



**HAL**  
open science

## Persistence of clonal azole-resistant isolates of *Candida albicans* from a patient with chronic mucocutaneous candidiasis in Colombia

Andrés Ceballos-Garzón, Luz M Wintaco-Martínez, Norida Velez, Catalina Hernandez-Padilla, Alejandro De la Hoz, Sandra Liliana Valderrama-Beltrán, Carlos Alvarez-Moreno, Patrice Le Pape, Juan David Ramírez, Claudia Marcela Parra-Giraldo

### ► To cite this version:

Andrés Ceballos-Garzón, Luz M Wintaco-Martínez, Norida Velez, Catalina Hernandez-Padilla, Alejandro De la Hoz, et al.. Persistence of clonal azole-resistant isolates of *Candida albicans* from a patient with chronic mucocutaneous candidiasis in Colombia. *Journal of Global Infectious Diseases*, 2020, 12, pp.16-20. 10.4103/jgid.jgid\_74\_19 . hal-04719445

**HAL Id: hal-04719445**

**<https://hal.science/hal-04719445v1>**

Submitted on 3 Oct 2024

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



Distributed under a Creative Commons Attribution - NonCommercial - ShareAlike 4.0 International License

# Persistence of Clonal Azole-Resistant Isolates of *Candida albicans* from a Patient with Chronic Mucocutaneous Candidiasis in Colombia

Andrés Ceballos-Garzón<sup>1,2,3</sup>, Luz M Wintaco-Martínez<sup>4</sup>, Norida Velez<sup>1</sup>, Catalina Hernandez-Padilla<sup>2</sup>, Alejandro De la Hoz<sup>2</sup>, Sandra Lilibiana Valderrama-Beltrán<sup>2</sup>, Carlos A Alvarez-Moreno<sup>2</sup>, Patrice Le Pape<sup>3</sup>, Juan David Ramírez<sup>4</sup>, Claudia M Parra-Giraldo<sup>1,2</sup>

<sup>1</sup>Unidad de Proteómica y Micosis Humanas, Grupo de Enfermedades Infecciosas Departamento de Microbiología, Facultad de Ciencias, Pontificia Universidad Javeriana, <sup>2</sup>Grupo de Investigación en Enfermedades Infecciosas, Unidad de Infectología, Hospital Universitario San Ignacio, Pontificia Universidad Javeriana, Facultad de Medicina, Bogotá, Colombia, <sup>3</sup>Department of Parasitology and Medical Mycology, Faculty of Pharmacy, University of Nantes, Nantes Atlantique Universities, Nantes, France, <sup>4</sup>Grupo de Investigaciones Microbiológicas-UR, Programa de Biología, Facultad de Ciencias Naturales y Matemáticas, Universidad del Rosario, Bogotá DC, Colombia

## Abstract

**Purpose:** The present article describes retrospectively a case of a patient with chronic mucocutaneous candidiasis (CMC) who presented recurrent *Candida albicans* infection since he was 6 months old. We obtained 16 isolates recovered during a 4-year period. Our purpose was to determinate the susceptibility, genotyping, and the pathogenicity profile in all the isolates. **Methods:** Sixteen *C. albicans* were isolated from a 25-year-old male with several recurrent fungal infections admitted to Hospital. The isolates were recovered during 4 years from a different anatomical origin. We typed them by multilocus sequence typing, also we evaluated susceptibility to fluconazole, itraconazole, voriconazole, posaconazole, isavuconazole, caspofungin, and amphotericin B by microdilution method and we also test the pathogenic capacity in the *Galleria mellonella* model. **Results:** Genotyping of all clinical isolates showed the persistence of the same diploid sequence type (DST). Isolates changed their susceptibility profile over time, but there were no significant statistical differences in pathogenicity. **Conclusion:** Herein, a persistent clonal isolates of *C. albicans* (DST 918) in a patient with CMC, showed changes in its susceptibility profile after several antifungal treatments acquiring gradual resistance to the azole drugs, which did not affect their pathogenicity.

