Pseudozyma aphidis fungaemia with invasive fungal pneumonia in a patient with acute myeloid leukaemia: case report and literature review

Hyonsoo Joo,1 Yeon-Geun Choi,1 Sung-Yeon Cho,1,2 Jae-Ki Choi,1,2 Dong-Gun Lee,1,2,3 Hee-Je Kim,3 Irene Jo,4 Yeon-Joon Park4 and Kyo-Young Lee5

1Division of Infectious Diseases, Department of Internal Medicine, College of Medicine, The Catholic University of Korea, Seoul, Korea, 2Vaccine Bio Research Institute, College of Medicine, The Catholic University of Korea, Seoul, Korea, 3The Catholic Blood and Marrow Transplantation Centre, College of Medicine, The Catholic University of Korea, Seoul, Korea, 4Department of Laboratory Medicine, College of Medicine, The Catholic University of Korea, Seoul, Korea and 5Department of Hospital Pathology, College of Medicine, The Catholic University of Korea, Seoul, Korea

Summary

Pseudozyma species rarely cause invasive diseases in humans, which are usually isolated from plants. There have been anecdotal reports regarding Pseudozyma species infections in patients with underlying diseases or in neonates. However, clinical data and the pathogenicity in humans are still insufficient. We experienced a case of Pseudozyma aphidis fungaemia with invasive fungal pneumonia that developed during reinduction chemotherapy in a 51-year-old male with acute myeloid leukaemia (AML). P. aphidis was suspected based on the morphology of the yeast isolated from the blood and was confirmed via rDNA gene sequencing analysis. The patient successfully underwent stem cell transplantation with continuing antifungal treatment and finally completely recovered from both the AML and infectious complications. Here, we report a case of P. aphidis infection that developed during neutropenia in an AML patient and review the global literature.

Key words: Acute myeloid leukaemia, fungaemia, neutropenia, pneumonia, Pseudozyma aphidis yeasts.

Introduction

Rare fungi have recently been implicated in human infections ranging from colonisation to invasive fungal infections (IFIs) in immunocompromised patients, accounting for <10% of all isolated fungal pathogens.1 Pseudozyma species (spp.) are basidiomycetous yeast classified in the family Ustilaginaceae, and its members are close relatives of Ustilago maydis and other smut fungi. At least 20 Pseudozyma spp. are recognised, most of which are environmental pathogens.2 Pseudozyma spp. infections in humans have rarely been reported after the first description as a human pathogen in 2003.3 Data regarding the clinical characteristics and pathogenicity in humans remain insufficient. Recently, we experienced a case of Pseudozyma aphidis fungaemia with invasive fungal pneumonia after reinduction chemotherapy for acute myeloid leukaemia (AML). Here, we describe this case and review the global literature. The Institutional Review Board at Seoul St. Mary’s Hospital approved this case report and waived the need for patient consent (No. KC15RISI0570).

Case report

A 51-year-old man who was diagnosed with AML and had not experienced remission after the first induction chemotherapy had received consolidation chemotherapy as part of his treatment plan for acute myeloid leukaemia (AML). After relapsing, he received reinduction chemotherapy and was admitted to the hospital with symptoms of fever and neutropenia. Upon examination, the patient was found to have an infiltrate in the lower right lobe of the lung. Blood cultures were positive for Pseudozyma aphidis, and a chest CT scan revealed a consolidation in the lower right lobe of the lung. The patient was diagnosed with Pseudozyma fungaemia and invasive fungal pneumonia. He was treated with voriconazole and itraconazole, and his condition improved. After completing reinduction chemotherapy, he underwent stem cell transplantation, and his condition improved further. He finally recovered from both the AML and infectious complications.
Chemotherapy started reinduction chemotherapy with 100 mg m\(^{-2}\) cytarabine for 7 days and 90 mg m\(^{-2}\) daunorubicin for 3 days. On day seven after reinduction chemotherapy (D7), neutropenic fever (up to 38.0 °C) developed as measured using an axillary thermometer. No other symptoms were reported. Laboratory data included a white blood cell count of 220 \(\mu\)l\(^{-1}\) (absolute neutrophil count, 0 \(\mu\)l\(^{-1}\)) and C-reactive protein level of 8.60 mg dL\(^{-1}\). Empirical antibiotic therapy with ceftazidime (2 g twice a day) and isepamicin (400 mg once a day) was initiated after performing blood cultures. On D8, the results of the blood culture revealed presumptive growth of gram-positive cocci (GPC). Teicoplanin (12 mg kg\(^{-1}\) a day after loading with 12 mg kg\(^{-1}\) every 12 h for three doses) was added based on the culture results.

