TITLE: MOLECULAR CHARACTERIZATION OF *Paenibacillus antarcticus* IPAC21, A BIOEMULSIFIER PRODUCER AT LOW TEMPERATURES

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ABSTRACT:

Molecules produced in cold environments are interesting for several industries as they usually show high activity at low temperatures. The strain IPAC21, a bioemulsifier producer belonging to the species Paenibacillus antarcticus, was isolated from King Island soil, Antarctica. Bioemulsifiers are complex heteropolysaccharides, lipopolysaccharides, lipoproteins and/or proteins. These molecules can efficiently emulsify two immiscible liquids even at low concentrations although they are less effective in surface tension reduction. To better characterize the produced bioemulsifier, IPAC21 genome was sequenced and annotated using Illumina Hi-seq and Go Feat platform (Gene Ontology Functional Enrichment Annotation Tool). It was categorized in Coding DNA Sequences (CDS) using the Rast software (Rapid Annotation Subsystem Technology). The search for genes related to the production of bioemulsifier and other metabolic pathways was performed using KEGG (Kyoto Encyclopedia of Genes and Genomes). The IPAC21 strain has a genome with 8,810,884 bp and G+C content of 40.5%. Genes related to the biosynthesis of exopolysaccharides, melibiose sugar transporter, cold-shock proteins, ectoine and chaperone DNAk were found in its genome. The IPAC21 genome presents 4813 CDS distributed in 26 categories with the highest CDS values for carbohydrates (18.7%), amino acids and derivatives (11.5%) and protein metabolism (8.6%). Enzymes associated with EPS, terpene and 2,3 butanediol pathways were also present in IPAC21. Moreover, to gain knowledge about the bioemulsifier produced by IPAC21, the optimal conditions for the bioemulsifier production and its physicochemical properties were determined. Strain IPAC21 was cultivated in Tryptic Soy Broth (TSB) at two temperatures (5°C and 28°C) for up to 96 h and cell-free supernatants were evaluated for bioemulsifier production by the emulsification index (EI) using hexadecane, drop collapse test, oil displacement test and surface tension. The highest EI values were observed at $28^{\circ}\text{C}/48 \text{ h}$ (59% \pm 1.75) and at 5° C/72 h (42.2% \pm 6.7). The results of the drop collapse, oil displacement and surface tension tests at both temperatures were negative as expected for a bioemulsifier. Its characterization is extremely relevant because of the many potential applications remediation of oil polluted water and soil, enhanced oil recovery, heavy metal removal and formation of stable emulsions in food and cosmetics industries.

Keywords: Paenibacillus antarcticus, bioemulsifier, Antarctic, genome, low temperature

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