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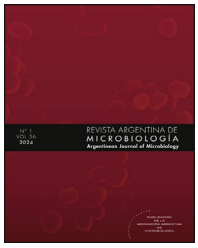

BRIEF REPORT

First documented isolation of *Diutina mesorugosa* (*Candida mesorugosa*) from a clinical sample in Argentina

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Abstract Recent advances in molecular phylogenetic and revisions to the International Code of Nomenclature have led to major changes in the fungal taxonomy. Genus *Candida* has been subject to extensive reclassification, with ribosomal DNA analysis demonstrating that *Candida rugosa* represents a cryptic species complex. Consequently, these taxa were reassigned to the novel genus *Diutina*. The *Diutina rugosa* complex includes species morphologically and physiologically indistinguishable: *D. rugosa*, *D. mesorugosa*, *D. neorugosa*, and *D. pseudorugosa*. We report the first case of *D. mesorugosa* diagnosed in Argentina in a 51-year-old man patient with complete spinal cord syndrome who developed osteomyelitis associated with sacrogluteal pressure ulcers. The yeast was isolated from bone tissue alongside ESBL-producing *Proteus mirabilis*. Fungal identification was confirmed using ribosomal DNA sequencing. *D. mesorugosa* is an emerging opportunistic pathogen that affects immunocompromised individuals. The patient responded favorably to a three-month course of oral fluconazole and combination antibiotic therapy (meropenem/ertapenem) for the bacterial co-infection.

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PALABRAS CLAVE
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Aislamiento de *Diutina mesorugosa* (*Candida mesorugosa*) en un material clínico. Primer caso informado en Argentina

Resumen La taxonomía fúngica ha cambiado significativamente por el uso de las herramientas moleculares y las actualizaciones del Código Internacional de Nomenclatura. Estudios de ADN ribosomal revelaron que *Candida rugosa* es un complejo de especies, reubicadas en el nuevo género *Diutina*. El complejo *Diutina rugosa* incluye especies morfológica y fisiológicamente indistinguibles: *Diutina rugosa*, *Diutina mesorugosa*, *Diutina neorugosa* y *Diutina seudorugosa*. Se describe el primer caso de *D. mesorugosa* en Argentina, diagnosticado en un paciente masculino de 51 años con síndrome medular completo por fractura vertebral, que desarrolló osteomielitis asociada a escaras sacro-glúteas. Se aisló la levadura del material óseo, junto con *Proteus mirabilis* BLEE. La identificación fúngica se confirmó mediante secuenciación del ADN ribosomal. *D. mesorugosa*, patógeno emergente en individuos inmunocomprometidos, debe ser considerado entre los posibles diagnósticos diferenciales en este grupo de pacientes. El enfermo fue tratado con fluconazol oral por tres meses y meropenem/ertapenem para la infección bacteriana concomitante, con evolución favorable.

The taxonomy and nomenclature of fungal species have undergone substantial revisions, primarily due to the incorporation of molecular phylogenetic tools in classificatory schemes and recent modifications to the International Code of Nomenclature (ICN) for algae, fungi, and plants. In particular, the genus *Candida* has been subject to numerous species reclassifications, with some species occasionally being placed in other or newly established genera. Molecular studies based on ribosomal DNA sequencing have enabled the distinction of cryptic species phylogenetically related to *Candida rugosa*, determining that *C. rugosa* actually represented a species complex comprising C. *rugosa*, *Candida mesorugosa*, *Candida neorugosa* and *Candida pseudorugosa*[1,5,10]. Subsequently, these species were transferred to the genus *Diutina*[3,7]. The complex subsequently included *Diutina rugosa sensu stricto*, *Diutina mesorugosa*, *Diutina neorugosa*, and *Diutina pseudorugosa*. These species are closely related to each other, and are morphologically and physiologically indistinguishable[9].

Although this complex is recognized as a pathogen in veterinary medicine[13], it is currently among the emerging human pathogens that must be considered in Latin America[6,12]. Infections caused by species in this complex are associated with immunosuppressed patients, particularly those undergoing prolonged hospitalizations, using intravenous catheters, or exhibiting other comorbidities[4,15]. The presence of virulence factors, such as aspartyl protease enzymes, phospholipases, esterases, DNAses, and hemolysins has been documented. These species have also demonstrated the ability to form biofilms[8].

Species of the *Diutina rugosa* complex can be found as colonizers in high-risk patients, comprising 0.6% of yeast isolates from clinical samples worldwide and 2.7% in Latin America[11]. In Argentina, there is limited epidemiological data on these cryptic species, as they are rarely isolated. Therefore, the successful identification of emerging pathogenic species is crucial for understanding regional epidemiology and informing therapeutic management strategies. Special emphasis must be placed on their accurate identification, particularly in reference laboratories, to ensure that these pathogens are not underdiagnosed or overlooked. Nevertheless, the technological infrastructure and costs associated with molecular fungal identification techniques pose significant barriers to their adoption as standard procedures in laboratories with limited resources. To our knowledge, this represents the first documented case of *D. mesorugosa* infection in Argentina.

A 51-year-old patient from Rosario – Argentina, with complete spinal cord syndrome secondary to a spinal cord injury from a fracture of the twelfth thoracic vertebra (T12) of traumatic etiology in the context of a fall from height, with a two year-course of history of the disease, was studied. The patient had a history of multiple hospitalizations for pressure ulcer infections treated with repeated antibiotic courses. Owing to his underlying condition, he received home care.

