treatment appears to be related with a response and associated with disease-free and overall survival. We recently demonstrated that circulating mitochondrial (mt) plasma DNA and RNA is a strong prognostic marker for survival in patients with prostate cancer (Clin Cancer Res 2007, Mehra et al.). The basis for this study was to assess whether these findings may be translated to other tumor types, and whether mtDNA/RNA can be seen as a pan-tumor marker.
Methods: We collected plasma from 198 cancer patients (prostate, head-and-neck, renal, and colorectal cancer) and 40 healthy subjects. Nucleic acids were isolated and mitochondrial and genomic nucleic acids were quantified using a PCR-based real-time detection and quantification method. The amplified mtDNA transcript encodes 16s rRNA, and the mtRNA transcript encodes cytochrome c oxidase subunit 1 (COX1). Using standardized cut-off points, mt nucleic acids were assessed as discriminatory marker for cancer, and as prognostic marker based on 2-year survival data.
Results: We demonstrate that mtRNA copies are increased in plasma of cancer patients compared to healthy subjects ($p = 0.001$). Patients with mtRNA copies above the normal range found in healthy controls, showed a trend to poorer survival after two-year follow-up (Log rank 3.21 with $P = 0.07$). The patients with highest mtRNA copies (above 50th and 75th percentile) showed significantly decreased survival, when compared to the patients with lower copy number (Log rank 5.05 with $P = 0.02$ for 75th percentile). We found no significant differences in survival based on mtDNA copies.
Conclusions: mtRNA copies in plasma of 198 cancer patients are increased compared to healthy controls. Patients with mtRNA copies above the normal range found in healthy controls, showed poorer survival. Standardized cutoffs for mtRNA could significantly discriminate between good and poor prognosis cancer patients, independent of cancer type. Plasma mtRNA is a prognostic factor that deserves further study as a pan-tumor marker.

P15

BRCA1 mRNA expression levels are associated with clinical responses to front-line docetaxel/gemcitabine in patients with lung adenocarcinomas in an expanded multicentre phase II study

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Background: Cis-platin based chemotherapy improves survival and symptoms control but its toxicity cannot be easily managed or prevented. Non-platinum-containing combinations offer similar survival times to the corresponding platinum-containing combinations. RRM1 plays a central role in the metabolism of gemcitabine and its overexpression in the tumor cell seems to offer resistance to the drug. BRCA1, a regulator of mitotic spindle assembly, is also associated with sensitivity to taxane. The efficacy of the docetaxel–gemcitabine (DG) regimen in patients with advanced lung adenocarcinomas in correlation with the expression of these two genes in the tumor cells was investigated.
Methods: Chemotherapy-naive patients, with locally advanced or metastatic lung adenocarcinomas and performance status (PS) $\leqslant 2$ (ECOG) received gemcitabine 1100 mg/m^2 (days 1 + 8) and docetaxel 100 mg/m^2 (day 8). rhG-CSF was given from day 9 to day 15. BRAC1 and RMM1 mRNA levels were determined by a quantitative Real-Time PCR, after RNA isolation from microdissected cells from the patients' primary tumors.
Results: Fifty-three patients (45 men and 8 women; median age 60 years) were enrolled. Amplification of at least one gene could be performed in 44. High levels of BRCA1 mRNA were significantly associated with response to treatment ($p = 0.024$), but not TTP and OS. For patients with BRCA1 mRNA levels in the upper quartile of expression a higher response rate ($p = 0.022$) and TTP ($p = 0.048$) but not OS ($p = 0.139$) was observed. Only patients with RRM1 mRNA levels in the bottom quartile experienced a benefit for the treatment with significantly prolonged TTP ($p = 0.044$) and OS ($p = 0.02$) and a trend for higher RR ($p = 0.62$). Response rate was, also, significantly higher for patients with high BRCA1/low RRM1 expression level in comparison with patients with low BRCA1/high RRM1 expression of both genes ($p = 0.016$).
Conclusions: BRCA1 and RRM1 expression is potentially an important tool for use in the management of patients with NSCLC and prospective studies are needed for the evaluation of their role for predicting differential chemosensitivity and tailoring chemotherapy in these patients.