**Keywords:** *Candida albicans*, chronic mucocutaneous candidiasis, drug susceptibility, *Galleria mellonella*, multilocus sequence typing

## INTRODUCTION

Chronic mucocutaneous candidiasis (CMC) is a persistent and recurrent infection mostly caused by the opportunistic pathogen *Candida albicans*. The most common lesions include whitish plaques associated to crusts and ulcers on the oral mucosa. This clinical entity starts during infancy for most of the patients. Defect associates with *Candida* recognition (e.g. CARD9 and Dectin 1), T helper 17 (Th17) differentiation (e.g. *STAT1* and *STAT3* mutations), and cytokine 17 (IL-17) signaling (e.g. anti-IL17 autoantibodies and TRAF3IP2 mutation) have been described.<sup>[1-3]</sup> CMC is frequently associated with inherited or acquired T-cell deficiencies such as autoimmune polyendocrine syndrome type 1 (APS-1), autosomal-dominant hyper IgE syndrome, or HIV infection which leads to disruption of the effective mucosal response.<sup>[4-6]</sup> Patients with APS-I have a

loss of function in the *AIRE* gene which results in high titers of autoantibodies against Th17. Patients with hyper IgE syndrome have a loss of function by *STAT3* gene mutations, which results in a reduction of Th17 cell number and impaired cytokine production IL-17, as also occurs in *STAT1* mutation;

**Address for correspondence:** Dr. Claudia M Parra-Giraldo, Director Unidad De Proteómica Y Micosis Humanas, Grupo de Enfermedades Infecciosas, Departamento de Microbiología, Facultad de Ciencias, Pontificia Universidad Javeriana Carrera7 #43-82 Ed52 Lab 404, Bogotá, Colombia.  
E-mail: claudia.parra@javeriana.edu.co

This is an open access journal, and articles are distributed under the terms of the Creative Commons Attribution-NonCommercial-ShareAlike 4.0 License, which allows others to remix, tweak, and build upon the work non-commercially, as long as appropriate credit is given and the new creations are licensed under the identical terms.

**For reprints contact:** reprints@medknow.com

**How to cite this article:** Ceballos-Garzón A, Wintaco-Martínez LM, Velez N, Hernandez-Padilla C, De la Hoz A, Valderrama-Beltrán SL, et al. Persistence of clonal azole-resistant isolates of *Candida albicans* from a patient with chronic mucocutaneous candidiasis in Colombia. J Global Infect Dis 2020;12:16-20.

**Received:** 27 June 2019 **Revised:** 11 September 2019

**Accepted:** 31 October 2019 **Published:** 19 February 2020

### Access this article online

#### Quick Response Code:



**Website:**  
www.jgid.org

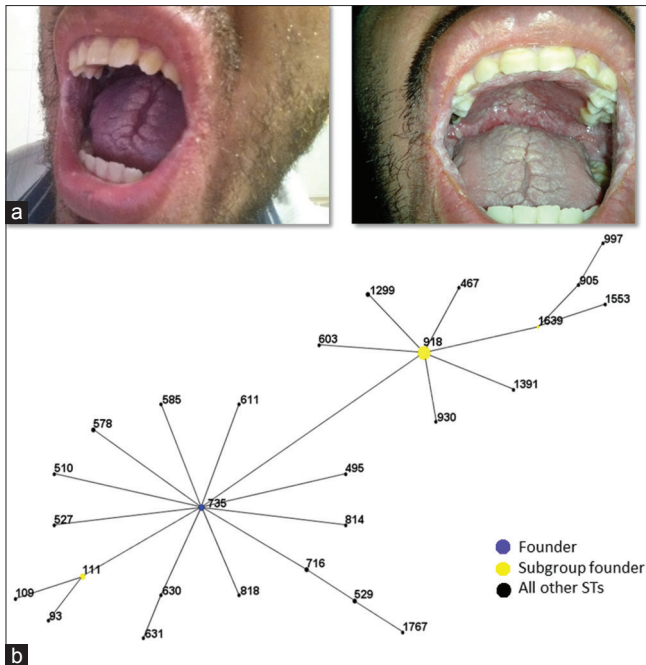
**DOI:**  
10.4103/jgid.jgid\_74\_19

*STAT1* gain-of-function mutations are associated with a variety of phenotypes including CMC, fungal infections other than candidiasis, bacterial and viral infections, mycobacterial infections, autoimmune disorders, and carcinomas and aneurysms.<sup>[2,7]</sup> *STAT1* is identified in approximately 40% of patients with CMC.<sup>[8]</sup>

