Neural Network Modeling and Optimization of Parameters for Production of Amylase from *Bacillus subtilis* using Agro Residuals as Substrates.

ABSTRACT

Research for improving the quality and quantity of everything useful to mankind is very essential. Most of the research is through physical experimentation. In the present day of globalization and liberalization with high level of competition, proper planning and careful utilization of all resources like time and materials to get the desired research result at less cost is of primary importance. The industry cannot afford expensive experimental research contributing to the increase in product cost. When developing a fermentation process, designing a fermentation medium is of critical importance because the medium composition and conditions can significantly affect product concentration, yield and productivity. It can also affect the ease and cost of down stream product separation. Appropriate and meaningful analysis of experimental data to reach purposeful conclusion, so that the experiments are conducted at the minimum possible time with optimally required materials and conditions, would greatly reduce the R & D cost. Many statistical and non-statistical research tools are now available for proper design of experiments.

The present study involved the utilization of agricultural residues like banana peel, wheat bran, rice bran and corn pith for α-amylase production using the bacterial strain *Bacillus subtilis* MTCC 441. The optimization of process parameters were done by the following three methods.
1. Taguchi method with orthogonal array experimental design and signal-to-noise ratio analysis with larger-the-better approach.

2. Response surface methodology with central composite design of experiments for second order polynomial model, response surface analysis and optimization.

3. Artificial neural network model with gblSolve version of global optimization algorithm DIRECT, in Matlab environment

Six parameters: substrate concentration, peptone concentration, pH, temperature, incubation time, inoculum level, at five levels were considered for this research study after considerable literature survey. Central points for the experimentation were decided through preliminary experiments.

The optimized results predicted by all the three methods were higher than the corresponding experimental design values, and were in agreement with the experimental values obtained by the corresponding optimal levels of the parameters. This proved the adequacy, accuracy and efficiency of all the methods. However, optimum levels predicted by artificial neural network gave the highest yield compare to Taguchi method and response surface methodology.

Maximum \( \alpha \)-amylase activity of 1832 U/ml was obtained by shake flask experiment conducted at the optimized levels of parameters determined by artificial neural network for wheat bran as substrate. It is closely followed by banana peel with 1742 U/ml. The corresponding values for rice bran and corn pith were 1287 and 558 U/ml respectively. These values were 41, 59, 51 and 51 percent more than the maximum activity obtained during designed experiments.
The α-amylase activity obtained by experiments conducted at the optimum levels determined by Taguchi techniques (orthogonal array experimental design) were 1544, 1580, 1212 and 530 U/ml for wheat bran, banana peel, rice bran and corn pith respectively which were 34, 41, 57 and 99 percent more than the corresponding values obtained during regular orthogonal array experiments. Optimized α-amylase activities by response surface methodology were 1326, 1321, 1123 and 503 U/ml for wheat bran, banana peel, rice bran and corn pith respectively, which were 7, 15, 32 and 36 percent more than the central composite design experimental values.

The present study shows that optimization by artificial neural network is better than Taguchi and response surface methodology, although each method has its own advantages and disadvantages and may not be suitable for all situations.

The experiments with 2L bioreactor at the optimal level of parameters points gave enhanced α-amylase activities of 2672, 2316, 1759 and 1325 U/ml which were 46, 33, 37 and 137 percent more than the corresponding shake flask experiments at the same optimized levels of the fermentation conditions. This shows the usefulness of conducting the experiments in a scaled up bioreactor of various sizes with aeration, agitation and proper control of other process parameters.