Over-expression of COX-2 in human prostate cancer

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Cyclooxygenases (COXs) are enzymes involved in the conversion of arachidonic acid to prostaglandins (PG). COX has two isoforms: COX-1, which is constitutively expressed and COX-2, which is inducible and shown to be up-regulated in various cancers. There is compelling evidence in the literature linking Cyclooxygenase-2 (COX-2) with some cancers. The aim of our study was to determine the expression profile of both COX-1 and 2 proteins in benign prostatic hyperplasia (BPH) and PC using immunohistochemistry (IHC) and immunoblotting.

30 BPH and 82 PC formalin-fixed paraffin-embedded samples were examined by standard 3-layer IHC with COX-1 and COX-2 specific mouse monoclonal antibodies. Western blotting was performed on a total of 13 fresh frozen samples (6 BPH and 7 PCs) and probed with COX-1 or COX-2 mouse monoclonal antibody.

In BPH, COX-1 was expressed primarily in fibromuscular stroma with occasional weak cytoplasmic expression in glandular epithelial cells. COX-2 immunostaining revealed membranous expression in luminal glandular cells with no expression in the stroma. In prostate cancer (PC), stromal expression for COX-1 and COX-2 remained unaltered. There was variable weak cytoplasmic expression of COX-1 in tumour cells. A significant increase ($p = 0.008$) in COX-2 expression was observed in tumour cells with an appreciable change in staining pattern from membranous to cytoplasmic ($p = 0.001$). There was also a significant increase ($p = 0.01$) in the intensity of COX-2 expression in high grade (poorly differentiated) cases compared to low grade (well differentiated). On immunoblotting upto 4 fold increased expression of COX-2 was detected in PCs compared to BPH, while COX-1 levels were similar in both. These results suggest that COX-2 may contribute to the differentiation state of the prostatic adenocarcinoma.

repp86, A Novel Proliferation-Associated Protein

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A novel cell cycle-regulated nuclear protein with protein kinase activity, repp86, is described. Its expression is restricted to the cell cycle phases S, G2, and M, and can be detected on paraffin sections by means of a specific monoclonal antibody. The protein is rapidly lysed at the end of cytokinesis, and therefore absent in G1 and noncycling cells. In a retrospective analysis of 371 lymph node-negative breast cancers, repp86 was found to carry exceptional prognostic relevance. Under physiological conditions, repp86 expression levels parallel those of Ki-67 at a ratio of approximately 0.35:1. This ratio may vary widely in neoplastic cell populations, indicating different rates of cell transit through the "restriction point". In mammary carcinomas, a high repp68:Ki-67 ratio coincided with deficiencies in cell cycle regulators, such as p53 mutation, Rb deletion, and reduced expression of $p21^{WAF1}$, $p27^{Kip1}$, and $p16^{INK4a}$. Also, the ratio increased in proportion to the number of concurrent cell cycle defects. In a series of 93 mammary carcinomas, the repp86:Ki-67 ratio emerged as a significantly more accurate predictor of prognosis than standard prognostic criteria, Ki-67, and the above mentioned cell cycle regulators considered individually or in combination. This suggests that the repp86:Ki-67 ratio may be regarded as a global indicator of cell cycle deregulation, which may encompass the inactivation of yet unidentified cell cycle regulators.

Consistent Chromosomal Translocations in Embryonal Rhabdomyosarcoma

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The discovery of specific chromosomal abnormalities in paediatric solid tumours has led to the development of a range of molecular techniques for the diagnosis and assessment of prognosis of these neoplasms. Although consistent chromosomal translocations have been identified in alveolar rhabdomyosarcoma, no such abnormalities have yet been described in the more common embryonal variant (embryonal rhabdomyosarcoma, ERMS).

Until recently, karyotypic analysis of tumours such as ERMS was limited by the low resolution of conventional cytogenetic techniques. We have used molecular cytogenetics to obtain a more definitive karyotype than previously possible of four ERMS cell lines. 24-colour chromosome painting was undertaken using spectral karyotyping and abnormalities were confirmed using 7-colour multiplex FISH and comparative genomic hybridisation.

Several recurring cytogenetic abnormalities have been demonstrated, including translocations between chromosomes 1 and 15 in all four cell lines and translocations between chromosomes 1 and 3 in three cell lines. Two of the lines show translocations involving 1p11-13, a region that has also been suggested by classical cytogenetics to be rearranged in ERMS. Further characterisation of translocation breakpoints may indicate mechanisms of development of ERMS and may enable the generation of novel techniques for the diagnosis of this tumour.

Clonal and Kinetic Profiles Correlate with Vascular Patterns in Adrenal Cortical Lesions.

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Background: Monoclonal adrenal cortical lesions have been characterized by an inverse correlation between proliferation and apoptosis, while polyclonal lesions reveal a direct correlation. Their relationship with the vascular pattern remains unknown in nodular hyperplasias (ACNH), adenomas (ACA), and carcinomas (ACC) of the adrenal cortex.

Methods: We studied 20 ACNH, 25 ACA, and 10 ACC, classified according to WHO's criteria, from 55 females. Representative samples were histologically evaluated and microdissected to study clonality by X-chromosome inactivation. Proliferation (PR) and apoptosis were evaluated in consecutive sections using slide and flow cytometry, as well as in situ end labeling (ISEL). Endothelial cells were stained with CD31 and the blood vessel area and density quantified by image analysis in the same areas. Appropriate tissue controls were run in every case. Regression analyses between kinetic and vascular features were performed in both polyclonal and monoclonal lesions

Results: Polyclonal pattern was observed in 14/18 informative ACNH and 3/22 informative ACA, and monoclonal in 4/18 ACNH, 19/22 ACA, and 9/9 ACC. A direct correlation between proliferation and apoptosis was observed in polyclonal lesions (PR= 29.32 ISEL - 1.93), while that correlation was inverse for monoclonal lesions (PR = - 9.13 ISEL + 21.57). A progressive increase in blood vessel area was observed in the transition ACNH – ACA – ACC, but statistically significant only between benign and malignant lesions (191.36±168.32 vs. 958.07±1279.86 μm^2, $p<0.0001$). In addition, case stratification by clonal pattern revealed significant differences between polyclonal and monoclonal benign lesions: 6% polyclonal vs. 57% monoclonal lesions had blood vessel area > 186 μm^2 ($p=0.0000008$). Vessel area inversely correlated with both apoptosis and proliferation in polyclonal lesions and with apoptosis in monoclonal lesions (Vessel Area = -5.50 ISEL +278.88). Proliferation showed a direct correlation with blood vessel area in monoclonal lesions (Vessel Area = 5.05 PR + 168.13).

Conclusions: The kinetic advantage of monoclonal adrenal cortical lesions (↑proliferation, ↓apoptosis) is maintained by an increased vascular area.