For the treatment of chronic infections caused by *C. albicans*, azoles are considered the mainstay of therapy, in particular fluconazole because of high bioavailability after oral administration.<sup>[9]</sup> Although treatment with azoles can be effective, long-term use of fluconazole has led to the emergence of *Candida* strains with decreased susceptibility.<sup>[7]</sup> The present article describes retrospectively a case of a patient with CMC who presented with recurrent *C. albicans* infection since he was 6 months old. Susceptibility testing was performed for all the *C. albicans* isolates recovered during a 4-year period. Genotyping was carried out using the multilocus sequence typing (MLST) methodology to identify the population structure.

**Case description:** A 25-year-old male was admitted to the Hospital in February 2012 with history of recurrent fungal infections of the oral cavity and bacterial infections of the skin and respiratory tract. He was treated intermittently as an outpatient with fluconazole and nystatin for 20 years. On admission physical examination, white cotton-like plaques

widely distributed in the oral cavity, hypertrophic mucosa, cheilitis [Figure 1a], and signs of onychomycosis were observed. Fluconazole was switched to itraconazole, leading to resolution of the lesions within 2 months. However, he had a new relapse 3 months later. A palate biopsy was performed, and cultures were done because an infection by a different yeast was suspected. The susceptibility tests showed fluconazole-, itraconazole-, and voriconazole-resistant *C. albicans* isolates. Posaconazole was started without improvement; therefore, caspofungin was indicated for 14 days, but there was no recovery. The infectious diseases medical board decided to initiate amphotericin B deoxycholate (1 mg/kg/day) for 14 days upon treatment failure with echinocandins. Amphotericin B was continued as suppressive therapy for 2 months with slight improvement but no remission. Caspofungin alone and in combination with posaconazole and amphotericin B were given subsequently with partial improvement. In a different institution, genetic studies for CMC were made, revealing the *STAT-1* mutation and confirming of immunocompromised related with diagnosis of CMC. Most antifungal therapies were empirical because at that time, the susceptibility profile of the isolates was not available. During a 4-year period, since his admission, samples from different lesions were cultured and the 16 recovered isolates were identified as *C. albicans* by matrix-assisted laser desorption/ionization-time of flight mass spectrometry (MALDI-TOF MS). Retrospectively antifungal susceptibility testing was performed using the Clinical and Laboratory Standards Institute broth microdilution method (CLSI-BMD) [Figure 2b].



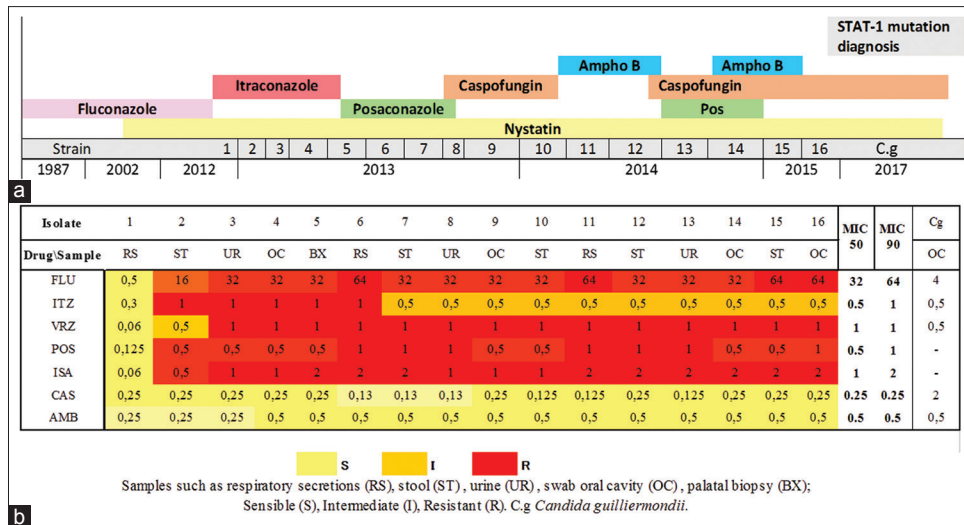
**Figure 1:** Patient's lesion and eBURST diagram. (a) Lesions of chronic mucocutaneous candidiasis patient: White cotton-like plaques widely distributed in the oral cavity, hypertrophic mucosa, and cheilitis. (b) The illustration shows the figure generated by eBURST 3.0 software, in which diploid sequence types are linked when they differ in the sequence of just one of the seven gene fragments used for MLST. The lengths of lines are not significant. The lines indicate differences between diploid sequence types that result from a loss of heterozygosity from the putative parent to the putative-derived isolate or from mutation, gene exchange, or another mechanism

