

Population genetics and microevolution of clinical *Candida glabrata* reveals recombinant sequence types and hyper-variation within mitochondrial genomes, virulence genes, and drug targets

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Abstract

Candida glabrata is the second most common etiological cause of worldwide systemic candidiasis in adult patients. Genome analysis of 68 isolates from 8 hospitals across Scotland, together with 83 global isolates, revealed insights into the population genetics and evolution of *C. glabrata*. Clinical isolates of *C. glabrata* from across Scotland are highly genetically diverse, including at least 19 separate sequence types that have been recovered previously in globally diverse locations, and 1 newly discovered sequence type. Several sequence types had evidence for ancestral recombination, suggesting transmission between distinct geographical regions has coincided with genetic exchange arising in new clades. Three isolates were missing MAT α 1, potentially representing a second mating type. Signatures of positive selection were identified in every sequence type including enrichment for epithelial adhesins thought to facilitate fungal adhesion to human epithelial cells. In patient microevolution was identified from 7 sets of recurrent cases of candidiasis, revealing an enrichment for nonsynonymous and frameshift indels in cell surface proteins. Microevolution within patients also affected epithelial adhesin genes, and several genes involved in drug resistance including the ergosterol synthesis gene *ERG4* and the echinocandin target *FKS1/2*, the latter coinciding with a marked drop in fluconazole minimum inhibitory concentration. In addition to nuclear genome diversity, the *C. glabrata* mitochondrial genome was particularly diverse, with reduced conserved sequence and conserved protein-encoding genes in all nonreference ST15 isolates. Together, this study highlights the genetic diversity within the *C. glabrata* population that may impact virulence and drug resistance, and 2 major mechanisms generating this diversity: microevolution and genetic exchange/recombination.

Keywords: *Candida glabrata*; genome sequencing; epidemiology; candidiasis; microevolution; mitochondria; drug-resistance; evolution

Introduction

Candida is the most prominent genus of the Debaryomycetaceae family, with over 400 genetically and phenotypically diverse species currently described (Daniel *et al.* 2014). Many of these species are harmless commensals of the mucous membranes and digestive tracts of healthy individuals. Approximately 30 *Candida* species are of clinical importance in humans. Most of these species that are capable of causing disease in humans belong to the CTG-Serine clade, including *Candida albicans*, *Candida dubliniensis*, *Candida tropicalis*, *Candida parapsilosis*, *Candida lusitanae*, *Candida guilliermondii*, and *Candida auris*, while others such as *Candida glabrata* and *Candida braccarum* belong to the genetically distant

Nakaseomyces clade (Daniel *et al.* 2014; Turner and Butler 2014). In adult patients, *C. glabrata* is the second most commonly isolated species after *C. albicans*, which together cause approximately 3 quarters of all systemic candidiasis (Perfrotth *et al.* 2007; Rajendran *et al.* 2018a). Infections caused by these species range from mild vulvovaginal candidiasis (VVC or thrush) to severe, drug resistant and difficult to treat invasive infections affecting single organs or the blood stream (candidemia) with or without dissemination to the heart, brain, kidneys, and other parts of the body (Mullick *et al.* 2006). Bloodstream infections caused by *Candida* spp. are associated with mortality rates of 30–60% (Hirano *et al.* 2015; Lamoth *et al.* 2018). Candidemia is associated

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Genetics Of Candida

Yi-Tong Ma



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