278. Biomarkers for lung cancer

P3234

Prognostic significance of Nestin in resected non-small cell lung cancer

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Background: Nestin was originally identified as a classVI intermediate filament protein that is expressed during early stages of development in progenitor cells of the central nervous system. This protein becomes re-expressed in pathological conditions such as brain injury, muscle injury and neoplasia. While Nestin has been detected in many kinds of tumors, no report concerning the relationship between the expression of Nestin and clinicopathological features in non-small cell lung cancer (NSCLC) have been published. Therefore, we aimed to investigate the relationship between the expression of Nestin and clinicopathological parameters and clinical outcome in NSCLC.
Methods: For this retrospective study, Nestin expression in cancer cells was immunohistochemically evaluated for 171 consecutive cases, of surgically resected NSCLC. Nestin expression was examined for an association with the clinicopathologic parameters. Analyses were performed to evaluate the prognostic significance of Nestin expression.
Results: Nestin expression was observed in 27 of 171 (15.8%) cases. Nestin expression was significantly associated with squamous cell carcinoma, poorly differentiation and vascular invasion. Nestin expression was significantly associated with poor prognosis ($p<0.001$). On multivariate analysis, Nestin expression (HR 2.611; 95% CI: 1.162-5.866; $p=0.02$) and TNM stage were significant risk factor for overall survival.
Conclusion: Present study suggests that Nestin expression may be a prognostic indicator for resected NSCLC. Further study will be need to clarify the role of Nestin in NSCLC.

P3235

***EGFR* and *hTERT* mRNA expression in non-small cell lung cancer patients – Diagnostic and prognostic implications**

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Objective: The early detection of NSCLC is of importance because it provides chances for better outcomes. The aim of the study was to explore the clinical utility of *EGFR* and *hTERT* mRNA expression as markers for diagnosis and prognosis of NSCLC.
Methods: *EGFR* and *hTERT* mRNA were quantified by quantative real time polymerase chain reaction in plasma of 45 non-small cell lung cancer (NSCLC) and 40 chronic obstructive pulmonary disease (COPD) patients, selected by certain spirometric characteristics that made them at high risk of developing lung cancer in future. RNA was isolated with TriZOL from 3ml plasma. To enhance the sensitivity a preamplification reaction with gene specific primers preceded the qPCR. TaqMan gene expression Assays for *EGFR* and *hTERT* were used. β-actin served as endogenous control. A TaqMan MGB probe (FAM) was used.
Results: The gene expression level of each gene was calculated as a relative quantity – RQ. *EGFR* was found in all lung cancer patients. Its mean level was RQ = 29.39. *hTERT* mRNA was detected in 88% of patients. Its mean expression was RQ=17.31.Only 50% of the high risk patients were positive for *EGFR* – RQ = 2.09. *hTERT* was detected in 17 (42.5%) of the high risk COPD patients – mean RQ=1.02. A statistically significant difference in *EGFR* and *hTERT* mRNA expression could be observed between the two groups of patients – p=0.0001. NSCLC patients were followed up for one year. None of the markers was associated with survival.
Conclusion: *EGFR* and *hTERT* mRNA are potential markers for lung cancer diagnosis, whose clinical importance should be replicated in a larger cohort of patients.

P3236

EGF-receptor mutation in non-small cell lung cancer – Experiences by routine molecular testing in a German lung cancer centre

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Introduction: Due to novel therapy strategies in EGF-receptor mutated patients, molecular analysis of the EGF-receptor genome has become crucial. A novel approach in this context is the routine testing of all consecutive patients with non-small cell lung cancer (NSCLC) as done in our clinic for the last six months.
Methods: 350 subsequent tumor biopsies obtained by routine bronchoscopy were histologically analyzed. In case of diagnosis NSCLC the EGF-receptor was analyzed by sequencing techniques, accessing exons 18, 19 and 21 for the presence of activating mutations by conventional Sanger sequencing after PCR, in most cases also parallel by pyrosequencing technique.
Results and discussion: In 350 patients EGF-receptor mutation analysis resulted in only 27 (7.1%) cases of mutation in one of the three exons. Exon 18 was mutated only once, whereas exon 19 was subject to a detected mutation in 15 cases. We found 9 mutations in the most common typical location delE746-A750. The other mutations were evenly distributed within the known genetic changes, whereas 1 mutation has not been described before. On Exon 21 only one type of mutation was found.
Conclusion: In serial screening EGF-receptor mutations appear less frequent than presumed. The variation of mutations on Exon 19 however is much more pronounced than originally expected. Evaluation should be done by accessing the full genomic sequence in order to detect unknown mutations or to avoid misinterpretation of silent mutations without effect on the decision for TKI-therapy.

