Case Report

Breast abscess caused by *Gordonia bronchialis* and the use of 16s rRNA gene sequence analysis for its definitive identification

Carme Vidal,¹ Emma Padilla,² Paula Alcacer,³ Enrique Campos,³ Fernando Prieto² and Carmen Santos¹

Correspondence

Carme Vidal

mariac.vidal@ssib.es

¹Sequencing Unit, Hospital Son Espases. Crta. Valldemossa 70, 07010, Palma de Mallorca, Spain

²Microbiology Unit, Manacor Hospital. Crta. Alcúdia s/n 07500, Manacor, Spain

³Gynecology Service, Manacor Hospital. Crta. Alcúdia s/n 07500, Manacor, Spain

Introduction: *Gordonia* spp. have been recognized as pathogens in immunocompetent and immunocompromised patients, although infections remain a rare event.

Case presentation: We describe the second reported case of recurrent breast infection caused by *Gordonia bronchialis* in an immunocompetent patient.

Conclusions: Careful observation of the Gram stain from the abscess raised suspicion, and final identification was achieved with 16S rRNA gene sequencing analysis.

Keywords: 16S rRNA analysis; breast abscess; *Gordonia bronchialis*.

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Introduction

Gordonia spp., previously classified as *Gordona*, are nocardioform actinomycetes isolated from soil and from the intestinal content of mammals (Conville & Witebsky, 2001). These organisms are emergent pathogens that have been isolated in infections from immunocompromised patients and, less frequently, from previously healthy individuals (Siddiqui *et al.*, 2012; Werno *et al.*, 2005; Riegel *et al.*, 1996).

There have been few reports of different infections caused by *Gordonia* spp. in recent years and, to our knowledge, only 13 cases caused by *Gordonia bronchialis*.

We present a case of recurrent breast abscess caused by *G. bronchialis* in an immunocompetent patient.

Case report

A 39-year-old healthy, immunocompetent woman presented in our emergency department with pain and swelling on her right breast extending to her right arm. The patient had no previous medical history or any other conditions that would predispose the formation of breast abscess. Upon examination, a palpable mass with marked swelling and erythema on the upper right area of the areola was observed. She was diagnosed with non-puerperal mastitis and treated empirically with cloxacillin (500 mg every 6 h), and followed up for additional tests. An initial ultrasound scan showed an area of acute inflammatory

changes and excluded abscess formation. Ten days later, she returned to the hospital without having experienced any improvement in her symptoms. Physical examination during the second emergency department visit revealed a palpable mass of about 10 cm in diameter with erythema of the overlying skin on the upper quadrant of the right breast, as well as enlarged axillary lymph nodes. A full blood count showed a high leukocyte count (12 000 cells μl^{-1}) with neutrophil predominance. A repeated ultrasound scan detected a poorly circumscribed mass compatible with an abscess. There was no mammographic evidence of malignancy. Consequently, the abscess was drained and the purulent material sent for microbiological study. Antibiotic treatment was switched to oral cefuroxime (500 mg every 8 h). A Gram stain of the abscess aspirate showed abundant polymorphonuclear leukocytes and Gram-positive bacilli. After 48 h of incubation, there was no growth under aerobic conditions, and metronidazole was added to the treatment due to the possibility of an anaerobic bacterial infection. Meanwhile, given the lack of improvement in the local inflammatory signs and pain, treatment was changed again to oral amoxicillin-clavulanate, after which a slow clinical improvement was noted. Six weeks later, the treatment was changed to oral ciprofloxacin (500 mg every 12 h), and, given the persistence of the abscess, a second surgical drainage was performed and new samples were obtained for microbiological and pathological studies. The antibiotic treatment was stopped after 19 uninterrupted weeks due to a marked improvement.

On day 4 of incubation, non-haemolytic, dry, creamy-white colonies were seen on aerobic cultures of pus aspirated at the time the abscess was first incised. The anaerobic cultures and the cultures taken from the second surgical drainage were both negative.

The isolate was Gram positive, pleomorphic and bacillary shaped, and formed rough white colonies that became yellow-coloured after several days of culture. The colonies were catalase and urease positive, oxidase negative and weakly acid-fast according to a modified Ziehl–Neelsen stain.

The isolate was identified initially as *Rhodococcus* sp. by a commercial identification system (API Coryne; bioMérieux) with a profile number of 3110004. When entered into the API database, this biochemical pattern identified the organism as *Rhodococcus* sp. (98 % identification). The software also suggested the possibility that the isolate was of the genus *Gordonia*, *Dietzia* or *Nocardia*.

