tumor biology and pathology

1688PD
THE COMBINED EXPRESSION OF CXCR7 AND ITS LIGAND CXCL12 IS A MARKER FOR UNFAVORABLE PROGNOSIS IN GASTRIC CANCER
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Background: Chemokines and their receptors have been shown to play a critical role in cancer growth and metastasis. In particular, recent data have suggested that the chemokine CXCL12 and its receptor CXCR7 (also known as RDC1), which has been recently identified as a chemokine receptor, have key functions in promoting tumor development and progression. However, there is little information regarding their expression and clinical relevance in gastric cancer. Here we investigated for the first time the effects of combined CXCR7 and CXCL12 expression on the prognosis of patients with gastric cancer.

Methods: We studied CXCL12 and CXCR7 protein expression in 221 specimens of primary gastric cancer using immunohistochemistry, and investigated the relationship between CXCL12/CXCR7 expression and clinicopathological features and clinical outcomes.

Results: Patients were categorized into four groups according to CXCR7 and CXCL12 expression: low CXCR7/low CXCL12, high CXCR7/low CXCL12, low CXCR7/high CXCL12, and high CXCR7/high CXCL12. No significant differences existed in age, gender, histology, tumor location, lymphovascular invasion among the four groups. However, high CXCR7/high CXCL12 expression in tumor cells was significantly associated with invasion depth of the tumor (T status; P < 0.001), lymph node involvement (N status; P = 0.002), higher tumor stage (P = 0.002), and proportion of tumor size >5 cm (P = 0.006) compared to tumors with low CXCR7/low CXCL12 expression or high CXCR7/low CXCL12-low CXCL12/high CXCL12 expression. Furthermore, patients with high CXCR7/high CXCL12 expression had the worst prognosis (5-year survival rate 30.6%; median, 2.3 years; range, 0.1 - 5.7 years) compared to those of other patient groups (5-year survival rate 52.4%; median, not reached; log-rank test, P = 0.008).

Conclusions: CXCR7 and CXCL12 are useful prognostic factors in gastric cancer, and the combination of high CXCR7 protein expression with high CXCL12 expression suggests a dismal prognosis.

Disclosure: All authors have declared no conflicts of interest.

1689P
CACHEXIA IN Pancreatic CANCERS. COMPARISON OF ADENOCARCINOMA AND NEUROENDOCRINE TUMORS
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Background: Patients with pancreatic adenocarcinoma (pCA) have high propensity for weight loss and cachexia. Patients with pancreatic neuroendocrine tumors (pNET) maintain their muscle mass for long period of time. Aims: We undertook a study to quantify the magnitude of cachexia in patients with pNET and compare it with pCA.

Methods: An audit of the nutrition clinic database from 2000 to 2011 was reviewed and identified patients with pNET or pCA who had full nutritional workup including subjective global assessment scores (SGA) and body mass index (BMI). Cachexia was defined as a SGC score = C or BMI <18.5 Kg/M2. Cachexia and other variables of pNET patients were compared with pCA.

Results: There were 328 pCA patients and 88 pNET patients. There were 286 males and 130 females with age ranging from 1 to 95 years (Mean 53 years for pCA and 48 pNET; p = 0.003). The SGA score were A= 39.5%; B= 55.3%; C= 5.3% in pNET group and A= 12.7%; B= 52.5%; C= 38.4% in pCA group. BMI below 18.5 was significantly higher in the pCA group. Malnutrition (SGA B + C) and Cachexia were significantly associated with pCA. Odds ratio for Cachexia in pNET subjects after adjusting for age and sex was 0.249 (p = 0.0089). Cachexia was absent in several pNET patients with multiple liver metastases. pNET patients developing cachexia had high grade tumors with MIB-1 score over 20%.

Conclusions: Patients with pNET are resistant to cancer cachexia while those with pCA are highly susceptible to cancer cachexia. Further research on the biochemical and molecular pathways will help to understand cancer cachexia in pancreatic cancers.

