Department of Oncology and Pathology

Post-translational modifications of proteins in human breast cancer: proteomics study of phosphorylation and nitration and implication of these PTMs in tumorigenesis

AKADEMISK AVHANDLING
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av

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Abstract

Breast cancer is the most common cancer in women. Even though improvements in diagnosis and treatment of breast cancer have been made, it is still the most common cause of cancer death in women. There is a great need to find biomarkers for early detection of the disease and novel drug targets to fight the cancer.

Cell immortalization is the prerequisite step in tumorigenesis, and identification of the biomarkers of immortalized cells may be helpful for early detection of cancer. In this thesis, we described identification of 71 immortalization-related proteins. We used proteomics and conditionally immortalized human breast epithelial cells. Identified proteins showed involvement in immortalization of such functional domains as cell proliferation and growth, death, cell assembly and organization, cellular movement, cell-to-cell signaling, and cell morphology. Kinase MAP2K3 was identified as down-regulated in immortalized cells. Overexpression of MAP2K3 in immortal human breast epithelial cells was sufficient to induce senescence. p38, p53 and pRB were modulated by MAP2K3. We also identified KSR2 as up-regulated in immortalized human breast epithelial cells and in human breast tumors.

Twenty-four proteins affected by hyperthermia of human primary breast epithelial cells were also identified. Among the proteins, TGF-β2 was found up-regulated. It induced HSP27 expression, and protected cells from cell death.

Aberrant protein tyrosine nitration has been associated with different diseases, including cancer. We explored changes in protein tyrosine nitration during the cell cycle, and observed that tyrosine nitration affected a number of cell cycle regulators.

Cross talk of different regulatory pathways may contribute to the resistance to the anti-cancer treatment. Targeting of multiple pathways has been regarded as a novel anti-cancer strategy. We showed that combined action of TGF-β1 and EGF involves changes in phosphorylation of 47 proteins. We observed that the convergence components of TGF-β1 and EGF, e.g., MEK1, CK1, can influence cell proliferation in the context of TGF-β1 and EGF signaling. Interestingly, we observed that the strongest inhibitory effect of Gefitinib (Iressa), EGFR kinase inhibitor, would be only when both EGF and TGF-β are highly active, and MEK1 and CK1 are inhibited. ZAK kinase was identified as a convergent target of TGF-β and EGF signaling, and was found contributing to the positive feedback regulation of cell migration upon combined TGF-β and EGF action.

We also studied effects of a long term exposure to EGF and estrogen on tumorigenesis of breast epithelial cells. We observed that the long-term exposure to EGF and 17β-estradiol may affect proliferation rate, colony formation, vessel formation, and stem cell features of human breast epithelial cells.

Thus, our findings provided insights into different mechanisms of tumorigenesis, and impact of cross-talk of signaling pathways on tumor development.

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