

EVALUATION OF PHAGE DISPLAY SYSTEM AS A TOOL FOR SELECTION AND EXPRESSION OF THE SMALL TRYPSIN INHIBITOR BASED ON BvTI - *Bauhinia variegata* TRYPSIN INHIBITOR

Souza, A.F., Sampaio, C.A.M., Tanaka, A.S.

Departamento de Bioquímica, UNIFESP-EPM, Rua 3 de Maio 100, 04044-020
São Paulo, SP, Brazil. E-mail: afsouza-leoni@uol.com.br

Recently, we described the cloning of BvTI cDNA fragment and its expression by *E. coli* cells using pET-14b vector, in an active form. (de Souza et al., 2005). Furthermore, we also expressed BvTI and its reactive site region (RS-BvTI) as fusion proteins in a phage display system to generate a small inhibitor based on a Kunitz type inhibitor. Our results showed a RS-BvTI displayed on the M13 surface in an active form while BvTI was not expressed in an active form or it was expressed in low concentration not detectable by ELISA assay. In the present work, we are going to express the RS-BvTI in different expression systems to obtain enough material for kinetic studies. Our first result using the *E. coli* HB2151 strain to express RS-BvTI as recombinant protein showed no expression of this protein using the same vector construction selected by phage display system. In the moment, we constructed new pair of oligonucleotides to clone the RS-BvTI into the pET-14b vector successfully used to express the BvTI. The perspectives of this work after the functional expression of RS-BvTI, is to use, this inhibitor and phage display system to construct a RS-BvTI variants library. Supported by FAPESP and CNPq.