Although the patient had no respiratory symptoms, infiltration was suspected in the right lower lung field on the chest radiograph on D9. Low-dose computed tomography of the lung was performed, which showed consolidation with surrounding ground glass opacity at the medial segment of the right middle lobe (RML) (Fig. 1). The serum galactomannan assay performed twice a week was negative. The patient received fluconazole instead of posaconazole as the primary antifungal prophylaxis; because he had participated in a clinical trial using a thrombopoietin receptor agonist during the same period, a drug interaction was possible. Then, the antifungal prophylaxis was empirically changed to caspofungin (50 mg a day after the loading dose) for the possibility of fungal pneumonia according to the revised definition of IFI from the European Organization for the Research and Treatment of Cancer/Mycoses Study Group (EORTC/MSG)\(^4\).

The GPC from the blood culture was identified as vancomycin-resistant Entero\(\)coccus faecium (3 of 4 bottles), using Vitek 2 (bioMérieux, Hazelwood, MO, USA). Teicoplanin was changed to linezolid (600 mg twice a day). The follow-up blood culture performed on D11 revealed negative conversion of vancomycin-resistant Entero\(\)coccus bacteraemia. However, the neutropenic fever persisted, and yeast-like organisms were noted from central blood culture (1 of 2 bottles) performed on the same day (D11). This culture result was reported after 2 days of incubation, using BD BACTEC FX blood culture system (BD Diagnostics, Sparks, MD, USA).

Colonies observed on the blood agar plate were white, dry or wrinkled (Fig. 2a). Microscopically, they appeared as yeast with branching pseudohyphae (Fig. 2b). The species could not be identified using either Vitek 2 or API 20C (bioMérieux). Therefore, further identification was performed using rDNA gene sequencing analysis with the following primers: internally transcribed spacer (ITS)-1 (forward primer [5'–TCC GTA GGT GAA CCT GCG G-3'] and reverse primer [5'–GCT GCG TTC ATC GAT-3']), ITS-2 (forward primer [5'–GCA TCG ATG AAG AAC GCA-3'] and reverse primer [5'–TCC TCC GCT TAT TGA TAT-3']), and D1/D2 domain (forward primer [5'–GCA TAT CAA TAA GCG GAG-3'] and reverse primer [5'–GGT CCG TGT TTC AAG ACG G-3']).\(^5\) The isolated gene sequence showed 100% concordance with the P. aphidis strain (GenBank accession number: KF443199.1 and KF443201.1). Because the fungaemia developed during the caspofungin therapy and pneumonia was aggravated with persistent neutropenic fever, caspofungin was changed to liposomal amphotericin B (5 mg kg\(^{-1}\) per day) on D15. The Hickman catheter was removed. The culture result of the tip of the removed Hickman catheter showed no growth.

The follow-up blood culture performed on D14 showed no growth. However, pneumonia was aggravated despite the broad-spectrum antifungal agent. The findings of the bronchoscopy performed on D23 showed no endobronchial lesion. Transbronchial lung biopsy and bronchial washing and brushing were performed at the RML bronchus to identify the pathogens of fungal pneumonia. Pathology specimens showed inflammatory changes with necrosis and dichotomous hyphae with a septum, suggestive of organising

![Figure 1](image_url) Low dose computed tomography of the lung. Consolidation with surrounding ground glass opacity at the medial segment of the right middle lobe.
pneumonia with a fungal infection (Fig. 3). However, we could not identify the genus level due to tissue damage. Culture with bronchial washing fluid revealed no growth of fungal organisms. As the neutropenia recovered, the patient improved clinically and was discharged with itraconazole capsules (200 mg twice a day). At the outpatient clinic, chest radiography revealed little change in the RML consolidation despite 3 weeks of continued itraconazole. Considering the possibility of both proven invasive pulmonary aspergillosis and fungal pneumonia due to *Pseudozyma* spp., itraconazole was changed to voriconazole (4 mg kg\(^{-1}\) twice a day). The patient sequentially received consolidation chemotherapy and allogeneic stem cell transplantation (SCT) while continuing the voriconazole. During the voriconazole treatment, serum level of voriconazole was within the therapeutic range. The pneumonia resolved after 4 months of voriconazole treatment, with complete remission of AML after successful SCT.

**Figure 2** Colonies of yeast on blood agar and microscopic morphology. (a) Yeast-like colonies that are dry, creamy, brightly coloured, and glabrous in texture; (b) Gram stain showing yeast with branching pseudohyphae (arrow).

