and AAbs may considerably increase the efficacy of SERPA method to identify relevant cancer biomarkers.

PP 91

MicroRNA-mediated breakage of tumor cell differentiation

V. Halytskiy. *Palladin Institute of Biochemistry of the National Academy of Sciences of Ukraine, Kiev, Ukraine*

Background: Tumor growth is tightly associated with regular shifts in microRNA (miRNA) expression pattern. Expression of several miRNAs, e.g. miR-21, miR-23a/b, miR-100, miR-146a, miR-155, mir-181, miR-206, miR-221 and miR-222, is up-regulated in leukemia cells. This investigation aims to identify how abnormalities in miRNA network contribute to the arrest of tumor cell differentiation.
Materials and Methods: miRNA targets within gene transcripts were predicted in silico using TargetScan software.
Results: miRNA mir-21 silences genes encoding transcription factors Meis1 and Sox2 (as well as nuclear factor NFIB that inhibits NF-kappaB, a key element of antiapoptotic pathway). miR-125b targets transcript of genes encoding NFIB, transcription factors Stat3, IRF4, Ets1 and IL-6 receptor. Transcripts of genes encoding transcription factors EBF1, CEBPB, Ets1, Meis1 and PU.1 carry miR-155 binding sites. miR-181 can target transcripts of genes encoding transcription factors Ets1, Foxp1, Runx1, MITF, Bcl6 and Blimp1. Also, mir-23a/b can suppress genes encoding MITF and Blimp1. miR-150 and miR-23a/b can target transcript of gene encoding transcription factor IRF8. miR-29b suppresses gene encoding T-bet (TBX-21). miR-29b, miR-146a, miR-206 and miR-219−5p silence gene encoding transcription factor Bcl11a. miR-206 targets transcripts of genes encoding transcription factors EBF1 and Lef1 as well as retinoic acid receptor beta RARB. miR-221 and miR-222 silence gene encoding receptor c-Kit, transcription factors Ets1 and Fos.
Conclusion: Leukemia cells up-regulate expression of miRNAs that silences genes encoding key elements of cell differentiation network. EBF1 is a master regulator for B-cell development, as well as T-bet is for Th1-cell differentiation. Transcription factors Bcl11a, Ets1, Foxp1, Runx1, MITF, Bcl6, IRF4, IRF8, Blimp1 and IL-6 receptor are responsible for some stages of lymphoid cell differentiation and for recombination in immunoglobulin gene loci. Factors CEBPB and Meis1 are required for myelopoiesis. Illegitimate miRNA expression can directly repress these stage-specific genes; thereby leukemia cells can lose the normal cytokine susceptibility. As a result, the course of cell specialization proves to be complicated, requiring a high concentration of cytokines, or appears to be impossible at all, and transformed cells proliferate and accumulate, forming a tumor.

PP 46

In vivo imaging of modulation of IGF-1R expression in breast cancer models

S. Heskamp, O.C. Boerman, W.J.G. Oyen, J.D.M. Molkenboer-Kuenen, W.T.A. van der Graaf, H.W.M. van Laarhoven. *Radboud University Nijmegen Medical Centre, Nijmegen, The Netherlands*

Background: The insulin-like growth factor 1 receptor (IGF-1R) is a new target for breast cancer treatment. In vitro studies have shown that IGF-1R expression can predict response to IGF-1R targeted therapy. In vivo, other factors affect targeting of antibodies to tumors, such as vascular density, vascularity and interstitial pressure. Therefore, uptake of anti-IGF-1R antibodies in a tumor may be a better predictor for response to IGF-1R targeted treatment than immunohistochemical analysis of IGF-1R expression. The aim of the study was to determine whether immunoSPECT with radiolabeled R1507, an antibody directed against IGF-1R, can be used to measure IGF-1R expression and accessibility in vivo.
Materials and Methods: BALB/c nude mice with MCF-7 xenografts, were implanted subcutaneously with estradiol pellets. Three days later, mice were injected with 20 MBq 111In-R1507. Alternatively, mice were treated with tamoxifen and after seven days of treatment, 111In-R1507 was administered. In a third experiment, mice with SUM149 tumors were treated with a single dose of bevacizumab. Four days after treatment, mice received 111In-R1507. In all experiments, three days after injection of 111In-R1507, SPECT images were acquired and the biodistribution was determined ex vivo. IGF-1R expression was analyzed with immunohistochemistry.
Results: Uptake of 111In-R1507 in the tumor was significantly higher in the estradiol treated mice compared to non-treated mice (14.2 versus 10.9%ID/g ($p = 0.016$)). Differences in tumor uptake were visualized with immunoSPECT and correlated with IGF-1R expression as determined immunohistochemically. Tamoxifen did not affect tumor uptake of 111In-R1507, although on immunohistochemistry membranous IGF-1R expression was decreased. Bevacizumab treatment significantly decreased tumor uptake of 111In-R1507 (19.9 versus 26.6%ID/g for treated versus non-treated mice($p = 0.002$)), while immunohistochemically IGF-1R expression was unaltered.
Conclusion: ImmunoSPECT with 111In-R1507 is a sensitive method to measure modulations in IGF-1R expression caused by estradiol treatment. However, as illustrated by the results of tamoxifen and bevacizumab treatment, tumor uptake of 111In-R1507 does not necessarily correlate with IGF-1R expression. These data underscore that target availability also affects tumor targeting by antibodies. Therefore, immunoSPECT of IGF-1R expression with 111In-R1507 could be a better predictor of response to anti-IGF-1R antibodies than immunohistochemical analysis of IGF-1R expression.

