**TITLE:** STATISTICAL SCREENING OF MEDIUM COMPONENTS USING PLACKETT-BURMAN DESING FOR PROTEASE PRODUCTION IN SUBMERGED FERMENTATION BY ENDOPHYTIC FUNGUS ISOLATED FROM BRAZILIAN CERRADO.

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**ABSTRACT:**

Proteases are responsible for catalyzing the hydrolysis of protein peptide bonds and degrading them in amino acids and peptides. They are produced by almost all living organisms. Proteases are one of the most important groups of enzymes that are used in industries and their use is quite old. In recent years, endophytic fungi have proved to be a source of great potential to produce secondary metabolites with promising applications in agriculture, the environment, the pharmaceutical industry and the food industry. In this work, the endophytic fungus code as OH 03 was selected to evaluate the optimization of protease production against some independent variables. A Plackett-Burman design was used to determine the effects of 5 variables on the production of protease. The variables evaluated were yeast extract, peptone, $(NH_4)_2SO_4$, temperature and pH. The variables were screened in 15 trials at two levels for each variable (+1 and -1), with a triplicate of the center point. The proteolytic activity was determined using azocasein as substrate. The digestion of this substrate by a protease results in the formation of an orange component soluble which is analyzed in a spectrophotometer at 430nm. The Plackett-Burman design showed that the concentration of $(NH_4)_2SO_4$ and temperature were the variables that showed significance above 95% confidence level. The temperature were the variables that presented positive effect, while the $(NH_4)_2SO_4$ concentration had a significant negative effect on protease activity. In total, 15 runs were performed and the maximum activity (26,13 IU/mL) was found at second run of Plackett-Burman design. The result found was important due to the high production of proteases and could serve as a precursor for the large-scale production of proteases of interest to the industries.

**Keywords:** Endophytic fungi; microbial enzymes; protease; submerged fermentation; Plackett-Burman design.

**Development Agency: Fapdf, CAPES**