CHARACTERIZATION OF THE MECHANISMS OF ACTION OF PLUMIERIDE NATURAL COMPOUND IN CRYPTOCOCCUS GATTII

Silva, A.C.¹,²; Barcellos V.A.¹,²; Fernandes C.L.²; Corá, R.K.²; Staats, CC²; Schrank, A²; Von Poser, G. L.³; Kmetzsch L²; Vainstein, MH².

¹Programa de Pós-Graduação em Biologia Celular e Molecular (PPGBCM), Universidade Federal do Rio Grande do Sul, Rio Grande do Sul, Brazil.
²Centro de Biotecnologia, Instituto de Biociências, Universidade Federal do Rio Grande do Sul, Rio Grande do Sul, Brazil.
³Departamento de Matéria Prima, Faculdade de Farmácia, Universidade Federal do Rio Grande do Sul, Rio Grande do Sul, Brazil.

Cryptococcus neoformans and Cryptococcus gattii are etiologic agents of cryptococcosis. These yeasts belong to the phylum Basidiomycota. Infection proceeds via inhalation and dissemination to the central nervous system, to cause meningoencephalitis. These fungal pathogens cause one million cases of cryptococcal meningitis per year among people with HIV/AIDS, resulting in nearly 625,000 deaths. Treatment of invasive fungal infections is still a challenge due to some limitations such as nephrotoxicity, hepatotoxicity and resistance development, which justify the search for new drugs. Our group identified the antifungal activity of two iridoids - plumieride and plumieridine - isolated from Allamanda polyantha (Apocynaceae). In order to elucidate the molecular mechanisms of plumieridine, a C. gattii mutant library with 8,000 mutants is being screened to find susceptible and resistant mutants.

C. gattii mutant library obtained by Agrobacterium tumefaciens-mediated transformation with approximately 8,000 hygromycin resistant mutants is being screened using higher and smaller concentrations based on plumieridine MIC value for C. gattii. MIC and MFC determination are realized following NCCLS-M27A protocol for broth microdilution methods.

Until now 3 susceptible and one resistant mutants were found and the classical virulence factors were evaluated. Plumieridine compound decreases polysaccharide capsule and cell size. No changes were observed in melanin production or ability to grow at 37°C.

Plumieridine compound affects the major virulence factor in C. gattii, indicating that some step in the capsule biosynthesis pathway could be target of plumieridine. The interrupted locus in the selected mutants will be determined by inverse PCR and sequencing to provide an outline of plumieridine compound mechanism of action.

Financial support: CAPES

Mutant library, Cryptococcus gattii, plumieridine.