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1690P
HER SIGNALING EFFECTS ON THE NK CELL-MEDIATED CYTOTOXICITY VIA REGULATION OF MHC CLASS I-RELATED CHAIN A/B IN CANCER CELLS
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Background: Overexpression of the HER receptors is associated with a poor prognosis in several types of cancer. Currently the HER receptor-targeted therapies are in clinical practice or evaluated within clinical trials, including treatment with monoclonal antibodies mediating activation of antibody induced innate or adaptive cellular immunity. A better understanding of how HER signaling in tumors influences cellular immune mechanisms is therefore warranted. We previously reported that the HER3 signaling enhanced the expression of MHC class I-related chain A and B (MICA/B), resulting in a phenotype promoting tumor escape from innate immunity. Here, we demonstrate that HER3 signaling in breast cancer (BC) cells increases MICA/B expression via AKT pathway, while EGFR signaling in non-small cell lung cancer (NSCLC) cells decreases MICA/B expression.

Material and methods: A possible influence of HER signaling on MICA/B expression in BC and NSCLC cell lines was investigated. To assess the consequences of HER activation, cells were either treated with the HER3 ligand NRG or EGFR ligand EGF, or left untreated. NK cell-mediated cytotoxicity against tumor cells was assessed by 51Cr release assay.

Results: Among the major pathways activated by HER3 signaling, the PI3K-AKT pathway was shown to predominantly regulate MICA/B expression in BC cells. Treatment with NRG promoted MICA/B expression, in a process that was antagonized by pharmacological and genetic interference with HER3 but not by ATM-ATR pathway inhibitor. These observations further emphasize that HER3 signaling directly, and not via genotoxic stress, regulates MICA/B expression. In contrast, treatment with EGF decreased MICA/B expression in NSCLC cells. Among the major pathways of EGFR signaling, the MAPK pathway was shown to predominantly attenuate MICA/B expression. As expected, activation of HER3 signaling enhanced MICA/B induced NK cell cytotoxicity, while activation of EGFR signaling attenuated it.

Conclusions: We conclude that HER signaling directly regulates the expression of MICA/B and that this may influence the recognition of tumor cells by the innate immune system.

Disclosure: All authors have declared no conflicts of interest.

1691P
THE SIGNIFICANCE OF EPHRIN RECEPTOR A4 (EPHA4) EXPRESSION IN PRIMARY PULMONARY NEOPLASMS
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Background: Ephrin receptors (Ephs) are frequently overexpressed in a wide variety of human malignant tumors, being associated with tumor growth, invasion, metastasis and angiogenesis. The present study aimed to evaluate EphA4 expression in lung cancer.
Material and methods: 101 patients who underwent surgical resection due to lung cancer were included in this study. None of them received any kind of treatment prior to surgery. 86 were men and 15 women with a mean age of 62 years old. The tumors were classified histologically as adenocarcinoma in 48, squamous cell carcinoma in 33, large cell carcinoma in 11 and small cell carcinoma in 9 cases. Using tissue microarray technology, 101 paraffin-embedded tissue samples were core, re-embodied to the final recipient block and used for Epha4 protein immunohistochemical expression. All analyses were performed by SPSS for Windows Software (SPSS Inc, Chicago, IL, USA) and a p value less than 0.05 was considered the limit of statistical significance.

Results: Epha4 positivity was noted in 36 out of 101 (36%) cases. Epha4 staining intensity was classified as mild in 11 (31%), moderate in 23 (64%) and intense in 2 (5%) out of 36 positive cases. Increased Epha4 positivity and moderate/intense Epha4 staining intensity were noted in adenocarcinoma and squamous cell lung carcinoma cases compared to large cell and neuroendocrine carcinoma ones (p = 0.0049 and p = 0.0170, respectively). Epha4 positivity and staining intensity were associated with the presence of lymph node metastases (p = 0.0349 and p = 0.0404, respectively). Epha4 staining intensity was also correlated with tumor histopathological stage (p = 0.0145). No association of Epha4 positivity nor staining intensity was noted with tumor histopathological grade, size and organ metastasis.

