A CASE OF MYCOTIC SEPSIS DUE TO *TRICHOSPORON COREMIIFORME* IN A TRAUMA PATIENT IN MHAT-SHUMEN

S. Voycheva[1], E. Bozova[1], I. Filipova[2], L. Boyanova[3]

[1]Microbiology laboratory, Multispecialty Hospital for Active Treatment – Shumen,
[2]National Reference Laboratory of Sexually Transmitted Diseases, National Center of Infectious and Parasitic Diseases,
[3]National Reference Laboratory of Mycoses, Department Microbiology National Center of Infectious and Parasitic Diseases, Sofia

ABSTRACT

Trichosporon spp. are yeast-like fungi,a rare cause of mycotic sepsis. We share a case report of a thirty-three year old man with polytrauma after a car accident, who died after a long hospitalization and treatment in intensive care unit. Four blood samples for blood culture were submitted to the Microbiology laboratory in MHAT-Shumen, which were all positive for fungi. The tests available and performed in our laboratory were insufficient for species identification. Pure culture was sent to National Reference Laboratory of Mycology and identified as *Trichosporon coremiiforme*. Mycotic sepsis remains a challenge in terms of diagnosis and therapy and often has a fatal outcome.

Keywords: Trichosporon spp., *Trichosporon coremiiforme*, mycotic sepsis, blood culture, intensive care

ADDRESS FOR CORRESPONDENCE:
Stanislava Voycheva
Microbiology laboratory
MHAT-Shumen
63 Vasil Aprilov str, 9705 Shumen, Bulgaria
e-mail: stanislava_voycheva@abv.bg

INTRODUCTION

Mycotic sepsis is a life-threatening condition, which is often fatal. Prolonged hospitalization, prolonged treatment with corticosteroids and broad-spectrum antibacterial agents in intensive care units increase the risk of mycotic sepsis. Fungal isolates from blood culture are a challenge for clinicians as well as for microbiologists, due to species diversity and specific therapy needed. The described clinical case aims to expand our knowledge and outline the difficulties in identifying the causative agents of mycotic sepsis, considering Trichosporon spp.

Trichosporon spp. are widespread yeast-like fungi of the phylum Basidiomycota, a frequent part of human skin and gastrointestinal tract microbiota. The taxonomy of genus Trichosporon has changed over time, with an increasing number of species, seventeen of which are currently clinically significant. [1]

Species of the genus Trichosporon are characterized by the formation of radially symmetrical colonies on Sabouraud agar, which may be white to cream colored, with a smooth, velvety or rough surface. Microscopically rarely septate hyphae, abundant arthroconidia and blastoconidia are observed. A main characteristic of the genus is the ability of the species to hydrolyze urea. [2] This distinguishes them from the genus Geotrichum, whose species also form arthroconidia, but are urease test negative. [3]

Trichosporon spp. is known as the causative agent of the superficial infection “white piedra”, but is also the second most common causative agent of invasive infections from phylum Basidiomycota, after Cryptococcus spp. [4]

Invasive trichosporonosis is an opportunistic infection observed in the settings of immune deficiency, in cancer patients, neutropenic patients, HIV-positive patients and those requiring prolonged intensive care. Due to the ability of Trichosporon to form a biofilm, cases of catheter-related fungemia have also been reported. [3, 5, 8] Despite the adequate treatment, invasive trichosporonosis is often fatal. [1, 2, 6]

Azoles are the recommended therapy for invasive Trichosporon spp infection according to Sanford Guide voriconazole 6 mg/kg i.v/p.os every 12 h on the first day, then – 4 mg/kg i.v/p.os every 12 h;

Amphotericin B, posaconazole or isavuconazole as an alternative), since the genus shows intrinsic resistance to echinocandins. [2, 6, 7, 8].

Until recently, *Trichosporon coremiiforme* was considered as a variety of *Trichosporon asahii*, due to their genetic relatedness. Therfore the reported cases of invasive trichosporonosis with this species are few. [7, 8]

CLINICAL CASE

A thirty-three year old man was transported to the emergency department of MHAT-Shumen, severely injured in a car accident. He was hospitalized in the intensive care unit (ICU) with intracranial and chest trauma. During the first seven days of hospitalization combined antibacterial therapy with ceftriaxone (4g/d) and metronidazole (1g/d) was started. A 12-day therapy with Dexamethasone was also administered for 12 days (12mg/d which were titrated down until discontinuation). The patient underwent weekly microbiological monitoring. At the end of the first week hospital strains of *Acinetobacter baumannii complex* and *Klebsiella pneumoniae* were isolated from the intubation cannula. The antibacterial therapy was changed to meropenem (3g/d), followed by colistin (9MU/d). During the therapy with colistin, antimycotic treatment with fluconazole (200 mg/d) was also started by the attending physician, and was discontinued in fourteen days.