## METHODS

A 4-year (2012–2016) retrospective study was conducted to recover yeast isolates in a CMC patient admitted to the Hospital.

### Culture and identification

The samples were seeded on CHROMagar *Candida* plates (CHROMagar *Candida*, France) and Sabouraud dextrose agar (SDA, Merck, Germany). The cultures were incubated at 35°C for 36 h, and the identification of yeast was performed by MALDI-TOF MS. Protein extraction was performed with the formic acid/ethanol method, according to the Bruker Daltonics protocol. Briefly, two or three colonies were mixed with 300 µl of ultrapure water until homogenization and 600 µl of 100% ethanol was added (Sigma-Aldrich, St. Louis, MO). After centrifugation at 15,000 g for 2 min, the pellet was dried at 25°C and reconstituted in equal volumes of 100% formic acid (Sigma-Aldrich) and acetonitrile (Sigma-Aldrich) (in 20 µl volumes), mixed all right. A microliter of supernatant was placed in a 96-point steel plate (Bruker Daltonik) and allowed to dry at room temperature before the addition of 1 µl of the  $\alpha$ -cyano-4-hydroxycinnamic acid matrix. Each sample was tested in duplicate. A score was obtained according to the manufacturer's technical specifications as follows: correct genus and species



**Figure 2:** Characteristics of *Candida* spp. isolates. (a) Antifungal treatment over time. (b) Antifungal susceptibility results

identification ( $\geq 2.0$ ), secure genus identification (1.7–2.0), and no reliable identification ( $< 1.7$ ).

### Multilocus sequence typing

*C. albicans* isolates were typed using MLST with the internal regions of seven housekeeping genes (*AAT1a*, *ACCI*, *ADPI*, *MPIb*, *SYA1*, *VPS13*, and *ZWF1b*) as described previously.<sup>[10,11]</sup> Sequencing was performed on both strands using an ABI Prism 3130 xl genetic analyzer (Applied Biosystems, Foster City, CA, USA). All data were entered into the MLST online database (<http://calbicans.mlst.net>). For the analysis, we used the algorithm eBURST which divides an MLST data set of any size into groups of related isolates and clonal complexes, predicts the founding (ancestral) genotype of each clonal complex, and computes the bootstrap support for the assignment.<sup>[12]</sup>

### Antifungal susceptibility testing

Antifungal susceptibility testing was carried out using the (CLSI-BMD), following the M27-A3 guidelines<sup>[13]</sup> for triazoles (fluconazole (FLU), itraconazole (ITZ), voriconazole (VRZ), posaconazole (POS), and isavuconazole (ISA)), caspofungin (CAS), and amphotericin B (AMB). The minimum inhibitory concentrations (MICs) were visually determined as the lowest concentration of drug that caused a significant diminution (MIC – 0 or 0% amphotericin B/MIC – 2 or  $\geq 50\%$  others) compared with that of the drug-free growth control after 24 h of incubation for caspofungin and amphotericin B, and after 48 h for triazoles. Quality control was ensured by testing the CLSI-recommended strains *Candida parapsilosis* ATCC 22019 and *Candida krusei* ATCC 6258.<sup>[14]</sup>

