Production of Levan Using Immobilized Techniques

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ABSTRACT

This study is regarding the immobilization of Bacillus subtilis natto Takahashi, a commercial natto spore, on alginate and the fermentation of immobilized cells in the medium containing sucrose. The stability and the levan production of the immobilization cell particles during the fermentation were also studied.

The stability and the levan production of the immobilization cells in the process of fermentation were affected by the medium pH, metal ions and agitation speed. The immobilized cell particles were stable and the levan production was high at slightly acidic condition (pH 5.0-6.0). The integrity of the immobilized cell particles was maintained when the certain level of metal ions was kept, while the high metal ions concentration inhibited the cell growth and thus the levan production. High stability of cell particles and high levan production were observed when the agiation speed was kept between 100 to 175 rpm; however, the cell particles collapsed at 225 rpm. When the immobilized cells was cultivated in 300 ml of SM medium (Sucrose：250 g/L, $MgSO_4 \cdot 7H_2O$ ：0.5 g/L, $NaH_2PO_4 \cdot 2H_2O$：3 g/L, $Na_2HPO_4 \cdot 12H_2O$：3 g/L, $CaCl_2$ 0.2%, $AlCl_3 \cdot 6H_2O$ 0.1%) at optimal pH (5.6-6.8), temperature (37 ℃) and agitation speed (150 rpm), high leva production (60-70g/L) was obtained.

In the repetitive cultivation of the immobilized cells in the SM medium, it was found that after the second batch the cell activity and the levan production declined dramatically. However, the levan production was elevated when the initial pH was adjusted to 5.6-5.8 and 10% of organic nitrogen source was added at the beginning of each batch fermentation. It is obvious that the presence of organic nitrogen source is critical for high levan production. At the optimal medium contents (Sucrose：250 g/L, $MgSO_4 \cdot 7H_2O$ ：0.5 g/L, $NaH_2PO_4 \cdot 2H_2O$：3 g/L , $Na_2HPO_4 \cdot 12H_2O$：3 g/L, $CaCl_2$ 0.2 %, $AlCl_3 \cdot 6H_2O$ 0.1 %, 10 % NB medium compound) and the optimal culture condition (pH 5.6-5.8, 37℃, 100 rpm), high levan production (60-70 g/L) was obtained after 3 d of fermentation. The immobilized cells were still active and stable even after 5 repetitive fermentation, and 55-70 g/L of levan were obtained for each batch.

It is shown that immobilized Bacillus subtilis natto Takahashi has the advantages of high levan production even after long term and repetitive fermentation.

Keywords : Bacillus subtilis natto、poly fructose、levan、alginate、immobilized

Table of Contents

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