During the first week of hospitalization, three blood samples for blood culture were taken as a part of the ICU monitoring, two of them were sterile and one was contaminated. During the third week of the ICU stay, *Staphylococcus epidermidis* MRS was isolated from another blood sample and treatment with teicoplanin (400mg/d) was administered. In the settings of ongoing treatment, on the twenty-fifth day of his stay in the ICU, the patient presented with high temperature 38°C to 39°C. Subsequent laboratory tests revealed increased white cell count (13.34 x 10^9/L) and C-reactive protein values (CRP; 90,4 - to 115,2 mg/L). A fungus was isolated from the following four blood samples taken for blood culture. Despite restarting treatment with the locally available antimycotic fluconazole (400 mg/d), fevers became more frequent, the patient's condition deteriorated and he died five days after the first fungi positive blood culture.

The microbiological assessment was made in the Microbiology laboratory of MHAT-Shumen. Microscopic slides were prepared from the first positive for fungi blood culture showing elongated spores and hyphae. Therefore, in addition to standard blood agar (HiMedia, India) and MacConkey (HiMedia, India) agar, cultures on Sabouraud (BioLab, Hungary) and Chrom (HiMedia, India) agar were prepared. The same was done for the subsequent three blood cultures as well. After twenty-four hours, white colonies with a smooth surface and a creamy consistency were observed on Sabouraud agar, but forty-eight hours later they increased in size and took on a rough appearance. Substrate mycelium became creamy to brown. On microscope slides prepared from the colonies rare septate hyphae and numerous arthroconidia as well as spores were observed.

After twenty-four hours on Chrom agar for Candida (HiMedia, India), the colonies were white, and forty-eight hours laterbthey became pale pink with a velvety surface. Upon subsequent cultivation they became grayish-purple inside with a dirty gray back. In a liquid medium, a wide film on the walls of the tube above the solution was presented. The urease test performed was positive. Identification with Integral System Yeasts (Liofilchem, Italy) was dubitous and no resistance to the antimycotic agents available in the test was observed. The culture was further examined with VITEK 2 YST automated identification system (BioMerieux, France). The result was *Cryptococcus laurentii* (93% probability), which was not a convincing result given the macroscopic and microscopic morphology .

After a pure culture of the same species from the last four consecutive blood samples was obtained, the isolate was sent according to the standard protocol to the National Reference Laboratory of Mycoses at the NCIPD, Sofia. We received a confirmatory result for *Trichosporon coremiiforme*, based on validated laboratory methods (MALDI-TOF, Biotyper Bruker).

The isolate was tested for susceptibility to antimycotic agents. The minimum inhibitory concentration (MIC, Himedia, India) values were as follows: Voriconazole - 0.06 mcg/ml, Posaconazole and Itraconazole - 0.25

