

Case Report

Isolation of *Fereydounia khargensis* from continuous ambulatory peritoneal dialysis fluid – A rare case report

Sukanya Sudhaharan¹, Umabala Pamidimukkala¹, Nikhi Verma¹, Ruqaiya Begum¹, Gangadhar Taduri²

Departments of ¹Microbiology and ²Nephrology, Nizam's Institute of Medical Sciences, Hyderabad, Telangana, India.

*Corresponding author:

Sukanya Sudhaharan
Department of Microbiology,
Nizam's Institute of Medical
Sciences, Hyderabad,
Telangana, India.

sukanyavimala@gmail.com

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ABSTRACT

In recent years, the infections caused by rare fungi have been increasing due to the increase in immunocompromised conditions. *Fereydounia khargensis* is usually an environmental pathogen and rarely causes infection in humans. To our knowledge, the present case is the world's third case and India's first case of *F. khargensis* identified by sequencing of the internal transcribed spacer (ITS) region. Unlike *Candida* spp., these yeasts are resistant to echinocandins. Hence, identifying the yeast is important, as it would help in the proper management of the patients.

Keywords: DNA sequencing, Echinocandin resistance, Immunocompromised

INTRODUCTION

Fereydounia khargensis is an emerging new yeast in the order *Urocystidales*, subphylum *Ustilaginomycotina*. The yeast was first isolated from plant remnants on Kharg Island in the Persian Gulf of Iran in 2014. In the order *Urocystidales*, a new lineage is represented by *F. khargensis* of the *Ustilaginomycotina* subphylum. In general, the members of the *Ustilaginomycotina* subdivision produce two phases. The first phase is known as the saprobic haploid yeast phase, and the second is called the parasitic dikaryotic hyphal phase; hence, it is known to be dimorphic. Predominantly, they are found in countries with temperate climates as compared to tropical countries.^[1] Identification of the yeast is difficult by conventional phenotypic and automated methods^[2] and can be confirmed by molecular methods. Therefore, the identification of the yeast was successfully done using Sanger sequencing of the internal transcribed spacer (ITS) and a less variable large subunit (LSU) region in the *Ribosomal ribonucleic acid (rRNA)* gene. There were only two published case reports regarding the human infections caused by the yeast. We report, to our knowledge, the world's third case and India's first case of continuous ambulatory peritoneal dialysis (CAPD) peritonitis caused by *F. khargensis*. Antifungal susceptibility testing was also carried out using Clinical Laboratory Standard Institute (CLSI) guidelines M27-A3 against two azoles, three echinocandins, and a polyene, namely fluconazole, voriconazole, caspofungin, micafungin, anidulafungin, and amphotericin B.

CASE REPORT

The present case was of a 56-year-old female with autosomal-dominant polycystic kidney, known hypertensive with chronic kidney disease, and end-stage renal disease (ESRD) since

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2019. She was started on maintenance hemodialysis in July 2023. After 12 days, due to multi-access failure, bilateral internal jugular vein thrombosis, and bilateral arteriovenous fistula primary failure, she was started on CAPD. She had a history of left lower-limb deep vein thrombosis on oral apixaban. One month later, she came to the hospital with complaints of decreased and further no ultrafiltrate (UF) with nausea, vomiting, decreased appetite, and decreased urine output for 2-3 days. On examination, the patient had pedal edema and pallor. Peritoneal dialysis (PD) fluid was turbid, and effluent was sent for analysis and culture. The patient was given automated PD for 2 days. PD catheter position was confirmed. The total cell count of the PD fluid on the day of admission was 300 cells. She was continued on PD manual exchanges. Daily volume of fluid, UF, and renal charting were done. The patient was started on intravenous ceftazidime 1.5 g once daily and vancomycin 1 g in 100 mL of normal saline over 1 h. The patient's symptoms did not improve. Aerobic and fungal culture showed growth of yeast, which was unidentified by Vitek 2C and later identified as *F. khargensis* by DNA sequencing. Due to access failure, the catheter was not removed to salvage it. The patient was started on oral fluconazole 200 mg once daily for 5 days. She improved symptomatically and was advised to continue the treatment for 14 days. The PD fluid total counts were reduced to 75 cells. She was discharged with advice to follow-up after 1 week. The course of the hospital stay is given as a flowchart in Figure 1.

Microbiological processing

The sample was inoculated on Sabouraud's dextrose agar (SDA) and incubated at 30°C. After 48 h of incubation, there was the growth of a dry, wrinkled colony [Figure 2a]. Gram stain of the colony showed elongated, irregular yeast cells [Figure 2b]. In Vitek 2C, the organism could not be identified. ITS (ITS 4 and ITS 5) and the D1/D2 region in the LSU of the *rRNA* gene were amplified using universal primers and sequenced, which showed *F. khargensis* with homology of 99.7% (ITS). The gene accession ID of the isolate is PQ100145. Antifungal susceptibility testing was done by broth microdilution as per CLSI guidelines.^[3] As there are no clinical breakpoints for this yeast, the minimum inhibitory concentration (MIC) could not be interpreted. The *in vitro* antifungal susceptibility of the drugs is mentioned in Table 1. The yeast showed a lower MIC to azoles and a high MIC to amphotericin B and echinocandins.

Table 1: *In vitro* antifungal susceptibility test results.