This unusual isolate in this clinical setting prompted us to send the strain to a reference laboratory where it was identified as *Gordonia bronchialis* by amplification and sequencing of the 16S rRNA gene with a previously described procedure (Lane, 1991).

According to disk diffusion, the organism was susceptible to gentamicin, amikacin, amoxicillin-clavulanate, cefotaxime, penicillin G, vancomycin, ciprofloxacin and tetracycline; intermediate to erythromycin; and resistant to clindamycin and cotrimoxazole (breakpoints used for the interpretation of the resulting zone sizes were those established by the Clinical and Laboratory Standards Institute for *Staphylococcus* spp.; CLSI, 2012). Susceptibility testing was carried out before the definitive identification.

Discussion

Gordonia spp. are non-motile, Gram-positive, weakly acid fast rods classified as aerobic actinomycetes. *Gordonia* spp., previously classified within the genera *Rhodococcus* and *Gordona*, have been isolated from the environment and from the intestinal content of mammals (Conville & Witebsky, 2001). These organisms rarely cause disease in humans, although they have been isolated in infections from immunocompromised patients and, less frequently, from previously healthy persons (Siddiqui *et al.*, 2012; Werno *et al.*, 2005; Riegel *et al.*, 1996). However, their true prevalence could be underestimated due to their fastidious growth requirements, slow growth in conventional cultures and confusing morphology. Isolation of *Gordonia* spp. requires prolonged incubation for at least 3–4 days, and this delay may cause false-negative culture reports resulting in infections caused by this genus being overlooked (Gil-Sande *et al.*, 2006). Moreover, *Gordonia* spp. may also be incorrectly identified as other morphologically similar bacteria, such as commensal species of actinomycetes or mycobacterial species.

A recent literature review (Siddiqui *et al.*, 2012) described several types of infection caused by *Gordonia* spp. including a sternal wound infection after heart surgery, external otitis, bronchitis, skin and soft tissue infections, bacteraemia and catheter-associated infections.

G. bronchialis, the species isolated in this case, was initially identified from samples of soil and sputum from patients with pulmonary disease (Conville & Witebsky, 2001; Johnson *et al.*, 2011; Siddiqui *et al.*, 2012). Only 13 cases of infection with this species of actinomycete have been described (Siddiqui *et al.*, 2012), including a reported recurrent breast abscess in an immunocompetent patient with no previous medical history (Werno *et al.*, 2005) that shared many features with our case. Another case of granulomatous mastitis in an immunocompetent patient caused by other species of *Gordonia* (*Gordonia terrae*) has also been reported, although the patient had a previous history of nipple piercing (Zardawi *et al.*, 2004).

Recurrence and a protracted clinical course are common characteristics of infections with *Gordonia* spp. (Werno *et al.*, 2005). Many patients, as in our case, require 6–12 weeks of therapy with different antibiotics, often along with appropriate surgical debridement (Johnson *et al.*, 2011; Siddiqui *et al.*, 2012; Sng *et al.*, 2004; Werno *et al.*, 2005). This prolonged course could be explained by the ability of *Gordonia* spp. to form sessile communities where antimicrobial penetration is poor (Werno *et al.*, 2005). There are no standardized recommendations for treatment of infections caused by *Gordonia* spp. These species are susceptible to a wide range of antibiotics *in vitro*. Patients described in the literature were successfully treated with either monotherapy or a combination of different antibiotics such as amoxicillin-clavulanate, quinolones, cephalosporins, carbapenems, cotrimoxazole, azithromycin, clindamycin and vancomycin (Werno *et al.*, 2005).

In our case, careful observation of the Gram stain performed on the abscess material and extended incubation of cultures beyond the protocols for *Staphylococcus aureus* (the most common etiological agent) were crucial in guiding the techniques used to establish the aetiology. 16S rRNA gene sequencing allowed us to definitively identify the species involved and differentiate it from other phylogenetically related species such as *Rhodococcus*, *Streptomyces* and *Nocardia*. Molecular methodologies are increasingly being used to identify actinomycetes, and 16S rRNA gene sequencing has become a valuable tool for this purpose in many laboratories (Blaschke *et al.*, 2007).

In summary, we would like to emphasize that, although *Gordonia* spp. infections are rare, they nonetheless appear in young, healthy and immunocompetent people. This is the second case of breast abscess caused by *G. bronchialis* described in the literature, with significant clinical similarities to the first case. Whilst careful execution of simple tests such as Gram staining and a modified acid-fast stain were key to determining the aetiology and patient

management, we recommend additional tests such as 16S rRNA gene sequencing for the accurate identification of these organisms.

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