



## ANALYSIS OF CULTURES DEVELOPED BY *TRAMETES VERSICOLOR* IMMOBILIZED ON CALCIUM ALGINATE

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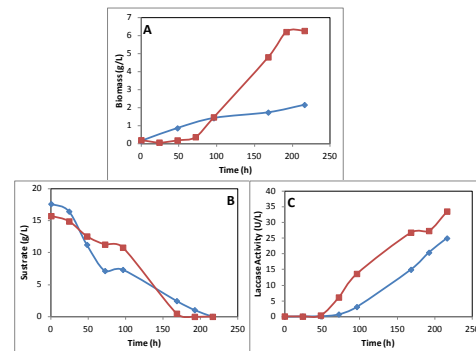
**Introduction.** *Trametes versicolor* is a white rot fungus very important in nature and at industrial level too, because of its ability to produce laccases, enzymes that degrade different compounds<sup>1</sup>. The development of some cell immobilization techniques has allowed the maximum exploitation of the oxidative capabilities of this fungal specie, for been applied in several processes<sup>2</sup>. However, the behavior of cultures developed with immobilized *T. versicolor* cells can be affected by several factors, including those related with the rheology and physicochemical characteristics of culture<sup>3</sup>.

The objective of this work was to establish the better conditions for the immobilization of *T. versicolor* in calcium alginate and analyze the effect of specific factors on the behavior of the immobilized cells at bioreactor level.

**Methods.** Mycelium produced after the incubation of *T. versicolor* on mineral medium<sup>4</sup> during 10 days at 30°C and 200 rpm was recovered by filtration and resuspended in saline solution. For knowing the mycelium concentration (5, 10 or 15 g/L) and calcium alginate percentage (2 or 3%) that must be mixed for obtaining pearls with the best characteristics to develop cultures at bioreactor level, a 3X2 factorial design was made. After that, a 2<sup>3</sup> factorial design was developed, in order to know the effect of pH (values among 3 and 6), C:N proportion (values among 6 and 10) and  $K_{La}$  (values among  $1.025 \times 10^{-8}$  and  $4.119 \times 10^{-9} \text{ L/m}^2 \text{ s}^{-1}$ ) on cultures with *T. versicolor* free (F) and immobilized (I) cells. Finally, batch cultures were developed in bioreactors developed under the best operational conditions, which were inoculated using 1 g/L of F or I cell and incubated at ambient temperature during 10 days sampling every 24 h. Biomass was quantified by dry weight technique, substrate consume using DNS and laccases production by means guayacol oxidation in a spectrophotometer ( $A=$  ).

**Results.** The 3X2 factorial design showed that the best immobilization condition was reached after mixing 10 g/L of biomass with 2% calcium alginate. ANOVA of 2<sup>3</sup> factorial

design indicated that only C:N proportion and  $K_{La}$  were significant for laccases production in F, meanwhile for I all factors were significant (data not shown), however in both cases the best operational condition for operating a batch reactor were the same. So, after comparing the behavior of these batch cultures, it was observed that the growth of I was considerably lower, although in both cases cells showed the same substrate consumption profile. On the other hand, although immobilization process only caused a small decrease on laccases production by cells, the specific production values was duplicated (Fig. 1). This showed the advantage of the immobilization of *T. versicolor* for developing several processes of environmental and industrial importance in which laccases activities are involved.



**Fig.1** Biomass (A), substrate consumption (B) and laccase production (C) by *T. versicolor* F (■) and I (◆) on reactor with pH de 6.0, C:N proportion of 10 and  $K_{La}$  of  $1.51 \times 10^{-9} \text{ L} \cdot \text{m}^2 \cdot \text{s}^{-1}$

**Conclusions.** Immobilization of *T. versicolor* on calcium alginate means an alternative to favor the microbial action on several processes in which laccases action is required.

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