Catheter-related bloodstream infection by *Lindnera fabianii* in a neutropenic patient

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*Lindnera (Pichia) fabianii* (*Candida fabianii* teleomorph) is a yeast species that is an uncommon cause of invasive human infections. This report describes what we believe to be the first human case of a catheter-related *L. fabianii* bloodstream infection in a neutropenic patient. The Clinical and Laboratory Standards Institute guidelines do not offer antifungal breakpoints in this neutropenic case and empirical chemotherapy was considered. Sharing our experience, we will discuss the choice of an effective antifungal agent in this uncommon clinical situation.

**Introduction**

The incidence of human infection by uncommon invasive fungi is increasing. Among uncommon fungi that can infect humans, *Lindnera fabianii* (*Candida fabianii* teleomorph) is known to be a rare cause of invasive human infection. Since the first case, to the best of our knowledge, of *L. fabianii* prostatitis in a patient with chronic lymphocytic leukaemia was reported in 1989, five more cases have been reported over the past 10 years. Interestingly, neutropenia, an important risk factor for invasive *Candida* infection, was not mentioned in any of these cases (Bhally *et al.*, 2006; Dooley *et al.*, 1990; Gabriel *et al.*, 2012; Grenouillet *et al.*, 2010; Hamal *et al.*, 2008; Valenza *et al.*, 2006). In treating invasive *Candida* infection it is important to consider whether the patient is neutropenic, as the antifungal breakpoints for *Candida* species in the Clinical and Laboratory Standards Institute (CLSI) guidelines were determined based on data from non-neutropenic patients (CLSI, 2008a). Furthermore, there are no available guidelines for antifungal breakpoints for rare *Candida* species such as *L. fabianii*.

Here, we describe what we believe to be the first case of a catheter-related *L. fabianii* bloodstream infection in a neutropenic patient and we discuss the best empirical choice of antifungal medication for this species, which is often misidentified as *Candida utilis* or *Candida pelliculosa* in commonly used biochemical kits (Gabriel *et al.*, 2012; Grenouillet *et al.*, 2010; Hamal *et al.*, 2008; Valenza *et al.*, 2006).

**Case report**

A 47-year-old Korean woman visited the emergency room due to severe lower back pain that was aggravated for 3 days. She had been diagnosed with plasma cell myeloma 1 year previously, and underwent autologous peripheral blood stem cell transplantation and chemotherapy. Her blood oxygen saturation in room air was 94% and other laboratory findings were as follows: white blood cell count, 4180 μl⁻¹ (absolute neutrophil count, 2730 μl⁻¹); haemoglobin, 8.8 g dl⁻¹; platelet count, 56 000 μl⁻¹; aspartate aminotransferase/alanine aminotransferase, 46/14 U l⁻¹; creatinine, 0.73 mg dl⁻¹; and lactic acid, 4.16 mmol l⁻¹. Whole-spine magnetic resonance imaging revealed that plasma cell myeloma involved both iliac bones and the sternum, with severe central canal stenosis. Lenalidomide and high-dose dexamethasone with radiation therapy were started to control refractory plasma cell myeloma and spinal cord compression. On day 23, she was moved to the intensive care unit (ICU), owing to respiratory failure, and at the time of ICU admission, her complete blood count showed pancytopenia, with a white blood cell count of 1980 μl⁻¹ (absolute neutrophil count of 1330 μl⁻¹), haemoglobin of 10.1 g dl⁻¹ and a platelet count of 4000 μl⁻¹.

On day 33, three blood cultures were reported to be positive in a BacT/ALERT automated detection system (bioMérieux). A small, white colony was isolated after 24 h of incubation on a blood agar plate; microscopic examination of Gram-stain samples revealed yeast-like structures. The colonies appeared white on CHROM agar medium (Becton Dickinson). Subcultures on Sabouraud dextrose agar at 37 °C produced white, glistening, round colonies with the production of an ester-like odour. The organism was identified at the species level using the VITEK-2 YST (bioMérieux) as *C. utilis* (probability 93.33%). Upon *in vitro* susceptibility testing using a VITEK-2 YST card, MIC values of amphotericin B, 5-fluorocytosine, voriconazole and fluconazole were 0.5, <1, <0.12 and 2.0 μg ml⁻¹, respectively. With the diagnosis of candidaemia, intravenous amphotericin B treatment was initiated. On day 34, a tip culture from the removed central catheter grew more than 100 colonies of *C. utilis*. Despite
an 8-day course of intravenous amphotericin B, blood cultures remained positive for the same fungus. This failure of first-line antifungal treatment and the rarity of *C. utilis* fungaemia led us to perform a molecular identification of the yeast, suspecting that the previous identification by the automated system was incorrect. Finally, we identified *C. fabianii* (*Lindnera fabianii*) using molecular methods.

