Expression of Recombinant ADAM33 Protein Coding the Cysteine Rich Domain

Manica, G.C.M.1; Ramos, E.A.S.1; Klassen, L.M.B.1; Oliveira, M.A.S.2; Pedrosa, F.O.2; Souza, E.M.2; Klassen, G1.

1 Dep. de Patologia Básica, Laboratório de Epigenética, 2 Dep. de Bioquímica e Biologia Molecular, UFPR, PR, Brazil

Breast cancer is the most common malignant tumor among women and it is the leading cause of female cancer death. It is classified into subtypes by its morphology, molecular characteristics and clinical outcome. Diagnosis and prognosis is performed using established immunohistochemical protein markers such as estrogen and progesterone receptors that indicate the therapy to be recommended. The invasive lobular carcinoma (ILC) accounts for up to 15% of the cases with atypical morphology and difficult classification. The loss of E-cadherin expression reflects the histological morphology of the ILC subtype. However, only 10 to 56 % of this type of tumor shows loss of expression of E-cadherin, and up to now there is no specific marker for ILC. ADAM33 has decreased expression in the ILC due to hypermethylation of it promoter. Therefore, this protein could be used as a new marker for differentiation of ductal and lobular carcinomas. The DNA region coding has amino acids 1586 to 2197 of the ADAM33 (611 bp), that contains part of the disintegrin and cysteine rich domains, was cloned into pGEMT®easy, the plasmid obtained was digested with EcoRI and the insert subcloned into the vector pET28a. The induction of the truncated ADAM33 protein (25,7 kDa) was performed in E. coli BL21Ai using arabinose and confirmed by western blot with anti-poly-histidine. The protein was purified using the HisTrap Ni-Chelating column with a yield of 0,4 µg/µl of soluble protein. Anti-ADAM33 polyclonal antibodies are expected to differentiate between lobular and ductal breast cancer tumors immunohistochemically.

Word Keys: ADAM33, invasive lobular carcinoma (ILC), immunohistochemical protein markers
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