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Regulation, Clinical and Biological Significance of Cathepsin D in Breast Cancer

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The lysosomal protease, pro-cathepsin D, is overexpressed and secreted by human breast cancers. In estrogen-responsive breast cancer cell lines, estrogens and growth factors stimulate cathepsin D expression through distinct mechanisms. Clinical studies indicate that high cathepsin D concentration in primary breast cancers is correlated with an increased risk of metastasis and particularly useful to orientate node-negative tumors towards an adjuvant therapy.

Key words: Breast cancer, Cathepsin D, Estrogens, Prognosis marker, Metastasis.

The major characteristics of human breast cancers are their selective responsiveness to estrogen and their elevated incidence of metastasis. In an attempt to understand the mechanism by which estrogens stimulate cell proliferation and mammary carcinogenesis, estrogen-induced proteins have been detected and studied in metastatic breast cancer cell lines. These estrogen-regulated genes include growth factors, steroid receptors and proteases. Our laboratory has studied a 52 Kda glycoprotein secreted by breast cancer cells whose regulation was associated with cell growth (15). This protein has been purified to homogeneity using monoclonal antibodies and identified as the secreted precursor of a cathepsin D bearing mannose-6-phosphate signals and routed to lysosomes *via* mannose-6-phosphate receptors (3, 10). The corresponding cDNA has been cloned and its sequence demonstrated a nearly complete homology with cathepsin D gene from normal tissue which has been localized on chromosome 11 by *in situ* hybridization (1).

The regulation of cathepsin D gene

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expression in human breast cancer cells appeared to be a complex phenomenon. Estrogens specifically and directly stimulate transcription of cathepsin D. Antiestrogens which antagonized the estrogen-induction in antiestrogen-sensitive cells (MCF_7 cells), induced cathepsin D mRNA in several antiestrogen-resistant cell lines (5). Other mitogens in MCF_7 cells, i. e. EGF, IGF1 and basic FGF also induce cathepsin D mRNA and protein but their effects depend upon *de novo* protein synthesis suggesting a mechanism different to that of estradiol (6). In estrogen-receptor negative cell lines MDA-MB231 and BT20, the cathepsin D mRNA and protein are also over-expressed. Finally, in all breast cancer cells, the production of pro-cathepsin D is largely enhanced and the cellular processing altered as compared to normal mammary epithelial cells in culture (4).

Clinical studies also indicate that the over-expression of this protease is correlated to proliferation of mammary ductal epithelium and to the metastatic potential of breast cancers. Using immunohistochemistry or two-site solid phase immunometric assay of breast cancer cytosol, we showed that the concentration of total cathepsin D (52 Kda + 48 Kda + 34 Kda forms) is increased in proliferative ductal mastopathies and in invasive breast carcinoma where it varied markedly and independently to the presence of steroid receptors (7-9).

Several retrospective clinical studies indicate a significant correlation between high cathepsin D concentrations in the cytosol of primary breast cancers and poor prognosis. An increased risk of metastasis and death is associated to tumors with high cathepsin D level (11-13). This marker is independent of other prognostic markers including lymph node invasion, tumor size, Scarff and Bloom histological grade, *neu-erbB* 2 and *int*-2 oncogene amplifications, estrogen and progesterone receptors. The prognostic value of cathepsin D appears particularly useful for node-negative tumors to orientate towards an adjuvant therapy.

The biological significance of the correlation between high cathepsin D concentration and metastasis is not yet known. The enhanced production of this protease could be an incidental consequence or a causely related event of the multistep process facilitating tumor cell invasion and metastasis. The two biological activities of the purified cathepsin D demonstrated *in vitro*, i. e. its capacity to digest extracellular matrix and several membrane proteins (2) and its mitogenic activity on MCF_7 human breast cancer cells (14) suggest that over-production of this protease by and around tumor cells may influence their growth autonomy and facilitate the metastatic process.

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Resumen

La proteasa lisosomal, pro-catepsina D, está sobreexpresada y secretada por los cánceres de mama humanos. En líneas celulares de cáncer de mama sensibles al estrógeno, los estrógenos y los factores de crecimiento estimulan la expresión de catepsina D a través de distintos mecanismos. Estudios clínicos indican que una concentración alta de catepsina D en cánceres de mama primarios está relacionada con un riesgo incrementado de metástasis y es particularmente útil para orientar tumores de nodos negativos hacia una terapia auxiliar.

Palabras clave: Cáncer de mama, Catepsina D, Estrógenos, Metástasis.

References

1. Augereau, P., García, M., Mattei, M. G., Cavaillès, V., Depadova, D., Derocq, D., Ca-

pony, F., Ferrara, P., Rochefort, H.: *Mol. Endo.*, 2, 186-192, 1988.
2. Briozzo, P., Morisset, M., Capony, F., Rougeot, C. and Rochefort, H.: *Cancer Res.*, 48, 3.688-3.692, 1988.
3. Capony, F., Morisset, M., Barrett, A. J., Capony, J. P., Broquet, P., Vignon, F., Chambon, M., Louisot, P., Rochefort, H.: *J. Cell. Biol.*, 104, 253-262, 1987.
4. Capony, F., Rougeot, C., Montcourrier, P., Cavaillès, V., Salazar, G., Rochefort, H.: *Cancer Res.*, 49, 3.904-3.909, 1989.
5. Cavaillès, V., Augereau, P., García, M., Rochefort, H.: *Nucl. Acids Res.*, 16, 1.903-1.919, 1988.
6. Cavaillès, V., García, M. and Rochefort, H.: *Mol. Endo.*, 3, 552-558, 1989.
7. García, M., Lacombe, M. J., Duplay, H. *et al.: J. Steroid Biochem.*, 27, 439-445, 1987.
8. García, M., Salazar-Retana, G., Pagès, A., Richer, G., Domergue, J., Paguès, A. M., Cavalié, G., Martín, J. M., Lamarque, J. L., Pau, B., Pujol, H. and Rochefort, H.: *Cancer Res*, 46, 3.734-3.738, 1986.
9. Maudelonde, T., Khalaf, S., García, M., Cavaillès, V., Derocq, D., Delarue, J. C. Freiss, G., Duporté, J., Benatia, M., Rogier, H., Paolucci, F., Simony, J., Pujol, H., Pau, B. and Rochefort, H.: *Cancer Res.*, 48, 462-466, 1988.
10. Rochefort, H., Augereau, P., Briozzo, P., Capony, F., Cavaillès, V., Freiss, G., García, M., Maudelonde, T., Morisset, M. and Vignon, F.: *Biochimie*, 70, 943-949, 1988.
11. Spyratos, F., Maudelonde, T., Brouillet, J. P., Brunet, M., Defrenne, A., Andrieu, C., Hacene, K., Desplaces A. and Rochefort, H.: *The Lancet*, 8.672, 1.115-1.118, in press, 1989.
12. Tandon, A., Clark, G., Chirgwin, J., McGuire, W. L.: *Proc. Amer. Assoc. Cancer Res.*, 30, 252, 1989.
13. Thorpe, S. M., Rochefort, H., García, M., Freiss, G., Christensen, I. J., Khalaf, S., Paolucci, F., Pau, b., Rasmussen, B. B. and Rose, C.: *Cancer Res.*, 49, 6.008-6.014, 1989.
14. Vignon, F., Capony, F., Chambon, M., Freiss, G., García, M. and Rochefort, H.: *Endocrinology*, 118, 1.537-1.545, 1986.
15. Westley, B., Rochefort, H.: *Cell*, 20, 352-362, 1980.