Title: Analysis of the presence of *Cryptococcus* sp. in tree hollows in the main public square of Alfenas -MG

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Abstract:

Cryptococcus sp. is a capsulated yeast that causes cryptococcosis in humans and animals, and inhabits environmental sources, such as avian droppings and tree hollows. The objective of this study was to investigate the presence of Cryptococcus and other yeasts in tree hollows in Getúlio Vargas Square, located in the city of Alfenas, MG, determine the profile associated with virulence and test the strains isolated to Ocimum basilicum. Samples from 30 trees were collected in Getúlio Vargas Square due to the large amount of pigeons and passers-by. Plates with Niger agar were inoculated with sterile saline suspensions of the samples with 300mg/ml of chloramphenicol. After five days of incubation at 25°C, twelve samples (40%) showed dark brown colonies. These colonies were sub cultivated for morphophysiological identification. After the identification by the classic method, none of the isolated samples were characterized as Cryptococcus. It was possible to identify 13 yeast samples of the genera: Candida (4/30.77%); Rhodothorula (1/7.7%); Trichosporon (8/61.53%); and one (3.33%) tree presented the growth of two different species of Trichosporon (brassicae and lactis). The genus Candida presented one (1/7.69% each) strain of each species: blankii, castunsis, fennica and silvanorum, as well as Rhodothorula glutinis (1/7.69%). The genus Trichosporon presented the following species: brassicae (3/23.07%), gracile (3/23.07%) and lactis (2/15.38%). All the strains isolated were submitted to the virulence tests of enzymatic activities of lipase, phospholipase and protease. None of them presented activity of phospholipase and proteinase (Pz 1) and the lipase activity was classified as moderate due to the minor Pz 0.64. We conducted the test of Minimum Inhibitory Concentration using Ocimum basilicum, according to the CLSI, applying the concentrations of 1.0; 0.5; 0.250; 0.125; 0.0625; 0.0313; 0.0156; 0.0078 and 0.0039 mg/ml. All strains that presented MIC ranging from 0.0039 to 0.5 mg/ml were inhibited, excepting Candida blankii, which presented MIC 1mg/mL. Although the strains did not confer factors associated to virulence and show good sensitivity to the tested essential oils, this does not exclude its ability to develop diseases in humans. The absence of Cryptococcus in the analyzed samples suggests the need to improve on classical techniques of isolation, or the use of molecular biology to ensure the presence or absence of this agent in these samples analyzed.

Keywords: Cryptococcus sp. Yeasts. Hollow trees.