CASE REPORT

Total knee arthroplasty infection due to *Abiotrophia defectiva*

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The first documented case of knee alloarthroplasty infection due to *Abiotrophia defectiva*, formerly known as nutritionally variant streptococci (NVS) and *Streptococcus defectivus*, is presented. The microbiology of this bacterium is discussed and clinical features of previously reported cases of infections by NVS are reviewed briefly.

Introduction

Abiotrophia defectiva is part of the normal human flora [1–3] and is a resident of the oral cavity [1, 4], and the genitourinary and intestinal mucosae [4, 5].

Abiotrophia spp. have previously been referred to as nutritionally variant streptococci (NVS) because of their fastidious nutritional growth requirements. Frenkel and Hirsch [6] first isolated these organisms from patients with endocarditis and otitis media. The organisms tend to form satellite colonies around *Staphylococcus aureus* and other bacteria, including some Enterobacteriaceae and other streptococci. They were described as ‘ungroupable’ viridans streptococci and required pyridoxal hydrochloride (vitamin B₆) analogues for growth and produced a chromophore, pyrrolidonyl arylamidase and a bacteriolytic enzyme [2–4, 7, 8].

Bouvet *et al.* [2] proposed two new species names for the NVS, *Streptococcus defectivus* and *S. adjacens*, on the basis of DNA–DNA hybridisation studies. Analysis of 16S ribosomal RNA (rRNA) sequences showed that these two species were not related to other members of the genus *Streptococcus*. Consequently, they were placed in the new genus *Abiotrophia* as *A. defectiva* and *A. adjacens* [9]. *A. defectiva* differs from *A. adjacens* by the presence of α - and β -galactosidases and the absence of β -glucuronidase [7].

Recently, a third species, *A. elegans*, has been added to the genus *Abiotrophia* [10, 11]. Lawson *et al.* [12] described a fourth species, *A. balaenopterae*, isolated from the lung of a minke whale. Kanamoto *et al.* [13] proposed the existence of *A. para-adjacens*.

Recent 16S rRNA gene sequencing studies have demonstrated that the genus is not monophyletic. Phylogenetically, the genus *Abiotrophia* consists of two distinct lines, *A. defectiva* and another group consisting of *A. adjacens*, *A. balaenopterae* and *A. elegans*. Therefore, it was proposed by Collins and Lawson that the genus *Abiotrophia* be restricted to *A. defectiva* and that *A. adjacens*, *A. balaenopterae* and *A. elegans* be reclassified in a new genus, *Granulicatella* gen. nov., as *G. adjacens* comb. nov., *G. balaenopterae* comb. nov. and *G. elegans* comb. nov. [14].

To draw attention to their clinical significance and difficulties in diagnosis and treatment, this paper describes the first documented case report of an infected total knee alloarthroplasty caused by *A. defectiva*.

Case report

In April 2000, a 65-year-old woman was admitted to this hospital with progressive pain and swelling in her right knee, which had undergone a total knee arthroplasty (TKA) 4 years earlier. Her medical history included a nephrectomy because of glomerulonephropathy, non-insulin-dependent diabetes mellitus and alcohol abuse in the past.

She was in good general health and reported no fever. Physical examination revealed no abnormalities, in

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particular no port of entry for bacteraemia nor stigmata of endocarditis. Scintigraphy showed evidence of loosening of the prosthesis, and a conventional radiograph was normal. Laboratory data showed a normal white blood count (5500 cells/ml) and elevated C-reactive protein (CRP) (41 mg/L). Aspiration of the right knee yielded 10 ml of turbid fluid. The specimen was injected into agar vials (Venturi Transsystem®, Nehren, Germany). A Gram's stain failed to reveal any micro-organisms. Nevertheless, on the basis of the clinical impression, an infection of the total knee arthroplasty was diagnosed.