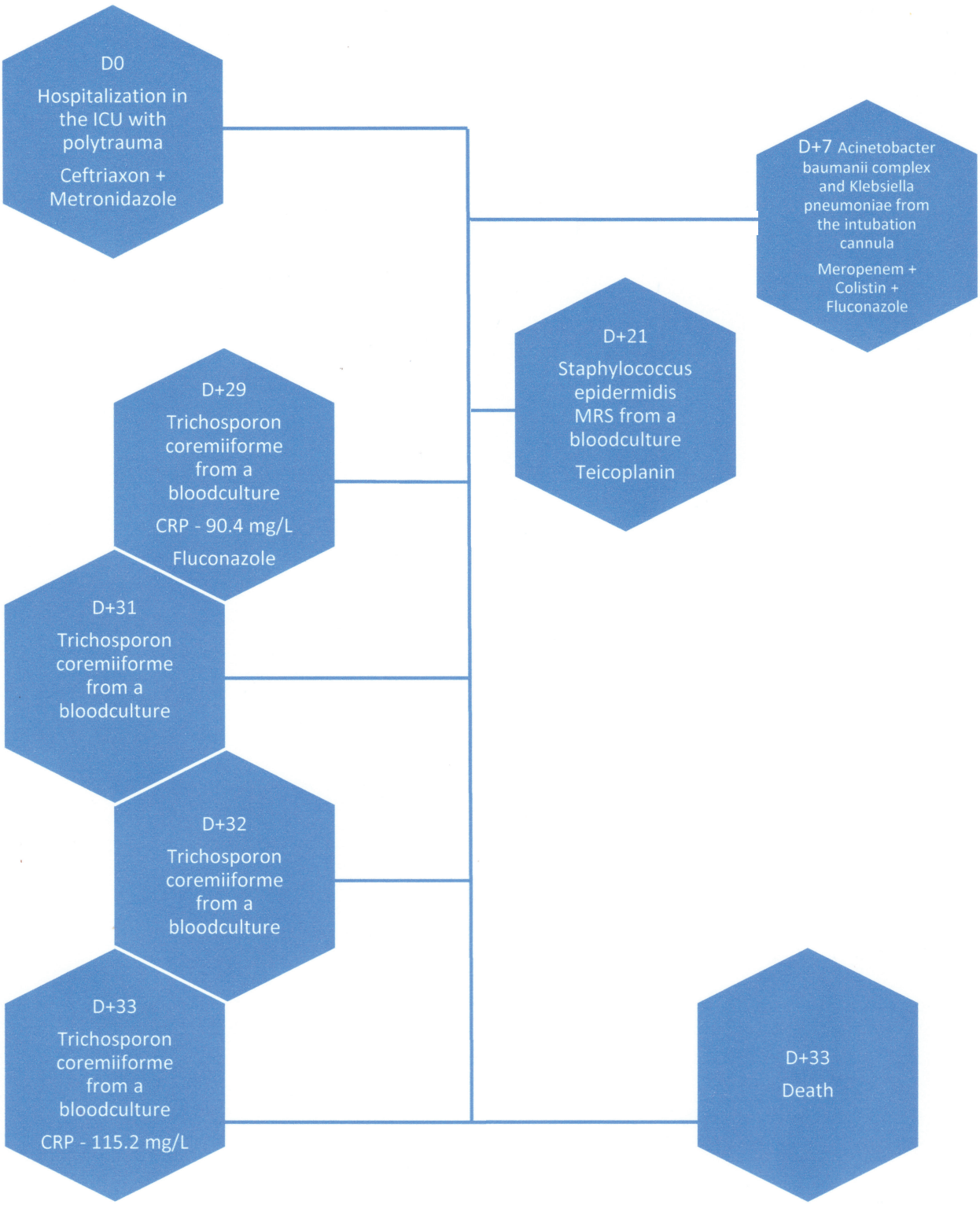

Figure 1. Case progression – schematic presentation.

mcg/ml, Fluconazole - 4 mcg/ml, Caspofungin - 4 mcg/ml, Micafungin and Anidulafungin - 8mcg/ml, Amphotericin B – 4mcg/ml. There are no available criteria for interpretation for this species. The Sanford Guide recommendations for invasive infection are Voriconazole as primary regimen, and Amphotericin B or Posaconazole as alternative regimens. [9]

DNA sequencing of the ITS region was performed as previously described. [10] The DNA sequence of the isolate was 100% identical with the type strain *T. coremiiforme* CBS2482.

The isolate was deposited at the National Center of Infectious and Parasitic Diseases culture collection as *T. coremiiforme* 454-23 and the DNA sequence

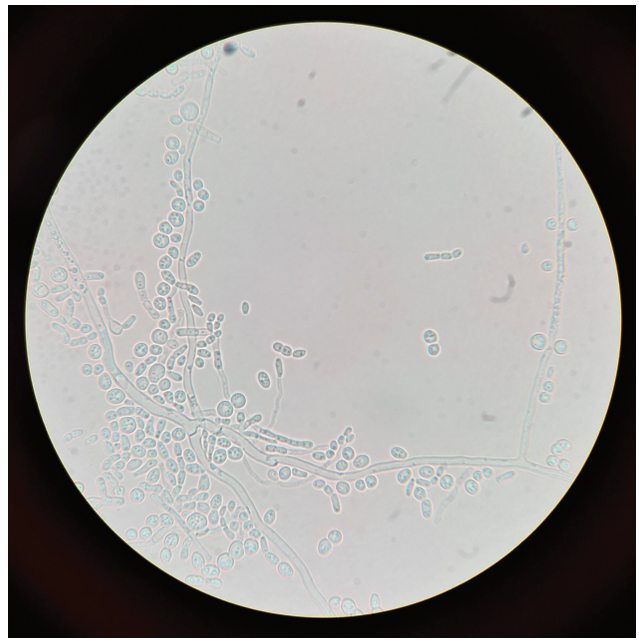

Figure 2. *Trichosporon coremiiforme*: Colony morphology on Sabouraud agar (1); Arthroconidia (2)

was submitted to Genbank (accession number: PP455503).

DISCUSSION

Hospitalization of patients in ICU is a risk factor for occurrence of mycotic sepsis. [11] Those are usually, polymorbid or trauma patients in a critical general condition who are subjected to broad-spectrum antibacterial therapy, intubation, catheterization and other invasive procedures. Some of them require a long corticosteroid treatment, which also increases the risk of mycotic sepsis. The most common causative agent of invasive mycotic infections are Candida spp., but given the specific therapy and high mortality from mycotic sepsis, microbiologists should consider a wider range of fungi. Despite the availability of new and diverse automated methods for accurate pathogen species identification, microscopic examination and other routine microbiology methods retain their importance for adequate therapy choice.

