

TITLE: PROTEOMIC ANALYSIS OF *Synechococcus* sp. CACIAM 66 IN DIFFERENT NITROGEN CONDITIONS IN GLUCOSIDASE INHIBITORS PRODUCTION.

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**ABSTRACT:** Cyanobacteria are oxygenic photosynthetic microorganisms, which make them suitable to be explored in renewable studies and applications. Furthermore, their innumerable species show a great metabolic diversity making them fit for prospection of compounds of industrial and biotechnological applications. One important metabolite of medical and environmental interest is the glucosidase inhibitors. They can be used to treat diabetes and Gaucher disease, and also show antifungal and antiviral activities. However, it is still necessary a better understanding how these compounds are synthesized in cyanobacteria. In previous works, the cyanobacteria *Synechococcus* sp. CACIAM 66 presented improved inhibitory activities against beta-glucosidase when cultivated in BG11 supplemented with 0.230g/L of  $(\text{NH}_4)_2\text{SO}_4$  (BG11<sub>N+</sub>). Thus, this work aimed to identify the homologous proteins of GutT1, GutB1 and YktC1, which was previously described as responsible for the synthesis of nojirimycin and its derivatives, by proteomics approach. This cyanobacterium was cultivated in BG11 and BG11<sub>N+</sub> for 60 days. The biomass was collected by centrifugation at 16.000g for 5min at 4°C, and pellet washed with Tris-HCl buffer (1M, pH 8) and re-centrifuged in the same parameters. The cells were submitted to a lysis buffer and sonication. The solution was mixed with phenol and re-centrifuged, removing the supernatant. The pellet was washed with ammonium acetate, acetone and ethanol and then dried. The proteins were prepared and submitted to digestion by trypsin. The protein solutions were then analyzed by mass spectrometry, and the data were extracted and analyzed by softwares. A total of 75 proteins were identified in both conditions, which 38 were common in both, 15 exclusively in BG11 and 22 in BG11<sub>N+</sub>. Regarding their distribution considering physiological roles, proteins related to photosynthesis and central metabolism were identified in more quantities in BG11, and more proteins related to protein synthesis and iron assimilation were identified in BG11<sub>N+</sub>. This behavior indicates a possible stress in BG11<sub>N+</sub>. However, exclusively in this condition was possible to identify a homologous of GutB1: Quinone-oxidoreductase. These results are consistent with previous work, which cells cultivated in BG11<sub>N+</sub> showed improved inhibitory effects. However, it is still necessary to test different conditions aiming to improve the inhibitor productions by this organism.

**Keywords:** proteomics, cyanobacteria, amazon, glucosidase inhibitors.

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