Conclusions: The current study supports evidence for possible Epha4 participation in lung cancer biological behaviour, implying also its potent role as a therapeutic target. However, further molecular and clinical studies are required in order to define the significance of Ephs and their ligands (ephins) in lung cancer prognosis and management.

Disclosure: All authors have declared no conflicts of interest.

Disclosure: A. Scott: A. S is an inventor of a patent for mAb806, and a consultant to efficacy of mAb806 in NSCLC are warranted.

Background: Ephitelial-to-mesenchymal transition (EMT) was originally proposed as a process of organogenesis. In recent years, the association between EMT and cancer invasion and metastasis has been advocated and actively investigated in various cancers. In addition, chemoresistance and cancer stemness could be involved in EMT, and the elucidation of this association might contribute to improved outcomes of hepatocellular carcinoma (HCC). We utilized surgical specimens of HCC from our department to examine the clinical implications of EMT.

Methods: One hundred and one patients with hepatocellular carcinoma, who underwent resection in our department between 1994 and 2003 were analyzed. The mRNA expression of E-cadherin and Vimentin were measured by quantitative real-time PCR and EMT status of each patient was determined as follows: Vimentin/ E-cadherin < 2 = Epithelial (E), Vimentin/E-cadherin ≥ 2 = Mesenchymal (M).

Moreover, transcription factors which are involved in EMT (Twist, Snail, Slug, Zeb-1, and Zeb-2) and also IL-6 and its receptor IL-6R were measured. The correlation between these values and clinicopathological factors and prognosis were analyzed statistically.

Results: 1) AFP values were significantly higher in the epithelial group than in the mesenchymal group (P = 0.029). There was no difference in overall survival, but a significant difference was found in disease-free survival (P = 0.042), which showed that patients with a mesenchymal tumor were more prone to have early recurrence than those with an epithelial tumor. 2) All transcription factors were more highly expressed in mesenchymal than in epithelial tumors, and in particular, Twist and Zeb-2 were significantly overexpressed (P = 0.0012, P = 0.0017, respectively). 3) IL-6 expression was also significantly higher in mesenchymal than in epithelial tumors, but there was no difference in IL-6R expression.

Discussion: Our study using resected surgical specimens suggest that EMT could be involved in cancer invasion and metastasis at the clinical level. In particular, Twist and Zeb-2 might be important for inducing EMT in HCC and patients with mesenchymal tumors are more prone to have early recurrence or metastases after resection. In addition, IL-6 might be an important factor for HCC EMT and could be a potential therapeutic target.

Disclosure: All authors have declared no conflicts of interest.
expression, but not in Gli3-undetectable HCT116 or DLD-1 cells. Silencing of endogenous Gli3 down-regulated colony formation and proliferation in HT29 and SW480 cells. However, truncated Gli3 (Gli3-R; repressor isoform) transduction had no effect on these properties although Gli3 expression was inhibited. After implantation of Gli3- or mock-transfected DLD-1 cells into immunodeficient SCID mice, tumor formation was observed in only Gli3-transfected DLD-1 group but not in mock control. In surgically resected colorectal cancer specimen, Gli3 expression was heterogeneously detected and Shh expression was highly observed.

Conclusions: Gli3-FL and Shh signals induce tumorigenicity in Gli3 independent manner, and Gli3-FL may be molecular targets for refractory colorectal cancer.

Disclosure: All authors have declared no conflicts of interest.

1695P FIBROBLAST INDUCED EPITHELIAL TO MESENCHYMAL TRANSITION (EMT) IN A NOVEL NON-SMALL CELL LUNG CANCER (NSCLC) MODEL

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Introduction: Different molecular processes lead to metastatic spread and the occurrence of tumour cell resistance to therapeutic interventions. Among them, the epithelial to mesenchymal transition (EMT) process plays a key role. During EMT, epithelial tumour cells lose the expression of specific proteins and adopt the phenotype of mesenchymal cells. These structural conversions are substantially dependent on the tumour microenvironment. EMT of tumour cells can induce drug resistance and metastasis. Thus, EMT inhibition may offer a new strategy for overcoming tumour progression.

