



### Exploitation of olive oil mill wastewater for esterase production

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Development of bio-sustainable and renewable resource technologies is extremely important on environmental contexts. Waste can contain many valuable substances and, through a suitable process or technology, this material can be converted into value-added products or raw materials that can be used in secondary processes. To reduce the environmental problems caused by olive oil mill wastewater (OOMW), which is the liquid effluent produced during the extraction process of olive oil, this waste can be applied to bioproducts production. The annual production of OOMW worldwide was estimated to be over 20 million m<sup>3</sup>, and its disposal represents a huge environmental problem. The main compounds found are sugars, residual oil (lipids), proteins, organic acids, minerals and recalcitrant compounds such as phenols and tannins. In particular, OOMW presents a suitable substrate to produce lipolytic enzymes as the esterase. Esterases (EC 3.1.1.1) are a group of carboxyesterases that catalyzing the cleavage and formation of ester bonds of an extensive range of substrates, preferentially short-chain fatty acids, no more than 10 carbons. Esterases have a broad spectrum of industrial uses including food and dairy products, ester production, detergents, pharmaceuticals, synthesis of optically pure compounds, degradation of pollutants and production of perfumes. The goal of this work was the esterase production using OOMW as substrate from *Aureobasidium pullulans* LABIOTEC 01 cultivation. The esterase enzyme was produced by submerged cultivation of *A. pullulans* LABIOTEC 01. For inoculum preparation, this strain was grown on Sabouraud 4% Glucose Agar at 25°C for 72 h. Cultures were scraped from the agar surface, added to a Sabouraud dextrose broth and grown during 24 h at 28°C. A homogeneous suspension of grown microorganism with O.D.<sub>620</sub> of 0.1 was added (10%, v/v) to the culture medium containing OOMW and incubated at 30 °C in a rotary shaker (200 rpm) for up to 168 h. Aliquots of the culture medium were collected at pre-determined intervals and clarified by centrifugation whereas the supernatant with the enzyme was used for monitoring the pH and esterase activity. Furthermore, the esterase productivity (U/mL/h) was determined from the maximum values of esterase activity. The OOMW was a suitable substrate for the production of esterase at 96-h submerged cultivation period, with the highest enzyme activity about 16.0 U/mL. In relation to the pH of the media, an increase from 5.5 to 9.0, approximately after a 168-h cultivation period, was observed. The productivity of esterase enzyme reached a maximum value of 0.15 U/mL/h at 96 h. Results showed that OOMW could be an interesting alternative to producing esterase enzyme, creating an interesting alternative to manage this waste.

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