

Dietzia papillomatosis Bacteremia

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The clinical significance of *Dietzia papillomatosis* is for the moment limited to the rare skin disease confluent and reticulated papillomatosis. We present a case of infection with *D. papillomatosis* in a 2-year-old boy with known syringomyelia. The microbiological diagnosis was done using 16S rRNA gene sequencing. This is the first report of bacteremia with *D. papillomatosis*.

CASE REPORT

A 2-year-old boy was approximately 5 months earlier diagnosed to have syringomyelia. As part of the syringomyelia elucidation, a cerebrospinal fluid (CSF) sample was obtained and tested for *Borrelia* IgG and IgM antibodies, and PCR was performed for herpes simplex and varicella-zoster; all were negative. In addition, a Gram stain was performed, but no bacteria were seen and the culture was negative. CSF leukocytes were at a level of $13 \times 10^6/\text{ml}$, but other parameters were reported normal. The patient had a ventriculoperitoneal shunt inserted for treatment of the syringomyelia. Due to malfunction of the shunt, the patient had an uncomplicated planned shunt revision. The day after the shunt revision, the boy was readmitted to the pediatric department due to high fever (39.2°C). He had a discrete truncal maculopapular exanthem but was not septic. He was suspected of having a viral infection. Chest X ray was normal, and there was no obvious focus of his infection. Two days later, the symptoms were unchanged, but the C-reactive protein rose from 9 to 108 mg/liter (normal value, <10 mg/liter), and his leukocyte count remained normal. A blood culture was obtained, and treatment with intravenous cefuroxime (100 mg/kg divided into three daily doses) was initiated. After another 2 days, the fever disappeared and the exanthem faded. Blood culture identified *Dietzia papillomatosis* by 16S rRNA gene PCR and sequencing. The boy was treated with cefuroxime for 4 days and then switched to oral amoxicillin, 36 mg/kg, for another 3 days and discharged. A blood culture, taken after antibiotic treatment was terminated, remained negative.

The *Dietzia* species is an aerobic, Gram-positive coccus or short rod belonging to the group of actinomycetes. On the basis of its colony appearance and its microscopic resemblance, it has been classified as a *Rhodococcus* species. Gram staining of positive aerobic blood culture obtained from a peripheral vein revealed Gram-positive bacteria with an alternating morphology of cocci in clusters and coryneform rods. Blood was plated on blood agar and chocolate agar (Statens Serum Institute, Copenhagen, Denmark). The bacteria grew poorly on the first day under aerobic conditions and were later incubated on chocolate plates in a CO₂ atmosphere where growth was significantly improved. Repeated microscopy of colonies showed small Gram-positive rods. Fluorescence *in situ* hybridization using peptide nucleic acid probes (PNA-FISH; AdvanDx, Vedbæk, Denmark) using the probe set for identification of *Staphylococcus aureus* and coagulase-negative staphylococci was negative. Matrix-assisted laser desorption ion-

ization–time of flight (MALDI-TOF) using the Bruker MALDI Biotyper was unable to provide any reliable identification. Antimicrobial susceptibility tests were performed using disk diffusion (Neosensitabs; Rosco, Taastrup, Denmark) on chocolate agar (Statens Serum Institute) in CO₂. The isolate was found to be susceptible to penicillin, ampicillin, tetracycline, meropenem, moxifloxacin, and vancomycin. Resistance to sulfonamides and azithromycin was detected. Since we did not have standardized interpretations of zone sizes, etc., for this organism, we used the interpretation from Gram-positive microorganisms (*corynebacteria* and *streptococci*).

The blood sample was cultured in Bactec Peds Plus/F flask (BD Diagnostics) using the Bactec alert system (BD Bactec FX; BD Diagnostics) for detection of an increase in CO₂ level. Cell lysate was obtained with PrepMan Ultra sample preparation reagent (Applied Biosystems, Life Technologies, Nærum, Denmark), and the ratio of sample to PrepMan reagent was altered 7-fold in order to yield a homogenous suspension before extraction. A region of 500 bp from the gene coding for 16S rRNA was amplified by PCR using the Fast MicroSEQ 500 16S rDNA kit (Applied Biosystems). The template for PCR was diluted 1:40 in nuclease-free H₂O (Ambion, Life Technologies) or diluted 1:2 in 2% bovine serum albumin (BSA) prepared solution (Sigma) to a final concentration in the PCR of 0.5% BSA. The PCR product was incubated with hydrolytic enzymes exonuclease I and shrimp alkaline phosphatase (ExoSAP-IT; GE Healthcare, Brøndby, Denmark) in order to remove unwanted deoxyribonucleotides and primers before the sequencing reaction. Sequencing was performed in both forward and reverse directions (Applied Biosystems), with temperature and time definitions in accordance with the manufacturer's instructions. Before running capillary electrophoresis on a genetic analyzer (Applied Biosystems), the sequencing product was purified from unwanted reaction components (BigDye Terminator version 1.1 and primers) through a gel filtration system (Performa DTR gel filtration cartridges; EdgeBio, Sopachem BV, Ochten, The Netherlands). Software used for data analysis was MicroSEQ ID 16S rDNA 500 Library version 2.2 (Applied Biosystems). Se-