The patient was taken to surgery for a radical open debridement and replacement of all polyethylene components of the prosthesis. Because no micro-organism had been isolated pre-operatively and no signs of loosening of the prosthesis were seen intra-operatively, the prosthesis was left in place.

Intra-operative aspirates were placed in Bactec plus aerobic and anaerobic culture vials (Becton Dickinson, Sparks, MD, USA). The sample was plated on to Columbia agar supplemented with defibrinated sheep blood 5% and chocolate agar and incubated aerobically and in an anaerobic atmosphere containing CO₂ 5% at 37°C. Several small α -haemolytic colonies were noted on the blood-agar plate after incubation for 24 h. Gram's stain revealed pleomorphic gram-positive coccobacilli. The bacterium was not identified with a rapid identification system for gram-positive rods (API Coryne; bioMérieux, Marcy l'Etoile, France) after incubation for 12 h, nor was it identified by rapid identification systems for streptococci (Rapid ID 32 STREP, bioMérieux) after incubation for 4 h. The bacterium was identified with a universal PCR system, with primers targeting part of the 16S rDNA [15]. The amplified PCR product was sequenced and a BLAST search at the NCBI was performed [16]. *A. defectiva* was identified, with a homology of 99.9%.

Antimicrobial treatment included cefazolin i.v. for 10 days, followed by ciprofloxacin orally for 26 days until CRP levels and the physical examination returned to normal.

In July 2000 the patient presented again with pain and swelling as signs of an infected knee prosthesis. Gram's staining of the aspirated intra-articular fluid showed pleomorphic gram-positive coccobacilli and samples plated on to Columbia agar with defibrinated sheep blood 5% grew small α -haemolytic colonies.

As *Abiotrophia* was considered to be the possible causative agent, a rapid ID 32 STREP test (bioMérieux) was performed. After 12 h the biochemical reactions exhibited the characteristic patterns of *A. defectiva*. This prolonged incubation (12 h instead of

4 h) was necessary because of the long generation time of *Abiotrophia*.

A two-stage revision arthroplasty was performed. After debridement and explantation, antibiotic treatment was initiated with ciprofloxacin i.v. As the patient developed profuse diarrhoea, the regimen was changed to penicillin i.v. and subsequently to flucloxacillin orally until the CRP level returned to normal. In December 2000 a new alloarthroplasty was implanted. At present the patient has good function of the TKA.

Discussion

A. defectiva and *A. adjacens* account for 5–6% of cases of streptococcal endocarditis and some authors have suggested that they are an important cause of blood culture-negative endocarditis [17–21]. Bouvet [22] isolated two strains of *S. adjacens* and five strains of *S. defectivus* from 91 strains of viridans streptococci that caused endocarditis. Some authors noted that the clinical course was often more severe than in cases of endocarditis caused by other viridans streptococci or enterococci [23, 24].

Ormerod *et al.* [25] described four cases of infectious crystalline keratopathy caused by NVS. Recently, it has been shown that infiltrative keratitis can be associated with *A. defectiva* [26]. Wofsy [27] reported a case of joint infection caused by NVS as a complication of endocarditis. These organisms have also been implicated in pancreatic abscess, otitis media, wound infections and iatrogenic meningitis [28, 29].

Therapeutic failure occurred in 41% and relapse in 17% of endocarditis cases, despite treatment with antibiotics that appeared to be active *in vitro* [23]. Stein and Nelson [23] hypothesised that the difficulties in treatment may be caused by the slow growth rate and suggested that a longer course of antibiotic therapy may be required for successful treatment of cases of NVS endocarditis.

Exopolysaccharide production by NVS in later stages of rabbit endocarditis has been reported. It was suggested that nutrient limitation within vegetations could account for altered ultrastructural morphology and that exopolysaccharide might enhance NVS pathogenicity [4]. The frequent colonisation and infection of cardiac valves with *Abiotrophia* spp. suggests an affinity for avascular tissue, as found around an alloarthroplasty.