### Invertebrate *Galleria mellonella* model

Killing assays were performed in *Galleria mellonella* as described by Fallon, 2012.<sup>[15]</sup> Briefly, larvae of late stages (fifth and sixth) between 250 and 330 mg and a length of approximately 2 cm were selected. A group of ten larvae was used for each of the controls: absolute control,

disinfection, and inoculation. To compare mortality, three biological replicates were performed with ten larvae for each isolate evaluated. The strain *C. albicans* SC5314 was used as a control of pathogenicity. The isolates were grown in Sabouraud dextrose agar and incubated for 48 h at 27°C. Suspensions, adjusted to  $3.10^5$  UFC/ml using Neubauer chamber, were used to inoculate ten larvae per isolate, each with 10  $\mu$ l of inoculum by injection into the last left proto-leg using a 0.5 ml gauge insulin syringe. After inoculation, larvae were placed in Petri dishes and incubated in darkness at 37°C, and the number of dead larvae was recorded daily.<sup>[10]</sup> Survival analysis was performed using the Kaplan–Meier method in Graph Pad Prism version 5.0, Software Inc., La Jolla, CA, USA..

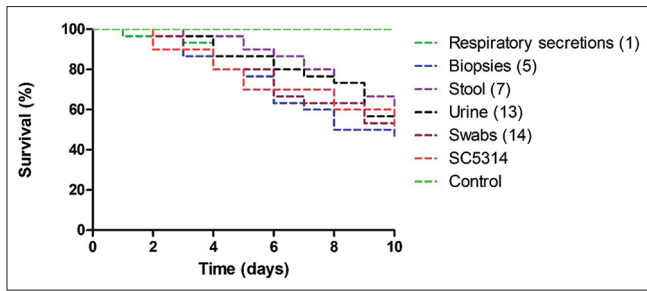
## RESULTS

### Culture and identification

The retrospective study allows us to recover 16 *C. albicans* isolates from different anatomical sites including oral cavity ( $n = 4$ ), respiratory secretions ( $n = 3$ ), stool ( $n = 5$ ), palate ( $n = 1$ ), and urine ( $n = 3$ ). A *Candida guilliermondii* was also isolated from the oral cavity in 2017.

### Multilocus sequence typing

The analysis identified the same diploid sequence type (DST) in all the 16 isolates of *C. albicans* recovered from the patient, which was the 62/3/3/3/3/39/95 allele profile that corresponds to the DST918 and clade 9. This indicates that these successive infectious episodes by *C. albicans* correspond to recurrences by the same DST and are not reinfections by different types of DSTs. Moreover, we employed 84 DSTs representative of clade 9 to which the previously identified DST 918 belongs. These DSTs, retrieved from the database, originated from different sources, including blood, oropharynx, oral cavity, and animals from France, USA, Australia, Guyana, Ireland, Kuwait, Israel, and Brazil reported in different years (2000, 2001, 2002, 2007, 2008, 2010, and 2013). To perform this analysis, we took a representative cluster of clade isolate



**Figure 3:** Survival curve of average according to anatomical origin. Kaplan–Meier plots of *Galleria mellonella* survival after infection with *Candida albicans*  $3.10^5$  UFC/ml. inside the brackets the number of samples

9 which evidence the cluster clone 1 based on founding genotype in blue circle (DST 735) as the putative founding type, subgroup founder yellow circle DST 918 to which the identified genotype belongs in the patient and 5 singleton DSTs are illustrated in Figure 1b.

### Antifungal susceptibility testing

Through determination of the resistance profiles of the 16 *C. albicans* and *C. guilliermondii* isolates [Figure 2b], we observed that the initial isolate 0218 PUJ/HUSI (Isolate 1) was susceptible to all antifungal tested, whereas the other isolates were susceptible to CAS and AMB but resistant or nonwild type to azoles. We found a minimal decrease in the MIC to itraconazole after it was stopped [Figure 2]. In contrast, the *C. guilliermondii* isolate exhibited susceptibility to all azoles tested.