Considering the clinical course of the patient and the review of peer experience describing invasive *L. fabianii* infection, amphotericin B was changed to caspofungin to control fungaemia. Caspofungin was continued for another 2 weeks until all signs of blood infection had disappeared. However, the patient died on day 64 due to multi-organ failure, septic shock and lactic acidosis due to refractory plasma cell myeloma.

**Molecular identification and antimicrobial susceptibility**

For molecular identification, the internal transcribed spacer (ITS) domain and the D1/D2 domain of the large subunit (28S) of the rRNA gene were amplified using primers of ITS1/ITS4, ITS5/ITS4 (White et al., 1990) and D1/D2 (Kurtzman & Robnett, 1997). The ITS gene sequence showed 100 % (588/588) and 99.3 % (583/587) similarity to that of *C. fabianii* (*Pichia fabianii*, accession no. HQ651909) and *Candida mississippensis* (*P. mississippiensis*, accession no. GQ340433) in the GenBank database (http://blast.ncbi.nlm.nih.gov/Blast.cgi), respectively, and 100 % (624/624) similarity to that of the *C. fabianii* (*Lindnera fabianii*, CBS 5481) type strain in the CBS yeast sequence database (http://www.cbs.knaw.nl/). The next closest sequences of other *Candida* species had less than 95 % similarity in both databases. The D1/D2 domain sequence showed 100 % (601/601), 99.0 % (595/601) and 99.1 % (579/584) similarity to those of *C. fabianii* (*P. fabianii*, accession no. HQ651909), *C. veronae* (*P. veronae*, accession no. DQ409160) and *C. bimundalis* (*P. bimundalis*, accession no. EF550329) strains in the GenBank database, respectively. In the CBS yeast sequence database, it showed 100 % (590/590), 100 % (570/570) and 99.14 % (579/584) similarity to the sequences of the type strain of *C. fabianii* (*Lindnera fabianii*, CBS 5640), *C. veronae* (*P. veronae*, CBS6591) and *C. bimundalis* (*L. bimundalis*, CBS 5642), respectively. The percentage similarity to other *Candida* species was less than 99 % in the two databases. Based on molecular identification using ITS and D1/D2 domain genes, we concluded that *C. fabianii* (*Lindnera fabianii*) was the best species match.

In *vitro* antifungal susceptibility testing was performed by using the CLSI M27-A3 broth microdilution method and MICs were determined after 24–48 h of incubation (CLSI, 2008b). The results are shown in Table 1.

**Discussion**

Until now, some non-neutropenic patients with multiple risk factors were reported with *L. fabianii* fungaemia (Bhally et al., 2006; Dooley et al., 1990; Gabriel et al., 2012; Grenouillet et al., 2010; Hamal et al., 2008; Valenza et al., 2006). The risk factors include long stays in the ICU, acute renal failure treated by haemodialysis, long-term use of broad-spectrum antibiotics, and a long-term indwelling catheter, especially vascular access (Concia et al., 2009; Eggimann et al., 2003; León et al., 2009; Leroy et al., 2009). The patient in our report had all of the above risk factors with persistent neutopenia.

Initially, the biochemical identification of the fungus in this patient was *C. utilis* using the VITEK-2 YST (bioMérieux) identification card. *C. utilis* is also an uncommon species that can cause invasive human infection. Only a few cases of invasive human infection including fungaemia, fungal keratitis and urinary tract infection have been reported in the literature (Alsina et al., 1988; Bougnoux et al., 1993; Hazen et al., 1999; Lukić-Grlić et al., 2011; Shih et al., 1999). Based on a literature review and an *in vitro* susceptibility test, intravenous amphotericin B therapy was initiated for the patient. Furthermore, since this *Candida* species is not easily identified or is often misidentified as other species (Gabriel et al., 2012; Grenouillet et al., 2010; Hamal et al., 2008; Valenza et al., 2006), molecular diagnosis was very helpful in this case study.

