

# Etude comparative entre la cinétique de l'oxydation du phénol par la tyrosinase libre et immobilisée dans le gel d'alginate de calcium

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**Abstract :** Phenol is known as a toxic compound; therefore, it is necessary to develop an efficiency method to remove phenol from wastewater of industrial effluent. Removing of phenol and its derivatives can be made by different biological and physico-chemical processes. The biological treatment of water containing phenol is currently made by enzymes such as tyrosinase, laccase and peroxidase. The objective of our work is to study the kinetics oxidation of phenol by free tyrosinase (EC: 1.14.18.1) and encapsulated tyrosinase in calcium alginate gel. First of all, tyrosinase is extracted from mushroom (*Agaricus bisporus*) and its phenolase activity was measured spectrophotometrically at 400 nm. Mushroom tyrosinase catalyzes efficiently phenol oxidation in solution. A comparative study is made between the different kinetic parameters of free and immobilized tyrosinase. The Michaelis-Menten constant ( $K_m$ ) for the immobilized enzyme (0.9 mM) is approximately twice higher than that of the free enzyme (0.55 mM), suggesting that alginate matrix limits the rate diffusion of substrate and product. In contrary, the maximal velocity ( $V_{max}$ ) of the enzyme immobilized is 20-fold lower than that of the free enzyme. The effects of pH, temperature, enzyme and substrate concentration, alginate beads size and operational stability on the initial rate of phenol oxidation were studied. The pH and temperature optima are shifts from 7.6 to 5.6 and from 45 to 55°C, for free and immobilized tyrosinase, respectively. The immobilized enzyme seems to be more thermostable than the free enzyme and its activity is maximal when immobilized in alginate beads with a diameter of 2.6 mm.

**Keywords :** *Agaricus bisporus*, Tyrosinase, Kinetics, Phenol, Oxidation, alginate, Immobilization.