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Clinical experiences with therapeutic derivatives of the anti-ED-B fibronectin immunoprotein L19

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Background: One avenue towards the development of more selective anti-cancer drugs consists of the targeted delivery of bioactive molecules to the tumor environment by means of binding molecules specific for tumor-associated markers. The use of antibodies specific to markers of neoangiogenesis is particularly attractive, in view of the ready accessibility of these structures within the solid tumor mass and the pathophysiological relevance of angiogenesis in cancer. The human monoclonal antibody L19 is specific to the extra-domain B (EDB) of fibronectin, which represents one of the best characterized and validated antigens associated with neoangiogenesis. This antibody has been produced in several formats (e.g., scFv, scFv fusions, SIP, IgG), which have been shown to preferentially localize at neoplastic sites in rodent tumor models and in patients with cancer using nuclear medicine techniques.
Methods: Phase I-II trials with L19IL2 (a fusion protein consisting of scFv(L19) and of recombinant human interleukin-2) and 131I-L19-SIP (a radioiodinated version of the L19 antibody in SIP format) have been conducted in European countries in patients with solid tumors.
Results: From November 2005 to March 2007 a Phase I trial of L19IL2 has been carried out in 21 patient with solid tumor. We explored five dose levels (5, 10, 15, 22.5, 30 Mio IU IL2 equivalent) in a modified Fibonacci dose-escalation study. L19IL2 was safely administered in an outpatient modality. All toxicity was manageable and reversible. Two dose-limiting toxicities occurred in the upper level dose (30 Mio IL2 equivalent): a Grade 2 increase of creatinine level during the first cycle and hypotension requiring vasopressor support. We identified 22.5 Mio IU IL2 equivalent as the recommend dose for further phase II study. Seven patients experienced disease stabilization (confirmed in two cases): 4 patients with renal cell carcinoma (RCC), one patient with biliary tract adenocarcinoma, one with peritoneal mesothelioma. A disease-oriented study in patients with renal cell carcinoma is still ongoing. The L19 antibody both in scFv and SIP formats has been studied in more than 70 patients with cancer in a clinical trial featuring the administration of radiolabeled product for dosimetric calculation. The SIP format showed a clearly superior targeting capability and therefore has been chosen for radioimmunotherapy in those patients featuring a tumor radiation dose which was at least ten-fold higher compared to the dose delivered to the bone marrow. The study is ongoing at the European Institute of Oncology and at two additional centers.
Conclusions: The human immunoprotein L19 represents a good-quality validated agent, which can be used for the construction of innovative anti-cancer therapeutic agents selectively directed against tumors by targeting markers of neoangiogensis. Studies using L19IL2 in combination with gemcitabine for patients with pancreatic cancer and of L19 fused to TNFa for a variety of different malignancies are ongoing.

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BRCA1 and BRCA2 polymorphisms and intronic variants: which pathological role?

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Background: Genetic polymorphisms are variants in individual genomes which could contribute to variability in both pharmacokinetic and pharmacodynamic drugs. The aim of our study was to evidence the possible pathological role of polymorphisms and intronic variants of BRCA1 and BRCA2 genes in familial breast cancer of Apulia population.
Methods: 110 patients affected by familial breast and/or ovarian cancer have been consecutively enrolled according to pathological features, family history and BRCA mutation risk. All of them came from Genetic Counselling Program of National Cancer Institute of Bari and were nominated for BRCA1 and BRCA2 genes genetic testing. DNA extracted from blood sample was amplified and used in pre-screening analysis by dHPLC. DNA sequencing was performed on both strands of two independent PCR products by cycle sequencing.
Results: In the present series, BRCA1 resulted mutated in 14% (15/110) while BRCA2 in 4% (5/110) of cases. We have found four different type of BRCA1 mutations: 5382insC, 4647delA, 172delC and R1495M, and five different mutations in BRCA2 gene: 2024del5, 6024delTA, 6714delACAA, Lys3326Stop and 6696delTC. We also studied the most frequent polymorphic alterations identified in both genes: in particular