Karolinska Institutet

Institutionen för Biovetenskaper och Näringslära

Estrogen receptor β signalling in mammary epithelial and breast cancer cells

AKADEMISK AVHANDLING
som för avläggande av medicine doktorsexamen vid Karolinska Institutet offentligen försvaras i Hörsalen, plan 4, Novum, Huddinge

Fredagen den 17 december, 2010, kl 09.15

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Stockholm 2010

ABSTRACT

Estrogens are key players in the etiology and progression of breast cancer, and mediate their effects through the estrogen receptors (ERα and ERβ). ERα plays important roles in proliferation and progression of breast cancer, whereas a distinct function of ERβ in the initiation and development of breast cancer is not yet clearly established. The general aim of this thesis was to increase our understanding of the molecular and cellular mechanisms of estrogen signalling in the normal and cancerous breast, focusing on the potential anti-tumourigenic effect of ERβ. Using cell lines with endogenous expression or inducible expression of ERβ we have characterised possible pathways of how ERβ could mediate its anti-tumourigenic effects.

The role of ERβ in cell proliferation and cell cycle regulation has been characterised, mainly *in vitro*. In **paper I** we investigated how ERβ re-expression would affect breast cancer cells in *in vivo*. Presence of ERβ in breast cancer xenografts reduced tumour growth and the number of intratumoural blood vessels. Expression of the pro-angiogenic growth factors vascular endothelial growth factor and platelet-derived growth factor β were also reduced upon ERβ expression, both *in vitro* and *in vivo*. These findings suggested an anti-tumourigenic role for ERβ by inhibiting growth and angiogenesis.

Studies in ERβ$^{-/-}$ mice have suggested a role for ERβ in the regulation of cell adhesion. In **paper II** we looked at cell-cell adhesion with a focus on E-cadherin. We reported that decrease of ERβ in mammary epithelial cells was associated with a decrease of E-cadherin protein levels through different posttranscriptional regulatory mechanisms, including protein shedding, internalisation and degradation. This correlated with an increase in β-catenin transcriptional activity and impaired morphogenesis on Engelbreth-Holm-Swarm matrix. This study suggests that ERβ has an important role in maintaining cell adhesion and a differentiated phenotype.

In **paper III** we analysed the effects of ERβ on cell-extracellular matrix adhesion. We found that integrin α1 and integrin β1 levels increased in breast cancer cells following ERβ expression. Also, the formation of vinculin containing focal complexes and actin filaments was enhanced, correlating to a more adhesive potential as seen by adhesion to ECM proteins. Furthermore, the migratory potential of the breast cancer cells was decreased upon ERβ expression. This study indicates that ERβ affects integrin expression and clustering and consequently adhesion and migration of breast cancer cells.

ERβ has been implicated as an indicator of endocrine response in breast cancer. In **paper IV** we investigated if ERβ could modulate pathways implicated in endocrine resistance development. Expression of ERβ in human breast cancer cells resulted in a decrease in both active Akt, as well as its upstream regulator, the epidermal growth factor receptor 2 and 3 (HER2/HER3) dimer. Expression of the tumour suppressor and important inhibitor of Akt signalling, PTEN was increased upon expression of ERβ. Further, ERβ expressing breast cancer cells had also an increased sensitivity to tamoxifen. In all, these data provide a possible mechanistic insight into how ERβ may contribute to endocrine sensitivity.

In conclusion, the studies presented in this thesis contribute to the knowledge of ERβ function in normal and cancerous breast, and highlight several possible anti-tumourigenic mechanisms for ERβ. Although the mechanisms have not yet been fully characterised, in breast cancer, ERβ seems to affect growth, adhesion, angiogenesis and sensitivity to endocrine therapy. These studies highlight the importance of ERβ as a prospective prognostic marker with potential as a target in the treatment of breast cancer.

ISBN 978-91-7457-171-4