Methods: To evaluate EMT in a non-small cell lung cancer (NSCLC) model a 2D and a 3D culture system was applied using both the human lung cancer cell line (A549) and the human lung fibroblast cell line (SV-80). For generating 3D cell spheroids, a novel system was established consisting of 96-well hanging drop microtiter plates (InSphero AG, Zürich, Switzerland). 2D co-culture assays were performed in transwell filter inserts (Costar). EMT was induced with transforming growth factor-β (TGF-β) or co-cultivation with fibroblasts in 2D/3D. The switch from epithelial to mesenchymal cells was monitored by Western Blot (WB) analyses of e-cadherin, vimentin and n-cadherin. Furthermore, immunohistochemistical analyses of e-cadherin, vimentin, α-smooth muscle actin, fibronectin, Ki-67 and CA-IX were done on paraffin embedded spheroids.

Results: EMT could be induced in the 2D not only by incubating tumour cells with TGF-β but also by co-culturing them with fibroblasts in transwell filter inserts. In A549 cells a change in morphology as well as in protein expression defined by WB analysis (down regulation: E-cadherin; up regulation: Vimentin, n-cadherin) could be detected. When cultivated in the 3D system, A549 cells showed an up regulation of the mesenchymal protein vimentin without TGF-β stimulation. Furthermore, a significant up regulation of vimentin, Ki-67, CA-IX, and a slight down regulation of e-cadherin could be measured compared to monocultures.

Conclusion: 3D culture represents a model to study EMT in tumour cell lines without addition of growth factors and thus reflects in vivo conditions closer than 2D culture.

Disclosure: All authors have declared no conflicts of interest.

1696P CANCER STEM CELL MARKERS IN CLINICAL PANCREATIC CANCER: IMPACT OF CD44+/CD24-/EpCAM+ EXPRESSION ON HISTOLOGY AND PROGNOSIS

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Purpose: Emerging evidence suggests that the capability of a tumor to grow and propagate is dependent on a small subset of cells within it, termed cancer stem cells (CSCs). In pancreatic cancer, CD44+/CD24-/EpCAM+ cells have been reported to be CSCs, however, the histological and clinical importance of these cells has not yet been investigated. Here we clarified the characteristics of CD44+/CD24-/EpCAM+ cells in clinical specimens of pancreatic cancer using immunohistochemical assay.

Materials and methods: We used surgical specimens of pancreatic ductal adenocarcinoma from 30 patients. In view of tumor heterogeneity, we randomly selected 10 high-power fields per case, and triple-positive CD44+/CD24-/EpCAM+ expression was identified using our scoring system. The distribution, histological characteristics, and prognostic importance of CD44+/CD24-/EpCAM+ cells were then analyzed.

Results: Among a total of 300 assessed fields, 41 (14%) were evaluated as triple-positive. The distribution of CD44+/CD24+/EpCAM+ cells varied widely among the 30 cases examined, and CD44+/CD24+/EpCAM+ expression was correlated with poor glandular differentiation and high proliferation (high Ki-67 labeling). Analysis of the three markers individually showed that CD44 and CD24 were also correlated with poor differentiation and high proliferation, while EpCAM was not. Survival analysis showed that CD44+/CD24+/EpCAM+ expression was not correlated with patient outcome; however, CD44 and CD24 each appeared to be correlated with poor prognosis.

Conclusion: In pancreatic cancer, CD44+/CD24+/EpCAM+ cells overlapped with poorly differentiated cells and possess high proliferative potential. In particular, double-positive CD44+/CD24+ cells seem to have relevance when considering clinical aspects.

Disclosure: All authors have declared no conflicts of interest.