P3237

Detection of k-ras mutations in cytologic specimen of lung cancer patients as predictive factor of targeted therapy

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Introduction: The k-ras protein is part of signalling cascade of the epidermal growth factor (EGF) and plays in important role in cancer genesis including NSCLC.
Aims and objectives: It is unclear, how the mutation status of k-ras interferes with the therapeutic effect of EGF-tyrosin kinase inhibitors like erlotinib. Cytologic specimens of NSCLC were tested for k-ras mutations in relation to the outcome of Erlotinib therapy.
Methods: We recruited patients with NSCLC (n=167), patients with pulmonary metastasis from cancer of non-pulmonary origin (n=6), patients with SCLC (n=8), and bronchitic patients (n=8). Samples were gathered by cytologic material, e.g lymph node biopsy or samples from pleural fluid. NSCLC patients were grouped to erlotinib and standard chemotherapy. K-ras mutations were detected using specific PCR-RFLP.
Results: 81 of the NSCLC patients had a mutation in k-ras codon 12, one had a mutation in codon 13 and two had a mutation in codon 61. At the reference group (n=22) we found no mutation in any of the examined codons. In the erlotinib group (n=50), 37 had a k-ras mutation in codon 12. Patients in tumour stages III or IV had significant higher mutation rates (58%) than those with lower stages (20%, p<0,01). We were unable to prove significant differences in therapeutic effect in our cohort.
Conclusion: We confirmed the feasibility to detect k-ras mutation in cytologic specimen. If the k-ras mutation status of a patient with NSCLC correlates with the benefit of therapy with Erlotinib, is not clear. The use of cytologic probes for diagnostic k-ras research is an extraordinary effective method, which could be established as technique for clinical needs.

P3238

Her/2 neu and p53 expression in patients with non-small cell lung cancer predict shortened survival

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The outcomes of patients with different stage of non-small-cell lung cancer (NSCLC) vary greatly. The aim of this study was to evaluate potential interaction of two molecular markers that may aid in predicting prognosis in patients with NSCLC. Immunohistochemical staining of Her-2/neu and p53 was performed on paraffin-embedded sections from 87 NSCLC patients who were treated with radiotherapy and/or chemotherapy and were followed up at least 24 months. The prevalence of Her/2 neu and p53 expression is 36,8% and 52,9% respectively. Except to the histological type of tumors, there was no correlation of p53 and Her/2 neu expression with other clinicopathologic features. Differences in survival rates were evaluated by log rank test. Patients whose tumors were both Her-2/neu and p53 positive had the worst outcome, and all of them died before 24 months. The survival rate were 40% in patients whose tumors were both Her-2/neu and p53 negative (HR=3,026, p<0,000). In patients who were Her-2/neu positive but p53 negative, survival rate was15%, and 10% in those who were p53 positive and Her-2/neu negative. Multivariate analysis controlling for TNM stage, histological type of tumors and performing status showed that overexpression of p53, Her/2neu, especially combination of their expression and performance status were an independent factors predicting shortened survival (HR=2,97, p<0,000). Thus, the analysis of the co-expression of Her-2/neu and p53 in NSCLC patients may identify patients with poor prognosis who may benefit from more aggressive therapy.

