**TITLE**: DETECTION AND PARTIAL EVALUATION OF PROTEOLITIC ACTIVITY PRESENT IN CYANOBACTERIA FROM AMAZON REGION

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## **ABSTRACT**

Cyanobacteria are microorganisms with photosynthetic abilities and are also producers of different bioactive compounds. These microorganisms are found in several habitats, resulting in metabolic diversity and a high potential to bioprospect compounds of biotechnological interest, such as pigments, inhibitors and enzymes. The proteases are enzymes that hydrolyze peptidic bonds and have been used in several industrial processes, for example: food and textile industries, and detergent formulation. The objective of this work was to identify the proteolytic activity of crude extracts and aqueous fractions from seven lineages of cyanobacterias from the genera Limnothrix, Nostoc and Synechococcus, obtained in Amapá State-Brazil (collection of cyanobacteria from CGBS-UFPA). The strains were cultivated in BG-11 medium with a controlled photoperiod. The biomass was obtained by centrifugation and its pellet was subjected to cell lysis, achieving the crude extract. The extract was fractioned in a column containing DIAION HP-20 resin to obtain the aqueous and metanolic fraction. Then, the crude extract and the aqueous fraction obtained were tested regarding their proteolytic activity using the synthetic substrate azocasein. The protease classes were investigated by their residual enzymatic activity in the presence of specific inhibitors (EDTA 1mM, PMSF 1mM and iodoacetamide 4mM). The crude extract and aqueous fraction from all samples were able to hydrolyse the substrate. The analysis using these classical inhibitors demonstrated the presence of serine proteases in three strains of Limnothrix and in one strain of Synechococcus; cystein proteases in two strains of Limnothrix, and metalo proteases in three strains of Limnothrix, one in Nostoc and one in Synechococcus. The aqueous fraction obtained from the Synechococcus sp. GFB01 strain, whose genome has been sequenced, was also evaluated using different EDTA concentrations (1-7 mM), and showed a higher inhibition (59%) using the concentration of 4 mM. Then, the current study has demonstrated that the cyanobacteria strains of Limnothrix, Nostoc and Synechococcus genera have different protease classes of industrial interest and, consequently, they can be used as alternative sources of prospection of proteolytic enzymes aiming to apply in biotechnology.

Keywords: cyanobacteria, proteases, biotechnology

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