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Capecitabine (X) and etoposide (E) for patients (pts) with locally advanced or metastatic gastric cancer: a Mexican Oncology Study Group phase II trial

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Background: Palliative chemotherapy is the mainstay of treatment for >60% for pts with non-operable/metastatic gastric cancer and for the 80% of cases with recurrent disease after surgery. Response rates for combined chemotherapy range from 0–70%, but half of the pts die within 1 year. Toxicity of the most popular schedules remains high. Therefore, schedules for elderly and poor performance pts are needed. The ELF combination was designed with this aim and produced good results: 53% response rate and 17-month survival in locally advanced responders. X monotherapy is active in the general gastric cancer population with response rates of approximately 30%. This study was conduced to evaluate the XE combination in a poor-prognosis population.
Materials and methods: pts with locally advanced non-resectable gastric cancer or recurrent disease, without previous systemic treatment, were treated with X 1000 mg/m^2 twice daily on days 1–14 + E 120 mg/m^2/day x 3 days, every 3 weeks. Primary objective was response rate (by RECIST criteria) and secondary aims were safety, quality of life, PFS and OS.
Results: Baseline characteristics of the 65 pts enrolled were: male/female (35/30); median age 53 years; ECOG PS 0/1/2 (25%/58%/17%). Main metastatic sites were: stomach (66%), lymph nodes (21%), liver (9%), other (20%). Median number of delivered cycles: 5 (range 1–14). Safety is shown in the table.

% of pts	All grades	Grade 3/4
Anaemia	78	3
Neutropenia	31	14
Thrombocytopenia	8	0
Hand–foot syndrome	45	2
Mucositis	45	0
Diarrhoea	11	0
Vomiting	21	4

Overall response rate was 21% (including 4 pts with CR, all with single site tumour activity), with stable disease in 21% of pts, disease progression in 31%, and ongoing treatment in 27% of pts. Median survival time is 13 months (95% CI, 7–20 months). Quality of life was measured during each cycle: global health status improved to double the basal score at cycle 3 and remained at this level until cycle 5 ($p = 0.006$); treatment reduced fatigue to half that observed at baseline and remained low until cycle 5 ($p = 0.015$); pts perception of nausea/vomiting disappeared from cycle 2 until cycle 5 ($p = 0.05$) and none of the cycles were worse than the baseline level; other symptom scales did not change over time. Physical, emotional ($p = 0.014$) and social functioning improved from cycle 2 onwards.
Conclusions: treatment was well tolerated, dose intensity maintained, quality of life improved in almost all domains and median survival was comparable with that observed with more intensive combination regimens in pts with a less-poor prognosis.

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Gemcitabine plus oxaliplatin (GEMOX) in advanced hepatocellular carcinoma (HCC): results of a phase II study

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Background: New therapies are clearly needed to improve the prognosis of patients (pts) with advanced HCC. The rationale to develop the gemcitabine/oxaliplatin combination in HCC is based on: (i) the synergy between these two drugs, (ii) the clinical activity of Gemcitabine alone and the FOLFOX regimen in HCC according to preliminary phase II results, (iii) the lack of renal or hepatic toxicity of oxaliplatin in cirrhotic pts. Recently, we conducted a pilot study to evaluate two distinct bimonthly schedules of gemcitabine plus oxaliplatin in advanced HCC. We have therefore selected the most promising schedule to perform a phase II trial in non pre-treated advanced HCC patients.
Methods: 34 pts with non pre-treated advanced HCC were prospectively enrolled. They received gemcitabine 1000 mg/m^2 d1 and oxaliplatin 100 mg/m^2 d2 (GEMOX). Treatment was repeated every two weeks until disease progression or limiting toxicity. Eligibility criteria were: pathologically proven advanced HCC or alpha-fetoprotein (AFP) levels over 250 ng/ml associated with a radiological liver tumor, PS (ECOG) 0–2, age >18, measurable disease, adequate hematological and renal functions, compensated Child score <9, and written informed consent.
Results: Thirty two patients are currently evaluable for efficacy and 33 for toxicity. Patient's characteristics, were mean age: 58 (37–82), sex (M/F): 28/6, PS (0/1/2): 8/19/7. 271 cycles of treatment were performed. No toxic death occurred. Hematological grade 3–4 toxicities consisted in thrombocytopenia (27%), anemia (9%) and neutropenia (24%), with 2 febrile neutropenias and no bleeding event. Grade 1, 2 and 3 neurotoxicity occurred in 19, 7 and 3 pts, respectively. No other grade 3–4 toxicity was observed. Five objective and 6 minor responses and 15 stable diseases were observed. Leading to a disease control rate of 77%. One pt have had a curative resection following GEMOX treatment.
Conclusion: gemcitabine plus oxaliplatin seems to be a relatively active regimen with manageable toxicity in non pre-treated cirrhotic patients with advanced HCC.