P3239

Diagnostic markers of lung cancer in bronquial fluid: Development of inmunoassays and validation

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Background: We have previously identified some proteins that have differencial expression in patients with lung cancer using proteomic techniques.
The aim of our study is to validate these markers using highly sensitive inmunassays.
Methods: We have included bronchial aspirates samples from 204 patients diagnosed with lung cancer. (63 microcitic, 59 adenocarcinoma and 82 epidermoide) and 48 control patients with other non malignant pathologies.
Inmunoassays have been performed based on xMAP technology (luminex R) for each specific molecular marker. In each case, it's necessary to obtain a pair of antibodies that link simultaneously to the marker.
Once the assay has been designed and optimized, the amount of each marker has been quantified in each sample. The comparison between both groups has been done using t-student test and the diagnostic capacity of each marker has been evaluated by ROC curve.
Results: Inmunoassays have been performed and optimized, so far, for two markers (M1 and M2) that showed an overexpression in patients diagnosed with lung cancer using these proteomic techniques (M1: 1668±70 *vs.* 770±146 p<0.0001; M2: 2850±125 *vs.* 920±163 p<0.0001). The calculated ROC area was 0,79 for M1 and 0,85 for M2.
Conclusions: Among all the possible candidates obtained previously by proteomic, two proteins have been validated as diagnostic protein markers of lung cancer and both of them with a high diagnostic capacity.
Clinical validation is needed in a larger population and less invasive samples such as serum. Thus, we could obtain a diagnostic test to apply in clinical routine.

P3240

Contribution of genetic polymorphism of stromal cell-derived factor-1 (SDF-1) and its receptor, CXCR4, to the susceptibility and severity of non-small cell lung cancer

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Background: Stromal cell derived factor-1 (SDF-1), a CXC chemokine that play important roles in tumor growth, angiogenesis and metastasis of tumor cells.The aim of this study was to evaluate the relations of SDF-1 and its receptor, CXCR4, gene variants on non-small cell lung cancer (NSCLC) risk.

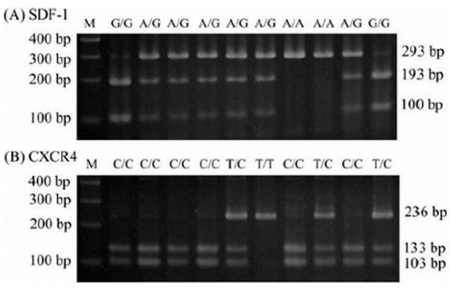

Figure 1. Polymerase chain reaction–restriction fragment length polymorphism of SDF-1 and CXCR4 gene.

Methods: A total of 247 NSCLC patients were recruited into this study, together with 328 age- and gender-matched healthy smokers acting as control. Polymorphisms of SDF-1 and CXCR4 gene were analyzed using PCR-RFLP technique on genomic DNA isolated from peripheral lymphocytes.
Results: The distribution of the genotype frequencies of SDF-1 and CXCR4 were significantly different between lung cancer patients and controls, and also different between patients of various stages. Logistic regression analysis revealed that higher odds ratios (ORs) for lung cancer were seen for individuals with SDF-1 AA against GG/GA genotypes (an OR of 1.95, 95% CI 1.08-3.50,p = 0.035), and for individuals with CXCR4 TT against CC/TC genotypes (an OR of 4.71, 95% CI 1.99-11.2, $p < 0.0001$). The patients carrying a homologous AA genotype at SDF-1, or a homologous TT genotype at CXCR4, had a tendency to advanced disease.
Conclusions: A significant association between the polymorphisms of SDF-1 and its receptor, CXCR4, and the susceptibility to and severity of NSCLC was demonstrated.

