PURIFICATION OF FRUIT BROMELAIN FROM PINEAPPLE (*Ananas comosus*) PEROLERA VARIETY

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Bromelain, pineapple proteolytic enzyme, shows pharmacological activity for several diseases, such as respiratory disturbs, allergic rhinitis, treatment of burns, anti-inflammatory and anti-tumoral activities and ulcerative colitis. The objective of this work was extrating and purifying the fruit bromelain (E.C. 3.4.22.33), a glycoprotein, from *Ananas comosus* (pineapple) perolera variety. The fruit pineapple extract was concentrated with ammonium sulphate 80% (w/v) and dialysed, showing a purification factor of 2.45-fold with 173.5% of activity recovery. The molecular exclusion chromatography in ÄKTA-FPLC system (column TSK4000SW) showed two peaks. The first peak was in 104.15 mAU containing 8 fractions but there was no proteolytic activity and the second one in 359.49 mAU containing 5 fractions (F-6, F-7, F-8, F-9, F-10) with proteolytic activity. The major proteolytic activity was found in the fraction F-6 (40.73 U/mg proteins). Total activity of the second peak was 95.44 U/mg protein representing 18.5 fold of purification compared to concentrated extract. The concentrated extract and the 5 fractions pool were analysed by electrophoresis SDS-PAGE showing two bands about 26 kDa and some bands of low-molecular-mass proteins (14 kDa), which both showed proteolytic activity according to zymogram assay. The pineapple fruit extract showed various active protein fractions, which will be analysed using mass spectroscopy (MALDI-TOF) for determining their molecular weight and peptide composition after digestion by specific enzymes.

Key words: Bromelain, *Ananas comosus*, proteolytic
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