**TITLE:** STATISTICAL SCREENING OF MEDIUM COMPONENTS BY PLACKETT-BURMAN DESIGN FOR L-ASPARAGINASE PRODUCTION IN SOLID STATE FERMENTATION BY FUNGUS ISOLATED FROM BRAZILIAN CERRADO

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**ABSTRACT:**

L-asparaginase is an enzyme clinically accepted as an antitumor agent to treat acute lymphoblastic leukemia. The treatment with L-asparaginase leads to a reduction of asparagine levels, resulting in cytotoxicity to leukemic cells. Asparaginase is widely distributed in nature and microorganism like fungi and bacteria have proved to be very efficient and inexpensive sources of the enzyme. The use of solid state fermentation (SSF) process is being reported by several researchers as an alternative to submerged fermentation. The objective of the present work was to enhanced production of L-asparaginase from fungus isolated from Brazilian Cerrado using wheat bran which is a cheap agricultural residue byproduct in SSF. The Plackett-Burman (PB) design was used to determine the effects of 11 variables in the production of L-asparaginase by fungus encoded 2DSST1. The variables were screened in 12 trials at two levels for each variable (+1 and -1), with triplicate repeats at the center point. The variables evaluated were the L-asparagine ($X_1$), L-proline ($X_2$), the wheat bran ($X_3$), potato dextrose broth ($X_4$), ammonium sulphate ($X_5$), the temperature of incubation ($X_6$), the time of fermentation ($X_7$), the initial pH of the culture medium ($X_8$), yeast extract ($X_9$), sucrose ($X_{10}$) and the glucose concentration ($X_{11}$). Asparaginase assay was performed for the formation of β-aspartyl hydroxamate. One unit of asparaginase is defined as the amount of enzyme that formed 1 µmol of β-aspartyl hydroxamate in 1 min. The PB design showed that the concentration L-asparagine, L-proline, wheat bran, potato dextrose broth, yeast extract and glucose were the variables that showed significance above 95% confidence level. The L-asparagine, L-proline, potato dextrose broth and sucrose were the variables that presented positive effect, while the residue and yeast extract concentration had a significant negative effect on L-asparaginase activity. If the component showed significance at or above 95% confidence level and its effect was negative, it indicated that the component was effective in L-asparaginase production but the amount required was lower than the indicated as low (-1) concentration in PB design. If the effect was positive, a higher concentration than the indicated high value (+1) concentration was required during further optimization studies. Therefore, the fungus 2DSST1 exhibits potential for the production of L-asparaginase in SSF.

**Keywords:** L-asparaginase, solid state fermentation, filamentous fungi, Plackett-Burman design, Cerrado.

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