determine the mutational status and polymorphism frequency of KRAS gene in Mexico, we analyzed a large sample of mCRC.
Material and Methods: We retrospectively collected paraffin embedded tumours from 807 patients with mCRC diagnosis from different hospitals in Mexico. Mutation detection in KRAS codons 12 and 13 were determined. The tests were performed in a LightCycler® 2.0 system using the LightMix® Kit KRAS mutation (TIB-Molbiol, Germany).
Results: Four hundred ninety-five patients were KRAS wild-type tumours (61%) and 312 patients were KRAS mutated (39%); mutated tumours shown the following polymorphism: Gly12Ser (GGT>AGT) (47%; n = 148); Gly12Asp (GGT>GAT) (16%, n = 50), Gly12Val (GGT>GTT) (7%; n = 22); Gly13Asp (6%, n = 20), Gly13Cys (5%, n = 16), Gly12Arg (4%, n = 13), Gly12Cys (4%, n = 12), Gly12Ala (4%, n = 11).
Conclusions: KRAS mutation status frequency (39%) was not out of range by previous reports in others countries or regions (35−55%). However, the most common mutation polymorphism was Gly12Ser (GGT>AGT) (47%), in contrast with previous reports that indicated Gly12Asp (GGT>GAT) as the most common polymorphism. The mutational polymorphism associated to higher recurrence risk and mortality by RASCAL II study was Gly12Val (GGT>GTT); surprisingly, in our study it was of low frequency mutation type (7%). Outcome measures in KRAS wild-type patients treated with a cetuximab-based regimen at the moment are not evaluated because of the early treatment stage.

1102 POSTER

High-sensitive D-dimer Determination for the Prediction of Chemotherapy-associated Venous Thromboembolism in Lung Cancer Patients

P. Ferroni[1], F. La Farina[1], S. Mariotti[2], C. Retrosi[2], S. Riondino[1], F. Martini[1], G. Massimiani[2], I. Portarena[2], F. Guadagni[1], M. Roselli[2]. *[1]IRCCS San Raffaele Pisana, Department Laboratory Medicine & Advanced Biotechnologies, Rome, [2]University of Rome Tor Vergata, Department Internal Medicine Medical Oncology, Rome, Italy*

Background: Venous thromboembolism (VTE) is an important cause of morbidity and mortality in lung cancer patients, especially during the first 3−6 months of chemotherapy. Several studies have been undertaken to identify novel candidate biomarkers to improve risk prediction, namely D-dimer (DD). However, despite its determination has become a cornerstone in the exclusion of clinically suspected VTE, its predictive use in cancer yielded controversial results. The recent availability of a high sensitive (HS) assay for DD determination prompted us to investigate whether baseline HS-DD determination could be useful to predict VTE risk in lung cancer patients scheduled for chemotherapy (adjuvant or first-line).
Material and Methods: 108 consenting patients (65±9 years) with newly diagnosed (22) or metastatic (86) lung (11 SCLC, 97 NSCLC) cancer were enrolled. Exclusion criteria were: ECOG score >2, prophylactic or therapeutic doses of any heparin, use of anticoagulant drugs. HemosIL DD and DD HS 500 (Instrumentation Laboratory) immunoassays were performed on an ACL TOP coagulometer on citrated plasma samples obtained before chemotherapy.
Results In a median 6-months follow-up, VTE occurred in 16 (15%) of 108 patients with a median time-to-event of 1.8 months. Higher median DD (780 vs. 411 ng/ml, $p = 0.002$) and HS-DD (3507 vs. 1055 ng/ml, $p = 0.0002$) levels were found in patients who developed VTE. Despite a correlation between DD and HS-DD levels ($r = 0.837$, $p < 0.0001$), a significant proportion of negative DD patients scored positive for HS-DD (18%, $p < 0.0001$) using conventional cutoffs (280 and 500 ng/ml, respectively). ROC curves generated from continuously distributed test results showed a better area under the curve for HS-DD (0.816, SE = 0.05) than DD (0.762, SE = 0.07). Based on these results, a cutoff value of 1500 ng/ml was calculated for HS-DD resulting in a sensitivity 0.81, specificity 0.69, NPV 0.96 (OR for VTE: 9.4, $p < 0.0001$). Multivariate analysis showed that HS-DD ($p < 0.0001$), but not DD, was an independent predictor for VTE. Finally, Cox proportional hazard survival analysis demonstrated a RR of 10.1 ($p < 0.0001$) of developing VTE during chemotherapy for patients with baseline HS-DD levels >1500 ng/ml.
Conclusions: These results demonstrate that the use of an enhanced immunoassay for D-dimer determination prior to chemotherapy is able to predict VTE in lung cancer out-patients with a substantial gain in accuracy over conventional testing.

