Results: We included 344 colon cancer and 198 rectal cancer survivors and 1181 controls. In a global analysis, survivors reported a significant QOL decline in social functioning (QLQC30) at 5 years (−5.2 points; p = 0.005), and in diarrhea symptom (QLQC30) at 5, 10, and 15 years after diagnosis (+8.2, p < 0.0001; +10.2, p < 0.0001; +6.4, p = 0.006). In subgroup analyses, QOL of rectal cancer survivors were more affected than controls in the physical functioning (SF36) at 5 years (−9.4; p = 0.002), in the physical fatigue (MFI) at 10 years (+8.6; p = 0.01), and in mental fatigue at 5 years after diagnosis (+8.5; p = 0.006). On the assessment of reintegration, cancer survivors saw their marital relationship has strengthened (RR = 1.82 (1.21–2.75); p = 0.0002), attributing change in quality of this relationship (positive or negative) to their health (RR = 4.89 (2.09–11.44); p < 0.0001). As well, their health closely influenced professional activity more often than controls (RR = 4.50 (1.85–10.95); p < 0.0001). They met more difficulties in loan or insurance requests (RR = 3.83 (1.99–7.37); p < 0.0001) whatever tumour location and gender.
Conclusion: Colorectal cancer survivors may experience the effects of cancer and its treatment on QOL up to 10 years after diagnosis. They noted positive changes or less negative changes in life than controls. However, they still have to face to barriers that are keeping in job by avoiding early retirement and accessing to insurance or a bank loan. Clinicians, psychologist, and social workers must pay special attention for colorectal cancer survivors to improve overall management.

6018 POSTER DISCUSSION

EORTC 22921 Rectal Cancer Trial: Quality of Life (QoL) and Functional Outcome 5 Years After Treatment

J.F. Bosset[1], M. Puyraveau[2], L. Mineur[3], G. Calais[4], E. Bardet[5], P. Maingon[6], M. Bolla[7], L. Collette[8], M. Mercier[9]. *[1]University Hospital, Radiation Therapy Department, Besançon cedex, France; [2]University Hospital, Clinical Research Management Unit, Besançon, France; [3]Clinique Sainte-Catherine, Radiotherapy, Avignon, France; [4]University Hospital, Radiotherapy, Tours, France; [5]Centre René Gauducheau, Radiotherapy, Nantes, France; [6]Centre Georges François Leclerc, Radiotherapy, Dijon, France; [7]University Hospital, Radiotherapy, Grenoble, France; [8]EORTC, Statistics, Bruxelles, Belgium; [9]University Hospital, Statistics, Besancon, France*

Purpose: Long-term impact of preoperative (chemo)radiation (P(C)RT) on QoL bowel and sphincter function in patients with rectal cancer is unknown. Patient-reported health-outcomes in a cross sectional study attached to EORTC 22921 study are presented.
Patients and Methods: 167 French patients, free of disease and had a sphincter preservation had to complete, on time, two questionnaires (Q). The EORTC QLQC30 Q and the Anal Sphincter Conservative Treatment (ASCT), a validated patient-Q.
Results: 5 years after treatments (1−11 y), the QLQC30 Global Health (GH QoL) score was 73.1, similar with observed in a same age group of general population. CT (concurrent, postop or both) negatively affected QLQC30 social functioning (p = 0.06), GHQoL (p = 0.03) and diarrhoea complaints (p = 0.0003). On ASCT, nearly 60% of patients suffered faecal incontinence (any severity), urgency, soiling, modifications of social life. Faecal incontinence was associated with impaired social life measured by both Q.
Conclusion: Adding CT to PRT negatively affect social life. Patients reported high rate of sphincter dysfunction. These results are similar with those previously reported after short course PRT (5 × 5 Gy).

Poster Presentations (Sun, 25 Sep, 09:30−12:00) **Gastrointestinal Malignancies − Colorectal Cancer**

6019 POSTER

No Association Between Dukes' Stage and Genetic − Epigenetic Markers in Colorectal Cancer

B. Metzger[1], M. Dicato[1], G. Mahon[1], S. Obertin[1], J. Kayser[1]. *[1]Centre Hospitalier de Luxembourg, Recherche Sur Le Cancer et Maladies du Sang, Luxembourg, Luxembourg*

