tumourigenesis in this subgroup remain poorly described. We have analysed copy number aberrations and mutation profile of known target genes in tumours from early and late onset CRC patients.
Material and Methods: High resolution array CGH analysis (385 000 features), was performed in 23 patients with CRC diagnosis at young age (range: 28−53 years, median: 44 years), and 17 patients with CRC diagnosis at old age (range: 69−78 years, median: 79 years). Furthermore, mutation profiles were analysed in a larger series of carcinomas stratified according to microsatellite instability analysis (MSI) status. These patients included 45 young-at-onset (range; 27−50 years, median; 43 years), and 69 old-at-onset (n = 69, range; 71−93 years, median; 81 years). A panel of five genes, TP53, KRAS, BRAF, PTEN and PIK3CA, were investigated for mutations by sequencing.
Results: The overall genome copy number profiles were similar between carcinomas from patients in the two age groups. However, some chromosomal stretches were found to have statistically significant ($p < 0.05$) more aberrations in the young patients compared to the old-at-onset group (not visa versa); DNA sequences within 2q, 10q, 19q, were more often gained, and sequences within 1p, 1q, 2q, 4p, 4q 10p and 19p, were more frequently lost. KRAS and PTEN mutations were distributed equally between the patient groups, whereas the mutation frequencies of TP53 and PIK3CA differed between the groups. BRAF mutations were not significantly correlated with MSI in the young-at-onset group.
Conclusions: We have identified genomic and gene specific differences in colorectal carcinomas related to time of disease onset. The somatic genomic changes that occur preferentially in tumours from young patients pinpoint potential genetic risk loci that will be further examined.

801 Downstream targets of the TMPRSS2-ERG rearrangement in prostate cancer: cysteine-rich secretory protein-3 (CRISP3) is strongly up-regulated in fusion-positive carcinomas

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Background: A large percentage of prostate cancers harbor *TMPRSS2-ERG* fusions, leading to the aberrant overexpression of the transcription factor *ERG*. The target genes deregulated by this rearrangement, however, remain mostly unknown, precluding additional therapeutic strategies directed at the downstream effectors.
Material and Methods: Genome-wide RNA expression analysis on a subset of 24 prostate carcinomas with (n = 16) and without (n = 8) *ERG* rearrangements provided a list of candidate targets significantly associated with the fusion event. RNA expression data for the top-most deregulated genes was validated on an independent series of 200 tumours using Real-time PCR, whereas protein levels were assessed in an extended series of clinical samples comprising morphologically normal prostate, benign hyperplasic tissue, and prostate carcinomas (n = 77).
Results: Within the group of genes significantly over-expressed in fusion-positive lesions, Cysteine-rich secretory protein-3 (*CRISP3*) showed a striking 38-fold increase in expression when compared to fusion-negative carcinomas, being almost absent in normal and benign prostate tissue. In the independent validation series, *ERG* and *CRISP3* expression levels were strongly correlated ($r_s = 0.84$, $p < 0.001$), and a median 43-fold increase was observed for *CRISP3* in *ERG*-positive tumours. Imunohistochemistry results showed a marked overexpression of *CRISP3* in 66% of the carcinomas, but no clear distinction could be made between fusion-positive and fusion-negative lesions at the protein level.
Conclusions: *CRISP3* mRNA is strongly up-regulated in *ERG*-rearranged tumours, providing a good surrogate marker for the TMPRSS2-ERG fusion.