1103 POSTER

Overexpression of MiR-141 and MiR-126 Distinguishes Metastatic Castration Resistant Prostate Cancer (mCRPC) From Localized Prostate Cancer (PCa) and Controls in Human Plasma

R.J. Macfarlane[1], A. Watahiki[2], Y.Z. Wang[2], K.N. Chi[1]. *[1]British Columbia Cancer Agency, Medical Oncology, Vancouver BC, [2]British Columbia Cancer Research Centre, Basic Science, Vancouver BC, Canada*

Objective: MicroRNAs (miRNAs) are small, non-coding RNA molecules implicated in the pathogenesis of several malignancies. Several miRNAs exhibit dysregulated expression in PCa, but little is known about the clinical utility of this. We sought to establish a miRNA signature in PCa by comparing levels of expression of miRNAs in the plasma of men with varying stages of PCa.
Methods: Plasma samples were prospectively collected from 75 subjects (25 controls, 25 localized PCa, 25 mCRPC). Candidate miRNAs were chosen based on increased expression in the mCRPC group compared with the local and control groups from a miRNA panel qPCR analysis on the pooled samples of each group. Expression of 8 candidate miRNAs was validated using qPCR performed on individual subject samples.
Results: From the pooled analysis 8 candidate miRNAs were chosen: miR-141, miR-375, miR-200a, miR-9, miR-126, miR-152, miR-200c, and miR-21. qPCR performed on individual samples revealed that overexpression of miR-141, miR-375, and miR-126 was seen in mCRPC and that the combination of miR-141/miR-126 overexpression was able to consistently distinguish between mCRPC vs. local or controls. Among mCRPC samples marked overexpression of miR-126 was observed in those patients who had not been treated with docetaxel. Of the 3 patients who were treated with abiraterone acetate (AA) as part of an open label clinical trial, all had clinical responses and all showed marked overexpression of miR-141 and miR-126.
Conclusions: In this exploratory study, candidate miRNAs that have been associated with prostate cancer pogression were detectable in human plasma that distinguished between mCRPC vs. local/control samples. miR-126, associated with angiogensis and inflammation, was upregulated in patients with progressive mCRPC who had not previously been exposed to docetaxel. Overexpression of miR-141, which has been associated with androgen receptor upregulation, may be a potential predictive biomarker for next-generation antiandrogen therapy. Further studies are planned.

1104 POSTER

Sphingosine Kinase 1 Correlates With a Neuroendocrine Phenotype in Breast Cancer in Vivo and in Vitro

E. Jackson[1], L. Righi[2], E. McGowan[1], A. Sapino[3]. *[1]Royal Prince Alfred Hospital, Centenary Institute, Sydney, Australia; [2]University of Torino, Department of Clinical and Biological Sciences, Torino, [3]University of Torino, Department of Biomedical Sciences and Human Oncology, Torino, Italy*

Background: Neuroendocrine differentiated (NE) breast tumours are diagnosed by their diffuse expression of the NE markers synaptophysin (SYP), chromogranin A (CgA) and neuron-specific enolase (NSE). However the genesis of differentiation to this phenotype in the breast has not been uncovered. Sphingosine kinase 1 (SK1) is a lipid kinase whose bioactive product, sphingosine-1-phosphate has been associated with tumour growth and proliferation, negative prognosis and refraction to endocrine therapy in breast cancer. Recently it was suggested that SK1 is involved in the NE differentiation of LnCAP prostate cells. We have investigated the correlation of SK1 expression with NE differentiation.
Materials and Methods: MCF7 breast tumour cells were stably transfected with SK1 and pellets were formalin fixed and paraffin embedded. 50 formalin fixed NE breast tumour samples were collected from patients admitted for surgery to San Giovanni Battista and San Luigi Hospitals of Torino (Italy). SK1 and the NE markers SYP, CgA and NSE were detected using immunohistochemistry and immunofluorescence.
Results: MCF7 cells do not express detectable levels of the NE markers, SYP or NSE. Over-expression of SK1 in MCF7 cells resulted in an induction of expression of both SYP and NSE. Investigation of NE breast tumours revealed that they display a unique, diffuse SK1 expression pattern when compared to other breast tumours and that SK1 colocalises with CgA.
Conclusions: Results from this case study indicate that SK1 is a potential marker of NE differentiation in breast cancer. SK1 over-expression is sufficient to induce SYP and NSE expression in MCF7 breast tumour cells, suggesting that SK1 has a causative role in NE differentiation.