1697P SEMULOPARIN EFFICIENTLY INHIBITS THROMBIN GENERATION TRIGGERED BY PANCREAS ADENOCARCINOMA CELLS BXPC3. DISTINCT ROLES OF ANTI-XA AND ANTI-IIA ACTIVITY

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Background: Venous thromboembolism (VTE) is a common complication in patients with cancer receiving chemotherapy. Currently no anticoagulant is approved for VTE prophylaxis in this setting. Semuloparin is an ultra-LMWH generated through a highly selective depolymerization of heparin which protects the antithrombin (AT) binding site in order to improve the benefit/risk ratio compared to existing anticoagulants.

Aims: We studied in vitro the mechanism of action of semuloparin on the inhibition of thrombin generation (TG) of human platelet poor plasma (PPP) triggered by human pancreatic cancer cells BXPC3. We compared the antithrombotic efficiency of semuloparin to that of enoxaparin and the specific AT-dependent factor Xa inhibitor fondaparinux.

Materials and methods: BXPC3 cells were suspended in PPP spiked with clinically relevant concentrations of semuloparin, enoxaparin and fondaparinux. The endogenous thrombin potential (ETP) and the mean rate index (MRI) of the propagation phase of TG were monitored with the CAT assay (Stago France) as described previously (Gerotziafas et al Thromb Res 2011). Anti-Xa and anti-IIa specific activities were assessed assays obtained from Diagnostica Stago.

Results: Both semuloparin and enoxaparin, at the concentration of 0.4 anti-Xa U/ ml, completely abrogated TG. Total inhibition of TG occurred in the presence of 0.002 anti-IIa U/ml of semuloparin and 0.05 anti-IIa U/ml of enoxaparin. Fondaparinux, even at concentrations higher than 2 µg/ml, reduced the MRI but did not completely affect the ETP and did not completely inhibited TG. The IC50 for the ETP of the anti-IIa activity of enoxaparin was 37-fold higher as compared to that of semuloparin.

Conclusion: In a cancer cell model of hypercoagulability, semuloparin reduced efficiently thrombin generation. The unique anticoagulant profile of semuloparin based on its high AT-affinity differentiates it from enoxaparin and fondaparinux. The residual anti-IIa activity of semuloparin amplifies its antithrombotic efficacy. This profile is expected to translate into an improved benefit/risk ratio.

Disclosure: All authors have declared no conflicts of interest.

1688P DO THE WELL-KNOWN RISK FACTORS OF BREAST CANCER HAVE THE SAME IMPACT ON DEVELOPMENT OF MOLECULAR SUBTYPES?

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Background: Although clinical differences between breast cancer (BC) subtypes have been well-described, etiologic heterogeneity have not been fully studied. The aim of this study was to assess the associations between risk factors and molecular subtypes of BC.

Methods: 1884 invasive BC cases were retrospectively analyzed. The odds ratios (OR) and 95% confidence intervals (CI) were estimated using multiple logistic regression analysis.

Results: 1249 patients had luminal A, 234 had luminal B, 169 had HER-2
overexpressing and 232 had triple negative BC. The age of ≥40 years was found to be a risk factor for luminal A (OR 1.41 95% CI 1.15-1.74; p = 0.001) and HER-2 overexpressing subtype (OR 1.51, 95% CI 1.01-2.25; p = 0.04). Women who were multiparous (OR 1.25 95% CI 1.03-1.23; p = 0.04) and who had their first full-term pregnancy ≥30 years (OR 1.25 95% CI 0.83-1.88; p = 0.04) were at increased risk of luminal A, whereas women with >2 children had a decreased risk (OR 0.68, 95% CI 0.47-0.97; p = 0.03). Breast-feeding was a protective factor for luminal BC (OR 0.74, 95% CI 0.53-1.04; p = 0.04). We found increased risks for postmenopausal women with HER-2 overexpressing (OR 2.20, 95% CI 0.93-5.17; p = 0.04) and luminal A (OR 1.87, 95% CI 0.93-3.90; p = 0.02) BC, who used hormone replacement therapy for ≥25 years. Overweight and obesity increased the risk of triple negative BC (OR 1.89 95% CI 1.86-3.37; p = 0.04) and OR 1.90 95% CI 1.30-3.61; p = 0.03), on the contrary, decreased the risk of luminal BC (OR 0.63 95% CI 0.43-0.95; p = 0.02 and OR 0.50 95% CI 0.32-0.76; p = 0.002, respectively) in premenopausal women. There were no significant differences between risk of BC subtypes and early menarche, late menopause, family history, postmenopausal obesity, oral contraceptive use, smoking, in vitro fertilization and blood groups.

