Biotechnology potencial estudy of Leguminoseae proteases

Telles, K.F.\textsuperscript{1}, Gonçalves, R.N.\textsuperscript{2}, Caldeira, A.S.P.\textsuperscript{2}, Silva-López, R.E.\textsuperscript{2}

\textsuperscript{1}Universidade Federal do Rio de Janeiro, Rio de Janeiro, Brasil; \textsuperscript{2}Instituto Oswaldo Cruz, Fiocruz, Rio de Janeiro, Brasil;

**Introduction:** *Canavalia ensiformis* or “Jack bean” is a legume with wide distribution in the tropical regions of the world. This plant is used as a green cover and also serves as food for human and animal. Its seeds are rich sources of proteins with biotechnological interest such as ureases and proteases and, Concanavaline A. **Objectives:** Study the biochemical and kinetic characteristics of *C. ensiformis* steam extract and isolate and partial characterization of serine proteases from this extract. **Material and Methods:** Fresh steams were processed, in a blender, using destilled water. The material was centrifuged and lyophilized obtaining an aqueous steam (CE-CA) extract which as dialyzed against water and submitted to affinity chromatography on Aprotinin-Agarose. Protein measurement, eletrophoretic analysis and kinectic assays were performed. **Results and Discussion:** CE-CA exhibited 34.21\% of protein and good activity against L-\textalpha;-Tosyl-arginil-methyl-ester and protein substrates such as serum albumin bovine, casein, gelatin and native collagen I. CE-CA showed peaks of maximal activity was found at pH 5.0, 7.5 and 9.0 and the purified enzyme has the maximum activity at pH 7.5 and 9.0. SDS - PAGE analysis of CE-CA showed different profiles under reducing and non-reducing conditions with major bands of about 80-28 kDa under reduction and, 80-34 kDa under non-reducing conditions. Furthermore, proteins of about 140 to 75 kDa under non-reducing conditions and, about 85 to 65 kDa proteins under reducing conditions, exhibited important proteolysis in SDS-PAGE-gelatin. Purified serine protease showed about 90 kDa and 135 kDa under reducing and non-reducing conditions, suggesting that has more than one polypeptide chain. **Conclusions:** Investigation of plant proteases is substantial to know their functions and evaluate the possibility of their employment for biotechnological purposes.

**Key-words:** *Canavalia ensiformis*, serine protease, steam extract.