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Identification of hedgehog-related downstream genes in pancreatic cancer cell

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Background: Several types of cancer have been linked to a disruption of the hedgehog (Hh) signaling pathway, which is crucial to the normal development of many organs. The Hh signaling might also be an important mediator in the development and maintenance of human pancreatic cancer. However, little is known about the function of activated Hh signal and the Hh signaling-related downstream genes in pancreatic cancer. The purpose of this study is to identify the biological role of the Hh gene and to explore the potential target genes of Hh signal in pancreatic cancer.
Material and Methods: Two human pancreatic cancer cell lines, MiaPaCa-2 and Panc-1, were used. MiaPaCa-2 and Panc-1 were cultured and treated with Hh signal inhibitor, cyclopamine (5 μM, 10 μM, 20 μM, 40 μM) or tomatidine (5 μM), as a control. After 72 hours, the degree of apoptosis was measured by fluorescence activated cell sorting (FACS). Expressions of the Hh signal-related proteins were detected by Western blot analyses for Shh (sonic Hh) and Ptch (patched). To identify Hh signal-targeted genes, the oligo microarray containing a set of 22,746 human oligo was used. Expression and activity of cathepsin B, which was identified from our experimental results as one of the target genes of Hh signal, were examined by Western blotting and fluorometric assay, respectively.
Results: Cyclopamine treatment increased apoptosis dose-dependently in Panc-1 ($p < 0.05$) but not in MiaPaCa-2. Shh and Ptch levels were highly expressed in Panc-1 but not in MiaPaCa-2. However, Cyclopamine suppressed the expression of Ptch in Panc-1. After cyclopamine treatment in Panc-1, 138 genes were down-regulated by 2 folds or more and 24 genes by 3 folds or more. These down-regulated genes were cancer-related genes including Cathepsin B. Cyclopamine suppressed the expression and activity of cathepsin B in Panc-1.
Conclusions: Hh signal activation is associated with anti-apoptosis in some pancreatic cancer cells and this effect can be related with the activation of several target genes such as cancer-related genes including cathepsin B, which might be responsible for late mediator of pancreatic cancer.

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Tissue-specific transcription factors network in hepatocellular carcinoma progression

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The progression of epithelial tumors is closely associated with cell dedifferentiation. While the key role in the maintenance of hepatocyte

differentiation and the control of liver-specific gene expression is attributed to hepatocyte nuclear factors (HNF), the role of this class of transcription factors in hepatocarcinogenesis is relatively poorly understood.
Using the experimental model of mouse one-step HCC progression in which a slow-growing differentiated tumor (sgHCC) rapidly gave rise *in vivo* to a highly invasive dedifferentiated fast-growing variant (fgHCC), we have investigated the fundamental mechanisms underlying HCC progression and the role of HNFs in this process.
The progression from sg to fgHCC variant was accompanied by a complete loss of cell polarity, a decrease in cell-cell and cell-matrix adhesion, activation of telomerase, extinction of liver-speci?c gene expression, ability to proliferate rapidly in the culture, invasion and metastasis. These alterations were coupled with a reduced expression of several liver transcription factors including HNF4, a nuclear receptor essential for hepatocyte differentiation. Studies of the collection of chemically induced mouse HCCs of independent origin and human HCC clinical samples revealed strict correlation of HNF4 expression with tumor differentiation status.
Forced expression of HNF4 in cultured fgHCC cells partially re-established epithelial morphology, hepatic gene expression, induced the decrease of proliferation rate and dramatically inhibited tumor growth *in vivo*. Thus HNF4 reexpression can promote the reversion of invasive HCC toward a less aggressive phenotype.
HNF4 promoter was found to be inactive in fgHCC, providing the strong evidence for the existing of HNF4 upstream mechanisms responsible for tumor progression. Some candidate genes were identified by microarray analysis of gene expression profiles in one-step HCC progression model. Investigation of the interplay of HNFs network with signaling pathways conducting the control of cell proliferation and morphology is now in progress.
These data indicates that deregulation of tissue-specific transcription regulation network might be a crucial step of epithelial tumors progression.
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Role of interleukin-1 alpha in hepatic metastatic potential in pancreatic carcinoma cells

