significance as it was observed in human brain tumor biopsies of various grade.

Conclusions - Caveolin-1 plays a critical role in the aggressiveness of glioblastoma. Caveolin-1 effects are achieved through $\alpha5\beta1$ integrin. Mediator of caveolin-1 effects, $\alpha5\beta1$ integrin is also a marker for glioma aggressiveness and an efficient target for the treatment of glioma especially the ones exerting the highest aggressive phenotype.Caveolin-1 /$\alpha5\beta1$ integrin are diagnostic and prognostic markers for glioma and might be predictive of the response to future anti-$\alpha5\beta1$ integrin therapies.

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Tumor cell NG2 proteoglycan controls cancer progression through its interaction with host Collagen type VI

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Soft-tissue sarcomas are highly aggressive and heterogenous tumours that remain largely incurable. As for most types of tumours, the presence of metastasis at diagnosis, or the evolving of such lesions with time, catastrophically reduces the probability of survival. Factors predicting the formation of metastasis in soft-tissue sarcoma patients are not known and similarly obscure remains the modes through which metastases form in these individuals. We find that NG2 and its putative ECM ligand collagen type VI (Col VI) are highly upregulated in metastases of soft-tissue sarcoma patients and that highly enhanced expression of NG2 in these lesions adversely correlate with patient survival. Relative expression levels of NG2 on sarcoma cells, as determined by qPCR and immunostaining analyses, define their malignancy degree and subpopulations of immunosorted highly enriched NG2+ cells exhibit a strongly aggressive behaviour. However, growth and dissemination of NG2+ cells is strongly impaired in Col VI knock-out mice suggesting that the NG2-Col VI interplay dictates tumour progression in vivo. Adhesion and migration of sarcoma cells expressing intact or truncated variants of NG2 and confronted with purified Col VI tetramers or Col VI+ and Col VI- native matrices isolated from wild type and Col VI null mice corroborate the importance of NG2 in collagen recognition and allowed the pinpointing of the reciprocal binding domains within the two molecules. Thus we demonstrated that NG2 cell surface proteoglycan represents a novel independent prognostic factor in certain types of soft-tissue sarcomas where its relative expression levels in primitive lesions strongly predict future appearance of metastases. Global gene profiling of NG2+ versus NG2-, siRNA treated, cells reveals that the proteoglycan confers a malignant and potentially metastatic phenotype independently of previously identified metastasis-associated gene signatures. We also identify some of these signalling pathways that are activated upon NG2-collagen type VI interaction and propose that in addition to serving as a prognostic biomarker, the NG2-collagen type VI interplay and its downstream effectors may constitute novel therapeutic targets in soft-tissue sarcomas and other tumours where NG2 is upregulated/de novo expressed. Taken together these findings highlight a crucial role of NG2 and its interaction with Col VI in the regulation of tumour progression and metastasis formation, providing the first molecular explanation for its uniqueness as a prognostic/therapeutic tool in soft-tissue sarcomas

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COX-2 transgenic mice as models for epithelial neoplasms

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Epidemiologic, pharmacologic, clinical, and experimental studies document the importance of prostaglandin (PG) signaling in epithelial cancer development. First of all, enzymes involved in PG biosynthesis, such as cyclooxygenase (COX)-2 and/or membrane prostaglandin E synthase (mPGES)-1, are overexpressed in a wide range of premalignant and malignant epithelial tumors, including those of the skin, breast, esophagus, stomach, colorectum, pancreas, prostate and urinary bladder. On the other hand, 15-hydroxy-prostaglandin dehydrogenase (15-PGDH), which is involved in the degradation pathway of PG including PGE2, thus counteracting the activities of COX-2 and PGES, was found to be down-regulated in human epithelial tumors, indicating a tumor suppressor activity of this enzyme.

Transgenic mouse lines with keratin 5 promoter-driven overexpression of cyclooxygenase (COX)-2 develop spontaneously pre-invasive epithelial neoplasms. These were diagnosed by human pathologists to be early-stage lesions in skin epidermis, prostate, and pancreas. In addition, in urinary bladder transitional cell carcinomas were observed. The pre-invasive neoplasms and carcinomas in COX-2 transgenic mice resemble not only on the histological level but also on molecular level (e. g. COX-2-, Her-2, VEGF expression) defined progression stages of human neoplasms. COXibs, selective inhibitors of COX-2-mediated PG synthesis representing a class of an approved prescription drug in human medicine have been found to suppress the transgene-induced phenotype, indicating the cause-and-effect relationship between aberrant COX-2 overexpression and the development of the neoplasms.

Moreover, the chronic systemic excess of PG induced by transgenic COX-2 overexpression caused severe white adipose tissue wasting in these mice. The molecular mechanism leading to this phenotype may explain cachectic body wasting in human cancer patients.

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Understanding the complex crosstalk between p53 and the estrogen receptors at a polymorphic variant of the VEGF receptor Flt-1 promoter

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Recently we established that a C>T single nucleotide polymorphism (SNP) in the Flt-1 promoter generates a functional half-site p53 response element (RE-T). We also showed that p53 is required but not sufficient for Flt-1 transactivation and that there is cooperative interaction with ligand-bound estrogen receptors (ER) via an ER half-site response element (ERE) located 225nt upstream the p53 RE-T. Disruption of the ERE in a reporter construct containing a 1kb fragment of the Flt-1 promoter resulted in loss of p53 responsiveness in HCT116 (p53 wt, weakly ERbeta positive) and U2OS (p53 wt, negative for ER) cells. Surprisingly, we have now observed that disruption of the ERE has no impact on transactivation in MCF7 cells (p53 wt, ERalpha and ERbeta positive) treated with doxorubicin (doxo) to induce p53. Searches for transcription factor binding sites revealed another putative half-site ERE in the promoter fragment located 145bp downstream the p53 RE. Using site-directed mutagenesis, we showed that while the mutation of this second site has no impact, mutation of both EREs greatly reduced transactivation. Over-expression of ERalpha or ERbeta in HCT116 phenocopied the MCF7 results in terms of the EREs contribution. To induce p53 in MCF7 cells we also used the thymidylate synthase inhibitor 5-FluoroUracil (5FU). Although 5FU was similar to doxo in stabilizing the p53 protein and inducing the p21 target gene, there was minimal transactivation of the Flt1-T construct, suggesting that doxo might have a specific impact on the p53, ER transcriptional cooperation or might enlist additional transcription factors/cofactors that contribute to the activation of the promoter. Using HCT116 cells (p53 wt and p53-null clones), which are heterozygous for the C>T SNP, we are also examining the expression of the endogenous Flt-1 gene, using qPCR. The Flt-1 transcript undergoes alternative splicing resulting in a soluble form of the receptor. These experiments are confirming the p53-dependent regulation of the Flt-1 gene and the different impact of doxo and 5FU. Notably, we are also observing an additional layer of complexity in the regulation of the gene, as the relative abundance of the two splice variants is differentially affected by the doxo treatment. This observation is currently being followed up with the development of assay systems probing stress-dependent stability of the two Flt-1 mRNAs, which have distinct 3'UTRs, as well as relative efficiency of alternative splicing.

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Beta endorphin produced by melanoma cells promotes tumor growth and immune escape

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Beta endorphin (BE) is an endogenous agonist peptide for the mu opioid receptor (MOR); its major role consists in releaving the sensation of pain at proximal nerve endings but can also inhibit immune responses. Interestingly, BE has also been found to be secreted in high amounts by several tumors of neuronal and non-neuronal origin where its role remains unclear. This project intended to investigate if BEsecreted by tmelanoma