After reviewing all documented cases of invasive *L. fabianii* infection (Bhally et al., 2006; Dooley et al., 1990; Gabriel et al., 2012; Grenouillet et al., 2010; Hamal et al., 2008; Valenza et al., 2006), intravenous amphotericin B was found to be a successful choice for invasive *L. fabianii* infection in several cases (Bhally et al., 2006; Dooley et al., 1990; Hamal et al., 2008). As for azoles, resistance to fluconazole and voriconazole and cross-resistance to azoles developed rapidly, despite the fact that both antibiotics looked fair based on initial MICs (Hamal et al., 2008). So the management using intravenous amphotericin B was thought to be effective at the time of molecular identification of this fungus. However, an 8-day course of intravenous amphotericin B did not eradicate the pathogen from the patient’s blood, although the *in vitro* MIC of amphotericin B was stable with a value of 0.5 μg ml⁻¹ during management.

<table>
<thead>
<tr>
<th>Antifungal</th>
<th>MIC (μg ml⁻¹)</th>
<th>Broth microdilution</th>
<th>VITEK-2</th>
</tr>
</thead>
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<tr>
<td>Amphotericin B</td>
<td>0.5</td>
<td>1.0</td>
<td>0.5</td>
</tr>
<tr>
<td>5-Fluorocytosine</td>
<td>Not done</td>
<td>Not done</td>
<td>&lt;1.0</td>
</tr>
<tr>
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<td>0.06</td>
<td>&lt;0.12</td>
</tr>
<tr>
<td>Fluconazole</td>
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<td>4.0</td>
<td>2.0</td>
</tr>
<tr>
<td>Caspofungin</td>
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<td>Not done</td>
</tr>
<tr>
<td>Micafungin</td>
<td>0.125</td>
<td>Not done</td>
<td>Not done</td>
</tr>
</tbody>
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(Bhally et al., 2006; Dooley et al., 1990; Gabriel et al., 2012; Grenouillet et al., 2010; Hamal et al., 2008; Valenza et al., 2006). The risk factors include long stays in the ICU, acute renal failure treated by haemodialysis, long-term use of broad-spectrum antibiotics, and a long-term indwelling catheter, especially vascular access (Concia et al., 2009; Eggimann et al., 2003; León et al., 2009; Leroy et al., 2009). The patient in our report had all of the above risk factors with persistent neutopenia.
In managing this fungaemia, we considered two clinical aspects of this case study. Firstly, *L. fabianii* is known as a yeast capable of producing biofilms (Hamal *et al.*, 2008), which may explain the resistance of this strain to most of the antifungal agents described, with the exception of echinocandins and lipid amphotericin B formulations (Chandra *et al.*, 2005). Especially if the patient has a central venous catheter or receives total parenteral nutrition, the tendency of biofilm production in the majority of non-*C. albicans* Candida species can increase significantly (Shin *et al.*, 2002). *Candida* biofilms are related to enhanced resistance against most antifungal drugs, except lipid amphotericin B formulations and echinocandins (Mukherjee & Chandra, 2004). Secondly, the antifungal breakpoints provided by the CLSI are based on data from non-neutropenic patients (CLSI, 2008b), so in this patient the clinical efficacy of the antifungal agent could be different from the data presented in the CLSI guidelines; it could be lower than it is in a non-neutropenic patient.

Based on the two points mentioned above, amphotericin B was changed to caspofungin, which finally converted the positive blood culture result to negative within a few days. In this patient who had multiple risk factors for *L. fabianii* fungaemia, caspofungin seemed to overcome the effect of biofilm production or the alternation of the effectiveness of antifungal agents due to neutropenia. Besides, caspofungin has good levels of activity in treating *C. pelliculosa* infection (Diekema *et al.*, 2009) the identity of which can be confused with *L. fabianii* in commercial biochemical identification methods. Liposomal amphotericin B could be an alternative option, but the clinical efficacy was not certain in our case.

In summary, this is the first case report of a *L. fabianii* catheter-related infection in a neutropenic patient, to the best of our knowledge. We share our experience to support the best empirical choice of antifungal agent in treating invasive infection by *L. fabianii*, which can be commonly confused with *C. utilis* or *C. pelliculosa* in commercial biochemical kits. We also emphasize the role of molecular assays in confirming the identities of these uncommon fungal species.

**Acknowledgements**

We thank Dr Jong Hee Shin, Department of Laboratory Medicine, Chonnam National University Medical School, for performing antifungal susceptibility testing based on the CLSI broth microdilution method.

**References**


