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Fascin, an actin-bundling protein associated with cell motility, is upregulated in hormone receptor negative breast cancer

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Loss of hormone receptor (HR) status in breast carcinomas is associated with increased tumor cell motility, invasiveness and poor prognosis. However, molecular mechanisms leading to this more aggressive tumor cell phenotype are still unknown. In an immunohistological study of 58 primary breast cancers, estrogen (ER) and progesterone (PR) receptor levels were inversely correlated with the expression of fascin, an actin-bundling protein associated with cell motility ($p < 0.0001$ and $p = 0.0019$, respectively). In addition, flow cytometry analysis revealed that fascin negative tumors were more likely diploid (26 of 43 (60.5%)) than non-diploid (17 of 43 (39.5%)), a difference that reached statistical significance ($p = 0.03$). No significant correlation was found between histological tumor grading and ploidy ($p = 0.12$). Immunohistochemically, fascin positivity appeared as cytoplasmic staining with a marked enhancement in areas of tumor–host interaction in most samples. In summary, the upregulation of fascin in HR-negative breast cancers may contribute to their more aggressive behavior.

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Overexpression of the keratin 18 gene results in reduced malignancy of human breast cancer cells

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Purpose: In a retrospective study we could show that a high expression of the cytoskeletal protein keratin 18 (K18) in the tumor is correlated with a favorable prognosis for the patients. Experiments with cultured breast cancer cells revealed similar results. To prove the principle we now transfected the K18 gene into a metastatic and invasive cell line that shows only weak K18 expression.

Methods: The complete human K18 gene (pGC 1853, R. Oshima) was transfected (liposomal transfer with pFx-2, Invitrogene) into MCF7-LCC2 cells. Several permanently overexpressing clones were established.

Results: (i) The anchorless growth of the transfected cells in soft agar was dramatically reduced. (ii) This effect was "dose dependent": the higher the K18 expression, the lower the proliferation rate. (iii) The expression of plakoglobin (=γ-catenin) a key protein of epithelial adhesion structures was enhanced in the transfected clones.

Conclusion: The intermediate filaments in epithelial cells are formed by keratins and K18 is a marker of well differentiated luminal cells in the breast epithelial tissue. The loss of K18 seems to be part of a general loss of differentiation along with the metastatic event. A reversal of the K18 depletion by gene transfer may reverse this process in part and, thus, result in reduced aggressiveness of the cancer cells.

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Dihydropyrimidine dehydrogenase and thymidylate synthase in relation to 5-fluorouracil sensitivity of breast cancer

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Thymidylate synthase (TS) and dihydropyrimidine dehydrogenase (DPD) has been investigated to understand 5-FU sensitivity and to develop a biochemical modulator. It is reported that both TS and DPD activity were significantly correlated to 5-FU effectiveness in several cell lines. It is also reported that tumoral/normal DPD activity ratio was significantly lower in complete responder to 5-FU chemotherapy than in partial or non responding head and neck cancer patients. In this study, we measured TS level, DPD activity and in vitro sensitivity to 5-FU using breast cancer as a sample to assess a correlation between the enzymes and 5-FU sensitivity of a breast cancer.

Methods: This study was conducted on 23 female patients undergoing surgery for breast cancer. Immediately after resection, portions of viable tumor and adjacent normal tissue were removed and properly stored for subsequent analysis. In both normal and tumor tissues, TS level and DPD activity was measured. In vitro sensitivity of breast cancer to various concentrations of 5-FU was measured with the use of a collagen gel droplet embedded culture drug sensitivity test.

Results: TS level was significantly higher in tumor than in normal tissue which was 12.6 ± 12.5 pmol/g-tissue and 2.5 ± 1.1 pmol/g-tissue, respectively ($p < 0.05$). DPD activity was significantly higher in tumor than in normal tissue which was 75.5 ± 25.5 pmol/min/mg-protein and 39.5 ± 16.9 pmol/min/mg-protein, respectively ($p < 0.0001$). No significant correlation was observed among TS level, DPD activity and in vitro sensitivity of tumor to various concentrations of 5-FU.

Conclusion: This study provides the first analysis of DPD activity in breast cancer. Contrary to previous study using cell lines, TS level and DPD activity were not correlated to 5-FU sensitivity. However, elevated TS level and DPD activity in tumor encourage a use of biomodulator in the treatment of breast cancer, such as DPD inhibitor and 5-formyl-tetrahydrofolate.

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Somatic mutations in bilateral breast carcinomas

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Purpose: The molecular pathogenesis of various categories of breast cancer (BC) has been well described, but surprisingly few reports have appeared on analysis of somatic mutations in bilateral BC. This study analysed common genetic lesions in paired bilateral carcinomas, with an especial emphasis on 2 questions: 1) is there a concordance or discordance between two tumours within the same patients? 2) does bilateral BC show distinct features as compared to regular, monolateral BC?

Methods: PCR-driven analysis of common losses of heterozygosity (LOH) was performed for 23 cases (46 tumours) from patients diagnosed with bilateral BC.

Results: LOH was observed in 15/46 (33%) informative cases for chromosome 1p, 7/40 (18%) for 5q, 12/44 (27%) for 11q, 15/40 (38%) for 13q, and 7/32 (22%) for 17p. These values are within the range of interlaboratory variations reported for monolateral BC. There was no strong evidence for concordance of LOH within the same patient for any chromosomal loci tested. Atypical for breast carcinomas, 7/46 (15%) tumours accumulated a high frequency (ranging from 11% to 29%) of shortened dinucleotide CA repeats, implying microsatellite instability (MI). Further analysis with the highly informative BAT-26 marker allowed for the classification of 2 of these cases as replication error positive (RER+) phenotype, whereas the remaining 5 tumours harboured so-called border-line MI.

Conclusion: An involvement of both RER+ and border-line microsatellite instability appears to be a distinct feature of bilateral breast carcinomas as compared to monolateral lesions.

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The influence of c-myc gene expression on disease-free interval in breast cancer

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Purpose: The current studies show the correlation between the amplification of c-myc oncogene and a shorter patient survival as well as a shorter disease-free interval. This especially concerns patients with the axillary lymph nodes clear from metastases, but with positive estrogene receptor and negative progesterone receptor. The aim of our study was to examine this correlation, however, in patients with the metastatic axillary lymph nodes.

Methods: We qualified 26 patients with the same degree of clinical advancement T1-2N1M0 (the number of metastatic lymph nodes ≤ 3), with positive estrogene receptor, aged 54–68, (average age 58) for the study. All the patients had been treated with hormonotherapy until the first symptoms of the disease recurrence appeared. The presence of c-myc oncogene expression was examined by means of the 'In situ' hybridization method, and its intensity was read in the microscope and evaluated in the four-degree scale.

Results: The considerable correlation was shown between c-myc oncogene expression and disease-free interval. 16 patients (61.53%) with short disease-free intervals (21.6 months on average) were found to have very intensive expression (III and IV degree), however, in women with long disease-free intervals (over 5 years) c-myc oncogene transcription was weak, and was evaluated to reach I and II degree of hybridization signal intensity.