Results: In patients treated with gefitinib who responded, median time to response was 6.1 weeks (ITT; n = 262) and 6.0 weeks (evaluable *EGFR* mutation-positive subgroup; n = 94); median duration of response was 9.7 and 8.7 months, respectively. Change from baseline in tumour size demonstrated evidence of substantial tumour shrinkage with gefitinib (waterfall plots will be presented). Incidence of rash was 76% and 68% in *EGFR* mutation-positive and -negative subgroups, respectively (ORR 71% v 1%). Percentage of patients overall with a deterioration in HRQoL and symptoms at 4 months, progressors *v* non-progressors: FACT-L 33.7% *v* 16.3%, odds ratio (OR) 2.59, 95% CI, 1.54–4.34; TOI 33.7% *v* 13.2%, OR 3.34, 95% CI, 1.95–5.70; LCS 31.7% *v* 15.5%, OR 2.51, 95% CI, 1.48–4.26.
Conclusions: In IPASS, patients who responded to first-line gefitinib experienced substantial tumour shrinkage and a rapid, durable response. The data does not support rash as a predictive marker of response. A higher percentage of patients who progressed *v* non-progressors had a deterioration in HRQoL and symptoms.

9135 POSTER

Immune-modulating Effects of Bevacizumab and Metronomic Platinum Based-chemotherapy in Advanced Non-small-cell-lung Cancer (NSCLC) Patients

E. Bestoso[1], C. Botta[1], S. Apollinari[1], G. Giorgi[2], M.G. Cusi[3], P. Correale[1]. [1]*"S. Maria alle Scotte" Siena University Hospital, Department of Oncology, Siena,* [2]*"S. Maria alle Scotte" Siena University Hospital, Department of Neurosciences Pharmacology Section, Siena,* [3]*"S. Maria alle Scotte" Siena University Hospital, Department of Molecular Biology Microbiology Section, Siena, Italy*

Background: Bevacizumab is a humanized IgG1 mAb to the VEGF, with anti-angiogenetic activity, used for the treatment of different malignancies, including NSCLC. VEGF exerts multiple functional activities throughout the binding to three receptors expressed on different cell lineages, including endothelial precursors, CNS neurons, myeloid precursors, dendritic cells (DCs) and lymphocytes. We investigated the immune-modulating effects of bevacizumab in advanced NSCLC patients enrolled in the mPEBev phase I-II trial, who had received frontline biochemotherapy with cisplatin, oral metronomic etoposide and bevacizumab (mPEBev regimen).
Patients and Methods: Forty-eight patients (42 males and 6 females) with stage IIIB/IV NSCLC, and ECOG $\leqslant$ 2 were enrolled in this study (EUDRACT code #BEVA2007) and received iv. cisplatinum (30 mg/sqm, days 1−3), oral etoposide (50 mg, days 1−15) and bevacizumab (at dosage of 0; 2.5; 5; 7.5; and 10 mg/kg, day 3), every three weeks. We carried-out an immunological analysis on the PBMCs and serum of these patients taken at baseline and after 4 treatment courses.
Results: This regimen resulted moderately safe and very active with 66.7% (32 patients) response rate, 9 month median progression-free-survival, and 35% survival at 15 months. Our biological monitoring revealed a progressive decrease in monocyte and neutrophil counts, associated with myeloperoxidase activity decline [2133.3 ($\pm$162.2) vs 1413.2 ($\pm$110.05), P = 0.044]. Flow cytometry revealed a significant treatment-related decrease of inhibitory-myeloid cells and an increase of activated DCs ($CD11c^+CD14^+CD80^+CD83^+$) (2.48% vs 5.48%, P = 0.03) in patients' PBMCs, which were associated to a decline of the absolute number of peripheral $CD3^+CD4^+$ T cells, expressing an early memory ($CD3^+CD27^+$) and immune-suppressive regulatory ($CD4^+CD25^+FoxP3^+$) (T_{reg}) immune-phenotype. It was finally, observed an increase in activated cytotoxic-T-cell (CTL) effector ($CD3^+CD62L^+CD8^+$) number. In these patients there was a substantial decrease of Interleukin-10 levels [325.9 ($\pm$54) vs 187.5 ($\pm$24) ng/ml, P < 0.05] indicating an immunologic cytotoxic Th1-phenotype switch and impairment of inhibitory myeloid cells.
Conclusion: This bevacizumab-based regimen exerts immune-modulating effects in NSCLC patients, resulting in a major neutrophil and inhibitory-myeloid cell impairment, associated with a significant increase in activated DCs and CTLs able to trigger an efficient immune-response with potential anti-tumour activity.

9136 POSTER

Can Serum Be Used for Analyzing the EGFR Mutation Status in Patients With Advanced Non-Small Cell Cancer (NSCLC)?