### Invertebrate *Galleria mellonella* model

We compared the pathogenicity of 16 *C. albicans* isolates in a systemic infection model using the insect larval *G. mellonella* model. The concentration of  $3.10^5$  UFC/ml per larva did not exhibit significant differences in the 10 days of tracking ( $P = 0.2483$ ) [Figure 3 and Supplement Figure 1].

## DISCUSSION

A complete phenotypic and genotypic characterization of *C. albicans* isolates associated to a complicated case of CMC was conducted in the 2012–2016 period. Samples collected over this a period of 4 years, allow us to identify the same DST918 which belongs to clade 9, being this DST widely reported worldwide. It is important to highlight that among the 22 isolates reported to date with the same DST, none had been previously associated with CMC (<https://pubmlst.org/calbicans/>).<sup>[16,17]</sup> *C. albicans* quickly adapts to new microenvironments, and our analysis shows that a clonal population with the same DST was stable over time and was present in all the sampled anatomic sites, whereas, the first three sequential isolates exhibited very different resistance profiles, no difference in pathogenicity or fitness was demonstrated in the invertebrate model *G. mellonella*. Interestingly, this clone was maintained through large periods, yet it did not disappear or was not replaced regardless of all the administered antifungals.

The Th17 cells are the most important source of IL 17, an important cytokine for defense against *Candida* that induce antimicrobial peptides production. The perturbation of this aspect can lead to overgrowths of *Candida* which may be damaging to the host.<sup>[3,18,19]</sup> Herein, we corroborate it with a patient who has a mutation in STAT1. It is well known that STAT1 is involved in tumor genesis and tumor suppression with reports of reduced levels in transformed cancer cells and poorer outcomes if it is absent. Patients with CMC have an increased risk of both oral and esophageal cancers, the majority developing squamous cell carcinomas of skin, larynx, and gastrointestinal tract.<sup>[21]</sup> To date, the patient from this study does not present anything related to cancer.

We believe that immunosuppression in the patient plays an important role in the maintenance of a clonal population in the tissue throughout the years, and granulomatous lesions classically described in the CMC clinical entity could explain the yeast persistence.<sup>[20]</sup> Some studies have reported successful outcomes of azole refractory CMC after treatment by amphotericin B or echinocandins. We highlight these results as the first evidence in Colombia and South America *Candida* isolates resistant to antifungal therapy remains a concern.<sup>[21]</sup> In the Toubiana series, resistance to at least one antifungal was observed in 39% of patients treated with long-term antifungal therapy and in 15% of patients treated intermittently, with azole resistance being the most common.<sup>[8]</sup>

The robust characterization of isolates was pivotal for understanding the outcome of our patient. The stability and persistence of *C. albicans* has been highlighted previously in MLST studies using isolates taken at serial time points in a range of clinical settings.<sup>[22]</sup> However, the MLST methodology has some limitations such as the fact that only seven regions of constitutive genes varying between 350 and 490 bp were analyzed. This can explain why isolates with identical DSTs may differ significantly between them, through large genomic rearrangements in other regions that are not included in the analyzed loci.<sup>[23]</sup> Hence, further follow-up and characterization of the isolates are needed to reach accurate conclusions. In addition, since the analyzed gene sequences are not related to genes implicated in antifungal resistance or pathogenicity, we must continue the search of new molecular markers, which allow associating the phenotypic and molecular characteristics with the clinical presentation of the disease.

Whereas, the first three sequential isolates exhibited very different resistance profiles; no difference in pathogenicity or fitness was demonstrated in the invertebrate model *G. mellonella*.

### Acknowledgments

We would like to thank the patient for giving his consent for this research, and the Colombian Science, Technology and Innovation Department (COLCIENCIAS) for sponsoring PhD training in Colombia, within the framework of the National Program for the Promoting Research Training and the mobility program ECOSNord Colombia-France.

## Research quality and ethics statement

The authors of this manuscript declare that this scientific work complies with reporting quality, formatting and reproducibility guidelines set forth by the EQUATOR Network. The authors also attest that this clinical investigation was determined not to require Institutional Review Board/Ethics Committee review. The authors obtained informed consent from the patient. The corresponding protocol/approval number is not applicable. We also certify that we have not plagiarized the contents in this submission and have done a Plagiarism Check.

