model: B16F10 and B16Bl6 cells can both induce primary tumours after subcutaneous implantation of cells, but pulmonary metastasis are only found in the mice bearing a B16Bl6 tumour. We performed comparative proteomic studies on these two cell lines, using both cultured cells and subcutaneous tumours. 7 and 12 differentially expressed proteins respectively were identified by mass spectrometry. Especially, annexin A1 (ANXA1) was increased 1,5 to 2 fold in B16Bl6 cells as compared to B16F10 cells, in vivo and in vitro. In an attempt to characterize its role in melanoma B16 spreading, we showed that reducing ANXA1 protein level by siRNA in B16Bl6 cells decreased their in vitro invasion properties on Matrigel® coated chambers. This should be associated with the presence of formyl peptide receptors (FPR), which have been shown to activate invasion in an epithelial cell line SKCO-15 (Babbin et al, 2006). Indeed, we demonstrated by RT-PCR the presence of transcripts encoding for two FPR isoforms (FPR1 and FPRrs) in the two B16 lines without any reliable quantitative difference. These receptors seemed to be functional since B16Bl6 cells incubation with the FPR agonists (fMLP) or antagonists (tBOC) respectively enhanced or decreased Matrigel coated chamber invasion. Furthermore, preliminary data suggested that incubation of B16Bl6 cells with fMLP and tBOC lead to an increase or decrease of ANXA1 steady state level. In conclusion, our results showed that increased ANXA1 expression could be associated at least in vitro with an increased invasion capacity, that might be mediated by the FPR receptors.

308 Poster

Expression of alpha(1,6)fucosyltransferase in the early tumorigenesis of human colorectal cancer

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An elevated level of fucose content in glycoproteins is one of the cancer-related alterations found in patients with malignant diseases. In a recent study we have demonstrated that α(1,6)fucosyltransferase [α(1,6)FT], the enzyme which catalyzes the core fucosylation in these glycoproteins, was enhanced in tumoral colon when compared to healthy adjacent tissues from colorectal carcinoma (CRC) patients. Most CRC arise from neoplastic adenomatous polyps thus we developed the present work in order to determine weather this increase is an early event or it only occurs lately in tumorigenesis.

We investigated the α(1,6)FT expression by means of immuno-histochemistry in 81 adenomas, 13 inflammatory lesions and 9 healthy tissues of free-CRC patients. Tissue sections were stained using anti-human α(1,6)FT MAb and visualized with DAB (3,3´-diaminobenzidine). Negative controls were performed using PBS instead of primary antibody.

After the immunohistochemical assay, no positive expression was found in the healthy and inflammatory tissues. In the case of adenomas, 13 of the 81 polyps analysed (16%) were positive for α(1,6)FT expression whereas the percentage of positive expression in tumor tissues (that we previous described) was the 61.3%. After the statistical analysis, we found significant differences for tumour vs. adenomas, inflammatory lesions and control healthy tissues ($p<0.01$). We also analysed the possible association between the α(1,6)FT expression in the neoplastic polyps and the histological type or the grade of dysplasia in this polyps. A significant increase of expression was observed in the cases without dysplasia with respect to the dysplasic ones ($p=0.05$), whereas a correlation between the expression and the histology was not found.

In conclusion, the absence of α(1,6)FT immunohistochemical expression in the inflammatory and healthy tissues of patients without CRC clearly suggests that the alteration of the enzyme expression is specific of the malignant transformation and is not related to the inflammatory process normally associated to the cancer. On the other hand, the low rate of positive expression obtained in pre-cancerous lesions indicates that the α(1,6)FT expression increase is a late event in the tumorigenesis strongly associated with a total cell transformation of the colorectal tumor.