S. Kim[1], H. Jung[1], J. Sung[1], U. Jo[1], Y. Kim[1], S. Shin[1], T. Tanaka[2], K. Hagiwara[2], Y. Choi[1]. [1]*Korea University Anam Hospital, Medicine, Seoul, South Korea;* [2]*Saitama Medical University, Medicine, Saitama, Japan*

Background: Epidermal growth factor receptor (EGFR) mutations as prognostic or predictive marker in patients with non-small cell cancer (NSCLC) have been used widely. However, it may be difficult to get tumour tissue for analyzing the status of EGFR mutation status in large proportion of patients with advanced disease.
Patients and Methods: We obtained pairs of tumour and serum samples from 57 patients with advanced NSCLC, between March 2006 and January 2009. EGFR mutation status from tumour samples was analyzed by genomic polymerase chain reaction and direct sequence and EGFR mutation status from serum samples was determined by the peptide nucleic acid locked nucleic acid (PNA-LNA) PCR clamp.
Results: EGFR mutations were detected in the serum samples of 11 patients and in the tumour samples of 12 patients. EGFR mutation status in the serum and tumour samples was consistent in 50 (87.7%) of the 57 pairs. There was a high correlation between the mutations detected in serum sample and the mutations detected in the matched tumour sample (correlation index; 0.62 P < 0.001). Twenty-two of 57 patients (38.5%) received EGFR-TKIs as any line therapy. The response for EGFR-TKIs was significantly associated with EGFR mutations in both tumour samples and serum samples (p < 0.05). There was no significant differences in OS according to the status of EGFR mutations in both serum and tumour samples (p > 0.05).
Conclusion: Serum sample might be alternatively used in the difficult time of getting tumour tissue for analyzing the status of EGFR mutation status in patients with advanced NSCLC.

9137 POSTER

NF-kappaB Activation and Distinct Expression of Ubiquitin E3 Ligases in Skeletal Muscle of Patients With Non-small Cell Lung Cancer Cachexia

C. Op den Kamp[1], R. Langen[1], A. Schols[1], A. Dingemans[1]. [1]*Maastricht University Medical Centre, Department of Respiratory Medicine, Maastricht, The Netherlands*

Background: Clinical oncologists recently emphasized the need for attention of cancer cachexia and muscle wasting in clinical oncology because it decreases response to anticancer therapy and increases morbidity and mortality. Experimental research has shown that increased muscular Nuclear Factor kappa B (NF-κB) and subsequent ubiquitin (Ub) proteasome system (UPS) activity plays a causal role in cancer-induced muscle wasting but this needs verification in human cancer cachexia. The aim of this study was to investigate molecular changes (including NF-κB activity and expression of E3 Ub-ligases) in skeletal muscle of cachectic patients with advanced NSCLC.
Methods: In this prospective study, 14 cachectic patients (defined by >5% body weight loss in preceding 6 months) and 12 non-cachectic patients with newly diagnosed advanced stage NSCLC and 22 age and gender matched healthy controls were included. Body composition was assessed by dual energy X-ray absorptiometry (DXA) and biopsies were obtained from vastus lateralis muscle. IkappaBalpha (IκBα) and TNF-α mRNA expression levels were determined as indirect indices of NF-κB activity. UPS activity was evaluated by mRNA expression of E3 Ub-ligases: neural precursor cell expressed developmentally down-regulated 4 (NEDD4), Atrogin-1, Tripartite motif-containing protein 32 (TRIM32) and Muscle RING-finger protein-1 (MuRF1).
Results: Mean weight loss was 13.1$\pm$4.9% in cachectic patients compared with 2.1$\pm$2.0% in the non-cachectic group. Cachectic patients had significantly decreased skeletal muscle mass content of upper and lower extremities compared with non-cachectic patients (p = 0.02) and healthy controls (p < 0.001).
Muscle IκBα (p = 0.04) and TNF-α (p = 0.03) mRNA expression and NEDD4 (p = 0.03) were significantly increased in cachectic patients compared with healthy controls. No increase was observed in the other E3 Ub ligases. In contrast, TRIM32 (p < 0.01) showed a significant decrease in non-cachectic patients compared to both cachectic patients and healthy controls, whereas MURF-1 levels were unchanged.
Conclusions: These results indicate that NF-κB activity is increased in lung cancer cachexia but show that E3 Ub-ligases are expressed differently. As all assessed E3 Ub-ligases have shown to be important regulators in experimental models of cachexia, the observed distinct regulation in cachectic patients with NSCLC has implications for understanding the mechanism of human lung cancer cachexia. Further research is required to investigate the potential contribution of individual E3 Ub-ligases to human cancer cachexia in order to develop therapeutic interventions to prevent or reverse cachexia and increase survival of lung cancer patients.