Results: I.p. application of catumaxomab was well tolerated. MTD was defined at 10–20–50–200 μg for the 1st, 2nd, 3rd and 4th i.p. infusion. Most frequent adverse events of the first 17 patients >CTC grade 2 were nausea/vomiting (14), abdominal pain (12), fever (6), exanthema (4), elevation of liver enzymes (4) and cholangitis (2), which could all be successfully treated by conventional medication. Analysis of the peritoneal lavages showed a decrease/complete disappearance of tumor cells after trAb treatment in 7/8 patients. After a follow-up period of 15 months, 7/17 patients (41.2%) are alive. At present, the median survival is 9 months (mean 8.6) after treatment and 12 months (mean 12.2) after diagnosis of PC. The updated results at presentation will contain new information about safety (shorter infusion time, dose escalation with premedication, pk data) and survival.
Conclusion: I.p. application of the trifunctional antibody catumaxomab is safe and technically feasible and may represent a new concept for treatment of PC due to gastrointestinal cancer.

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Toward a circulating tumour cell analysis as an early marker for relapse in stage II and III colorectal cancer patients

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Introduction: Different technological approaches have recently used to evaluate the presence of circulating tumour cells (CTC) as a prognostic marker in cancer patients. Contradictory results can be easily found in the literature. However, CTC analysis in metastatic prostate or breast cancer patients resulted in a valuable tool for predicting progression free survival and overall survival.
Material and methods: A two-step design was made: (i) A comparative study was performed to assess the efficiency in the number of tumour cells obtained with four different molecular and cellular methods. We used two systems for tumour cell enrichment (immunomagnetic beads with anti-EpCAM and gradient of density), combined with two different methods to quantify tumour cells (flow cytometry with anti-CD45, anti-CK7 and anti-CK8 antibodies; and quantitative RT-PCR for CK20 gene expression). These experiments were performed in a model system using serial dilutions of HT29 tumour cell-line cells with lymphocytes (from 1 to 10000 HT29 cells in $5X10^6$ lymphocytes). The euclidean distance of the test curve to the perfect one was measured in order to determine the most efficient method along the different tumour cell dilutions.
(ii) CTC analysis using the technical approach selected in the first objective is being performed prospectively every four months, in blood samples (20 ml) from stage II and III colorectal cancer patients, after surgical resection of the primary tumour and informed consent.
Results: (i) Statistical analysis results showed that the immunomagnetic beads, as tumour cell enrichment method, followed by flow cytometry to quantify cells, was the most efficient combination (ED = 60.53); no significant difference was observed when compared to the perfect curve (p = 0.5).
(ii) The follow up of the patients recruited in the study is ranging from 12 to 34 months. Up to date, there are 20 patients with a minimum of six blood samples analysed in our study. In only two cases tumour relapse has been clinically documented. In both patients, we were able to detect a significant increase in the CTC number, five and six months earlier, respectively, to the date that relapse was clinically evidenced. An increase of CTC is also being observed in two other cases but there is not yet any clinical evidence of metastatic disease. Up to now, the rest of cases have a very low, or no detectable, number of CTC and no clinical evidence of relapse.
Conclusions: These preliminary results show that colorectal cancer CTC analysis is a promising tool to detect earlier tumour relapse when compared to conventional methods. More work needs to be done in order to confirm, and definitively conclude, the usefulness of CTC analysis for an early detection of tumour relapse in patients with stage II and III colorectal cancer.

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The effect of dihydropyrimidine dehydrogenase (DPD) activity and germline thymidylate synthase (TS) gene polymorphisms on the survival of colorectal cancer patients treated by adjuvant 5-fluorouracil

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Background: The antitumor activity of 5-fluorouracil (5-FU) is limited by various factors: i.e. the expression of its molecular target TS and the catabolic activity of the DPD. The TS gene is polymorphic, contains 5'-TSER and 3'-TSUTR polymorphisms, which influence its expression. In the present study we investigated the DPD activity and TS gene polymorphisms in the PBMCs of colorectal cancer (CRC) patients treated with adjuvant 5-FU and the relationship between the disease free (DFS) and overall (OS) survival of the patients and the studied prognostic factors.
Material and methods: 166 CRC patients receiving adjuvant 5-FU chemotherapy were involved in this study. Patients were followed-up for 19±14 (median±SD) months. DPD activity from the PBMCs was analysed by radioenzymological and TS polymorphisms by PCR-PAGE and RFLP methods on the DNA samples isolated from the PBMCs.
Results: Based on the DPD activity, patients were divided in four groups: ⩽10; 10–20; 20–30 and >30 pmol/min/10^6 PBMCs. The Kaplan-Meier survival analysis showed significant difference for both DFS and OS (p = 0.0197 and 0.0046, respectively) between the lowest (<10) and highest (>30) activity-groups indicating a significantly longer survival of patients with the lowest DPD activity. 5'-TSER 3R/3R homozygotes showed significantly longer DFS and OS (p = 0.048 and 0.009, respectively). At the same time the 3'-TSUTR genotypes were not significantly associated with DFS or OS although 0bp/0bp genotype-group showed higher hazard ratio compared to that of patients containing at least one 6bp allele. Combining the two TS polymorphisms eight groups were obtained. Evaluating the hazard ratios of the relapse, obtained by applying Cox regression analysis for the eight genotype combination patients were divided in two prognostic groups: "A" (3R/3R with any 3'-TSUTR genotype and 2R/3R with 6bp/6bp) with low (HR ⩽ 1) and "B" (all other genotype-combinations) with high (HR > 1) relapse risk, respectively. Multivariate Cox regression analysis demonstrated the following parameters as significant independent prognostic factors for DFS: tumor localisation, Dukes' stage, treatment type (bolus vs continuous infusion), DPD activity and TS polymorphism combination (p = 0.043, 0.028, 0.003, 0.044, 0.004, respectively).
Conclusion: DPD activity and TS gene-polymorphism combination of PBMCs are independent prognostic factors for DFS in adjuvant-treated CRC patients.
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Modification of the circadian rest/activity rhythm by 1st line oxaliplatin (l-OHP), 5-fluorouracil (5FU) and leucovorin (LV) in patients (pts) with metastatic colorectal cancer (MCC). An international study (EORTC 05963)

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Background: Circadian rest/activity rhythm correlates with several Quality of Life items and is an independent and strong prognostic factor of survival both in chemotherapy-naïve and in pre-treated MCC pts (ASCO 2005 Clin Cancer Res, 2000: 6; 3038). In this international study, we prospectively evaluated the effect of 4 courses of 1st-line chemotherapy (CHT) with biweekly infusional l-OHP, 5FU and LV on circadian rest/activity rhythm in MCC pts.
Methods: 77 MCC pts had rest/activity rhythm assessed for 3 days using a small wrist-watch (actigraph), which records the number of arm movements per minute, both before the beginning of the first course (C0) and after 4 courses (C4) of CHT with l-OHP (100 mg/m^2/course), 5FU (3000–3600 mg/m^2/course) and LV (600 mg/m^2/course). Three validated circadian rhythm parameters were calculated: mean activity (mAct), autocorrelation coefficient at 24 h (r24), indicating the robustness of the activity pattern over