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Laccase production by free and immobilized fungal mycelium of *Trametes versicolor***Sara C. Silvério¹, Sérgio Moreira², Solange I. Mussatto¹, Adriane M.F. Milagres², Eugénia A. Macedo³, José A. Teixeira¹**

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The production of laccase by free and immobilized mycelium of *Trametes versicolor* was evaluated. Fermentation experiments were carried out using a *Trametes* defined medium [1] supplemented with tween-80 (0.5%, w/v) and xyldine (30 µM) to stimulate and induce the secretion of extracellular enzyme. The support for the mycelium immobilization consisted in 0.3 cm³ cubes of synthetic fiber (Scotch Brite, 3M Spain, SA), which were added to the culture system at 1 g/100 mL of medium. Assays were performed in 250-mL Erlenmeyer flasks and in a 2 L stirred tank bioreactor. For the flasks fermentations, three 7 mm diameter plugs from the fungal monoculture plate were inoculated into 50 mL culture medium in the presence or not of the immobilization support. For the assays in bioreactor, the volume of inoculum necessary to obtain an initial cell concentration of 70 mg/L was transferred to the reactor containing 1 L of culture medium with or without the immobilization support. In both cases, the fungus was incubated at 28 °C and 180 rpm. During the experiments, samples were periodically withdrawn for laccase and glucose determinations. Synthetic fiber was used as immobilization support since this material was demonstrated to be of great potential for fungi immobilization [2]. Additionally, many studies have demonstrated that fermentation systems with immobilized cells are able to increase the process productivity. However, the laccase production by *T. versicolor* (present study) did not show this performance. In both systems (Erlenmeyer flasks and bioreactor), the highest laccase production was obtained when using free mycelium. Additionally, the maximum laccase production obtained in bioreactor was lower than the maximum found in Erlenmeyer-flasks, suggesting that the conditions used in the bioreactor should be optimized to increase the laccase production results. Due to the great importance of the laccases in the industrial sector, more studies will be performed aiming to find a strategy to maximize the production of this enzyme by *T. versicolor*.

[1] Roy et al *Appl. Environ. Microbiol.* 59, 1855–1863, 1993. [2] Mussatto et al., *Carbohydr. Res.* 344, 795–800, 2009. Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq, Brazil) and Fundação para a Ciência e a Tecnologia (FCT, Portugal).