

Nisa Romsomsa 2014: Improvement of Silk Degumming Protease Production from *Bacillus* sp. C4 SS-2013. Doctor of Philosophy (Microbiology), Major Field: Microbiology, Department of Microbiology. Thesis Advisor: Assistant Professor Patoomporn Chim-anage, Dr.Eng. 184 pages.

Bacillus sp. C4 SS-2013 isolated from the wastewater of a silk factory is a promising silk degumming protease producer. The optimization of the aeration and agitation rates for protease production by batch culture using a central composite design and response surface methodology was investigated. Both the agitation and aeration rates significantly affected the protease production, specific protease production rate and K_{La} . The maximum protease activity of 1,890 U/mL and a specific production rate (q_p) of 21,412 U/g/h were obtained at the agitation and aeration rates of 400 rpm and 2 vvm which corresponded to a K_{La} value of 182.16 h^{-1} . These experimental values of protease production, q_p , μ and K_{La} were coincident with the predicted values and the models were proved to be adequate with the determination coefficient (R^2) of 0.950, 0.919, 0.931 and 0.881, respectively. The unstructured models provided the good approximation of cell growth, protease production and substrate concentration kinetic profile of the batch fermentation. The data fitted the proposed mathematical model very well. The R^2 values were found to be 0.908, 0.887 and 0.990 by Logistic model for cell growth, Luedeking-Piret model for protease production and Luedeking- Piret like model for substrate utilization, respectively. The improvement of protease production with pH-stat fed-batch culture resulted in an increase of the silk degumming protease production to 4,437 U/mL (2.35-fold) and q_p to 36,820 U/g/h (1.72-fold). Degumming of raw silk yarn by the enzyme obtained caused the weight loss of raw silk about $25.31 \pm 2.98\%$ which corresponded to $92.76 \pm 1.16\%$ sericin removal. The addition of 10 mM CaCl_2 into the crude protease and storage at 4°C for 8 weeks could maintain its activity at about 64.25% of the initial activity. According to these data, the improvement of production and stabilization during storage of silk degumming protease were achieved in our study and has potential for industrial applications.

Student's signature

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