3.4 Characterization of glycophospholipid/evolin-linked aspartyl proteases in Candida glabrata Role in pathogenicity

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3.5 Host of the Fungal Call Wall Glycan Can Mediate the Immune Response, September 23, 2022, 3:00 PM – 4:30 PM

Candida glabrata is the second most common fungal pathogen found in Candida bloodstream infections, depending upon the geographical location. C. glabrata, which belongs to the Nakasonea clade, possesses a distinct set of virulence attributes which include the ability to form biofilms and predominate in macrophages, adheres to hosts, and exhibits surface and secrete a wide range of toxins. Our research is focused on unravelling the strategies that C. glabrata employs to survive the nutrient-poor hostile host environment and evade immune host response. Toward this end, we are delineating the cellular processes that are responsible for the virulence of C. glabrata. One such strategy is the production of a glycan-auxiliary protein (GAP), which we have recently characterized in C. glabrata. GAP is a type 1 transmembrane protein that is involved in the regulation of the cell cycle and biofilm formation. GAP was shown to be essential for the survival of C. glabrata in the host environment. However, GAP was also found to be required for the induction of innate immune responses in the host, suggesting a novel role for GAP in host-pathogen interactions.

3.6 Population biology of fungal hagfish Trichoplusia erinacei

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4.4 Cases of animal mycoses, September 23, 2022, 3:00 PM – 4:30 PM

Trichoplusia erinacei is a main cause of dermatitis in hagfishes and is increasingly reported from human infections worldwide. It is found in wild European hagfish (Trichomycterus europeus) but also in the African four-road hagfish (Debelius albidus), which is a species per annual worldwide. Little is known about the taxonomy and population genetics of this parasite despite its increasing importance in clinical practice. Notably, there are different populations or even genetic species associated with different hosts or geographic regions is not known. To answer these questions, we collected 161 isolates, performed multilocus genotyping analysis, and characterized the population distribution and host specificity. Multiple phylogenetic and microsatellite analysis supported T. erinacei as a monophyletic species, in contrast to highly incongruent results from population structure analysis, one main complex mainly to snakes and the second to European hagfishes, were identified inside T. erinacei, and slight differences in the size of microsatellites and antifungal susceptibility were observed among them. Although the process of speciation into two lineages is ongoing in T. erinacei, there is still gene flow between these lineages, which might explain the observed molecular diversity. The data from wild hagfish hagfish indicated that unusual reproduction in T. erinacei and the new discovery of hagfishes from soil are probably rare events and that clonal horizontal spread strengthens diversity. The molecular approach used in this study was a powerful tool to detect the presence of new fungal species in both snakes and hagfishes.

5.5 MLST genotyping and phylogenetics of AD-hybrids

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5.7 Geographically of Cryptococcus neoformans and C. gattii, September 23, 2022, 3:00 PM – 4:30 PM

Objectives: In a previous study a set of new molecular-type-specific primers were designed to apply the standard DSMH consumes multi-locus sequence typing (MLST) scheme to Cryptococcus neoformans AD-hybrids. In the present study, we report the preliminary results of the investigations by MLST of a large number of AD hybrids with the aim to identify the circulating genotypes, their phylogenetics, and population genetics.

Methods: A set of 90 AD-hybrid isolates from different parts of the world and from different countries were generated by MLST. Minimum spanning tree (MST) was generated using the G2 MLST database. The genotypes from different countries were compared by using the G2 MLST database.

Results: Analysis identified 32 hybrid genotypes grouped in three distinct main clusters (CCUG1, CCUG2, and CCUG12) including 12 isolates each. Both CCUG1 and CCUG2 clusters included isolates from different countries and continents but the former group shared only with migrants from type A and the latter those from migrants with type A. Clusters CCUG12 included only from Europe. Hybrid genotypes of all clusters were compared in the same host environment. Phylogenetic diversity of AD hybrids is higher and survival AD hybrids can occur. Sequencing of further AD hybrids is in progress to confirm these findings.

5.8 Cryptococcosis neoformans and Cryptococcus gattii clinical isolates from Colombia develop heteroresistance to fluconazole at high concentrations

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5.9 Geographically of Cryptococcus neoformans and C. gattii, September 23, 2022, 3:00 PM – 4:30 PM

Introduction: Cryptococcosis is a worldwide mycosis caused by Cryptococcus neoformans and Cryptococcus gattii. Although resistance to antifungals is infrequent, isolates with decreased susceptibility to fluconazole have been reported globally, including Colombia, which may be due to: 1) heteroresistance, defined as the ability to adapt to increasing concentrations of antifungals; and 2) point mutations in the ERG11 gene encoding the fluconazole target enzyme, lanosterol 14α-demethylase.

Objective: To determine the development of heteroresistance to fluconazole in C. neoformans and C. gattii clinical isolates from Colombia and to analyze and quantify the ERG11 gene of the isolates to seek for mutations that might characterize resistant or heteroresistant phenotypes.

Methods: The minimum inhibitory concentration (MIC) to fluconazole was determined in 28 and 24 isolates of C. neoformans and C. gattii, respectively, using broth microdilution. Heteroresistance was evidenced by plating each isolate on YPD agar that contained fluconazole at concentrations equal to the MIC of each isolate. Heteroresistant clones were then replicated in inoculating concentrations of fluconazole, and MICs were determined.

Results: All isolates were susceptible to fluconazole with MICs of 1 μg/ml (n = 6), 2 μg/ml (n = 4), 4 μg/ml (n = 17), 8 μg/ml (n = 25), 32 μg/ml (n = 2), and 512 μg/ml (n = 1). However, all isolates developed heteroresistant clones, with increasing concentrations of MIC (26% of the isolates were resistant to 8 μg/ml, 33% of the isolates were resistant to 32 μg/ml, and 50% of the isolates were resistant to 512 μg/ml). The MIC of C. gattii 32 μg/ml was confirmed by the genotypes C. neoformans and 8.3% (33.3%) of C. gattii, grown up to 64 μg/ml of fluconazole, which is the MIC that defines resistance to this drug, and 1.2% (33.3%) of C. neoformans and 14.7% (50%) of C. gattii were also resistant to 64 μg/ml of fluconazole, with MICs of the two species ranging from 64 to 512 μg/ml. Clinical isolates of C. neoformans and C. gattii that develop heteroresistance to fluconazole in high concentrations in circulatory systems, which is important because this characteristic contributes to the collapse of cryptococcal therapy with this triazole.