

The purification and application of protease produced from shrimp and crab shell wastes by *Monascus purpureus* CCRC31499

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ABSTRACT

The production of useful protease by *Monascus purpureus* CCRC31499, using shrimp and crab shell powder as the major carbon source, was studied. The optimized conditions for protease production was found when the culture was shaken at 37 °C for 4 days in 100mL of medium (pH7) containing 2 % shrimp and crab shell powder, 0.15 % poly-peptone, 0.1 % yeast extract, 0.1 % K₂HPO₄ and 0.05 % MgSO₄ · 7H₂O. The protease of *Monascus purpureus* CCRC31499, produced under the optimized culture condition, the first step was precipitated and dialyzed by using ammonium sulfate. The further purification and separation procedures of the protease were processed by the use of DEAE-Sepharose ionic exchange chromatography、Sephacryl S-200 gel permeation chromatography and chromatofocusing. Purification was 25-fold with the crude enzyme solution. After purification and separation, the activity of the protease was still stable at pH 5-11, while the optimal temperature and pH for the enzyme reaction were at 50 °C and pH9, respectively. By SDS-PAGE electrophoresis and gel permeation chromatography, the molecular weight of the protease was identified as 40kDa. The activity of protease was strongly inhibited by PMSF, and showed as a Serine protease. A protease-producing microorganism *Monascus purpureus* CCRC31499 can use acid or alkaline treated shrimp and crab shell powder (SCSP) induced protease production than untreated SCSP. The enzyme activity was 21.8U/mL when HCl treated SCSP was used, as compared to 19.1U/mL when untreated SCSP was used. The properties of composts made by bio-fertilizer crab and shell wastes with *Monascus purpureus* CCRC31499 were studied. The effects of the composts on the growth of Chinese cabbage were observed. The length and fresh weight of the whole plant were grown enormously; they are 155 % and 521 % in comparison with the control. Expect the culture supernatant of *Monascus purpureus* CCRC31499 is good for field test.

Keywords : *Monascus purpureus* ; protease ; shrimp and crab shell waste ; bio-fertilizer

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