Down-regulation of the metastatic function of β- catenin by estrogen and estrogen receptor-α in HA22T hepatocellular carcinoma cells

Wei-Wen Kuo[1], Hwai-lee Wang[2], Chih-Yang Huang[2,3,4]*,

[1] Department of Biological Science and Technology, [2] Graduate Institute of Chinese Medical Science, [3] Graduate Institute of Basic Medical Science, China Medical University. [4] Department of Health and Nutrition Biotechnology, Asia University

Abstract

Reports indicate the incidence of hepatocellular carcinoma (HCC) is higher in men than in women. Our previous data show that estrogen and estrogen receptor α (ERα) significantly suppressed Hep3B cell proliferation. In addition to higher levels of β-catenin translocating into nucleus in tumor areas, and appearing more significantly at the late stages of HCC, β-catenin gene knocked down by antisense oligonucleotides resulted in the inhibition of cell migration and invasion in HA22T HCC cell line. To identify the anticancer effect of estrogen and ER-α to mediate HCC cell motility is through the suppression of β- catenin expression, we co-transfected pCMV- β- catenin and ER-α into HA22T cells, and determined the cell motility by wound healing, invasion and migration assays . Results show that estrogen and /or ER-α inhibited β-catenin gene expression and repressed HA22T cell motility. Similar data were also observed using the model of cells with ER-α stable clone. Moreover ,we examined the protein-protein interaction between ER-α and β- catenin by immunostain, co-immunoprecipitation and western blotting analysis, and found higher level of β-catenin exported from nucleus and bound to E3 ligase,βTrCP, with the presence of ER-α to promote β- catenin protein ubiquitination and degradation. Additionally, the binding of ER-α with SP-1 site on β- catenin promoter was also identified by EMSA and Chip assays, further providing the evidence of the modulation of ER-α on β- catenin gene expression. Taken together, the metastatic function of β- catenin is down-regulated by estrogen and ER-α via gene suppression and protein instability in HA22T HCC cell line.