**Figure 3** Histopathology of lung tissue from a transbronchial lung biopsy specimen. Fungal organisms on the background of necrosis and inflammatory infiltration. (a) Haematoxylin and eosin stain, \(\times 100\); (b) Haematoxylin and eosin stain, \(\times 400\); (c) Silver stain, \(\times 400\).
Discussion
Invasive fungal infections remain a major cause of significant morbidity and mortality in immunocompromised patients, especially those with hematologic malignancies. Of the IFIs, a rare fungus is difficult to treat in the aspects of the early diagnosis and appropriate treatment, which might be related to the severity of the patient’s condition and often challenging intrinsic susceptibility pattern of the pathogens. A Pseudozyma spp. was first described as a possible human pathogen in 2003, when it was identified from blood cultures from three Thai patients, as P. antarctica, P. parantarctica, and P. thailandica. The first case

Table 1 Literature review of Pseudozyma species infections in human.

| Case no. | Pseudozyma spp. | MIC (mg dL\(^{-1}\)) | Treatment | Antifungal agent | Duration | Outcome | Country, year [reference] |
|----------|-----------------|----------------------|-----------|------------------|----------|---------|--------------------------|
| 1        | P. antarctica   | 0.125                | N/A       | N/A              | 4        | Improved | USA, 2008 [8]            |
| 2        | P. parantarctica| 0.25                 | N/A       | N/A              | 4        | Improved | USA, 2008 [8]            |
| 3        | P. thailandica  | 0.03                 | 0.03      | 0.03             | 8        | Improved | India, 2013 [7]          |
| 4        | P. aphidis      | 0.25                 | 0.25      | 0.25             | 8        | Improved | USA, 2014 [12]           |
| 5        | P. aphidis      | 0.25                 | 0.25      | 0.25             | 8        | Improved | Brazil, 2013 [11]        |
| 6        | P. aphidis      | 0.03                 | 0.03      | 0.03             | 8        | Improved | Korea, 2010 [9]          |
| 7        | P. aphidis      | 0.03                 | 0.03      | 0.03             | 8        | Improved | China, 2011 [10]         |
| 8        | P. aphidis      | 0.03                 | 0.03      | 0.03             | 8        | Improved | India, 2013 [7]          |
| 9        | P. aphidis      | 0.03                 | 0.03      | 0.03             | 8        | Improved | USA, 2014 [12]           |
| 10       | P. alboamericana| 0.25                 | >16\(^{6}\) | >16\(^{6}\)      | 8        | Improved | France, 2015 [14]        |
| 11       | P. crassa       | 0.25                 | >16\(^{6}\) | >16\(^{6}\)      | 8        | Improved | Argentina, 2015 [15]     |
| 12       | P. siamensis    | 0.25                 | >16\(^{6}\) | >16\(^{6}\)      | 8        | Improved | This case                |
| 13       | P. amicus       | 0.03                 | 0.03      | 0.03             | 8        | Improved | USA, 2014 [12]           |
| 14       | P. aphidis      | 0.03                 | 0.03      | 0.03             | 8        | Improved | Argentina, 2015 [15]     |
| 15       | P. aphidis      | 0.03                 | 0.03      | 0.03             | 8        | Improved | This case                |

Table 1 Literature review of Pseudozyma species infections in human.

| Case no. | Age/sex | Underlying condition | Predisposing factors | Clinical presentation | Isolated specimen |
|----------|---------|----------------------|----------------------|----------------------|-------------------|
| 1        | N/A     | N/A                  | N/A                  | N/A                  | Blood             |
| 2        | N/A     | N/A                  | N/A                  | N/A                  | Blood             |
| 3        | N/A     | N/A                  | N/A                  | N/A                  | Blood             |
| 4        | 7/F     | Short gut syndrome   | +                    | Fever, chill, malaise, fatigue | Blood             |
| 5        | 78/M    | Astrocytoma          | +                    | Fever after brain surgery | Abscess of brain  |
| 6        | 51/M    | Farmer, chronic leg swelling | – – +   | Chronic mycetoma of leg | Leg\(^{6}\)        |
| 7        | 17/M    | Burkitt lymphoma, chemotherapy | + N/A | Neutropenic fever, lung infiltrates | Pleural fluid     |
| 8        | 0/M     | Hemolytic jaundice   | – –                  | Lethargy, poor feeding | Blood             |
| 9        | 52/F    | Crohn’s disease, total colectomy state | + + –  | Fever, headache, weakness | Blood             |
| 10       | N/A     | N/A                  | N/A                  | N/A                  | Blood             |
| 11       | N/A     | N/A                  | N/A                  | N/A                  | Blood             |
| 12       | N/A     | N/A                  | N/A                  | N/A                  | Blood             |
| 13       | 68/F    | Adenocarcinoma of ampulla of Vater | – N/A | Fever, chill | Blood             |
| 14       | 6/F     | Osteosarcoma with lung metastasis | + N/A | Neutropenic fever | Blood             |
| 15       | 51/M    | AML, reinduction chemotherapy | + – + – | Neutropenic fever, lung infiltrates | Blood             |

