

TITLE: BIOPROSPECTION OF BIOSURFACTANT PRODUCING BACTERIAL COLONIES VIA *EX-SITU* SOIL TREATMENT WITH VEGETABLE OILS

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ABSTRACT:

In the soil there are biosurfactants producing bacteria. Biosurfactants are important for the industries in oils tanks, therapeutic applications, agriculture, food industry, and to the environment for bioremediation of soil and contaminated water. Studies involving Amapá soils and biosurfactants producing bacteria were not found in the literature. This research aimed to select biosurfactants producing bacterial colonies from *ex-situ* soil sample collected at the floodplain ecosystem at the Environmental Protection Area of the Curiaú River. The sample was divided 4 and placed into disinfected pet bottles and treated every two weeks for six months by adding of 50ml of vegetable oil, such as: canola, sunflower and mixture of oils (equal amount of soybean, palm, canola, sunflower and olive oil). A soil sample was maintained untreated. The soil bacteria were cultivated for two weeks with pH 7 at 37°C in nutrient broth containing, respectively, 1% of canola oil, sunflower oil and oil mixture. After this period the cultures were subjected to a serial dilution and plated in triplicate on nutrient agar. Ten colonies were chosen from each sample, cultivated in 10ml of nutrient broth with 1% olive oil and incubate at 37°C for 5 days. From each culture 2ml were transferred to another glass together with 2ml of commercial kerosene and agitated with vigorously vortex for 2 minutes. To identify the biosurfactant production, it was obtained the emulsification index (EI) from an emulsification test. In which, the height of emulsion layer was divided by the height of the entire column and expressed in percentage. This EI was determined at 5 minutes and 24, 48 and 72 hours. The ratio between EI at 24, 48 and 72 hours and EI at 5 minutes showed the study of the emulsion stability on relation to time. Biosurfactant production occurred in 62% of the colonies and 47% of the biosurfactants were stable till 72 hours. In canola and oil mixture treated soils 90% of the colonies produced biosurfactant, in sunflower treated 50% and untreated 20%. The best EI was 60, 4% found in untreated sample and lowest EI was 7,7 % found in oil mixture treated. The most stable biosurfactant occurred in oil mixture with a value of 2.1 at 72 hours, and less stable biosurfactant occurred in canola oil treated with a value of 0,3. This study showed that *ex-situ* soil treatment induced the appearing of biosurfactant producing bacterial colonies.

Keywords: Bioremediation. Emulsification Test. Kerosene. Nutrient Broth. PET Bottle.