802 The epidermal growth factor receptor (EGFR) is frequently overexpressed in uveal melanoma as a consequence of chromosome 7 polysomy and miRNA128b downregulation

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Background: Intraocular (uveal) melanomas represent 5% of all melanomas and about 40% of patients develop metastases, usually in the liver, during the first 5 years after diagnosis. Despite advances in the prognostic assessment through cytogenetics and molecular genetic techniques, current therapies are inefficacious for metastatic disease. In order to identify drug targets, we are analyzing gene expression profiles of primary uveal melanomas.
Methods: Expression profiling was performed on 40 samples using Affymetrix HGU133Plus2 arrays. Microarrays for miRNA screening were produced using the Exiqon library version 10.0. Differentially expressed miRNA and mRNAs were validated by qRT-PCR. Copy number of chromosomes 3, 8 and 7 was assessed by fluorescent in situ hybridization (FISH) on histological sections. Array based comparative genome hybridization (CGH) was performed on selected samples using Affymetrix 250K SNP arrays.
Results: mRNA expression analysis identified highly differential expression of several drug targets among which EGFR, a target of specific kinase inhibitors and therapeutic antibodies. We found a significant inverse correlation between the expression miR-128b and its target, EGFR. EGFR overexpression alone does not significantly correlate with recurrence yet the combination with miR-128b does. Polysomy of chromosome 7 also contributes to the overexpression of the receptor. Array CGH and FISH analyses reveal the presence of uveal melanoma subtypes with polysomic chromosomes 7.
Conclusions: EGFR expression, chromosome 7 polysomy and miR-128b expression may contribute to prognosis in uveal melanoma. EGFR is an interesting target for personalized therapy since over-expression is observed in a defined subset of patients who might benefit from therapy with inactivating antibodies.

803 PI3K signalling pathway is activated by PIK3CA gain and overexpression in prostate tumours, but PI3KCA, BRAF, KRAS AND AKT1 mutations are an infrequent event

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Background: The PI3K-AKT and RAS-MAPK pathways are deregulated in a wide range of human cancers by gain- or loss-of-function in several of their components, including *PIK3CA, KRAS, BRAF* and *AKT1*. Our purpose has been to identify genetic alterations in members of these pathways in prostate cancer.
Material and Methods: Eighty-one prostate tumours, 58 from prostate cancer alone (group G1) and 23 from bladder and prostate cancer patients (G2) are the subject of this study. In 20 of these 23, the bladder tumours were also analysed. *PIK3CA, KRAS, BRAF* and *AKT1* hot spot codons and the surrounding exon regions were studied by PCR and direct sequencing, and *BRAF* also by pyrosequecing. *PIK3CA* mRNA expression was tested by qRT-PCR in 19 prostate tumours, and 32 samples were analyzed by FISH to test copy number gain of *PI3KCA* gene. Immunohistochemistry for AKT and pAKT was performed in 46 and 20 prostate tumours respectively.
Results: Five of 19 (26.3%) prostate tumours with Gleason score $\geqslant 7$ showed *PI3KCA* mRNA overexpression, and *PI3KCA* copy gain was detected in 9 of 32 (28%) prostate tumours. Three of 20 (15%) matched bladder tumours, displayed mutations in *PI3KCA*, *KRAS* and *AKT1*, the corresponding prostate tumours being *wt.* We also detected a not previously described *PIK3CA* polymorphism (IVS9+91) in two prostate tumours. Thirty-four percent of samples overexpressed AKT protein, and there is a statistical association ($p = 0.013$) between strong immunostaining and mRNA overexpression and/or copy number gain of *PI3KCA* gene.
Conclusions: *PI3KCA* gene is deregulated by mRNA overexpression and DNA gain in about 26−28% of prostate tumours and the presence of these gene alterations is statistically related to AKT protein overexpression ($p = 0.013$). There is an association between mRNA overexpression and high-grade tumours ($p = 0.040$), but not with FISH status or AKT protein. *PI3KCA*, *BRAF*, *KRAS* and *AKT1* mutations are a very infrequent event in prostate tumours. Gene is deregulated by mRNA overexpression and DNA gain in about 26−28% of prostate tumours and the presence of these gene alterations is statistically related to AKT protein overexpression ($p = 0.013$). There is an association between mRNA overexpression and high-grade tumours ($p = 0.040$), but not with FISH status or AKT protein and mutations are a very infrequent event in prostate tumours.
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