Perez White, Bethany; Zhao, Huiping and Tonetti, Debra A.

Overexpression of PKCα in Breast Cancer Cells Induces Migration Through p120-Catenin Transcriptional Downregulation

Several factors have been identified that influence epithelial-mesenchymal transition (EMT) in breast cancer cells. We previously reported that overexpression of PKCα in breast cancer predicts tamoxifen resistant secondary tumors (Tonetti, 2003). In breast cancer cells, stable overexpression of PKCα (T47D/PKCα) leads to hormone-independent growth as well as tamoxifen resistance (Chisamore, 2001). In our current study we report that PKCα overexpression increases the metastatic potential of breast cancer cells through induction of a nonclasssical EMT. T47D/PKCα cells are morphologically distinct from T47D/neo cells and do not retain the cuboidal structure of epithelial breast cancer cells. Overexpression of PKCα resulted in significantly enhanced migratory capabilities in the Boyden chamber assay. PKC activation by phorbol 12-myristate 13 acetate further enhanced migration in T47D/PKCα cells. Pharmacological inhibition with the classical PKC inhibitor Go6976 reduced the migratory capacity of T47D/PKCα cells. Transient siRNA-mediated knockdown of PKCα (120 h) significantly reduced both basal and NIH3T3 fibroblast conditioned media-induced migration in T47D/PKCα cells. Levels of adherens junction proteins E-cadherin, α-E-catenin, β-catenin and p120-catenin were significantly downregulated in T47D/PKCα cells compared to T47D/neo cells as determined by western blot. Levels of E-cadherin protein were restored by more than 50% after 168 h of PKCα knockdown in T47D/PKCα cells. E-cadherin transcripts analyzed by SYBR green RT-qPCR were significantly higher in T47D/PKCα cells compared to T47D/neo control cells while there was no change in expression of β-catenin or α-E-catenin transcripts. Only p120-catenin transcript levels were significantly lower. Treatment with proteasomal inhibitor MG132 induces accumulation of E-cadherin but not p120-catenin. Taken together, these data suggest that PKCα may be responsible for EMT in breast cancer cells through upstream signaling that leads to transcriptional inhibition of p120-catenin and subsequent degradation of E-cadherin. Further PKCα expression may not only be predictive of tamoxifen-resistance but of increased potential for metastasis.