P3241
Serum napsin A as a tumor marker for primary lung adenocarcinoma
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Background: Napsin A serves as a marker specific to primary lung adenocarcinoma in immunohistochemistry. However, significance of napsin A as a serum tumor marker has not been examined in patients with primary lung adenocarcinoma.
Objective: The aim of this study is to investigate the potential of serum napsin A as a tumor marker in patients with primary lung adenocarcinoma.
Method: Thirty patients with primary lung adenocarcinoma (alveolar cell carcinoma (n=5) and non-alveolar cell carcinoma (n=25)) were enrolled who visited our institution from April 2007 to April 2008. Twenty healthy volunteers were included as a control group. Serum CEA and napsin A were measured by ELISA. The sensitivity, specificity, and diagnostic accuracy of CEA and napsin A were calculated.
Results: The cut off value for napsin A was set to 55 ng/ml from our previous study. The sensitivity, specificity, and diagnostic accuracy of napsin A (vs. CEA) for diagnosis of primary lung adenocarcinoma were 13% (vs. 23%), 95% (vs.100%), and 50% (vs.54%). However, in cases of alveolar cell carcinoma, the sensitivity, specificity, and diagnostic accuracy of napsin A (vs. CEA) were 40% (vs. 0%), 95% (vs. 100%), and 84% (vs. 80%).
Conclusion: These results suggest that napsin A is not more useful than CEA as a tumor marker for primary lung adenocarcinoma, but may useful for diagnosing alveolar cell carcinoma.

P3242
Tissue expression of alpha crystallins and plasma levels of antibodies against them in NSCLC patients – Prognostic significance
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Objective: The assessment of the prognostic significance of various biologic markers could provide risk stratification of patients and improve survival.
Aim: To determine the tissue expression of alpha-B antigens and plasma levels of alpha-crystallin antibodies in NSCLC patients and to study their prognostic role.
Materials and methods: A total of 48 consecutive NSCLC patients participated in the study.
Immunohistochemistry with primary polyclonal rabbit anti-alpha crystallin antibody and ELISA for detection of circulating antibodies were performed. The plasma detection of the marker was confirmed by Western blot. Kaplan-Meier and log-rank test evaluated the 1-year survival rate and the prognostic significance of the marker.
Results: The level of alpha-crystallin antibodies was higher (0.523) in patients with lymph node metastases compared to those with no mediastinal spread (0.3695) – (p=0.045). Early stage NSCLC patients (IA, IB, IIB) had significantly lower plasma levels of antibodies in comparison to IIIA-IIIB stage – p=0.038. In addition to a p-stage the plasma levels of alpha-crystallin antibodies could subdivide patients in risk subgroups with statistically significant difference in the survival rate (p=0.036). There was a trend for a negative association between alpha crystallin tissue expression and plasma levels of the specific antibodies.
Conclusion: The plasma levels of alpha-crystallin antibodies in NSCLC patients are potential biomarkers for the metastatic spread of the tumor and the disease progression. They could also be used to stratify the patients into risk subgroups, but this data needs further investigation.

P3243
Impaired expression of telomerase activity in bronchoalveolar lavage fluid cells in patients with lung cancer: Preliminary results
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Introduction: Telomerase is a reverse transcriptase enzyme contributing to the maintenance of the telomeric structure by adding telomere repeat sequences to chromosomal ends, thus compensating for its shortening. To avoid the attrition of telomeres, germ-line cells and some somatic cells produce telomerase, an enzyme that catalyzes DNA synthesis to maintain telomere length. Telomerase reverse transcriptase (TERT) uses the telomerase RNA component (TERC) as a template to synthesize telomere DNA.
Aim: In humans, studies are often limited by the necessity to measure telomeres in leukocytes, the uncertain significance of telomere length in tumor cells and the difficulty of performing longitudinal assessments. Nevertheless, telomere length has been linked to several types of cancer. We aimed to evaluate telomerase activity (mRNA expression of both subunits TERT and TERC) in Bronchoalveolar Lavage Fluid (BALF) of patients with Lung Cancer.
Methods: As the control group, 11 healthy subjects were also included. We studied prospectively 16 patients with Lung Cancer. mRNA expression was measured by Real-Time RT-PCR.
Results: Human hTERT mRNA transcripts were detected in 3/16 (18.7%) cases of Lung Cancer and in 4/11 (36.3%) of the controls subjects. TERC mRNA transcripts were detected in 9/16 (56.25%) cases of Lung Cancer and in 5/11 (45.45%) of the controls subjects.
Conclusion: These results showed decreased expression of TERT in BALF cells of patients with Lung Cancer. This finding is in alignment with genomewide association studies that have shown polymorphisms in the *TERT* gene at a higher frequency than normal in patients with cancer.

