

## Endophytic fungi: lipase activity and Biotechnological Applications

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**Purpose of study:** To isolate endophytic fungi from vegetal species of the Restinga de Jurubatiba Shoal, RJ State, Brazil, aiming to apply in biotechnological processes with optimized conditions. These microorganisms apparently do not produce any damage to their host [1] and, may represent important sources of promising enzymes, including lipases. Monoacylglycerols (MAGs) have many biotechnological applications, especially on the pharmaceutical industry. They are currently produced by alkaline glycerolysis oil, resulting in low yields and numerous by-products. The use of enzymatic processes catalyzed by lipase, may lead to a more ecological approach [2]. This work aimed to investigate the lipase activity of endophytic fungi as well as their application as biocatalysts in esterification reactions for biodiesel and MAGs production.

**Methods:** Initially, a screening for the lipase activity was held with fungi from *Tocoyena bullata* (TB) and *Humiria balsamifera* (HB) vegetal species by the Rhodamine B (0,001%) method, at 365 nm of absorbance by UV detection. Selected lipolytic fungi were then inoculated into a pre-fermentation medium (30°C, 120 rpm, 120 h) and then transferred into a fermentation medium (PDB) to stimulate the lipase production at the following conditions: 48h, 30°C and 180 rpm of stirring. From the fermentation medium it was performed the method based on the *p*-nitro phenyl palmitate hydrolysis by measuring the *p*-nitro phenol production at 410 nm by spectrophotometer. Finally, an esterification assay for biodiesel production was performed by the Lowry-Tinsley [3] method. The production of the ethyl oleate was monitored each 24 h until 120 h and, after 15 days of incubation. **Results:** The HB13 strain presented the highest fluorescence halo after 24h of inoculation, confirming the lipase activity. For the *p*-nitro-phenol production, the best result was obtained after 48 h of incubation with HB13 (92 U/mL) and TB1 (85 U/mL) strains. Both fungi showed excellent results for producing ethyl oleate ester, since the conversion achieved higher than 90% after 48 h (Table 1).

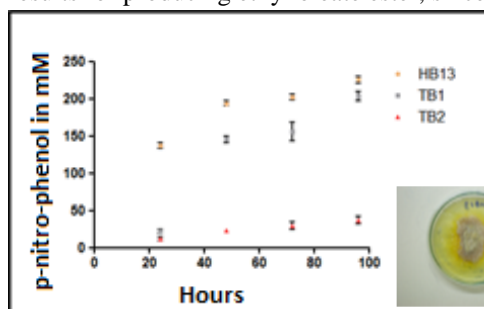


Table 1. Ethyl oleate conversion rates (%) under different incubation times

FUNGI	Different incubation times					
	24 h	48 h	72 h	96 h	120 h	15 days
TB1 (%)	73,02	95,40	89,56	95,55	95,59	94,95
HB13 (%)	81,31	92,89	92,81	95,30	95,6	95,32

**Conclusions:** For the Rhodamine B, pPNN and Lowry-Tinsley methods TB1 and HB13 strains showed to be promising for hydrolysis and esterification reactions. The microorganisms are going to be investigated for their MAG production, in optimized conditions, by using a mathematical experimental model.

### References:

[1] Schulz, B.; Boyle, C. (2005). *Mycol Res* 109:661–686; [2] Junior, I.I.; Flores, M.C.; Sutili, F.K.; Leite, S.G.F.; Miranda, L.S.; Leal, I.C.R. de Souza, R.O.M.A. (2012). *Journal of Mol Cat. B, Enz.*, 77:53– 58. [3] Lowry, R.R; Tinsley J.I. (1979). *J Am Oil Chem Soc*, 53:470-472.