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Molecular cytogenetic analysis of chromosome 7 and 17, c-erbB2 and TP53 genes in urothelial cell carcinoma of the bladder: clinical implications

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Purpose: Over 80% of urothelial bladder carcinomas (UCC) are superficial papillary lesions. The recurrences in these tumours are the rule and about one third progress. Aberrations of chromosomes 7 and 17 were defined as important alterations in UCC, but their significance in disease progression is not well established. Alterations of TP53 and C-erbB2 genes are associated with more aggressive tumours. Their role in papillary UCC is still controversial. It was our aim to evaluate the clinical significance of these alterations.

Methods: We analysed 49 superficial papillary primary UCCs and their recurrences divided in two groups: group A (n=27) consisting of patients with more than 45 years of age in diagnosis date, and group B (n=27) consisting of patients with 45 years and younger. Both groups were studied by FISH, using centromeric probes for chromosomes 7 and 17 (Oncor) and unique sequence probes specific for TP53 and c-erbB2 genes (Oncor). Normal mucosas (cadaveric donors) were used to establish the limits of aneuploidy.

Results: We found similar results in both series which let us analyse all patients jointly. Thus, we found that cases with monosomy of TP53 presented a poor recurrence-free survival (p=0,002). This alteration is significantly related with UCCs recurrences (p= 0,002). We did not find any case with c-erb B2 amplification. Alterations in chromosome 7 did not related with prognosis.

Conclusion - These results suggest that the measurement of TP53 gene monosomy by FISH, predict recurrences in patients with pTa/pT1 UCC tumours at diagnosis. This finding may identify patients with risk of recurrences. (Projecto CFCIS- MS -83/97)

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KI67: is it a prognostic index in node negative breast cancer patients ?

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Aim: the study assesses the prognostic significance of Ki-67 related to other conventional markers such as age, tumour size, grading, ER and PgR status, in a group of patients with lymph node negative breast cancer. The aims of this work is identifying more reliable markers, which allow a better prognostic stratification.

Patients and Methods: in a retrospective design we evaluate 147 breast cancer patients without lymph nodal involvement with 5 years of follow-u. All patients were diagnosed at Spedali Civili di Brescia from 1992 to 1995. The median age was 58 years (range: 26-80); the median tumour size was 1.6 cm (range: 0.4-3.9), the median number of lymph nodes resected and histologically evaluated was 14 (range: 5-30).

Results: the univariate analysis of discrete variables showed: p=0.0006 for grading, p=0.0004 for ER negative status, p=0.001 for PgR negative status and p=0.0001 for positive Ki-67. Ki67 appears to be an important prognostic factors in the determination of disease free survival and overall survival. In our data ploidy doesn't reach any statistic significance.

The continuous variables (age and tumour size) don't correlate with the prognosis.

At the multivariate analysis Ki-67 is the most predictive parameter of DFS and OS (RR=4.13).

Conclusions: Ki-67 could add an important contribution to actual and available prognostic model in node-negative breast cancer and, in our data, it appears to be the most significative marker.

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Systemic inflammatory response and survival in advanced cancers

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Purpose: The assessment of prognosis in patients with advanced cancer is problematical. The value of C-reactive protein concentration in this context has not been clearly defined.

Experimental design: Patients with a diagnosis of colorectal (n=182), gastric (n=87), breast (n=99) or bronchogenic (n=404) cancer and who had measurements of C-reactive protein were identified.

Results: Median survival, from the time of sampling, ranged from 478 days in the colorectal cancer patients to 60 days in patients with bronchogenic cancer. On univariate analysis there was, in each tumour type, a significant relationship between the duration of survival and both log10 C-reactive protein and albumin concentrations ($p<0.0002$). On multivariate analysis, in each tumour type, log10C-reactive protein remained a significant independent predictor of survival ($p<0.0002$). The hazard ratio for a tenfold increase in C-reactive protein concentration varied from 1.5 in bronchogenic cancer to 3.3 in colorectal cancer.

Conclusions: The results of the present study demonstrate that patients with advanced colorectal, gastric, breast and bronchogenic cancer have evidence of an inflammatory response and the magnitude of that response predicts the duration of survival.

Tumour and cell biology

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Suppression of urokinase expression and invasiveness by bikunin is mediated through inhibition of protein kinase C-dependent signaling pathway

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Introduction: Bikunin (Bik), a Kunitz-type protease inhibitor, interacts with cells as a negative modulator of the invasive cells. Bik forms membrane complexes with bik-binding proteins (bik-BPs) on the cell surface. It has been established that tumor cells expresses two types of bik-BPs; a 40 kDa bik-BP (bik-BP40), which is identical to link protein (LP), and a 45 kDa bik-BP (bik- BP45). First, we characterize binding properties of bik-BPs-bik complexes in the cells. We next asked whether the carbohydrate moieties and core protein are required for uPA suppression. Finally, we assessed the signal transduction cascade of bik on the suppression of uPA expression Materials and Methods: Human chondrosarcoma cell line HCS-2/8 and human ovarian cancer cell line HRA were cultured. In vitro ligand-blot, cell-binding and competition assays, Scatchard, and the functional analyses were carried out. We obtained bik derivatives (enzymatically degraded truncated bik proteins and recombinant bik). Results: Both bik-BP40 and bik-BP45 bind radiolabeled-bik. A deglycosylated form of bik (NG-bik), from which the chondroitin-sulfate side chain has been removed, binds only to bik-BP40. The chondroitin-sulfate side chain of bik is required for its binding to bik-BP45. Low affinity binding sites are the bik- BP40 (which can bind NG-bik) and the high affinity sites are the bik-BP45. Preincubation of the cells with bik reduced the ability of PMA to trigger the uPA expression at the gene level and at the protein level. PMA translocation of PKC from cytosol to membrane was inhibited by bik, indicating that bik inhibits the activation cascade of PKC. When cells were preincubated with bik, we could detect suppression of phosphorylation of MEK/ERK/c-Jun proteins. Conclusion: In conclusion, bik requires either the N-terminal extension with the O-linked carbohydrate moiety (chondroitin-4-sulfate sugar side chain; Ala1 to Lys21 residues) or the Kunitz-domain I (Lys22 to Arg77 residues) of bik to bind to cells, but the uPA expression was inhibited only by the O-glycoside linked core protein without the N-glycoside side chain. In addition, bik markedly suppresses the cell motility possibly through negative regulation of PKC- and MEK/ERK/c-Jun-dependent mechanisms, and that these changes in behavior are correlated with a coordinated down-regulation of uPA which is likely to contribute to the cell invasion processes.