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quencing data (4 PCRs and sequencing reactions) showed a high-percentage match (range of 99.69% to 99.74%) to *D. papillomatosis* (DSM 44961), with a high average consensus quality value (specimen score) (range of 41 to 43; values above 30 indicate high quality according to the manufacturer). The consensus length from the sequencing reaction was in accordance with the library entry length for the species with the applied primers (range of 436 to 462 nucleotides compared to the theoretical value of 461). We were able to distinguish our result from the second closest match, which was a strain of *D. maris* with a sequence divergence of more than 1%.

Dietzia species is an aerobic, Gram-positive coccus or short rod belonging to the group of actinomycetes. Our observations of altered life cycle morphology of *D. papillomatosis* is in accordance with a previous report that describes *D. papillomatosis* as having forms of both rods and cocci (1). We propose a novel habitat for *D. papillomatosis* in blood. In comparison to the current literature, the only previously known presence of this bacterium in humans is from skin scrapings in patients with confluent and reticulated papillomatosis (CARP) (1). Besides a reaction to bacterial infection, CARP has also been linked to several other causes, including a reaction to *Malassezia*, a reaction to UV light, and an eruption related to endocrinopathy. Electron microscopic studies show CARP, at least in some instances, to include a defect in keratinization, with signs of abnormal keratinocyte differentiation and maturation (i.e., an increased transition cell layer, increased involucrin expression, and increased lamellar granules in the stratum granulosum) (2).

The likelihood of cross-contamination of the blood culture sample with bacteria potentially located on skin is likely to be small because of the routine use of aseptic techniques when the blood sample was taken from the patient and the fact that no other skin-associated bacteria (e.g., *S. epidermidis*) were grown in the bottles. Clearing of some of the mentioned symptoms after treatment with cefuroxime and amoxicillin is hypothetically in accordance with the theory that a bacterial agent was involved in the patient's illness.

It is worth noting that although *Dietzia* species are most commonly found in connection with environmental sampling, they

have previously also been shown to be involved in human disease, including sepsis. In an immunocompromised patient, a related species, *D. maris*, was detected in connection with catheter-related bacteremia (3). Recently, two other strains of *Dietzia* species, *D. cercidiphylli* and *D. natronolimnaea* (99.5% sequence similarity to *D. papillomatosis*), have been reported in a clinical blood isolate (4).

Viruses, especially those with a dermal habitat, such as Epstein-Barr virus (EBV), cytomegalovirus, varicella-zoster virus, human herpesvirus 6 (HHV-6), coxsackievirus, and parvovirus, as well as drugs, e.g., amoxicillin, are the two most common groups of activators of the immune system that are known to be associated with maculopapular exanthems (5, 6).

This case is the first report of a possible connection between bacteremia with *D. papillomatosis* and maculopapular exanthems. We consider the truncal maculopapular exanthem we observed in this patient to be a critical element in differential diagnosis of confluent and reticulated papillomatosis in future cases.

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REFERENCES

1. Jones AL, Koerner RJ, Natarajan S, Perry JD, Goodfellow M. 2008. *Dietzia papillomatosis* sp. nov., a novel actinomycete isolated from the skin of an immunocompetent patient with confluent and reticulated papillomatosis. *Int. J. Syst. Evol. Microbiol.* 58(Part 1):68–72.
2. Scheinfeld N. 2006. Confluent and reticulated papillomatosis: a review of the literature. *Am. J. Clin. Dermatol.* 7:305–313.
3. Bemer-Melchior P, Haloun A, Riegel P, Drugeon HB. 1999. Bacteremia due to *Dietzia maris* in an immunocompromised patient. *Clin. Infect. Dis.* 29:1338–1340.
4. Niwa H, Lasker BA, Hinrikson HP, Franzen CG, Steigerwalt AG, Whitney AM, Brown JM. 2012. Characterization of human clinical isolates of *Dietzia* species previously misidentified as *Rhodococcus equi*. *Eur. J. Clin. Microbiol. Infect. Dis.* 31:811–820.
5. Fernández TD, Canto G, Blanca M. 2009. Molecular mechanisms of maculopapular exanthema. *Curr. Opin. Infect. Dis.* 22:272–278.
6. Trcka J, Seitz CS, Bröcker EB, Gross GE, Trautmann A. 2007. Aminopenicillin-induced exanthema allows treatment with certain cephalosporins or phenoxymethyl penicillin. *J. Antimicrob. Chemother.* 60:107–111.