The reported incidence of knee alloarthroplasty infections ranges from 0% to 23% with an average of 5% [30] and is caused by direct spread or by haematogenous spread. Staphylococci are the major causative agents of this type of infection, followed in descending order by gram-positive aerobic streptococci, gram-negative aerobes and anaerobes [31]. The authors

believe that, in the past, arthroplasty infections with NVS have included *Abiotrophia* spp., which were uncharacterised at that time. The treatment of arthroplasty infections includes repetitive debridements [32], removal of the prosthesis and leaving an excision arthroplasty [33] and converting to an arthrodesis [34–36]. A two-stage revision arthroplasty leaving a temporary spacer of bone cement containing antibiotics has become more common [37, 38].

In the case reported here, the isolate of *A. defectiva* was susceptible *in vitro* (disk diffusion tests on Mueller-Hinton agar with defibrinated sheep blood 5%) to penicillin, clindamycin, cefazolin, cefpodoxime, cefuroxime, cefotaxime, gentamicin, ciprofloxacin and imipenem and resistant to tobramycin, doxycycline and aztreonam. The susceptibility of 27 *G. adjacens* and 12 *A. defectiva* isolates was investigated by a microdilution method with pyridoxal hydrochloride and Mueller-Hinton broth supplemented with lysed horse blood. According to NCCLS interpretative criteria for *Streptococcus* spp. not *S. pneumoniae*, the susceptibilities of *G. adjacens* and *A. defectiva* strains were, respectively: penicillin, 55% and 8%; amoxicillin, 81% and 92%; ceftriaxone, 63% and 83%; meropenem, 96% and 100%; and 100% for both species with clindamycin, rifampin, levofloxacin, ofloxacin, quinupristin/dalfopristin and vancomycin [39].

High-level resistance to aminoglycosides (MIC >500 mg/L), as encountered with enterococci and viridans streptococci, has not been reported for NVS. Synergy between penicillin or vancomycin in combination with an aminoglycoside has been observed both *in vitro* and in experimental animal models of endocarditis [40]. However, *in vitro* antimicrobial susceptibility testing of *Abiotrophia* is not standardised, and the results of *in vitro* susceptibility tests do not correlate well with clinical effectiveness.

In conclusion, *A. defectiva* is a relatively unknown micro-organism, which may cause serious infections including culture-negative forms of alloarthroplasty infection and endocarditis.

The extreme pleomorphic appearance of strains may be confusing and cause diagnostic problems [30], which could lead to prolonged morbidity and costly diagnostic and therapeutic procedures. If Gram's stain indicates gram-positive cocci in chains or pairs, the addition of pyridoxal-containing medium or cross-inoculation of the inoculated plate with *Staph. aureus* may increase detection of *Abiotrophia* isolates. Also, because of the slow growth rate of *Abiotrophia* spp., prolonged incubation for at least 72 h in an atmosphere containing CO₂ 5–10% is recommended.

Clinicians and microbiologists should be aware of this organism and its pathogenic potential.

