

Studies on NattoKinase Production by *Bacillus subtilis*

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ABSTRACT

Bacillus subtilis var. natto with the high activity of nattokinase (NK), which can degrade fibrin, was isolated from commercial natto foods of Japanese. It was found that the optimum composition of the culture medium for high NK activity was investigated to be 8.0% glucose, 3.0% soybean protein, 2.0% yeast extract, 0.4% K₂HPO₄ and 0.2% CaCO₃. Furthermore, the highest NK activity of the fermented supernatant liquid was observed as about 900~1000 FU/mL under the 5-L fermentation conditions of temperature at 37 °C, pH controlled at pH 6.5, agitation at 300rpm, aeration rate at 2vvm after 48h of fermentation. For the crude NK activity test of the fermented supernatant liquid, the optimal reaction temperature and pH was at 50~60 °C and at the range of pH 6.5~10, respectively. The thermal stability of the crude NK was at 30~40 °C. During storage of the crude NK at 0~4 °C for 30 days, it still had close 80% of activity. For the acute toxic test of animal security experiment, the powder of the fermented supernatant liquid with crude NK was carried out for the oral administration of SD mouse. There was not any obvious toxic symptom for SD mouse after its oral administration dosage up to 5000 mg/kg.

Keywords : *Bacillus subtilis* ; liquid fermentation ; nattokinase (NK) ; enzymatic activity ; acute toxic test of animal security experiment

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