Characterization of the interactome profile of human Stanniocalcin-1

Santos, M.T. [1,2], Trindade, D.M.[1,2], Yunes, J.A.[2,3] and Kobarg, J.[1,2]

[1] Centro de Biologia Molecular e Estrutural
Lab. Nacional de Luz Síncrotron (LNLS), Campinas, SP, Brasil

[2]Depto. de Genética e Biologia Molecular
Instituto de Biologia - UNICAMP, Campinas, SP, Brasil

[3] Centro Infantil Boldrini, Campinas, SP, Brasil

The human stanniocalcin-1 (STC-1) is a glycoprotein that has been implicated in different physiological process, including angiogenesis, apoptosis and carcinogenesis. STC-1 mRNA was identified in the blood serum of leukemia patients but the presence of the protein could not be confirmed. Since leukemia cells interact with the bone marrow (BM) microenvironment, providing them proliferative advantages, a microarray gene expression analysis was performed using BM stromal cells co-cultivated with primary ALL cells. This and Real -Time PCR analysis, showed that STC-1 has a significant increase of expression in BM stromal cells after co-culture with leukemia cells. Our current aim is to discover the STC-1 receptor and other interacting proteins for further functional characterizations. For this, yeast-two-hybrid-system screens of human cDNA libraries were performed and interesting interacting protein candidates were found. Pull-down assay confirmed some of the interactions and a mapping study, using STC-1 Carboxy- and Amino-terminals, is ongoing. These parts of the protein were cloned and expressed into BL21 bacteria strain (6xHis and GST fused). Since both had an insoluble expression, a refolding assay from inclusion bodies was performed. The full-length protein, expressed in the baculo-virus system, will be used for immunization and together with the refolded carboxy-terminal used for hybridom screening. The antibodies will then be used in diagnostic ELISA tests important for leukemia diagnostic and prognostic and for the confirmation of STC-1 protein in blood serum, which may indicate its possible hormone like actions.

KEY WORDS: Stanniocalcin-1; Leukemia; Protein Interaction
Financially supported by: FAPESP, CNPq and LNLS