References

- Georg RH. The isolation of symbiotic streptococci. *J Med Microbiol* 1974; **7**: 77–83.
- Bouvet A, Grimont F, Grimon PAD. *Streptococcus defectivus* sp. nov. and *Streptococcus adjacens* sp. nov., nutritionally variant streptococci from human clinical specimens. *Int J Syst Bacteriol* 1989; **39**: 290–294.
- Pompei R, Caredda E, Piras V, Serra C, Pintus L. Production of bacteriolytic activity in the oral cavity by nutritionally variant streptococci. *J Clin Microbiol* 1990; **28**: 1623–1627.
- Ruoff KL. Nutritionally variant streptococci. *Clin Microbiol Rev* 1991; **4**: 184–190.
- McCarthy LR, Bottone EJ. Bacteremia and endocarditis caused by satelliting streptococci. *Am J Clin Pathol* 1974; **16**: 585–591.
- Frenkel A, Hirsch W. Spontaneous development of L forms of streptococci requiring secretions of other bacteria or sulphhydryl compounds for normal growth. *Nature* 1961; **191**: 728–730.
- Bouvet A, Villeroy F, Cheng F, Lamesch C, Williamson R, Gutmann L. Characterization of nutritionally variant streptococci by biochemical tests and penicillin-binding proteins. *J Clin Microbiol* 1985; **22**: 1030–1034.
- Kanamato TH, Eifuku-Koreeda H, Inoue M. Isolation and properties of bacteriolytic enzyme-producing cocci from the human mouth. *FEMS Microbiol Lett* 1996; **144**: 135–140.
- Kawamura Y, Hou XG, Sultana F, Liu S, Yamamoto H, Ezaki T. Transfer of *Streptococcus adjacens* and *Streptococcus defectivus* to *Abiotrophia* gen. nov. as *Abiotrophia adjacens* comb. nov. and *Abiotrophia defectiva* comb. nov., respectively. *Int J Syst Bacteriol* 1995; **45**: 798–803.
- Roggenkamp A, Abele-Horn M, Trebesius K-H, Tretter U, Autenrieth IB, Heesemann J. *Abiotrophia elegans* sp. nov., a possible pathogen in patients with culture-negative endocarditis. *J Clin Microbiol* 1998; **36**: 100–104.
- Roggenkamp A, Leitritz L, Baus K, Falsen E, Heesemann J. PCR for detection and identification of *Abiotrophia* spp. *J Clin Microbiol* 1998; **36**: 2844–2846.
- Lawson PA, Foster G, Falsen E, Sjöden B, Collins MD. *Abiotrophia balaenopterae* sp. nov., isolated from the minke whale (*Balaenoptera acutorostrata*). *Int J Syst Bacteriol* 1999; **49**: 503–506.
- Kanamato T, Sato S, Inoue M. Genetic heterogeneities and phenotypic characteristics of strains of the genus *Abiotrophia* and proposal of *Abiotrophia para-adjacens* sp. nov. *J Clin Microbiol* 2000; **38**: 492–498.
- Collins MD, Lawson PA. The genus *Abiotrophia* (Kawamura et al.) is not monophyletic: proposal of *Granulicatella* gen. nov., *Granulicatella adjacens* comb. nov., *Granulicatella elegans* comb. nov. and *Granulicatella balaenopterae* comb. nov. *Int J Syst Evol Microbiol* 2000; **50**: 365–369.
- Greisen K, Loeffelholz M, Purhoit A, Loeng D. PCR primers and probes for the 16S rRNA gene of most species of pathogenic bacteria, including bacteria found in cerebrospinal fluid. *J Clin Microbiol* 1994; **32**: 335–351.
- Altschul SF, Madden TL, Schäffer AA et al. Gapped BLAST and PSI-BLAST: a new generation of protein database search programs. *Nucleic Acids Res* 1997; **25**: 3389–3402.
- Roberts RB, Krieger AG, Schiller NL, Gross KC. Viridans streptococcal endocarditis: the role of various species, including pyridoxal-dependent streptococci. *Rev Infect Dis* 1979; **1**: 955–965.
- Christensen JJ, Gruhn N, Facklam RR. Endocarditis caused by *Abiotrophia* species. *Scand J Infect Dis* 1999; **31**: 210–212.
- Poyart C, Quesne G, Acar P, Berche P, Trieu-Cuot P. Characterization of the Tn916-like transposon Tn3872 in a strain of *Abiotrophia defectiva* (*Streptococcus defectivus*) causing sequential episodes of endocarditis in a child. *Antimicrobial Agents Chemother* 2000; **44**: 790–793.
- Boutoille D, Reynaud AE, Leautez S, Ponge T, Raffi F. [Abiotrophia related endocarditis: contribution of molecular biology.] *Presse Med* 1999; **28**: 2149–2151.
- Bouvet A. Human endocarditis due to nutritionally variant streptococci: *Streptococcus adjacens* and *Streptococcus defectivus*. *Eur Heart J* 1995; **16**: 24–27.
- Bouvet A. Endocardite infectieuse: enquete en France 1990–1991 – Sensibilite aux antibiotiques des streptocoques et enterocoques. *Med Mal Infect* 1992; **22S**: 987–992.
- Stein DS, Nelson KE. Endocarditis due to nutritionally