In our case, species identification was not possible locally. As for *Trichosporon coremiiforme*, this would not have been possible without the use of MALDI-TOF MS technique and DNA sequencing due to its high similarity to *Trichosporon asachii*. There is a limited number of publications on *Trichosporon coremiiforme* as a causative agent of invasive infections. One possible reason is its misidentification as *Trichosporon asachii*. At the same time, due to advances in treatment, the survival rate of immunocompromised patients increases, which is a prerequisite for an increasing number of opportunistic infections. With the availability of modern identification methods, the knowledge about *Trichosporon coremiiforme* and its susceptibility to antimycotic agents is expected to expand, to which our clinical case aims to contribute.

REFERENCES

1. Guo L-N, Yu S-Y, Hsueh P-R, Al-Hatmi AMS, Meis JF, Hagen F, Xiao M, Wang H, Barresi C, Zhou M-L, de Hoog GS, Xu Y-C. 2019. Invasive infections due to Trichosporon: species distribution, genotyping, and antifungal susceptibilities from a multicenter study in China. J Clin Microbiol 57:e01505-18. https://doi.org/10.1128/JCM.01505-18
2. Montoya AM, Gonzбlez GM. 2013. Trichosporon spp.: an emerging fungal pathogen. Medicina Universitaria 2014;16(62):37-43
3. Mehta V, Nayyar C, Gulati N, Singla N, Rai S, Chandar J. A Comprehensive Review of Trichosporon spp.: An Invasive and Emerging Fungus. Cureus. 2021 Aug 21;13(8):e17345. PMID: 34567886; PMCID: PMC8451254. https://doi.org/10.7759/cureus.17345
4. Francisco EC, de Almeida Junior JN, de Queiroz Telles F, Aquino VR, Mendes AVA., de Andrade Barberino MGM, de Tarso O. Castro P, Guimar~aes T, Hahn RC, Padovan ACB, Chaves GM, Colombo AL. 2018. Species distribution and antifungal susceptibility of 358 Trichosporon clinical isolates collected in 24 medical centres. Clinical Microbiology and Infection 25 (2019) 909.e1e909.e5 https://doi.org/10.1016/j.cmi.2019.03.026
5. Duarte-Oliveira C, Rodrigues F, Gonçalves SM, Goldman GH, Carvalho A, Cunha C. The Cell Biology of the Trichosporon-Host Interaction. Front Cell Infect Microbiol. 2017 Apr 7;7:118. PMID: 28439501; PMCID: PMC5383668. https://doi.org/10.3389/fcimb.2017.00118
6. Shamshuzoha M, Menon I, Sowjanya H. A rare case of fatal sepsis caused by Trichosporon asahii in an immunocompetent chronic obstructive pulmonary disease patient. Indian J Case Reports. 2022;8(10):325-327. https://doi.org/10.32677/ijcr.v8i10.3649
7. Chagas-Neto TC, Chaves GM, Melo ASA, Colombo AL - 2008. Bloodstream Infections Due to Trichosporon spp.: Species Distribution, Trichosporon asahii Genotypes Determined on the Basis of Ribosomal DNA Intergenic Spacer 1 Sequencing, and Antifungal Susceptibility Testing. JOURNAL OF CLINICAL MICROBIOLOGY, Apr. 2009, p. 1074-1081 https://doi.org/10.1128/JCM.01614-08
8. Monzani V, Cordoba S, Vivot M, Arias B, Vivot W, Szusz W, Castellaro P, Appendino A, Taverna CG, A case report of catheter-related bloodstream infection due to Trichosporon coremiiforme in a patient with secondary neutropenia to HIV, Journal de Mycologie Medicale (2019) https://doi.org/10.1016/j.mycmed.2019.100916
9. https://www.sanfordguide.com/
10. Kouzmanov, A., Philipova, I., Ivanova, Z., Ivanov, I., & Kantardjiev, V. (2016). DNA sequencing of the ITS region for rapid and accurate identification of medically important fungi. Problems of Infectious and Parasitic Diseases, 44(2), 19-22.
11. Muskett H, Shahin J, Eyres G, Harvey S, Rowan K, Harrison D. Risk factors for invasive fungal disease in critically ill adult patients: a systematic review. Crit Care. 2011;15(6):R287. doi: 10.1186/cc10574. Epub 2011 Nov 29. PMID: 22126425; PMCID: PMC3388661. https://doi.org/10.1186/cc10574