## Declaration of patient consent

The authors certify that they have obtained all appropriate patient consent forms. In the form, the patient has given his consent for his images and other clinical information to be reported in the journal. The patient understand that name and initials will not be published and due efforts will be made to conceal identity, but anonymity cannot be guaranteed.

## Financial support and sponsorship

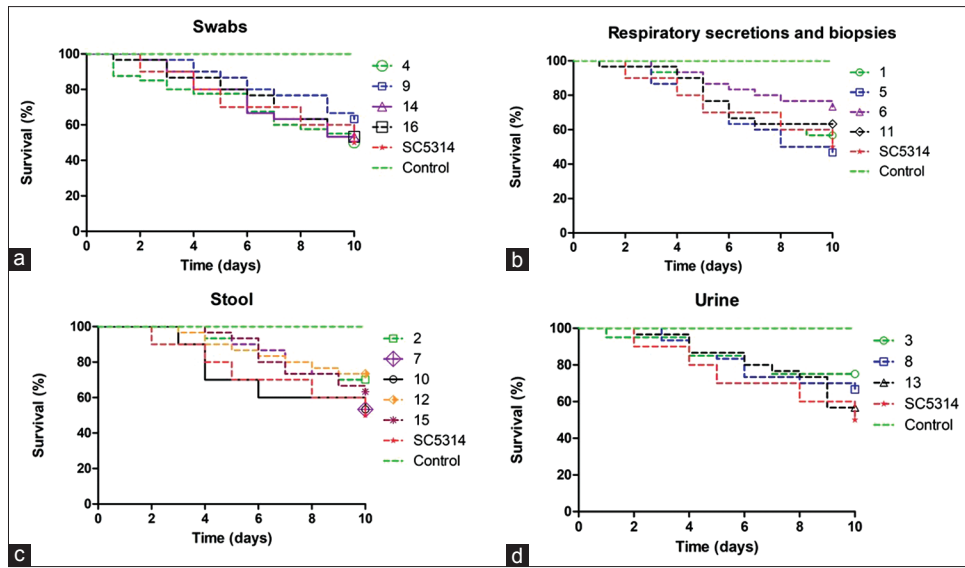
The Dirección de Investigación e Innovación, Universidad Del Rosario and vice rector of research at the Pontificia Universidad Javeriana Bogotá, Colombia, supported the research (grant no. 7710) and ECOS Nord mobility (Colombia-France).

## Conflicts of interest

There are no conflicts of interest.

