extensive in patients with HER2+ tumors in comparison with HER2- carcinomas. Surprisingly, patients with triple-negative tumors had the minimal metastatic involvement of CNS defined by size and number of lesions. Phenotype did not correlate with local response to therapy and OScns.

Conclusions: Our study has confirmed the dependence between primary tumor phenotype and the time of incidence of metastatic brain affection and character of their spread. Our results encourage the inclusion of CNS imaging examination (CT or MRI) into the regular restaging of patients with HER2 positive or triple-negative primary breast cancer, who are at high risk for early development of CNS dissemination after the first distant metastatic event have occurred. Especially, in case of triple-negative tumors, there is higher probability for early detection of limited CNS metastatic involvement. Supported by IGA Ministry of Health, CZ. Grant No.:NR/8335-3.

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Association analysis of XRCC1 and XRCC3 polymorphisms with normal tissue reactions after pelvic irradiation

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Background: The purpose of the study was to investigate the association of five polymorphisms in two DNA repair genes XRCC1 (Codon 194 Arg/Trp; Codon 280 Arg/His; Codon 399 Arg/Gln) and XRCC3 (Codon 241 Thr/Met; IVS5-14 17.893) with the development of acute side reaction after pelvic irradiation for gynecologic malignancy.

Materials and Methods: The sample included 125 women with cervical or endometrial cancer, recruited from 2005 to 2008. They received external beam radiotherapy as primary or adjuvant treatment after surgery. The acute normal tissue morbidity in the pelvic area was evaluated using the NCI CTCAE v3.0. DNA was isolated from venous blood and RFLP analysis performed for genotyping. The patients reactions were separated in two groups: "no or slight reactions" (grade 0 and 1) and "moderate and severe reactions" (grade 2 and 3). No grade 4 reactions were recorded. The side effects were subdivided into gastrointestinal and genitourinary. Moderate and severe gastrointestinal reactions were observed in 77 patients, while 48 patients had no or slight reactions. The moderate and severe genitourinary reactions were found in 48 patients and 77 patients had no or slight reactions.

Results: Significant association was found between XRCC1 Codon 280 Arg/His and moderate and severe genitourinary side effects. The genotype G/G has a protective role, while the presence of mutated allele enhance the radiosensitivity (p=0,0045). No significant difference was found for the other XRCC1 and the investigated XRCC3 gene polymorphisms.

Conclusion: The results of the present study support the contribution of XRCC1, but not XRCC3 gene for the occurrence of early genitourinary reaction after gynecologic pelvic irradiation.

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SPR label-free ranking of small molecule negative modulators of adrenomedullin

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Surface Plasmon Resonance (SPR) is one of the most informative technologies for generating binding data (kinetic, affinity, thermodynamic parameters, binding stoechiometry). Weak affinity interactions can be detected and quantified because complex formation is monitored in real-time. SPR is thus a promising tool not only for screening libraries of chemical compounds, but also for structure-activity relationship studies which require ranking of a series of related compounds for their binding properties.

Adrenomedullin (AM) is a 52 amino-acid peptidic hormone, whose dysfunction is related to several diseases, such as diabetes, hypertension, and cancer. A Surface Plasmon Resonance (SPR) biosensor (Biacore T100®, GE Healthcare Biacore) was used to screen against AM, a collection of 21 synthetic compounds generated from a previously identified AM negative modulator.

AM was immobilized on a sCM5 sensor chip surface. Compounds were injected over AM and reference surfaces at concentrations ranging between 25 and 200 µM. Binding data were obtained after reference subtraction, DMSO correction and molecular weight adjustment. Equilibrium SPR responses were low (between 1 and 14 resonance units), corresponding to binding affinities (Kd) in the 50-500 uM range.

The data generated were used to derive a three-dimensional quantitative structure-activity relationship (3D-QSAR) model which was useful to identify relevant features for an effective binding to AM. These compounds have potential interest as anti-angiogenic and anti-tumour agents.

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Genetic and epigenetic alterations in esophageal squamous cell carcinomas from Brazil

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Background: Esophageal cancer (Squamous Cell Carcinoma – ESCC) is one of the ten most common malignancies and it is the sixth cause of cancer-related death in the world. Epigenetic alterations, such as the hypermethylation of CpG islands, are important events in cancer development and are a common way of inactivating tumor suppressor genes.

Methods: In this study, we analyzed by real-time PCR the spectrum of the expression of genes involved in cell cycle and epigenetic regulation in 65 ESCC and normal adjacent mucosa from patients from Southeastern or South Brazil. We further tried to organize them in subgroups and to analyse the potential of these genes to be used as molecular markers for ESCC. The genes analysed were DNMT3B, MBD4, p14ARF, p16INK4a, HDAC1, HDAC2, p21waf/CIP1, TP53, KMT-6 and GADD45a. We also analysed the methylation status of the promoter region of p14ARF and p16INK4a.

Results: The methylation analysis revealed that p14ARF was methylated in 7.1% and p16INK4a was methylated in 35.7% (in 70% of those that presented a lower expression in the tumor when compared to the normal mucosa) of ESCC samples. We performed a cluster analysis of the data that showed that DNMT3B expression may be an important differentiator of tumors in relation to normal tissue in ESCC, and that patients from Rio de Janeiro and Porto Alegre show different profiles of gene expression.

Conclusion: Our results suggest that the low expression of p16INK4a is related to the methylation of its promoter region. Our results also suggest that a higher expression of DNMT3B in ESCC is an important event and that ESCC from patients from different regions of Brazil, and exposed to different etiological factors, may present different molecular profiles of gene expression.

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Use of a cocktail of biomarkers in serum and urine to improve detection of prostate cancer

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Prostate cancer (PCa) is one of the most common tumors in men. Early detection of PCa relies on the determination of PSA levels, digital rectal examination, and ultimately on analysis of prostate biopsy. Because PSA values over than 4 ng/mL are suggestive of PCa, patients over that threshold value have to be subjected to biopsy. A rising PSA (>0.75 ng/mL per year) is also suspicious for PCa, even if PSA is in the normal range. However, PSA can also be increased due to BPH and prostatitis and thus, a large proportion of men undergoing biopsy do not have PCa. Indeed, the PSA test has a high sensitivity (>80%), but lacks specificity (20%). This situation has prompted the search for novel non-invasive biomarkers that may predict which patients will not benefit from prostate biopsy. Because of the inherent molecular heterogeneity of PCa, measurement of a single new marker could underestimate the presence of malignant tissue. The purpose of our study was to quantify a cocktail of biomarkers in blood and urine samples with the goal of improving specificity in the diagnosis.

Urine after rectal massage, and serum samples were obtained from 113 men with ages between 50 and 78, and PSA levels from 0.4-23.3ng/mL. 15 corresponded to patients with normal prostates, 44 showed BPH, and 54 had PCa. Biomarkers analyzed in serum were the humoral response to AMACR, and MMP-2 levels (both of them by ELISA). Hypermethylation of GSTP1 and RASSF1a was evaluated in urine samples by MSP. Sensitivity and specificity were computed with Epiinfo v6.1; discriminant function analysis was performed with SPSS v15, and comparison between ROC curves areas using a Chi Square Test (computed with Stata v9 software).

Areas under the ROC curves were as follows: 0.476 for PSA; 0.532 for AMACR; and 0.706 for MMP-2. Sensitivity and specificity for methylation status was 53.3% and 47.7%, respectively. Discriminant function analysis