EXPRESSION AND PURIFICATION OF HUMAN SPI-C ETS DOMAIN

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The Ets proteins are a family of transcription factors containing a winged helixturn-helix DNA binding domain. This 85 amino acid region is composed of three ahelices and a four-stranded ß-sheet that recognizes a binding site with a GGAA/T core sequence. These Ets proteins act as positive or negative regulators of the expression of genes involved in cancer development. The Spi-C protein is a member of the Ets family expressed temporarily during B-cell development and in macrophages and seems to promote B cell differentiation, whose expression is confined to particular stages of B cell development. The DNA sequence encoding the human Spi-C Ets domain was constructed, cloned into the expression vector pET23a(+) and initial expression tests in *Escherichia coli* were carried out. Here we describe expression optimization and purification of the recombinant protein. The Spi-C Ets domain corresponded to 11% of the total soluble protein fraction using the *E.coli* BL21 (DE3) strain. We also present protein purification protocols involving nucleic acid precipitation and liquid chromatography to obtain homogeneous recombinant protein. The latter will allow DNA binding and structural analysis studies to be carried out.