**Note:** MIC values for micafungin.

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Mycoses, 2016, 59, 56–61

59
of *P. aphidis* human infection was reported in a 7-year-old girl with short gut syndrome in 2008.\(^7\)

*Pseudozyma* spp. is classified under the family Ustilaginaceae, phylum Basidiomycota, subphylum Ustilaginomycotina, class Ustilaginomycetes, and order Ustilaginales.\(^2,8\) Ustilaginales are plant pathogens that can infect corn plants to produce tumour-like galls. Of the *Pseudozyma* spp., *P. aphidis* is known as the most common human pathogen. However, due to the rare isolation of *P. aphidis* in human infections, this rare fungus species cannot be identified using commercial laboratories that are available in routine diagnostic laboratories. However, sequencing and phylogenetic analysis can help with the direct detection of a fungus from blood or tissues because yeast taxonomy is continually evolving.

We retrospectively reviewed the global literature to identify the possible risk factors, clinical presentation, and optimal treatment strategies for *Pseudozyma* infection; in addition to the present case, 14 case reports were found (Table 1).\(^3,7,12–15\) Since 2014 when previous literature review was reported by Prakash *et al.* [8], there were seven additional cases including our case. Therefore, we present a updated literature review with some modifications and adding recent cases. Of the 15 cases, seven were identified as *P. aphidis*, two were *Pseudozyma* spp. without spp. level identification, and the remaining six cases were non-*aphidis* *Pseudozyma* isolates that were all different spp. from Thailand. Of the nine cases with reported underlying medical conditions, three had gastrointestinal (GI) tract problems such as short bowel syndrome or intestinal surgery,\(^4,9,13\) and three had neutropenia,\(^7,14,15\) which can damage the gut mucosa. Of the seven patients for whom the presence or absence of a central venous catheter (CVC) was reported, six patients had a CVC. The risk factors are thought to be similar to those of other less common yeast infections including GI tract problems, the presence of a CVC, and neutropenia. We could not identify the exact social history or dietary history to determine potential exposure to plants or crops. However, crop exposure was identified in two cases: a farmer with mycetoma of the leg and a paediatric patient who had eaten corn chips.\(^8,10\) In the present case, the possible port of entry for the *P. aphidis* fungaemia is uncertain. While the fungaemia met the definition of a catheter-related bloodstream infection because *P. aphidis* was isolated only from central blood, the origin of *P. aphidis* is unclear.

The major manifestation of *Pseudozyma* infection is fungaemia (12 of 15 cases, 80%); others (*n* = 3) have been isolated from a brain abscess that developed after brain surgery, deep biopsy of mycetoma of the leg, and pleural fluid.\(^9–11\) In our case, we proved *P. aphidis* fungaemia with fungal pneumonia from lung tissue. However, our case has a limitation, in that we could not identify the fungal pathogen from the lung tissue. Fungus culture with fresh lung tissue and sequencing of the tissue was not performed. Galactomannan assay using bronchial washing fluid was not performed either. Antifungal susceptibility testing was performed in 11 cases of human infections caused by *Pseudozyma* spp., *P. aphidis* was susceptible to itraconazole, voriconazole, and amphotericin B; had varying susceptibility to fluconazole; and was resistant to echinocandines and flucytosine.\(^7,8,11,14,15\) Non-*aphidis* *Pseudozyma* spp. seemed to have susceptibility to amphotericin B only.\(^13\) In all reported cases, *Pseudozyma* spp. were resistant to echinocandines and flucytosine.

Here, we described a case of a 51-year-old male patient with AML who suffered from neutropenic fever during chemotherapy with a defined bacterial and fungal infection that was finally diagnosed as *P. aphidis* fungaemia and concurrent invasive fungal pneumonia without genus level identification. Many other fungal pathogens show subtle morphological differences between forms found in tissue and in culture. Further accumulation of data regarding rare fungi is needed. This case is worth reporting in the aspect that *P. aphidis* fungaemia developed during neutropenic fever with concurrent invasive fungal pneumonia in an AML patient.

**Conflict of interest**

We declare no conflicts of interest.

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Pseudozyma fungaemia with pneumonia

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Mycoses, 2016, 59, 56–61

61