



INDIGO BIOSYNTHESIS BY Pseudomonas monteilii P26 PLANKTONIC AND IMMOBILIZED CELLS

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Indigo is used as dye in the textile industry. Its production by conventional methods involves the use of toxic precursors, the consumption of high amounts of energy and the production of toxic waste. Indigo biosynthesis mediated by microorganisms could be an environmentally friendly and more efficient alternative for indigo production. The aim of this work is to study different conditions for indigo production using *Pseudomonas monteilii* P26 resting cells suspensions and to use the best condition obtained for the production of indigo by immobilized cells. Immobilization was carried out by biofilm formation using two methods: a continuous culture and a sequential batch reactor process. The carrier used was polyurethane foam (PUF). Results showed that the best condition for indigo production using cell suspensions was obtained when indole was added as aqueous solution at low cell concentration. Previous indigo biosynthesis induction by the addition of naphthalene resulted to be unnecessary. Indigo production by immobilized cells was also evidenced. This work is a first approach towards the optimization of indigo biosynthesis for the development of cleaner production processes.

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OPTIMAL CONDITIONS FOR THE PRESERVATION OF *Pseudomonas tolaasii* IEXb, A MICROOR-GANISM USED AS GRASS BIOFERTILIZER

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Phosphorus (P) is one of the most required inorganic nutrient for microorganisms and plants. Phosphate Solubilizing Bacteria (PSB) exert a relevant role in the P cycle on the soil, mobilizing deposits of insoluble phosphates. PSB belong to the group of Plant Growth Promotion Rhizobacteria (PGPR). In previous studies, PSB were isolated from Argentinian Puna (Salta and Jujuy) and characterized for their ability to solubilize tricalcium phosphate and hydroxyapatite. Other properties of PGPR were also evaluated, such as indole acetic acid and siderophore production, and ability to biocontrol against phytopathogen fungi. Pseudomonas tolaasii IEXb was selected for its ability to promote the plant growth and to increase performance of maize culture. The aim of this work was to evaluate different preservation treatments of P. tolaasii IEXb biomass, to formulate a product with shelf life according to its application on field. Biomass production was performed in a 10 L fermenter (New Brunswick Scientific Microferm Fermentor MF-214), using 5 L of a low-cost culture medium (1% whey permeate and 1% soybean meal), at 30°C, initial pH = 6.0, with agitation (200 rpm). Growth kinetic parameters were determined up to 8 h of culturing, through viable cell counts on agar plate. The produced biomass was harvested, washed and subjected to different preservation processes. A factorial experimental design 3 x 2 was applied to determine the effects of three formulations (cell suspensions in LB broth-20% glycerol, cell suspensions in LB broth-10% lactose, and lyophilized bacterial cells in 10% whey permeate-5% sodium glutamate) and two storage temperatures (4°C and 25°C) on the P. tolaasii IEXb viability during 150 days. Samples were taken during storage and the numbers of colony forming units (CFU) per mL (CFU/mL) were determined. Data were evaluated applying the general linear model of analysis of variance (MINITAB 17 statistical software). Maximal P. tolaasii IEXb biomass (6.2 x 108CFU/mL) was reached at 8 h of culture, with a growth rate of 0.87 h-1. All the factors evaluated significantly affected the P. tolaasii IEXb viability during storage. The increase of storage temperature exerted a marked negative effect on the bacterial survival in liquid formulations, mainly in the presence of lactose (absence of viable, cultivable cells at day 28, when storing at 25°C). However, high numbers of viable cells were recovered from lyophilized powders stored at 25°C (3.88 x 10 8 CFU/mL and 1 x 107 CFU/mL at days 42 and 96, respectively). After 150 storage days at 4°C, similar numbers of viable cells were recovered from cell suspensions in the presence of glycerol or lactose (around 5 x 108 CFU/mL), viability being lower than in lyophilized powders in whey permeate-sodium glutamate (1 x 109 CFU/mL). In conclusion, these results allowed the determination of the most suitable conditions for the preservation of *P. tolaasii* IEXb for its use as biofertilizer.