**TITLE**: SCREENING OF ENZIMATIC PRODUCTION BY ENDOPHYTIC FUNGI ISOLATED FROM *Myrcia guianensis* 

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## ABSTRACT:

Endophytic fungi are well known for their ability to produce biotechnological relevant enzymes. Among them, hydrolases have an important role in many industrial processes, reinforcing the need of their bio-sustainable process of synthesis. The present work aimed at evaluating the production of amylase, cellulase and xylanase by endophytic fungi preserved since April, 2009, after being isolated from Myrcia guianensis in the city of Santarém (Pará State, Brazil). The 28 strains preserved by the Castellani method were inoculated on Sabouraud agar (SDA) to test their viability and to provide the inoculums. For each enzymatic essay, a specific solid medium was prepared as follows: amylase starch 1%, agar 1,8%, 500 mL phosphate-citrate buffer 0,1M, pH 5,0; cellulase - carboxymethyl cellulose (CMC) 1%, agar 1,8%, 500 mL sodium-acetate buffer 0,1 M, pH 5,0; and xylanase - agar 1,8%, xylan 0,25%, 500 mL distilled water. All medium were sterilized at 121°C for 15 minutes. Each endophytic strain was inoculated as a 5 mm<sup>2</sup> fragment of mycelium at the center of a Petri plate containing a specific medium (3x/medium) and incubated at 28°C for 5 days. The plates were monitored daily and in the case of growing, a specific indicator was used: 0,1 M iodine solution for amylase, Lugol for cellulase, and for xylanase, a 0,5% Congo red/ethanol solution (w/v). The results were revealed with a 1 M NaCl aqueous solution. The hydrolytic effect of the enzymes should produce a halo around the growing mycelium. The enzymatic index (EI) of each strain was calculated by the ratio between the halo diameter and the colony diameter. 23 strains showed enzymatic activity: 7 (30,4%) with EI  $\geq$ 3 for amylase, 14 (60,8%) with EI  $\geq$ 3 for cellulase and 2 (8,7%) with EI  $\geq$ 3 for xylanase. Only 5 strains did not grow onto SDA. Our results demonstrate the strong ability of M. guianensis endophytic fungi to produce hydrolases even after 8 years of conservation, pointing out to the relevance of Castellani as long term preservation method of the fungal characteristics.

Keywords: hydrolases, enzymatic index, endophytic fungi

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