PURIFICATION AND PARTIAL CHARACTERIZATION OF AMINOPEPTIDASES FROM *Caesalpinia echinata* (PAU-BRASIL) SEEDS

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Aminopeptidases, enzymes that participate in the final stages of protein degradation, hydrolyze peptide bonds yielding amino acids from N-terminal peptides and aminoacyl-2-naphthylamides (AA-NA) yielding naphthylamines. The real importance of aminopeptidases from plant seeds is unknown probably they are involved in seed germination. The aim of this work is to study some aminopeptidases from *C. echinata* seeds (pau-brasil). Phosphate buffer (PB) 0.02M, pH 7.0 was added to ground seeds (5mL/g) and the extract was centrifuged at 4°C. The proteins were separated in a DEAE Cellulose Cellex D column equilibrated in the same buffer. Elution was performed with a linear gradient of PB, pH 7.0 (0.02-0.3M). The only protein peak, eluted at 710µS with activity on AA-NA (P_1), was purified in an octyl Sepharose fast flow column equilibrated with 0.02M PB, containing 3M KCl. Elution was performed with a linear gradient of KCl (3.0 to 0M) in the same buffer, followed by water and 30% i-propanol. Two protein peaks were obtained (P_1a and P_1b) in 15mS and 3.7mS, respectively. P_1a and P_1b were filtrated - Superdex 200 column in 0.02M PB, pH 7.0, containing 0.15M NaCl. Finally, two different protein peaks, P_{1a1} and P_{1b1}, with enzyme activity was obtained. Electrophoresis SDS-PAGE (10%) was performed and showed a molecular mass around 20 kDa to $P1_{a1}$ and 31kDa to $P1_{b1}$. These results shows that there are at least two aminopetidases in *C. echinata* seeds.

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