Background: The colorectal adenoma-carcinoma sequence goes with epigenetic and genetic modifications, which arise during oncogenesis. These modifications have an effect on the methylation status of different gene promoters and mutations in the K-ras and B-raf genes among others. Included are, as we have previously shown (ASCO 2011, abstract accepted): methylation of the gene promoters of H-cadherin, of MGMT deregulating removal of toxic methyl adducts on guanine bases and inducing the activation of K-ras through G to A point mutations, activation of B-raf gene by a V600E mutation, methylation of E-cadherin and PTEN inactivation by promoter hypermethylation in more than 200 patients.
Methods: DNA extraction was obtained by standard methods from resected tumour samples. PTEN methylation was analyzed by methylation-specific PCR, gel electrophoresis after Sybr green staining and UV-photography. From each individual patient we examined germline DNA from white blood cells as described above.
Results: Of the 95 out of 222 tumours (43%) with a PTEN hypermethylation, 77 (81%) were also methylated in CDH13, 52 (55%) were MGMT methylated, 35 (37%) had a K-ras gene mutation and were B-raf wild type as expected, their mutations being mutually exclusive. All results were tumour specific as all the sequenced blood controls were unmethylated respectively wild type. In 86 of these patients the Dukes' stage was determined and classified as early (Dukes' A & B) and late (Dukes' C & D).
Conclusions: The extremes of the correlations between Dukes' and the other markers ranged from −0.063 for Dukes' and MGMT (p = 0.40) to +0.078 for Dukes' and KRAS (p = 0.81). Thus no significant correlation was found between Dukes' and the other variables. The work is proceeding to include the additional 150 patient data available, but the p values are such that a modification of the conclusion cannot be expected.

6020 POSTER

KRas and Braf: Is a Predictor in Metastatic Colorectal Cancer Patients for Bevacizumab?

F. Selcukbiricik[1], O. Yildiz[1], D. Tural[1], M.A. Ozturk[1], G. Demir[2], M. Ozguroglu[1], S. Erdamar[1], E. Buyukunal[1], N.M. Mandel[1], S. Serdengecti[1]. *[1]Istanbul University, Cerrahpasa Medical Faculty, Istanbul, Turkey; [2]Bilim University, Medical Faculty, Istanbul, Turkey*

Aim: Unlike cetuximab, there is a paucity of biomarkers for bevacizumab to predict outcome of metastatic colorectal cancer (mCRC) patients. Therefore a research for potential biomarker is urgently needed. We aimed to search K-RAS and B-RAF status of our patients whether mutation would effect the outcome of metastatic colorectal cancer treated with first or second line bevacizumab combined with folfiri.
Methods: We retrospectively reviewed the clinical and pathological features of 166 mCRC patients treated in our outpatient clinic between the years 2000 and 2010. KRAS and BRAF mutations were analysed quantitatively by PCR after having extracted the DNA from tumour tissues.
Results: The median age of the patients was 60.5 (27−83). 62% of the patients were male and 37.2% female. Tumour locations were as follows: 36.6% rectum, 25% sigmoid, 16.3% left colon, 1.2% transverse colon, 9.9% right colon, 7.6% cecum. Forty four percent of the patients were KRAS mutant. Eighty patients had BRAF mutation analysis and 6 were found to be BRAF mutant (7.5%). Initial CEA and CA19-9 levels were not correlated with KRAS and BRAF mutations. All 6 patients who were found to have BRAF mutations had rectosigmoid tumours. On the other hand, 41.7% of rectosigmoid, 57% of left colon, 56% of transverse and right colon and cecum were KRAS mutant. Overall, 108 patients had liver metastasis (62.7%). Liver-only disese was 39%. Whereas 43 patients had lung metastasis (25%), 17 had lung-only disease (9.9%). Fourty-six percent of patients who had liver metastasis and 50% of patients who had lung metastasis were found to have KRAS mutation. When both liver and lung metastases were combined KRAS mutation rate rised to 61%. First or second line FOLFIRI and bevacizumab use was not affected by KRAS mutation or wild type status with respect to progression free survival.
Conclusion: KRAS or BRAF mutation was not observed as a potential biomarker in predicting progression free survival in patients with metastatic colorectal cancer who had been treated with first or second line FOLFIRI and bevacizumab. As KRAS mutation was found more frequently in combined lung and liver metastasis, it may represent a more virulant disease.

6021 POSTER

The Role of ABC Transporter Genes in Colorectal Cancer Resistance

I. Hlavata[1], B. Mohelnikova-Duchonova[2], R. Vaclavikova[3], V. Liska[4], P. Novak[4], L. Holubec[5], P. Pitule[4], O. Vycital[4], J. Bruha[4], P. Soucek[3]. *[1]Toxicogenomics Unit: National Institute of Public Health, Third Faculty of Medicine:Charles University in Prague, Prague, Czech Republic; [2]Toxicogenomics Unit: National Institute of Public Health, First Faculty of Medicine:Charles University in Prague, Prague, Czech Republic; [3]National Institute of Public Health, Toxicogenomics Unit, Prague, Czech Republic; [4]Teaching Hospital and Medical Faculty Pilsen: Charles University Prague, Department of Surgery, Pilsen, Czech Republic; [5]Teaching Hospital and Medical Faculty Pilsen: Charles University Prague, Department of Oncology, Pilsen, Czech Republic*

