LATEX OSMOTIN PURIFICATION BY IMMUNOAFFINITY CHROMATOGRAPHY AND ANTIFUNGAL ACTIVITY EVALUATION

Silva, M.Z.R.¹; Viana, C.A.¹; Moreno, F.B.M.B.²; Monteiro-Moreira, A.C.O.²; Freitas, C.D.T.¹; Ramos, M.V.¹

¹Departamento de Bioquímica e Biologia Molecular, UFC, Ceará, Brazil
²Núcleo de Biologia Experimental, UNIFOR, Ceará, Brazil

Introduction: Proteins that share similar primary sequences to the protein originally described in salt stressed tobacco cells have been named osmotins. Some of these proteins were reportedly induced under biotic or abiotic stresses. Objectives: The aim of this study was to investigate and isolate osmotins from Cryptostegia grandiflora (CgLP), Plumeria rubra (PrLP), Thevetia peruviana (TpLP), Himatanthus drasticus (HdLP) and Carica papaya (CapLP) latex and evaluate their antifungal activities. Materials and Methods: The osmotins were obtained through a new isolation method using immunoaffinity chromatography with anti-CpOsm (osmotin from C. procera latex - CpLP) antibodies. The osmotin-like proteins obtained were identified and characterized by ELISA, Dot blot, Western blot, SDS-PAGE and mass spectrometry and the antifungal activities were evaluated. Results: Osmotin-like proteins did not detect in Thevetia peruviana and Carica papaya latex. They were found in CgLP and PrLP by ELISA, Dot blot and Western blot using anti-CpOsm antibodies and were identified by mass spectrometry. The osmotin from HdLP was detected only by mass spectrometry. The osmotin from CpLP was co-purified with cysteine protease identified as Procerain B. The alignment and the 3-D structure analysis of Procerain B and CpOsm revealed the presence of a similar sequence in both proteins. This sequence might be an epitope which allows the anti-antibody recognition. The osmotins from CgLP, PrLP and HdLP didn’t show antifungal activity. Conclusions: Such as other plant osmotins, osmotins from latex fluids can be essential as osmotic or hydric regulators and not as defensive molecules. Further assays will be performed to study this hypothesis.

Keywords: PR-protein, laticifers plants, immune assays.
Acknowledgements: CNPq and CAPES.