EVALUATION OF PROTEASES AND INHIBITORS FROM THE SALIVA OF Amblyomma cajennense TICK (ACARI: IXODIDAE) RELATED TO BLOOD COAGULATION AND EXTRA CELLULAR MATRIX

<u>Simons, S.M.¹</u>; Oliveira, D.G.L.¹; Faria, F.¹; De-Sá-Júnior, P.L.¹; Silva-Ricci, M.E.¹; Valente, R.H.²; Leon, I.R.²; Perales, J.²; Chudzinski-Tavassi, A.M.¹.

Lab. Bioquímica e Biofísica. Instituto Butantan, São Paulo, SP¹. Dep. Fisiologia e Farmacodinâmica. Instituto Oswaldo Cruz, Rio de Janeiro, RJ².

Proteins isolated from the saliva of hematophages are useful tolls for the understanding of many physiological processes. In the study we characterize the cytotoxic and anti-coagulant activities of proteases and inhibitors present in the saliva of Amblyomma cajennense tick. The profile of saliva's proteins was also analyzed by 2D electrophoresis followed by mass spectrometry or, by ion exchange chromatography in a FPLC system. Fractions from the chromatographic steps were investigated concerning haemostatic parameters (coagulation, fibrinolysis and platelet aggregation) and cell survival. Saliva was also incubated with proteins (fibrinogen, FX, FXa, FII, thrombin, plaminogen, lisozyme, collagen, BSA, plasmin) and submitted to SDS-PAGE. The saliva induces fragmentation of fibrinogen, plasminogen and forms complex with FXa. 110 spots were revealed in the 2D (pl 3.5 and 9.5 and MW from 12 to 160 kDa) and have been analyzed by MS-MS. The peaks (I-VII) from ion exchange chromatography present independent activities on coagulation and cell cytotoxicity. The sequences found in the spots will be blasted against cDNA library data of A. cajennense constructed by our group, Genbank (EC778445 to EC780213). Key words: Amblyomma cajennense, inhibitors, proteases Supported by FAPESP, FINEP and CNPq.