Sr. No.	Antifungal agent	Minimum inhibitory concentration (MIC in µg/mL)
1	Fluconazole	2
2	Voriconazole	0.06
3	Amphotericin B	≥16
4	Caspofungin	≥16
5	Micafungin	≥16
6	Anidulafungin	≥16

MIC: Minimum inhibitory concentration

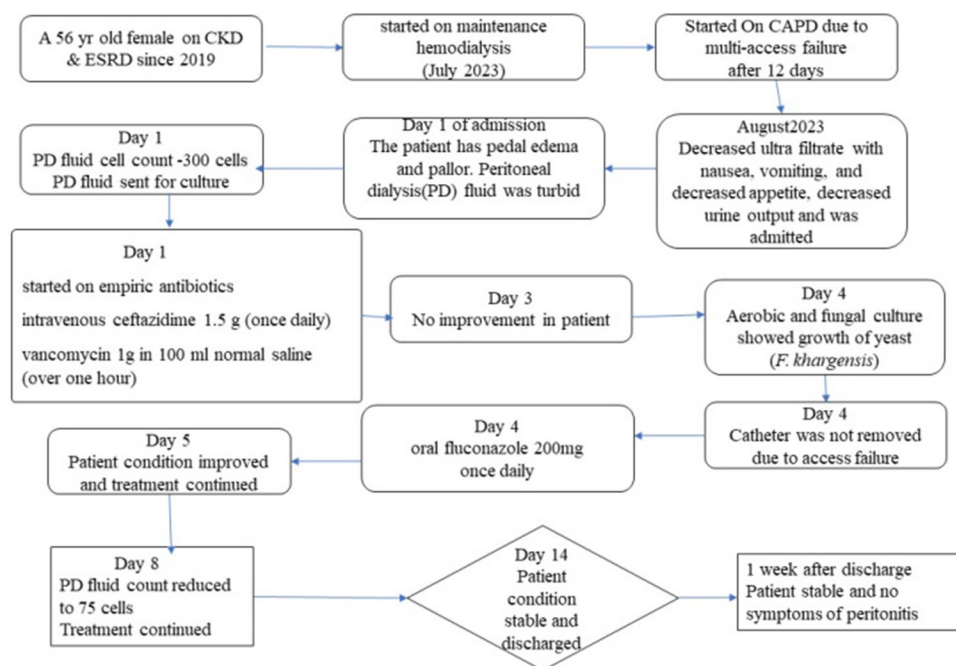


Figure 1: Course of hospital stay. CAPD: Continuous ambulatory peritoneal dialysis, CKD: Chronic kidney disease, ESRD: End stage renal disease

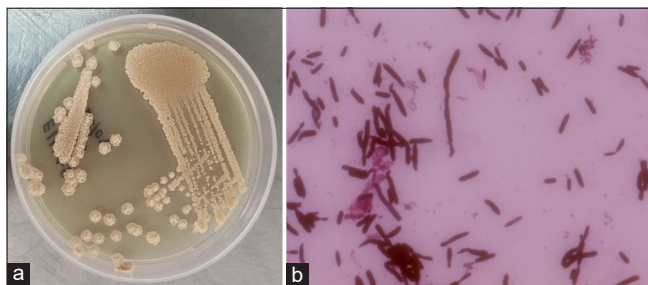


Figure 2: (a) Culture of *Fereydounia khargensis* on Sabouraud dextrose agar, (b) Microscopy-Gram stain of *Fereydounia khargensis* under oil immersion ($\times 1000$).

DISCUSSION

Species of *Ustilaginomycotina*, like *Pseudozyma*, are usually environmental pathogens and rarely cause infection in humans.^[4] To date, only two cases of infection in humans have been reported in the literature. Tap *et al.*^[2] have reported these first two cases of invasive infections caused by *F. khargensis* in immunocompromised patients in Malaysia.^[2] The patients were immunocompromised, a human immunodeficiency virus-positive patient in whom the yeast was isolated from blood culture, and an ESRD patient on CAPD in whom the yeast was isolated from peritoneal fluid. In immunocompromised patients, due to impaired host immune status, the site of entry could be due to either translocation of the pathogen from the gut or the presence of any catheters.^[5] In the present case, the patient was also an immunocompromised patient with ESRD on CAPD.

The yeast will grow on the conventional SDA media for fungal organisms, but identification of the yeast by conventional and automated methods is difficult. In the present case, the identification of the yeast was done by sequencing.

As it is a rare emerging yeast, further studies have to be done to know the pathogenicity, mode of transmission, and resistance mechanisms of the yeast.^[2]

The yeast showed a low MIC to azoles but a higher MIC to echinocandins and amphotericin B,^[2] similar to the present case.

In the two cases reported in the literature, one patient was treated with amphotericin B for 7 days. The patient did not respond and was switched to itraconazole for 14 days, for which he responded. The other patient was treated with fluconazole for 10 days. The present case is the world's third case and India's first case of *F. khargensis* identified by DNA sequencing ITS. In the present case, the patient was treated with fluconazole for 14 days and was discharged in stable condition.

CONCLUSIONS

F. khargensis is an emerging opportunistic yeast in immunocompromised patients. As the sensitivity differs from

other yeasts, which are usually susceptible to echinocandins, this yeast is resistant to echinocandins and is susceptible to voriconazole and itraconazole. So any yeast with varied morphology has to be identified by molecular methods to prevent the mortality associated with infections.

Author's contributions: SS: Conceptualization; data curation; formal analysis; validation; visualization; writing - original draft; and writing - review and editing. UP: Conceptualization; Writing - review and editing. NV: Investigation; methodology; resources; software; writing - review and editing. RB: Resources; writing - review and editing. GT: Resources; writing - review and editing.

Ethical approval: The research/study was approved by the Institutional Review Board at Nizam's Institute of Medical Sciences, approval number EC/NIMS/3038, dated 12th September 2022.

Declaration of patient consent: The authors certify that they have obtained all appropriate patient consent forms. In the form, the patients have given their consent for their clinical information to be reported in the journal. The patients understand that their names and initials will not be published and due efforts will be made to conceal their identity, but anonymity cannot be guaranteed.

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