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**Virulence not required? Albumin promotes pathogenicity of (non)-damaging *Candida* strains**

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The pathogenicity of the dimorphic yeast *Candida albicans* is associated with filamentation, adhesion, invasion, and production of the toxin Candidalysin. However, there are certain clinical isolates and other *Candida* spp., that cause infection independent of filamentation or the production of Candidalysin. Consequently, these strains and species are often non-damaging *in vitro*, this does not correlate with their potential to cause infection in patients. We hypothesize that specific host factors, which trigger pathogenicity, are absent in *in vitro* models, and thereby not reflecting the situation in the host.

To determine the impact of albumin, the most abundant protein in the human body, vaginal epithelial cells were infected with different *C. albicans* strains and *Candida* species. Interestingly, after prolonged infection (45 h) albumin increased the damage potential, even in otherwise non-damaging and non-filamentous strains. This included deletion mutants deficient in filamentation, als3 adhesin/invasin, thigmotropism, or Candidalysin production. Yet, the increased damage was likely not solely an effect of increased growth and nutrient competition between the fungus and epithelial cells. Reduced damage in presence of protease inhibitors and albumin hint toward the role of proteases in the utilization of albumin. Albumin enhanced *C. albicans* metabolism, by stimulating the utilization of various nitrogen sources. This metabolic adaption could explain the advantage and enhanced growth as a strain and species-independent feature.

Our data suggest that common host factors can impact *C. albicans* to cause damage independent of adhesion, invasion, filamentation, and toxin production. Possibly, also other host-derived factors can drive the pathogenic potential of fungi through unresolved mechanisms.

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**Phylogenetic and ecological overview of Onygenales**

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Objectives: To evaluate the general taxonomy and phylogeny of the order Onygenales using ecological, morphological, and molecular data, stimulate awareness of correct identification of neglected groups in the order, and contribute to the stabilization of the nomenclature.

Methods: In total 97 genera, 385 species, and 553 strains were analyzed in this study. The ITS, LSU, TUB, TEF3, and RP605 gene regions were amplified and sequenced. Sequences for the RPB1, RPB2, and TEF1 regions were retrieved from the NCBI nucleotide database. Whole genome data for 53 strains were also included in phylogenetic tree analyses. Ecology and ascomata morphology for the type species were retrieved from the literature. Phylogenetic trees were constructed using the maximum likelihood methods implemented in IQ-TREE software and MRBAYES v3.2.7 on the CIPRES portal. Additionally, relative divergence time within Onygenales was estimated based on the RelTime method implemented in MEGA 7.

Results: A total of 1667 sequences for LSU ( $n = 421$ ), ITS ( $n = 519$ ), TUB ( $n = 189$ ), RP605 ( $n = 123$ ), TEF1 ( $n = 119$ ), TEF3 ( $n = 144$ ), RPB1 ( $n = 71$ ), and RPB2 ( $n = 97$ ) were examined. The results of the combined data analysis yielded 14 clades with  $\geq 90\%$  support for Bayesian probability and  $\geq 80\%$  support for maximum likelihood analyses. Families, based on their type genera and type species, were resolved as *Ajellomycetaceae*, *Arthrodermataceae*, *Ascosphaeraceae*, *Eremasaceae*, *Gymnoasaceae*, *Onygenaceae*, and *Spiromastigoidaceae* (Fig. 1). Two families were newly introduced as *Malbrancheaceae* and *Neogymnomycetaceae*. The family *Nanniziospioidaceae* clustered amidst members of *Onygenaceae*. The ecological preferences were classified as soil/oligotrophic, soil/keratinophilic, dung/agricultural, skin/nail, hair/feather, insect/pollen, osmotic habitats, systemic, plant, and other/unknown (Fig. 2). Almost all families in the order have members that can be found on skin and nails, which can cause asymptomatic or symptomatic infections, or members that are able to grow at 37°C and cause systemic infections. Four main types of ascomata morphology were noted: cleistothecium, gymnothecium, spore cyst, and naked fruitbody. The results of RelTime analysis showed that the diversification of species in Onygenales occurred at 103 Mya. The earliest species of the order were found in *Gymnoasaceae*, while the most recent species were found close to *Arthrodermataceae*.

Conclusion: Determination of the borderlines in the order can be difficult because of the effects of chosen methods, number of samples, number of genes, and also the choice of outgroups. Taxon sampling and inclusion of both type species and related genera in analyses are particularly essential to minimize changes and stabilize nomenclature for longer periods. Providing molecular data for the isolates and making them publicly available is also important to prevent taxonomic disagreements. Significant ecological traits that determine evolution in Onygenales are osmophily, thermophily, cellulolysis, eutrophism, oligotrophism, keratinolysis, and thermal dimorphism. Morphological and physiological characteristics may be informative for habitat choice and evolutionary processes. Cellulolytic and osmophilic abilities might be ancestral characteristics in Onygenales. Even though most of the species are found in soil and are non-pathogenic, environmental and host alterations can lead to the emergence of new fungal pathogens among soil fungi. Therefore, Onygenales continues to deserve close attention.