P3244
Clinical significance of MMP-9 and their tissue inhibitor TIMP-2 in lung adenocarcinomas ongoing study
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Background: Evidence is existed that the matrix metalloproteinases (MMPs) and the tissue inhibitors of metalloproteinase (TIMPs) have been associated with tumor invasion and metastasis in many human cancers. The aim of our study was to investigate the association of expression of MMP-9 and TIMP-2 with time to progression and overall survival in lung adenocarcinomas.
Methods: A total of 32 patients with lung adenocarcinomas, stage IIIb-IV, were enrolled. The levels of total MMP-9 along with TIMP-2 enzymes activity levels were measured using enzyme-linked immunosorbent assay, twice: first at the time of diagnosis and secondly after the completion of chemotherapy, consisted of docetaxel 100mg/m^2 plus carboplatin AUC 5.5.
Results: After chemotherapy completion a negative correlation was observed between a. serum MMP-9 and survival (r=-0.461, p=0.023, Pearson correlation) and b.TIMP-2 and time to progression (r=-0.465, p=0.015).First measurements of serum MMP-9 and TIMP-2 had no significant impact on survival. A negative correlation was also observed between MMP-9 and TIMP-2 (r=-0.550, p=0.005).On the other hand, not any correlation was observed between the initial measurements of MMP-9 and TIMP-2.
Conclusion: Increased levels of MMP-9 and TIMP-2 are associated with decreased survival and time to progression. We suggest that determination of MMPs and TIMPs and their ratio may be helpful in the early identification of lung cancer patients with poor prognosis.

P3245
Presence of volatile organic compounds (VOC) in exhaled breath in patients diagnosed with lung cancer (LC) vs healthy patients. Preliminary data
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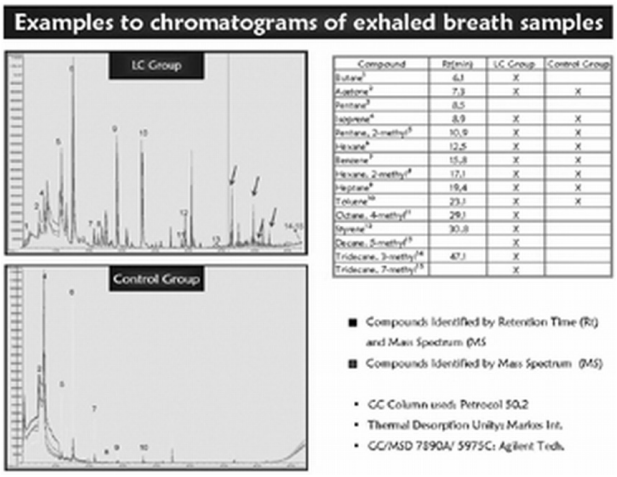
Introduction: Determination of VOC in exhaled breath as a LC tumorlike markers, is a new line of investigation applicable to the early diagnosis.
Objective: Determination of VOC in exhaled breath in patients with LC and healthy volunteers.
Method: Descriptive and observational study. LC Group: 10 patients with LC. Control group: 10 healthy voluntary subjects. Informed consent accepted.

Parallel collection of environmental air and exhaled breath by BioVOCTM, at FRC level. Analytical technique: Thermal Desortpcion- GC/MS
The list of VOC to determine are (see figure):



Results: 1. 91% of the studied VOC found in exhaled breath of patients with LC. In healthy patients the percentage found were 35%.
2. VOC >C10, were found in tumoral patients samples.
Conclusions: 1. The simultaneous and majority presence of all the VOC considered within the same sample appears to be correlated with the CP hystologic diagnose; consequently, no VOC is, by itself, a specific CP marker.
2. At the moment a threshold of concentration for different the VOC does not exist from which, the compound is considered as tumorlike marker.
3. The abundance of benzene and toluene has been similar in all the studied samples, environmental or alveolar. Therefore they do not seem to be useful as tumorlike markers.
4. The compounds with C>10 seem to be more specific of the pathological group.