309 Poster

Endo180 expression by tumour cells with an invasive phenotype correlates with prostate cancer progression

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The purpose of this study was to investigate whether stromal or epithelial expression of Endo180 (CD280; MRC2; urokinase-type plasminogen activator receptor-associated protein, uPARAP) can contribute to metastatic prostate cancer. The prognostic and functional roles of Endo180 were characterised using tissue microarray (TMA) and immunofluorescent costaining with pan-cytokeratin (pCk) to allow quantitative analysis of stromal (pCk-) and epithelial (pCk+) cellular expression. Expression of Endo180 and its function-associated partners in collagen remodeling and tumour cell migration and chemotaxis: membrane type-1 matrix metalloproteinase (MT1-MMP) and urokinase-type plasminogen activator (uPA)-uPA receptor (uPAR) respectively; were quantified in human prostate tissue clinically graded as benign prostate hyperplasia (BPH) (n=29) or with good (n=26), intermediate (n=96) or poor (n=18) tissue differentiation. Significant differences or correlations between categorical variables, including serum prostate specific antigen (PSA), were determined using two-sided statistical tests and 95% confidence intervals. Increases in Endo180+/pCk- and Endo180+/pCk+ cells confirmed both stromal and epithelial upregulation of Endo180 respectively. The increase in epithelial expression of Endo180 displayed linear correlation with advanced clinical grade and greater prognostic capability than serum PSA. The differential patterns of stromal and epithelial Endo180 coexpression with MT1-MMP and uPAR-uPA with increased clinical grade revealed the potential for a distinct sequence of their molecular and functional interplay during the different stages of prostate cancer progression. Taken together these data support the use of Endo180 as a potential biomarker for the prognosis of prostate cancer tissue biopsies and as a target to prevent tumour cell migration, chemotaxis, invasion and tissue remodelling during prostate cancer metastasis.

310 Poster

Candidate genes for the recurrence of glioblastoma multiforme identified by microarray

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Background: Glioblastoma multiforme (GBM) is the most aggressive and most lethal primary malignant brain tumor, correlated with survival rates of less than one year from the time of diagnosis. Current surgical procedure attempts to remove the bulk of the tumor mass, whereas GBM frequently recurs within 1-3cm from the primary tumor resection site. Molecular mechanisms involved in the recurrence of the tumor are still poorly understood. The aim of the study was to define the molecular signature of GBM surrounding white matter (WM) in order to better understand the molecular mechanisms involved with tumor relapse.

Material & Methods: Human GBM tumor bulk and surrounding tissue (1-3cm from the border of the tumor) were obtained from five patients who underwent total tumour resection, while normal white matter was harvested from patients who underwent surgical procedure for nonmalignant pathologies. Samples were processed for hybridization on the Affymetrix Human U133A arrays and data were examined with the GeneSpring analysis software. A subset of interesting genes was further validated by RT-PCR.

Results: Gene expression analysis of the samples was done in 2 independent steps. First, molecular profiling comparison of GBM surrounding WM and normal WM resulted in 59 genes differentially expressed between both tissues. Among these, numerous genes expressed by mature neural cells were down-regulated in GBM surrounding WM. Moreover, KLRC1, a specific natural killer receptor naturally involved in the activation of antitumoral cells was drastically repressed in GBM surrounding WM, suggesting that the antitumoral immune surveillance is compromised in this tissue.

Second, we focused our study on genes specifically regulated in GBM periphery respectively to GBM core.

The highest up-regulated gene in GBM surrounding tissue codes for DTX4, a regulator of NOTCH signalling pathway described for its key role in maintaining neural progenitors in an uncommitted state.

Conclusion: This study revealed unique molecular characteristics of GBM surrounding tissue, showing the dysregulation of genes involved in immune surveillance along with genes associated to stemness maintenance. All together, these data may help to understand the molecular mechanisms associated with GBM recurrence.

311 Poster

MYC inhibition of p27-induced erythroid differentiation is mediated by the repression of erythroid master genes and uncoupled from its cell cycle promoting activity

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Inhibition of differentiation has been proposed as an important mechanism for the tumorigenesis mediated by MYC, but the mechanisms involved are