

**TITLE:** ENZYMATIC HYDROLYSIS OF *SPIRULINA PLATENSIS* CARBOHYDRATES: SCREENING OF PROCESS VARIABLES

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**ABSTRACT:**

Sustainable alternatives have been required to replace fossil resources in the production of energy, chemicals and materials. Compared to other feedstock, microalgae can be a high-yield source of biofuels without compromising food supplies. Species of microalgae such as *Spirulina platensis*, *Chlorella vulgaris*, *Scenedesmus bijugatus*, among others, are being tested for bioethanol production due to their high starch content. The aim of this study was to investigate the significant variables in the saccharification process of the carbohydrates extracted from the *Spirulina platensis* biomass, by enzymatic hydrolysis, for future applications in the production of bioethanol. The defatted biomass was submitted to ultrasonic assisted extraction of proteins and carbohydrates, by a process previously optimized. A Fractional Factorial design  $2^{4-1}$  was applied to evaluate the enzymatic hydrolysis step of the extracted carbohydrates, to analyze the effects of four variables under the response of reducing sugars concentration determined by the Somogyi-Nelson method. The variables studied were:  $\alpha$ -amylase and amyloglucosidase (AMG) concentrations ( $10 - 30 \mu\text{L} \cdot \text{mL}^{-1}$ ), times of action of  $\alpha$ -amylase (1 - 3 h) and AMG (2 - 6 h)]. The reducing sugar concentrations ranged from 3.24 to 13.90  $\text{g} \cdot \text{L}^{-1}$  in runs 1 and 8, respectively. By the analysis of the effects it was observed that the concentrations of  $\alpha$ -amylase ( $p = 0.0002$ ) and AMG ( $p = 0.0000$ ), and time of action of AMG ( $p = 0.0350$ ) were significant ( $p \leq 0.10$ ). The effects of the variables were positive, consequently, an increase in reducing sugars released was observed with the increase of the variables, within the ranges studied. It was possible to conclude that the *Spirulina platensis* biomass is a feedstock with potential to be applied in the bioethanol production. Subsequent studies will be carried out by the application of statistical techniques of process optimization, with the aim of increasing the yield of the process in fermentable sugars and, consequently, the ethanol yield.

**Keywords:** bioethanol, amylolytic enzymes, enzymatic hydrolysis, *Spirulina platensis*

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