P3246
Elevated serum levels of interleukin-8 in advanced non-small cell lung cancer patients: Relationship with prognosis
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Introduction: Interleukin-8 (IL-8) is a pleiotropic cytokine that has also been shown to exert effects relevant to cancer growth and progression. Cancer progression is believed to be contributed to by the ability of this cytokine to promote angiogenesis and mitogenic effects. As IL-8 production at the tumor site may determine elevated serum levels of this cytokine because of hematogenous leakage, it is conceivable that patients with high IL-8 serum levels may have tumors actively producing this cytokine.
Methods i results: The aim of this study was, therefore, to assess IL-8 serum levels in 60 non-small cell lung cancer (NSCLC) patients undergoing chemotherapy and to correlate them with prognosis. IL-8 serum levels were found to be significantly elevated in cancer patients with respect to controls. Moreover, IL-8 serum levels were shown to be significantly increased in stage IV patients compared with stage III patients. When basal IL-8 serum levels in cancer patients were analyzed according to response to chemotherapy, responders were shown to have significantly lower IL-8 serum levels than nonresponders. On univariate analysis, the IL-8 serum level was included among the variables capable of affecting both overall survival (OS) and time to treatment failure (TTF). However, multivariate analysis failed to demonstrate an independent prognostic significance for IL-8 serum levels.
Conclusion: In conclusion, this study showed that IL-8 serum levels were elevated in advanced NSCLC patients and correlated with both OS and TTF, but they were shown not to be an independent prognostic factor.

P3247
Changes and significance of hepatoma-derived growth factor and vascular endothelial growth factor in serum and lung tissue from patients with lung cancer
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Objective: The purpose of this study is to determine the changes and significance of hepatoma-derived growth factor (HDGF) and vascular endothelial growth factor (VEGF) in serum and lung tissue from patients with lung cancer.
Methods: Fifty six newly inpatients with definite lung cancer were enrolled as the study group, fourteen patients with benign pulmonary diseases and fourteen healthy volunteers were selected for control groups. Enzyme-linked immumosorbent assay was used to detect the levels of HDGF and VEGF in serum of all subjects, and immunohisotchemical assay was used to detect the HDGF and VEGF expression in lung cancer tissues and matched normal lung tissues from ten patients with lung cancer.
Results: Squamous cell lung cancer had higher HDGF levels in serum compared to adenocarcinoma ($p<0.01$); Patients with poor-differentiated lung cancer had higher HDGF levels in serum than that with high-differentiated ($p<0.01$); Patients with NSCLC in II-IV stage had higher levels of HDGF and VEGF in serum than that in I stage ($p<0.05$); Lung cancer patients with lymph node metastasis had higher VEGF levels in serum than that without lymph node metastasis ($p<0.05$); There was a positive correlation between VEGF and HDGF level in serum form patients with lung cancer ($r=0.35$, $p<0.01$); HDGF and VEGF expression were dramatically increased in lung cancer tissues compared with in matched normal lung tissues ($p<0.01$, $p<0.05$); There was a positive correlation between VEGF and HDGF level in lung cancer tissues from patients with lung cancer ($r=0.75$, $p<0.05$).
Conclusions: HDGF and VEGF play an important role in tumor growth and tumor metastasis.

P3248
The prognostic significance of circulating neuroendocrine markers chromogranin A, pro-gastrin-releasing peptide and neuron- specific enolase in patients with advanced non-small-cell lung cancer
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Introduction: Chromogranin A (CGA), Pro-gastrin-releasing peptide (ProGRP) and neuron-specific enolase (NSE) are known as immunohistochemical tissue markers closely associated with neuroendocrine differentiation in non-small-cell lung carcinoma (NSCLC). The aim of thepresent study was to assess the value of serum levels of these markers in predicting response to chemotherapy and survival of patients with unresectable NSCLC.
Methods: The study included 84 patients with advanced NSCLC. Before treatment, serum levels of CGA, ProGRP and NSE were measured with commercial kits.
Results: Distribution of serum CGA differed significantly according to gender and histology with higher levels being found in men ($p = 0.01$) and in squamous cell carcinoma ($p = 0.01$). Serum ProGRP levels correlated with disease extent, (M1 vs M0; $p = 0.03$). On inclusion in multivariate Cox models, both CGA and ProGRP retained signifi cance with high levels showing an opposite effect on survival: CGA, relative risk (RR) –5.0; $p <0.001$, and ProGRP, RR –0.3; $p = 0.005$, and median cutoff points: CGA, RR –2.1; $p = 0.02$, and ProGRP, RR –0.6; $p = 0.04$. The combined use of CGA, ProGRP and NSE allowed for definition of two sets of patients with significantly different median survival times (23.1 vs. 10.6 months, $p <0.001$).
Conclusions: CGA and Pro-GRP appear to bear important information related to the prognosis for NSCLC
Keywords: chemotherapy, response, chromogranin A, neuron-specific enolase, non-small cell lung carcinoma, pro-gastrin releasing peptide