Conclusions: Reproductive and hormonal characteristics were associated with luminal BC. Obesity and overweight increased the risk of triple negative subtype, particularly in premenopausal women. Older age and use of hormone replacement therapy were related to the risk of HER-2 overexpressing BC. Our data suggest a significant heterogeneity in association of traditional BC risk factors and tumor subtypes.

Disclosure: All authors have declared no conflicts of interest.

1699P IDENTIFYING TRANS-ACTING COPY-NUMBER ALTERATIONS IN LUNG ADENOCARCINOMAS

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Background: One of the main challenges in genomic oncology is to identify somatic copy-number alterations (CNAs) that include critical genes driving either the initiation or the progression of cancer disease. Most integrated genomic/transcriptomic analyses have primarily focused on identifying cis-acting CNAs, whose copy gains or losses are significantly associated with gene expression changes for the genes they regulate. In this work, we do not restrict our analysis to cis-acting CNAs but focus on CNAs whose copy number changes are associated with large number of significant transcriptomic changes distributed over the entire genome. We hypothesize that CNAs influencing a large number of transcripts either by direct or indirect effect are likely to harbor candidate targetable genes.

Material and methods: A homogeneous series of 129 early stage lung adenocarcinomas, profiled using both high resolution array-based genotyping (SNP-array) and gene expression, was analyzed. After classical preprocessing, we selected recurrent CNAs that were exclusively amplified or deleted using a previously published latent class model analysis (BMC Med Genomics, 2009 Jul 14;2:43). For each genomic region, we computed genome-wide statistics measuring the relationship between transcriptomic changes over the different levels of copy number changes (copy loss/modal/copy gain). Then, we reported the number of significant associations taking into account multiple testing. We finally focused on “trans-acting CNAs” defined as those having extreme values.

Results: Main trans-acting CNAs were found on 1q, 4q, 5q, 9p, 14q, harboring several known oncogenes/tumor suppressor genes. In contrast, other classical recurrent CNAs (such as 8p amplification) were not selected, suggesting they were not associated with significant phenotype changes, as defined by transcriptomic analysis. The relationships between each “trans-acting CNAs” and clinic-pathological variables, as well as outcome is discussed.

Conclusion: Using an integrative high-throughput microarray analysis in a series of lung adenocarcinomas, we identify “trans-acting CNAs”. We show the interest of focusing on these CNAs which are expected to harbor potential targetable candidate genes.

Disclosure: All authors have declared no conflicts of interest.

