Watinee Kunpeuk 2011: Screening, Characterization and Optimization for Carboxymethylcellulase Production by Thermophilic Bacteria from Soil. Master of Science (Biotechnology), Major Field: Biotechnology, Department of Biotechnology. Thesis Advisor: Associate Professor Mangkorn Rodprapakorn, Ph.D. 122 pages.

A total of 121 thermophilic bacteria were isolated from 9 cellulose-decomposing soil samples screening on Nutrient agar with 1% (w/v) carboxymethylcellulose (CMC) at 50°C for 48-72 h of incubation. The isolates were selected in primary screening and examined their carboxymethylcellulase (CMCase) production by using a Congo red test. The result showed positive clear zone of 41 isolates on the Nutrient agar with 1% (w/v) carboxymethylcellulose (CMC). All isolates were observed for CMCase production in Nutrient broth containing 1% (w/v) CMC as a carbon source at 50°C for 48 h. The isolate PA1-1 showed the highest CMCase activity of 0.60 U/ml with Avicelase activity of 0.08 U/ml. Based on its morphology and molecular identification, the organism was classified as *Thermobifida fusca*. Study on the association between the growth of *T. fusca* PA1-1 and cellulase production in different medium was found that CMCase production induced in Basal medium 3 (Lima *et al*., 2005) at 0.95 U/ml which result was similar to Basal medium 1 whereas the highest Avicelase production was detected in this medium at 0.605 U/ml. Study on the effect of nitrogen sources to cellulase formation was carried out in medium 3. The result showed baker yeast was the most effective nitrogen source which gave the higher CMCase induction than yeast extract, peptone and malt extract respectively whereas yeast extract gave the highest Avicelase production. Characterization of CMCase activity showed the optimum pH was 4.0 and the highest relative activity was found at 60°C. The enzyme was stable in the pH range 4.0-10.0 and the residual activity retained up to 80% at 30-60°C. The optimization of CMCase production was studied using the response surface methodology with temperature and pH. It was found that the most significant factors influencing enzyme production were both temperature and pH ($p < 0.05$). The second order polynomial regression model was obtained with an R^2 of 0.8060 ($p < 0.05$). From the result of the optimization, maximum CMCase activity at 0.859 U/ml was achieved at temperature 50 °C and pH 8.0. To confirm te applicability of the model, *T. fusca* PA 1-1 was cultured at this condition in a flask as compared to a fermenter. Carboxymethylcellulase activities were measured at 0.977 and 1.21 U/ml respectively.

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