P3249
Plasma levels of alpha crystalline antibodies in NSCLC patients – Diagnostic significance
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Objective: The establishment of biomarkers in peripheral blood of cancer patients makes it useful for clinical application and cancer screening.
Aim: The aim of the study was to explore the diagnostic significance of alpha-crystallin antibodies as markers for diagnosis of NSCLC.
Materials and methods: Alpha-crystallin antibodies were detected with ELISA in 51 NSCLC patients and 52 age and sex matched healthy volunteers. Results were confirmed by Western blot. Alpha-crystallin IgG antibodies differed significantly between the groups of cancer patients and the healthy volunteers ($p<0.001$). A cut-off value of 0.317 discerned NSCLC patients with sensitivity 62% and specificity 72% among the control group. The assay was effective in distinguishing the patients with and without lymphogenic metastatic spread of the disease ($p=0.045$)-sensitivity 60% and specificity 70%. In NSCLC patients there was a trend for a negative association between the tissue and plasma levels of expression.
Conclusion: The clinical significance of this marker has a modest implication in lung cancer diagnosis. Its importance as a marker of disease recurrence and lymph node micrometastasis should be further explored.

P3250

Elevated serum levels of oxidative stress biomarkers in patients with lung cancer

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Background: Oxidative stress has been implicated in lung cancer oncogenesis.
Objectives: The aim of the present study was to determine the levels of an oxidant (8-isoprostane) and an antioxidant biomarker (CuZnSOD) in serum of patients with lung cancer and to investigate their association with clinicopathological factors.
Materials and methods: We enrolled 50 patients, median age 67 years (37/males and 13/females, 14/nonsmokers and 36/smokers), newly diagnosed with NSCLC stage IV and 25 healthy individuals (12/nonsmokers and 13/smokers). Serum levels of 8-isoprostane and CuZnSOD were analyzed by commercially available enzyme-immunosorbent assay kits (ELISA).
Results: 8-isoprostane and CuZnSOD serum levels were significantly higher in lung cancer patients compared to healthy controls ([median (min-max)] 38.6 (6.3-999.9) vs. 20.9 (10.4-82.7) pg/mL, p=0.003; and 131.4 (44.0-730.4) vs. 84.5 (34.6-299.4) ng/mL, p=0.0003, respectively). No significant differences to the levels of biomarkers related to clinicopathological parameters such as age, gender and histological type were observed. Additionally, serum concentrations of 8-isoprostane presented no significant difference between smokers and non-smokers lung cancer patients, while CuZnSOD levels were found significantly higher in nonsmokers compared to smokers patients (222.1 (76.6-604.0) vs. 122.0 (44.0-730.4) ng/mL, p=0.019).
Conclusions: Our study showed elevated serum levels of oxidant and antioxidant biomarkers in patients with lung cancer compared to healthy controls. However, lower antioxidant capacity was observed in smokers compared to nonsmokers lung cancer patients which potential leads to increased oxidative stress.