1700 WNT-2, BUT NOT WNT-1 EXPRESSION INCREASES DURING TUMORIGENESIS IN BREAST, PROSTATE, LUNG CANCER AND MELANOMA

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WNT-β/catenin pathway regulates cell cycle and proliferation. It is triggered by WNT ligands, and drives β-catenin regulated expression of cyclin D1, c-Myc, MMP7. Moreover β-catenin, along with E-cadherin, forms adherent junctions mediating cell adhesion. WNT-1 is a known oncogene and its expression occurs in several malignancies such as breast, prostate and lung cancers, while WNT-2 is a less known member of WNT ligands family. The aim of our study was to investigate the expression of WNT-1, WNT-2, β-catenin, E-cadherin and cyclin D1 in melanoma, breast, lung, prostate cancer in comparison to adjacent normal tissue and to further understand WNT ligands significance as a potential biomarker. Formalin-fixed, paraffin embedded samples were taken from 26 breast cancer patients, 22 non small cell lung cancers patients, 23 prostate cancers patients, 24 melanomas patients in I-III stage of the disease. Expression of WNT-1, WNT-2, β-catenin, E-cadherin and cyclin D1 was assessed by immunohistochemistry and analyzed by two independent histopathologists. Changes of studied proteins expression profiles between normal and malignant tissues were measured with Wilcoxon test, while comparison of expression of analyzed proteins between tumors was performed with Kruskall Wallis and post hoc test. A value of p < 0.05 was considered significant. The study received approval by the Local Bioethics Committee. In our study, WNT-1 cytoplastic expression was increased in breast cancer (p = 0.003) and melanoma (p = 0.0001), while in lung cancer it was decreased (p = 0.002). WNT-2 cytoplastic expression was increased in breast (p = 0.003), prostate cancer (p = 0.0002), melanoma (p = 0.0009), while in lung cancer WNT-2 increased expression showed a trend towards significance (p = 0.06). Moreover WNT-2 ligand was found in cell nuclei in all tumors and its expression level was increased in breast cancer (p = 0.007) and decreased in melanoma (p = 0.042). Our study revealed that WNT-2, but not WNT-1 expression was increased in all analyzed tumors. Moreover WNT-2 ligand was detected in cell nuclei, what could implicated its yet undiscovered role in gene expression regulation.

Disclosure: M. Wieszczyk, Maciej Wieszczyk is Chief Executive Officer of Celon Pharma Ltd.All other authors have declared no conflicts of interest.

1701 PROTEOMIC ANALYSIS OF HUMAN LUNG CANCER CELL LINES

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The causes and morphological appearances of human lung cancers are variable. We analyzed thirty seven seven lung cancer cell lines including thirty adenocarcinoma (ADC), three small cell carcinoma (SCLC), one large cell carcinoma (LCC) and one adenosquamous carcinoma (AdSc) with LC/MS and identified less than 500 proteins of each cell line. We compared proteomic profiles between EGFR mutations and K-Ras mutations, smokers and non-smokers, and non-small cell carcinoma (NSCLC) and small cell carcinoma (SCLC). Eleven proteins, CALR, KYNU, SLCA2, ALDH2, PGD, LAP, DLC1, KIAA0664, ILKAP, ABCA13 and PTGR1 are related to the relationship between cell lines with EGFR mutations and K-Ras mutations and some of them are associated to the signal network of cell cycle. Twelve proteins, PPP2R1B, RAP1A, RPS3, RNF113, TGM2, KIF22, UBA6, IFIT122, JIF16, AKR1C1/AKR1C2, RPS5 and AKR1C3 are related to the relationship between cell lines in from non/less-smokers and those from smokers and all proteins are associated to the signal network of cell morphology. Fifteen proteins, ANXA2, P4HB, ACTN4, MSN, ANXA5, IDH2, DPYS2, DPYS3, DPYS5, CKB, IPO8, MAP17, EFA4, TRIM22 and USP5 are related to the relationship between cell lines of NSCLC and SCLC, and some of them are associated to the signal network of cell cycle and some are associated to the signal network of cell morphology. Principle Component Analysis could separate NSCLC from SCLC distinctly. We also detected common proteins among each group as candidate markers of cancer stem cells. AKR1C1/AKR1C2 was detected as common proteins between EGFR mutations vs K-ras mutations and smokers vs non/less smokers and has been reported as one of cancer stem cell markers. These results suggested that this methodology was very useful to detect not only specific proteins of each group but also protein-related signal pathways.

Disclosure: All authors have declared no conflicts of interest.

1702 HISTOLOGY OF SYNCHRONOUS AND METACHRONOUS CONTRA-LATERAL BREAST CARCINOMA

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Background: Contra-lateral breast cancer (CBC) is considered to be the most frequent new malignancy after primary breast cancer (PBC). However, there are few reports about concordance of the histology and other major molecular characteristics between PBC and CBC. Also, to the best of our knowledge, there is no report about CBC histology according to the time of development from PBC.