PP 45

In vivo isolation of circulating tumor cells

A. Heyden, T. Tomasi, N. Zeni, S. Herold, P. Nowaczyk, A. Schmitz, T. Krahn, M. Zabel, D. Murawa, K. Luecke. *GILUPI GmbH, Potsdam, Germany*

Background: Circulating tumor cells (CTC) from cancers of epithelial origin frequently give rise to metastasis responsible for most cancer-related deaths. In addition, they can also serve as biomarker source to improve the management of cancer treatment. However, current technologies for isolation of these extremely rare cells are limited by their capability to detect sufficient cell numbers in the majority of cancer patients. In order to increase the sensitivity of CTC detection, GILUPI has developed a functionalized and structured medical wire (FSMW) that allows CTC isolation directly from the patients' blood stream.
Materials and Methods: In a clinical trial with 30 breast cancer patients, CTC were isolated by the medical wire that has been inserted into the patients' vein for 30 minutes. The medical wire mediates target CTC isolation by antibodies directed against the epithelial cell adhesion molecule (EpCAM). To confirm that the target CTC are bound to the wire, immunocytochemical staining against EpCAM or cytokeratin is performed as well as staining against CD45 for negative cell selection. In addition, 6 further patients are scheduled for two subsequent medical wire applications to evaluate the reliability of this method to detect comparable CTC numbers on the same day.
Results: Analysis of the breast cancer patients regarding the performance of the medical wire indicates besides very good biocompatibility and the absence of any side effects substantially higher CTC detection rates compared to the FDA-approved CellSearch method. This result proves the in vivo application of the medical wire technology with access to the whole blood stream being superior to methods isolating CTC from relatively small blood samples in vitro.
Conclusion: Increased CTC detection rates of the medical wire may serve to improve early detection, prognosis, and therapy monitoring of cancer patients in future. As this technology is an efficient method for tumor cell enrichment, subsequent molecular analysis of these cells have been initiated in collaboration with Bayer HealthCare and Prometheus to eventually establish more personalized treatment regimes.

PP 6

The metabolic response using FDG/PET for predicting tumor response and prognosis after pre-operative chemoradiotherapy (CRT) in patients with locally recurrent rectal cancer (LRRC)

M. Ikeda, M. Sekimoto, R. Handa, J. Nishimura, I. Takemasa, T. Mizushima, H. Yamamoto, Y. Doki, M. Mori. *Department of Surgery, Graduate School of Medicine, Osaka University, Suita, Japan*

Background: Local recurrence is the most common type of recurrence after resection of advanced low rectal cancer. Radical resection of recurrent tumor including adjacent tissues such as bladder, sacral bone is the only means of cure. Even R0 resection, incidence of local re-recurrence is 20 to 60%. In order to reduce the incidence of local re-recurrence, we have employed pre-operative CRT. The aim of the study was to predict tumor regression in pre-operative CRT and prognosis after radical resection using 18F-fluorodeoxyglucose-positron emission tomography/computed tomography (PET/CT) and serum carcinoembryonic antigen (CEA) in patients with LRRC.
Materials and Methods: Fourteen males and 6 females with median age of 61 (range 36 to 70) who had preoperative CRT and underwent R0 resection were evaluated. PET/CT was performed before and after three weeks of pre-operative CRT in all patients. Histological diagnosis was made based on resected specimen. The metabolic response of the tumor was assessed by determining the maximal standardized uptake value (SUVmax), absolute difference [ΔSUV(max)], and SUV reduction ratio (SUVRR) on pre- and post-CRT PET/CT scans. The serum CEA, absolute difference, and the CEA reduction ratio were also determined.
Results: Median pre- and post-CRT SUVmax was 7.8 and 3.1, respectively. The median serum pre- and post-CRT CEA was 12 ng/ml and 3.5 ng/ml, respectively. Ten patients (50%) were classified as responders