

Study on Enzyme Activities of Fermented Supernatants of Two Native Strains Isolated from Konjac Degradation

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ABSTRACT

The two better native strains of dyu-cs-3 and dyu-cs-6, isolated from konjac degradation, were used to preliminarily study their cultural conditions and enzymatic activities of the fermented supernatants for degrading (hydrolyzing) the commercial konjac gel. It was found that the optimal cultural temperature and initial pH of cultural media for the two strains were at 37°C and 8.0, while the suitable contents of cultural media with 3% konjac gel in 250mL flasks for dyu-cs-3 and dyu-cs-6 were 50mL and 75mL, respectively. For dyu-cs-3 and dyu-cs-6, the highest concentrations (0.15 and 0.16mg/mL, respectively) of reducing sugar were obtained at 48 h of cultivation. After 10 min centrifugation (10000 × g) at 4°C, the enzymatic activity of the fermented supernatants for degrading konjac gel was investigated. The optimal reaction temperature and pH were at 40°C and 8.0, respectively, while the activity decreased significantly at higher reaction temperature (50°C or higher) and lower pH (pH 7.0 or lower). The thermal enzyme stability of the dyu-cs-3 supernatants was only at 30°C, while that of the dyu-cs-6 ones was at 30~40°C. The thermal stability of the dyu-cs-3 enzyme seemed be better than that of the dyu-cs-6 one. For the enzymatic pH-stability test at room temperature, the pH-stability range of the dyu-cs-3 and dyu-cs-6 fermented supernatants was at pH 5.0~7.0, and their enzymatic activities retained about 70~80% for first 120 min treatment. The optimal releasing rate of reducing sugar from the substrate-konjac gel was at first 30 min for the dyu-cs-3 and dyu-cs-6 enzymes at pH 8.0 and 40°C. The storage temperatures of the enzymatic activities of dyu-cs-3 and dyu-cs-6 fermented supernatants were also investigated. During 7-day storage the dyu-cs-3 enzyme still had about 80-100% of activity at 6°C and 30°C, while the dyu-cs-6 enzyme had close 100% of activity at 30°C and had close around 80% at 6°C. For the reaction kinetics of the enzymes of the two fermented supernatants, the reaction constants for both the dyu-cs-3 and dyu-cs-6 enzymes were very similar. The reaction constants, Km and Vmax, for the dyu-cs-3 enzyme were 1.614mg/mL and 0.015mg/mL·min, while the reaction constants, Km and Vmax, for the dyu-cs-6 one were 1.617mg/mL and 0.014mg/mL·min, respectively.

Keywords : konjac ; reducing sugar ; enzyme activity ; microbial degradation (hydrolysis)

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