Aim: To evaluate and compare PBC/CBC histology type according to time of development CBC. Age at CBC and other major molecular characteristics (tumor grade, estrogen/progesterone receptors, HER2) have also been analyzed. Patients and
A cohort of 113 CBC patients, without distant metastases, has been prospectively registered during 28 months. Patients are divided in 2 groups according to the time of CBC diagnose: 1. Synchronous, if the CBC (S-CBC) was diagnosed either simultaneously or within 6 months after PBC; 2. Metachronous (M-CBC) if CBC was diagnosed > 6 months after PBC. (Table 1)

<table>
<thead>
<tr>
<th>Pts N = 113</th>
<th>Synchronous</th>
<th>Metachronous</th>
</tr>
</thead>
<tbody>
<tr>
<td>S-CBC N = 49</td>
<td>Median age at CBC diagnose (yrs)</td>
<td>57 (33-74)</td>
</tr>
<tr>
<td>M-CBC N = 64</td>
<td>Median time to CBC (months)</td>
<td>0 (0 ≤ 6)</td>
</tr>
<tr>
<td>Lobular/Ductal (%)</td>
<td>Lobular/Ductal (%)</td>
<td>76%</td>
</tr>
</tbody>
</table>

**Results:** Patient with S-CBC are median 7 years older than patients with M-CBC (p=0.007). S-CBC is more likely to be of the same histological type (76%) than M-CBC (56%) (P= 0.006). In the whole analyzed group, and each subgroup separately, lobular carcinoma is registered in higher percentage (41%) than expected. For all other characteristics (tumor grade, estrogen/progesterone receptors and HER2 status) there was no statistical difference.

**Conclusion:** According to these results, it seems that patients destined to develop S-CBC or M-CBC, as a first recurrence site, have a greater susceptibility to lobular carcinoma, because this histology type was confirmed in significantly higher percentage than expected. Whether this reflects different genetic susceptibility for S-CBC and M-CBC, and what could be implications on further prognosis, is yet to be analyzed.

**Disclosure:** All authors have declared no conflicts of interest.

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**Background:** Fluorouracil-based chemoradiotherapy (CRT) is regarded as a standard perioperative treatment in locally advanced rectal cancer. We investigated the efficacy and safety of substituting fluorouracil with the oral prodrug TS-1.

**Methods:** A multi-institutional (17 specialized centers), interventional phase II trial, was conducted from April 2009 to August 2011. This study is registered with UMIN-CTR, number C003396. For inclusion, patients must fulfill the following requirements before neoadjuvant CRT: (i) histologically proven rectal carcinoma; (ii) tumor located in the rectum (upper, lower); (iii) cancer classified as T3-4, N0-3 and M0; Two cycles of neoadjuvant CRT with TS-1 (100 mg/m² on days 1-5, 8-12, 22-26, and 29-33) is administered, and irradiation (total 45Gy/25fr, 1.8Gy/day, on days 1-5, 8-12, 15-19, 22-26, and 29-33) is performed. Total mesorectal excision with D3 lymphadenectomy is performed during the 4th and 8th week after the end of the neoadjuvant CRT. The primary end-point is rate of complete treatment of neoadjuvant CRT. Secondary endpoints are response rate of neoadjuvant CRT, short-term clinical outcomes, rate of curative resection, and pathological response (grade 2/3).

**Results:** This trial included 37 patients. A complete treatment of neoadjuvant CRT was found in 83.3% of patients (95%CI; 71.2-95.5%), and an adverse event (grade 3/4) occurred in 4 patients (11.1%). A rate of overall downstaging (PR/CR; RECIST 1.0) was 83.3% (95% CI; 71.2-95.5%), and a pathologic response rate was 50.0% (95% CI; 33.7-66.3%).

**Conclusion:** Our prospective phase-II study demonstrated that a neoadjuvant-synchronus TS-1 + RT for locally advanced rectal cancer was feasible in terms of pathological response and adverse events.

**Disclosure:** All authors have declared no conflicts of interest.