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Abnormal expression of let-7a miRNA in blood of lung cancer patient

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Background: The abnormal expression of let-7a miRNA in non-small cell lung cancer tissue has been reported. The objective of this study was to investigate the aberrant expression of let-7a miRNA in blood of lung cancer patients.
Methods: We analyzed let-7a miRNA in archived whole blood from 66 participants, 35 of whom did and 31 of whom did not have lung cancer by real-time PCR (RT-PCR). We also investigated the expressions of let-7a miRNA in lung cancer cells (A549, HCC 1588), normal respiratory epithelial cells (L132), and 40 human lung cancer tissues by RT-PCR.
Results: The $2^{-\Delta\Delta Ct}$ of let-7a miRNA in blood of normal subjects and lung cancer were 3242.49±355.28, 747.85±177.74, respectively. The relative expression of let-7a miRNA in A549 and HCC 1588 cancer cell lines were about 0.5 and 0.8 when compared with L132 normal epithelial cells. The $2^{-\Delta\Delta Ct}$ of let-7a miRNA in normal human lung tissues and human lung cancer tissues were 42.30±3.98, 27.73±3.86, respectively.
Conclusions: The let-7a miRNAs in cells, tissues, and blood of lung cancer patients were under-expressed compared with those of normal control. The possibility of using let-7a miRNA as serologic marker for lung cancer warrants further studies.

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Metabolomics in lung cancer

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Purpose: Some serum tumor markers of lung cancer are put to practical use, but they are unfit for early detection. Gene and protein expressions have been extensively profiled in cancers, but metabolites profiles have not been developed yet. The aim is to clear the metabolome profiling of human lung cancer by the latest metabolomics methods, and to investigate clinical biomarkers for early diagnosis and prognosis prediction of lung cancer.
Methods: First, serum samples, from the healthy non smoker without cancer (n=10), the healthy smokers without cancer (n=8), and the lung cancer patients (n=20) were analyzed with a combination of gas-chromatography and mass spectrometory (GC/MS). Next, obtained GC/MS metabolite data was statistical-analyzed by using principal component analysis, partial least squares discriminant analysis, and a two sample t-test to explore the potential metabolic biomarkers.Tissue samples from surgical specimen, one group is non-tumor lung tissue (n=7) and another one is tumor lung tissue (n=7), were analyzed in a similar way to serum samples.
Results: About 60 metabolites are identified in the serum samples and about 70 metabolites in the tissue samples. These metabolites were organic acids, amino acids and fatty acids and they were concerned with glycolysis or TCA cycles. In both the serum and tissue samples, some metabolites expressions were found to be different between lung cancer group and normal group ($p<0.05$), such as lactate and glutamate which are important for cancer cells to get energy for their proliferation.
Conclusion: The analysis of the pattern of metabolites expression can be one of the clinical biomarker of lung cancer diagnosis.

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Bone metabolic markers in detecting bone metastasis of lung cancer

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Aim: The aim of this study was to evaluate the role of bone metabolic markers in clinical evaluation of bone metastasis of lung cancer.
Material and methods: Sixty five male lung cancer patients with mean age of 64,07±8,7 were included into this trial. The presence of bone metastasis was investigated by whole body bone scintigraphy via Tc 99m mostly (80%) and in some cases, PET/CT which were taken for staging were used. Eighty percent of the patients were diagnosed as non small cell lung cancer and 20% were small cell lung cancer. Bone specific alkaline phosphatase (BALP) and osteocalcin were measured in serum as production markers of bone. N terminal telopeptide (NTX) and β form of C terminal telopeptide (β-CTX) were studied as bone destruction markers.
Results: There were 23 cases (35%) with bone metastasis. Single bone metastasis was detected in 3 patients (13%) and 20 (87%) cases had multiple bone metastasis. The cases were divided into two groups according to presence or absence of bone metastasis and biochemical parameters of two groups were compared. Serum levels of total alkaline phosphatase, BALP and NTX were significantly higher in the group with bone metastasis.Osteocalcin and β-CTX levels were not significantly different between two groups. According to ROC-curve analysis, at the threshold value of 22,38 μg/L, the sensitivity of bone specific ALP was 60,87% and the specificity was 69,05%. Similarly, at the threshold value of 25,69 nmol BCE, the sensitivity of NTX was 90,24% and the specificity was 43,4%.
Conclusion: Bone metabolic markers are considered noninvasive, useful and cost-effective. However, more prospective studies are needed to use them for evaluation of bone metastasis in lung cancer.