- deficient streptococci: therapeutic dilemma. *Rev Infect Dis* 1987; **9**: 908–916.
24. Roberts RB, Wilson WR, Bouvet A, Acar JF. Nutritionally variant streptococcal endocarditis. International Conference of Antimicrobial Agents and Chemotherapy, 1982: abstract no. 82.
  25. Ormerod LD, Ruoff KL, Meisler DM *et al.* Infectious crystalline keratopathy. Role of nutritionally variant streptococci and other bacterial factors. *Ophthalmology* 1991; **98**: 159–169.
  26. Keay L, Harmis N, Corrigan K, Sweeney D, Willcox M. Infiltrative keratitis associated with extended wear of hydrogel lenses and *Abiotrophia defectiva*. *Cornea* 2000; **19**: 864–869.
  27. Wofsy D. Culture-negative septic arthritis and bacterial endocarditis. Diagnosis by synovial biopsy. *Arthritis Rheum* 1980; **23**: 605–607.
  28. Carey RB, Gross KC, Roberts RB. Vitamin B₆-dependent *Streptococcus mitior* (*mitis*) isolated from patients with systemic infections. *J Infect Dis* 1975; **131**: 722–726.
  29. Bottone EJ, Thomas CA, Lindquist D, Janda JM. Difficulties encountered in identification of a nutritionally deficient *Streptococcus* on the basis of its failure to revert to streptococcal morphology. *J Clin Microbiol* 1995; **33**: 1022–1024.
  30. Insall JN (ed). *Surgery of the knee*. New York: Churchill Livingstone. 1984.
  31. Brause BD. Infected orthopedic prostheses. In: Bisno AL, Waldvogel FA (eds) *Infections associated with indwelling medical devices*. Washington, DC, American Society for Microbiology. 1989: 111–127.
  32. Morrey BF, Westholm F, Schoifet S, Rand JA, Bryan RS. Long term results of various treatment options for infected total knee arthroplasty. *Clin Orthop* 1989; **248**: 120–128.
  33. Falahee MH, Matthews LS, Kaufer H. Resection arthroplasty as a salvage procedure for a knee with infection after a total arthroplasty. *J Bone Joint Surg [Am]* 1987; **69**: 1013–1021.
  34. Chapchal G. Intramedullary pinning for arthrodesis of the knee joint. *J Bone Joint Surg [Am]* 1948; **30**: 728–734.
  35. Charnley JC. Positive pressure in arthrodesis of the knee joint. *J Bone Joint Surg [Br]* 1948; **30**: 479–486.
  36. Fahmy NRM, Barnes KL, Noble J. A technique for difficult arthrodesis of the knee. *J Bone Joint Surg [Br]* 1984; **66**: 367–370.
  37. Rand JA, Bryan RS. Reimplantation for the salvage of an infected total knee arthroplasty. *J Bone Joint Surg [Am]* 1983; **65**: 1081–1086.
  38. Windsor RE, Insall JN, Urs WK, Miller DV, Brause BD. Two-stage reimplantation for the salvage of total knee arthroplasty complicated by infection. *J Bone Joint Surg [Am]* 1990; **72**: 272–278.
  39. Tuohy MJ, Procop GW, Washington JA. Antimicrobial susceptibility of *Abiotrophia adjacens* and *Abiotrophia defectiva*. *Diagn Microbiol Infect Dis* 2000; **38**: 189–191.
  40. Mandell GL, Bennett JE, Dolin R. *Principle and practice of infectious diseases*, 5th edn. New York, Churchill Livingstone. 2000: 2172–2173.