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Increase of biosurfactant production by co-cultivation of producing bacteria with biofilm-forming strains

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Biosurfactants (BSF) are amphipathic compounds, produced by a vast range of microorganisms. BSF have applications in diverse areas, such as petroleum exploration and remediation of spills, control of biofilms and biocorrosion, and are also used as ingredients in therapeutic formulations, personal care products, and cosmetics. They represent an advantageous alternative to chemical surfactants, because they are less toxic than chemical surfactants, biodegradable, biocompatible and stable in extreme conditions of temperature and pH. However, mass production and application of BSF is still limited by the low production yield and high production costs. Considering that BSF are released as secondary metabolites, the hypothesis that co-cultivation with biofilm-forming strains would induce BSF synthesis was tested. BSF producing strains of *Bacillus licheniformis* and *Pseudomonas* sp. were cultivated with biofilm-forming (inducing) strains (*Pseudomonas aeruginosa* and *Listeria innocua*) as a way to stimulate the production of surfactin and rhamnolipids, respectively. Axenic cultures and co-cultures were tested as to the tensoactive effect by the oil spray method, the drop collapse assay, cultivation in CTAB-methylene blue medium and cultivation in blood agar. Surfactin and rhamnolipids were quantified by colorimetric methods and quorum quenching effect was tested in *Chromobacterium violaceum*. Both *Pseudomonas* sp. and *B. licheniformis* showed an increase in BSF production when co-cultivated with inducer strains. Results of the oil-spray assay and the CPC-BTB test (Figure 1) indicate that the stimulation of BSF production by *Pseudomonas* sp. was the highest in co-cultures with *Listeria innocua*. The results of the CTAB-methylene blue test indicate that BSF production in *B. licheniformis* was more stimulated by co-cultivation with *P. aeruginosa*. However, significant differences in the tensoactive effect of the cell free extracts were not observed. The results indicate that co-cultivation positively affects the efficiency of BSF production and that higher production yields can be attained by selecting convenient inducer strains.

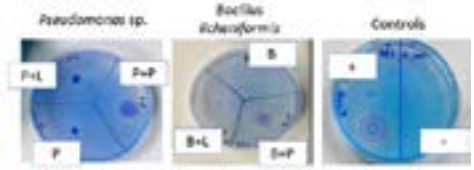

Figure 1: CTAB-methylene blue test performed on axenic cultures and co-cultures of *B. licheniformis* or *Pseudomonas* sp., and controls. A solution of 5% SDS was used as positive control and *E. coli* DH5 was used as negative control. P – *Pseudomonas* sp.; P+P - *Pseudomonas* sp + *Pseudomonas aeruginosa*; P+L - *Pseudomonas* sp + *Listeria innocua*; B – *Bacillus licheniformis*; B+P - *Bacillus licheniformis* + *Pseudomonas aeruginosa*; B+L - *Bacillus licheniformis* + *Listeria innocua*.

Biography

Angela Cunha is an Assistant Professor in the Biology Department of the University of Aveiro, Portugal and a Researcher at the Centre for Environmental and Marine Studies. She has been involved in lecturing graduate and post graduate courses in the fields of Microbiology and Genetics and is currently the Director of the Bachelor Course in Biology. Her main research field is the distribution and activity of microorganisms in the environment, microbial bioremediation tools and interactions of microorganisms with plants in the perspectives of disease control, plant growth promotion and microbe-assisted phytoremediation.

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