

## **Abstract**

Various sources such as soil, cereals (corn and oat), pulses (soybean), and vegetables (potato) were used to isolate a bacterial strain capable of producing L-asparaginase. The strain isolated from soybeans, referred to as MRS4, was found to produce the highest amount of the enzyme. MRS4 was characterized and found to have significant similarity with *Bacillus paramycoides* strain MCCC 1A04098 based on nucleotide homology and phylogenetic analysis. Various process parameters such as inoculum percentage, culture medium, pH, agitation speed, and aeration rate were optimized to maximize L-asparaginase production. The selected conditions resulted in the production of approximately 118 IU/mL of the enzyme. The research investigated the impact of different carbon sources (e.g., glucose), nitrogen sources (e.g., peptone), and minerals/metal ions (e.g., Di-Potassium phosphate) on L-asparaginase production. The optimal conditions for each were determined. RSM was used to optimize the operating parameters such as pH, temperature, fermentation time, inoculum percentage, and medium volume for maximum L-asparaginase production in batch scale. The RSM model showed good agreement with experimental results. The study also examined the effect of aeration rate, agitation speed, and medium volume in a laboratory-scale fermenter. These parameters were optimized to maximize L-asparaginase production. The enzyme was purified using ammonium sulfate fractionation, dialysis, and high-performance liquid chromatography (HPLC). The properties of the purified L-asparaginase, including pH and temperature stability,  $V_{max}$ ,  $K_m$ , molecular weight, and identification by SDS-PAGE and LC-MS, were determined. Application in Reducing Acrylamide in Foods: The purified L-asparaginase was tested for its ability to reduce acrylamide levels in various food samples (French fries, Nimki, fried fish, and fried chicken). The enzyme showed promise in reducing acrylamide levels, potentially mitigating the cancer risk associated with acrylamide in foods. The research provides valuable insights into the production and application of L-asparaginase for potential food safety and health benefits.