However, the development of skin and soft tissue infection and osteomyelitis associated with sacral-gluteal pressure ulcers required hospitalization. Following informed consent, surgical debridement, escharectomy, and bone resection of the affected region were performed. Samples of blood cultures, urine, pressure ulcer material, and bone material were sent to the bacteriology laboratory for analysis.

Blood was inoculated into BacT/Alert FA PLUS bottles (BioMérieux Marcy-l'Étoile®) and incubated in the BACT/ALERT® blood culture system. The culture results were negative. The urine sample was plated on chromogenic CPS medium (BioMérieux®) and blood agar (BA), while the escharectomy and bone samples were plated on cysteine-lactose-deficient in electrolytes (CLDE), chocolate agar (ChA), BA and thioglycollate enrichment broth (TG). After 24 h of incubation, the routine protocols for aerobic and anaerobic organisms were followed.

Microscopic examination of both urine and bone specimen cultures revealed Gram-negative bacilli.

Proteus mirabilis producing extended-spectrum beta-lactamase (ESBL) was isolated from both bone samples and urine cultures. Yeast development was also observed in the bone material cultured on BA, which was referred to the CEREMIC (Mycology Reference Center: Centro de Referencia de Micología) laboratory for species identification. The isolates were plated on Sabouraud glucose and Sabouraud glucose chloramphenicol. After 24 h of growth, rugose, yeast-like colonies were recovered. These were subcultured on CHROMagar *Candida*®, where they developed green-coloured colonies. The micromorphological analysis on milk agar-Tween medium revealed the presence of pseudomycelium with lateral branching, without chlamydoconidia, and with ovoid blastoconidia. Identification was performed using the commercial Vitek2® system (BioMérieux®) with YST tablets, resulting in a profile identified as *D. rugosa* with 99% probability (Bionumber 6102124040025551). Antifungal susceptibility obtained by the same method using Vitek2® AST-YS08 cards resulted in a minimum inhibitory concentration (MIC) of 2 μg/ml against fluconazole, while for voriconazole, caspofungin and amphotericin B, MIC of 0.12 μg/ml, 0.25 μg/ml and 1 μg/ml were obtained, respectively. Clinical breakpoints have not been established for species of the *D. rugosa* complex; therefore, it was not possible to determine the degree of susceptibility to the antifungals tested. However, for comparative purposes, using CLSI breakpoints for *Candida albicans*, the MIC values classified the yeast as susceptible to fluconazole, voriconazole, and caspofungin.

Given that *D. rugosa* is an emerging pathogen and part of a species complex that is physiologically indistinguishable, ribosomal DNA sequencing was performed to identify the species. ITS regions and the D1/D2 domain of 26S rDNA were sequenced. Primers ITS1/ITS4 and NL1/NL4 were used[1,10]. The sequences were analyzed using the NCBI BLAST tool, and both sequences showed 99% identity with the type strain *D. mesorugosa* CBS12656 (GenBank accession numbers PV553763 and PV553764).

Antimicrobial therapy for the ESBL-producing *P. mirabilis* consisted of intravenous meropenem for 10 days, followed by intramuscular ertapenem for at least six weeks. Empirical antibiotic treatment with vancomycin was also administered for 14 days post-surgery. For *D. mesorugosa* infection, oral fluconazole was prescribed at a dose of 400 mg/day for three months, with follow-up consultations to evaluate the evolution of the infection. The patient was discharged with antibiotic and antifungal treatment and all the hygienic-dietary measures recommended for medullary syndrome, with follow-up every 15 days.

Multicenter surveillance studies in Argentina have shown that more than 50% of yeast-related bloodstream infections are caused by species other than *Candida albicans*[14]. Since yeasts of the *D. rugosa* complex have been reported to have decreased susceptibility to azoles and amphotericin B[2,6,11], correct identification and antifungal susceptibility testing are crucial for effective therapy, preventing unnecessary drug exposure in patients with multiple comorbidities. The patient in this study successfully resolved the diagnosed infections, mainly thanks to adherence to treatment and regular medical monitoring.

It is of paramount importance to obtain regional data to understand and update the epidemiology of emerging pathogenic yeasts. This information is vital for healthcare teams to provide comprehensive patient care. In this case, the accurate diagnosis of the infectious etiologies was fundamental. Since patients with spinal cord syndrome often require frequent hospitalizations and multiple pharmacological therapies, the early and appropriate implementation of antimicrobial therapy is of utmost relevance for a positive patient outcome.

This research underwent ethical review by the Institutional Review Board of Hospital Provincial del Centenario (Comité de Ética de la Investigacion del Hospital Provincial del Centenario) – Register number7876/25 and Bioethics Committee. Faculty of Biochemical and Pharmaceutical Sciences, National University of Rosario [Comisión de Bioética, Facultad de Ciencias Bioquímicas y Farmacéuticas, Universidad Nacional de Rosario]. Identification Code 24996/2025.

Ethical considerations

This work does not involve animal experimentation and does not represent a clinical trial; as this is a patient case study, it was submitted for ethical review by Institutional Review Board of Hospital Provincial del Centenario (Comité de Ética de la Investigación del Hospital Provincial del Centenario) – Register number7876/25, and Bioethics Committee. Faculty of Biochemical and Pharmaceutical Sciences, National University of Rosario (Comisión de Bioética, Facultad de Ciencias Bioquímicas y Farmacéuticas, Universidad Nacional de Rosario). Identification Code 24996/2025.

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Conflict of interest

The authors declare no conflicts of interest that would preclude this publication.

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