Background: Worldwide, colorectal cancer (CRC) is the third most common malignancy. In terms of CRC incidence, the Czech Republic ranks

second in Europe and the number of new cases is rapidly growing. Due to systemic character of the disease, local recurrence or generalization are diagnosed in about 70% of patients. The individual sensitivity to anticancer drugs plays a key role in cancer therapy outcome. A large part of resistance of tumours to chemotherapy is caused by ATP-binding cassette (ABC) transporters, the ATP-dependent drug efflux pumps. The main aim of this study was to investigate expression levels of all, so far identified, human ABC transporter genes in tissue specimen from colorectal cancer patients and to follow their role in chemotherapy outcome.
Materials and Methods: Expression profile of 49 ABC transporter genes was evaluated in 19 pairs of tumour and distant unaffected mucosa tissues from patients undergoing predominantly the FOLFOX (based on 5-fluorouracil and oxaliplatin) palliative chemotherapy treatment. The analysis was performed by real-time PCR with TaqMan Gene Expression Assays. Stability of 24 reference genes was assessed and four reference genes were then used for normalization. Results were evaluated by REST2009 and SPSS programs.
Results: Significant differences in expression profiles of the examined ABC transporter genes between tumour and non-tumour tissues and between patients with remission vs. progression were observed. Significant upregulation of *ABCA12*, *ABCA13*, *ABCC1* and *ABCE1* gene expression in tumours vs. non-tumours suggested their possible role in outcome of the chemotherapy. More than 40% of ABC transporter genes were downregulated in tumours.
Conclusion: Our study suggests that ABC transporters may play an important role in outcome of colorectal cancer chemotherapy. Candidate genes will be further followed by a larger and more comprehensive study.
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6022 POSTER

Plasma Levels of Heparanase as Marker of Tumour Aggressiveness and Stage of Disease in Patients With Colorectal Cancer

E. Dudnik[1], A. Kuten[1]. [1]*Rambam Health Care Campus, Medical Oncology, Haifa, Israel*

Background: Heparanase enzyme upregulation was documented in large number of tumours, including colorectal cancer. The aim of the study was to evaluate plasma heparanase level in colorectal cancer patients, as a screening tool for diagnostic and disease monitoring purposes and to examine the correlation between plasma heparanase levels and clinical and pathological parameters, such as tumour burden and response to antineoplastic treatments, in patients with colorectal cancer.
Materials and Methods: Plasma heparanase was evaluated in 92 colorectal cancer patients, that were treated and followed-up in the Department of Oncology, Rambam Health Care Campus, Haifa, Israel. The patients were divided into 3 groups, according to their tumour burden. The 1st group was comprised of 47 patients with recurrent or metastatic disease. In this group of patients blood samples were collected at the start of the treatment and at restaging procedure. The 2nd group included 27 patients without evidence of disease up to 6 months after surgery. The 3rd group included 18 patients without evidence of disease at least for two years after surgery. Plasma heparanase levels were measured by enzyme linked immunosorbent assay. Tumour heparanase expression was evaluated by immunohistochemistry in 37 patients.
Results: The median and the mean serum heparanase concentrations in the first sample of the entire population of patients were 0 pg/ml and 179.6±595.3 pg/ml, respectively. In the 1st, 2nd and 3d group of patients the mean plasma heparanase levels were 221.9±703.8 pg/ml (n-47), 28.3±102.6 pg/ml (n-27), and 295.8±696.4 pg/ml (n-18), respectively. There was a trend for higher mean serum heparanase levels among the patients with active disease (1st group) in comparison with the patients without evidence of disease (2nd + 3d group), 221.9+703.8 pg/ml and 135.3+459.5 pg/ml, respectively, ($p = 0.1$). In univariate analysis, smoking history ($p = 0.004$), lymph node sampling ($p = 0.02$), and oxaliplatin-based chemotherapy ($p = 0.007$) were independent predictors of plasma heparanase levels. A trend for higher serum heparanase concentration among the patients with metastatic disease ($p = 0.2$), and high grade tumours ($p = 0.3$) was observed, also the trend for lower plasma heparanase concentration in oligometastatic disease ($p = 0.08$) was seen. Moreover, the non-significant correlation between response to oncological treatment and plasma heparanase alterations was observed ($p = 0.18$). No correlation was observed between tumour heparanase expression and serum heparanase concentration.
Conclusions: The positive, but non-significant correlation between plasma heparanase level and tumour aggressiveness and response to oncological treatment in patients with colorectal cancer was observed. Smoking history, lymph node sampling, and oxaliplatin-based chemotherapy were independent predictors of plasma heparanase level. Larger study is required in order to validate plasma heparanase as a marker of colorectal cancer aggressiveness.

