucose nutrient broth after seven days' incubation at 37°C. Thioglycollate nutrient broth produced no bacterial growth after 21 days' incubation at 37°C. Subcultures were made on blood agar incubated for two days at 37°C in an anaerobic jar containing 10% CO₂.

MORPHOLOGY

Both organisms were non-motile, non-sporing, non-acid-fast, Gram-positive rods which, when grown under optimum growth conditions, produced extensive branching. Under suboptimal conditions branching was less obvious and a more diphtheroid type morphology predominated. Films from growth in fluid media showed long, tangled, branching filaments.

CULTURAL CHARACTERISTICS

Optimum conditions for growth were obtained in an anaerobic jar containing 10% CO₂ at 37°C. Good growth was also obtained in a candle jar. Growth under anaerobic conditions but without added CO₂, and in air, did occur but was much reduced. Growth occurred on ordinary nutrient media but was slightly enhanced by the addition of blood.

When grown under optimal conditions colonies took several days to reach their maximum size of 1.5-2.0 mm diameter. They were white and of 'molar tooth' appearance, embedded in the medium, and difficult to emulsify and produced greening on blood agar. When colonies were grown under other conditions they became smooth, soft, opaque, and more easily emulsifiable. Growth in 0.2% glucose broth showed slight turbidity with a moderate granular deposit.

BIOCHEMICAL REACTIONS

These are given in the Table.

On these findings the two strains were presumptively identified as *A. viscosus*, the only member of the *Actinomyces* group recorded as giving a positive catalase reaction.

Both isolates were sensitive to penicillin, tetracycline, erythromycin, cephalosporins, ampicillin, streptomycin, and clindamycin.

Identification of these isolates as *A. viscosus* was confirmed by Miss J. M. Brown, of the Center for Disease Control in Atlanta, Georgia, who also reported that both strains fell into serotype 2.

Table Biochemical reactions of *Actinomyces viscosus* isolates

	Organism from case 1	Organism from case 2
Catalase	+	+
Oxidase	—	—
Hugh and Leifson's medium	Fermenter	Fermenter
Nitrate reduction	+	+
Starch hydrolysis	—	+
Gelatin hydrolysis	—	—
Indole	—	—
Glucose	A	A
Mannitol	—	—
Lactose	A	A
Sucrose	A	A
Salicin	—	—
Raffinose	—	A
Glycerol	A	A
Xylose	—	—
Gas from carbohydrates	—	—

A = acid production

Discussion

Animal isolates of *A. viscosus* have been shown to be capable of initiating periodontal disease in hamsters (Jordan and Keyes, 1964). Isolates from human dental plaque have been shown to produce periodontal lesions and fissure caries in hamsters and in gnotobiotic rats (Frank *et al.*, 1972; Jordan *et al.*, 1972). This ability may well be associated with the production of slightly soluble, extracellular polysaccharides (Rosan and Hammond, 1974) in the same way that the production of relatively insoluble polysaccharides by *Streptococcus mutans* established this organism as a probable cause of enamel caries (Gibbons, 1972; Scherp, 1971).

A. viscosus has been isolated from chronic abdominal abscesses in two dogs. In one case it was the sole isolate while in the other case it was found in conjunction with *Eubacterium lentum* and *Enterobacter cloacae*. Clinically, both cases bore all the signs of classical actinomycosis, and *A. viscosus* was suggested as the primary pathogen (Georg *et al.*, 1972). *A. viscosus* has also been isolated from the lungs of two pigs with enzootic pneumonia and from a prescapular lymph node in a goat, but the pathogenesis of the lesions in these cases remains unknown (Georg *et al.*, 1972).

Recently there have been two reports incriminating *A. viscosus* as an agent of human infection. The first of these (Lewis and Gorbach, 1972) describes the isolation of *A. viscosus* from a mass in the chest wall of a negro man together with *Mycobacterium tuberculosis* from the pleural fluid. The other report (Adeniyi-Jones *et al.*, 1973) describes the isolation of *A. viscosus* in pure culture from a branchial cyst in a

19-year-old youth, pathogenicity being supported by histological findings.

The isolations of *A. viscosus* from the two cases described in this paper are believed to be the first such isolations of the organism in this country. The identification of the isolates as *A. viscosus* serotype 2 agrees with the findings of Gerencser and Slack (1969), who reported that strains from hamster and human sources fell into two serotypes, serotype 1 containing the hamster strains and serotype 2 containing all the human strains. The strains of *A. viscosus* isolated from the two pigs, from one dog, and from the goat were similar to serotype 2, while the strain isolated from the other dog was similar to serotype 1 (Georg *et al.*, 1972).

The pathogenicity of *A. viscosus* in the two cases described here cannot be unequivocally affirmed but the reported ability of this organism to play an active role in both animal and human infections suggests that the isolation of *A. viscosus* in these two instances may have been significant.

We thank Miss J. M. Brown, of the Center of Disease Control, Atlanta, Georgia, for confirming the identity and serotype of the two isolates.

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