## REFERENCES

- Milner JD, Holland SM. The cup runneth over: Lessons from the ever-expanding pool of primary immunodeficiency diseases. *Nat Rev Immunol* 2013;13:635-48.
- Carey B, Lambourne J, Porter S, Hodgson T. Chronic mucocutaneous candidiasis due to gain-of-function mutation in STAT1. *Oral Dis* 2019;25:684-92.
- Bhattad S, Dinakar C, Pinnamaraju H, Ganapathy A, Mannan A. Chronic mucocutaneous candidiasis in an adolescent boy due to a novel mutation in TRAF3IP2. *J Clin Immunol* 2019;39:596-9.
- Chapel H. Classification of primary immunodeficiency diseases by the International Union of Immunological Societies (IUIS) Expert Committee on Primary Immunodeficiency 2011. *Clin Exp Immunol* 2012;168:58-9.
- Okada S, Puel A, Casanova JL, Kobayashi M. Chronic mucocutaneous candidiasis disease associated with inborn errors of IL-17 immunity. *Clin Transl Immunology* 2016;5:e114.
- Moorhouse AJ, Rennison C, Raza M, Lilic D, Gow NA. Clonal strain persistence of *Candida albicans* isolates from chronic *Mucocutaneous candidiasis* patients. *PLoS One* 2016;11:e0145888.
- Humbert L, Cornu M, Proust-Lemoine E, Bayry J, Wemeau JL, Vantyghem MC, et al. Chronic *Mucocutaneous candidiasis* in autoimmune polyendocrine syndrome type 1. *Front Immunol* 2018;9:2570.
- Toubiana J, Okada S, Hiller J, Oleastro M, Lagos Gomez M, Aldave Becerra JC, et al. Heterozygous STAT1 gain-of-function mutations underlie an unexpectedly broad clinical phenotype. *Blood* 2016;127:3154-64.
- García-Cuesta C, Sarrion-Pérez MG, Bagán JV. Current treatment of oral candidiasis: A literature review. *J Clin Exp Dent* 2014;6:e576-82.
- Bougnoux ME, Aanensen DM, Morand S, Théraud M, Spratt BG, d'Enfert C. Multilocus sequence typing of *Candida albicans*: Strategies, data exchange and applications. *Infect Genet Evol* 2004;4:243-52.
- Bougnoux ME, Morand S, d'Enfert C. Usefulness of multilocus sequence typing for characterization of clinical isolates of *Candida albicans*. *J Clin Microbiol* 2002;40:1290-7.
- Feil EJ, Li BC, Aanensen DM, Hanage WP, Spratt BG. eBURST: Inferring patterns of evolutionary descent among clusters of related bacterial genotypes from multilocus sequence typing data. *J Bacteriol* 2004;186:1518-30.
- Clinical and Laboratory Standards Institute (CLSI), Reference Method for Broth Dilution Antifungal Susceptibility Testing of Yeasts; Third Informational Supplement. CLSI Document M27-S3. Wayne, Penn, USA: Clinical and Laboratory Standards Institute; 2008.
- M27-S4 Reference Method for Broth Dilution Antifungal Susceptibility Testing of Yeasts; Fourth Informational Supplement an Informational Supplement for Global Application Developed through the Clinical and Laboratory Standards Institute; 2012.
- Fallon J, Kelly J, Kavanagh K. *Galleria mellonella* as a model for fungal pathogenicity testing. In: Brand AC, MacCallum DM, editors. *Host-Fungus Interactions: Methods and Protocols*. Totowa, NJ: Humana Press; 2012. p. 469-85.
- Alastruey-Izquierdo A, Mandelblat M, Ben Ami R, Perlin DS, Segal E. Multilocus sequence typing of *Candida albicans* isolates from candidemia and superficial candidiasis in Israel. *Med Mycol* 2013;51:755-8.
- Jacobsen MD, Boekhout T, Odds FC. Multilocus sequence typing confirms synonymy but highlights differences between *Candida albicans* and *Candida stellatoidea*. *FEMS Yeast Res* 2008;8:764-70.
- Mengeshu BG, Conti HR. The role of IL-17 in protection against mucosal *Candida* infections. *J Fungi (Basel)* 2017;3 (4):52.
- Khalsa KK, Yang Q, Shen X, Pasha MA, Celestin J. Immunologic characterization of patients with chronic mucocutaneous candidiasis disease. *Clin Case Rep* 2019;7:180-5.
- Misme-Aucouturier B, Albassier M, Alvarez-Rueda N, Le Pape P. Specific human and *Candida* cellular interactions lead to controlled or persistent infection outcomes during granuloma-like formation. *Infect Immun* 2017;85:1.
- Brown GD, Denning DW, Gow NA, Levitz SM, Netea MG, White TC. Hidden killers: Human fungal infections. *Sci Transl Med* 2012;4:165rv13.
- Magri MM, Gomes-Gouvêa MS, de Freitas VL, Motta AL, Moretti ML, Shikanai-Yasuda MA. Multilocus sequence typing of *Candida tropicalis* shows the presence of different clonal clusters and fluconazole susceptibility profiles in sequential isolates from candidemia patients in Sao Paulo, Brazil. *J Clin Microbiol* 2013;51:268-77.
- Saghrouni F, Ben Abdeljelil J, Boukadida J, Ben Said M. Molecular methods for strain typing of *Candida albicans*: A review. *J Appl Microbiol* 2013;114:1559-74.



**Supplement Figure 1:** Survival curves of *Galleria mellonella* inoculated with *Candida albicans*, (a) swabs isolates, (b) respiratory secretions and biopsies isolates, (c) stool isolates, and (d) urine isolates