6023 POSTER

Immunohistochemical Expression of CD133 is Associated With Tumour Regression Grade After CRT in Colorectal Cancer

K. Hongo[1], S. Kazama[1], E.I.J.I. Sunami[1], T. Nelson[1], H. Nagawa[1], J. Kitayama[1]. [1]*The University of Tokyo Faculty of Medical Sciences, Department of Surgical Oncology, Tokyo, Japan*

Background: The Cancer stem cell (CSC) model suggests that CSCs are involved in tumorigenesis, metastasis, resistance to treatment and poor prognosis. CD133 has been identified as a putative CSC marker in various cancers including colorectal cancer. We investigated the relationship between CD133 expression and the clinicopathological features as well as the survival of patients with colorectal cancer and those with rectal cancer after preoperative chemoradiotherapy (CRT).
Material and Methods: The expression of CD133 was immunohistochemically evaluated on surgical specimens of 225 patients with colorectal cancer who underwent curative resection as well as 78 patients with rectal cancer who received preoperative CRT followed by curative resection. The latter patients received 50.4 Gy irradiation with oral administration of the prodrug of 5-FU and leucovorin during the entire course of radiotherapy. Expression of CD133 was defined as positive when CD133 staining was found in more than 5% of the entire of the tumour. The correlation between the CD133 expression and the clinicopathological features, tumour recurrence as well as the overall survival was analyzed.
Result: Among the 225 colorectal cancer patients, 93(41.3%) were positive for CD133 expression. However, CD133 was positive in 47 (60.3%) of 78 cases receiving CRT, which was significantly higher than non-CRT specimens ($p = 0.05$). Positive expression of CD133 significantly correlated with the histological tumour regression grade ($p < 0.01$). By multivariate analysis, CD133 expression remained as the most important factor associated with the tumour regression grade ($p < 0.01$) in cases with CRT. However, CD133 expression was not significantly associated with either the recurrence-free or the overall survival in both groups.
Conclusions: CD133 expression may be one of the key factors associated with resistance to chemoradiotherapy in colorectal cancer.

6024 POSTER

E2F2 Transcription Factor as a Possible Genomic Marker in Colon Cancer Initiation/Progression: Impact of Its Altered Expression on a Human Colon Cancer Cell Line

M. Voegelin[1], E. Guérin[1], N. Hammelin[1], C. Nicolet[1], M.P. Gaub[1], D. Guenot[1]. [1]*Université de Strasbourg/CHU Hautepierre, Molecular Markers of Tumour Progression and Sensibility to Anti-Tumoral Drugs, Strasbourg, France*

Background: In order to identify molecular markers pronostic of initiation and/or progression of human colon cancer (CC), a genome-wide analysis was perfomed and highlighted a micro-deletion at the 1p36.11−12 region in 23% (n = 115) and 47% (n = 59) of adenomas and carcinomas, respectively. Within the micro-deleted region, a potential target gene, E2F2, is described as either oncogenic or tumour suppressor, depending on the tissue or cell type. E2F2 deletion incidence depends on tumour stages (60% in early stages whereas only 34% in metastatic stages of distal CC) and further clinical analysis showed that patients with deleted E2F2 had a lower rate of recurrence and a better overall survival. Also, RT-QPCR evidenced that E2F2 transcript expression level decreases in human CC. Thus, the aim of this study was to specify the functions of E2F2 in CC, and the impact of the E2F2 deletion in human CC process.
Material and Methods: E2F2 transcript expression was down-regulated by transitory transfection with siRNA in the human epithelial CC cell line Caco-2/TC7. Consequences were evaluated at the morphological level by immunocytochemistry for proteins involved in the cell architecture and in cell-cell and cell-matrix junctions, and at the expression level by RT-QPCR and Western Blot analyses. Functional analyses were assessed for the migratory potential with the wound healing assay, for proliferation with the MTS assay, and for adhesion on substrates such as laminin, collagen I and fibronectin.
Results: E2F2 down-regulation reduced proliferation and induced severe morphological modifications, associated with relocalization of structural members of adherens junctions (beta-catenin, APC), tight junctions (Claudin-1, ZO-1) and cytoskeleton (F-Actin, Cytokeratin-19). The integrins alpha5, alphaV, alpha2 and beta-1, were downregulated and the adhesion properties on laminin-111, but not on collagen I or fibronectin were lost. More interestingly, inhibition of E2F2 expression leads to a decrease of