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To study the mechanism of gene expression during formation of hepatic metastasis in pancreatic cells, we performed differential display assay of two pancreatic cell lines with highly metastatic potential (BxPc-3 and Sw1990) and two cell lines with non-metastatic potential (Capan-2 and Mia PaCa-2).
There were 39 different shifts in expression, 24 in the highly metastatic group and 15 in the non-metastatic group. Further DNA sequencing, homology research, Northern blotting, and/or reserve transcription-PCR results indicated that interleukin-1 alpha was among those up-regulated in highly metastatic group. An interleukin-1 receptor antagonist was also found to reduce hepatic metastasis in an intrasplenic metastatic assay using nude mice. Antisense cDNA of interleukin 1 alpha into SW1990 caused loss of metastatic potential in nude mice, while interleukin-1 alpha transfection into MIA PaCa-2 generated metastatic potential in nude mice. EMSA assay also demonstrated NF kappa B activation in highly metastatic carcinoma cells. These results indicate that interleukin-1 alpha and activation of the NF kappa B play an important role in the acquisition of metastatic potential in pancreatic cells.

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Interleukin 1 B gene polymorphisms and gastric adenocarcinoma in Oman – Preliminary results

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Gastric cancer (GC) is the most common malignancy in Sultanate of Oman. Interleukin 1beta (IL-1B) gene polymorphisms have been associated with increased risk of GC in Caucasian, Asian and Hispanic populations. No previous studies examined its role in Arab population. We tested the association between IL-1B-31, IL-1B-3954, IL-1RN-(2018) polymorphisms and GC in Omani Arab patients.
Method: Genomic DNA was extracted from peripheral blood of 175 healthy blood donors, 75 gastric cancer patients. The DNA samples were analysed using TaqMan real-time polymerase chain reaction and 5' nuclease assay. The frequency of carriage of the pro-inflammatory alleles were IL-1B-31*C, IL-1B-3954 *T, and IL-1B-RN*C were 76%, 42.3%, and 33.4% respectively in GC patients compared to 67%, 47.8%, and38.2% respectively in the controls. There was no statistical association between carriage of the pro-inflammatory alleles and gastric cancer; IL-1B-31*C (odds ratio [OR] – 1.53, 95% confidence interval [CI]-0.79–2.97, p = 0.2), IL-1B-3954 *T (OR – 1.56, 95% CI-0.56–4.5, p = 0.4), and IL-1B-RN*C (OR – 0.8, 95% CI-0.45–1.47, p = 0.5).
Conclusion: In these preliminary results, there is no association IL-1B-31, IL-1B-3954, IL-1RN-(2018) polymorphisms and GC in Omani Arab patients.

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Analysis of C-KIT mutations in gastrointestinal stromal tumors

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Gastrointestinal stromal tumors (GIST) is the unique model for the molecular based diagnostics, prognosis and treatment of gastrointestinal mesenchymal malignancies. GISTs typically express high levels of the KIT-receptor and carry activating C-KIT mutations, primarily of exons 11 and 9. We analysed C-KIT mutations by direct sequencing in 43 DNA samples from 36 GIST patients and 6 DNA samples from 5 leiomyomas and one leiomyosarcoma. All GISTs were CD117 positive and 65% GISTs were CD34 positive. Mutations of 11 exon were found in GISTs of stomach (68%, 17/25) and intestine (36%, 4/11). The most frequent deletions were located in the region of 551–563aa. with mutations of one of 557, 558 or 559 aminoacid. We did not found any correlation between this mutation and level of malignancy of GIST. In four GISTs with low malignancy we found insertions of different size in the region of 576–585aa of 11 exon. Point mutations of 11 exon were rare. Mutations of 9 exon (duplications of 502–503aa) were found exclusively in GISTs of intestine (45%, 5/11). Such tumors were CD34 negative, rather agressive and had poor prognosis. All DNA samples from 5 leiomyomas and one leiomyosarcoma were CD117 negative without c-KIT mutations in 11 and 9 exons. We conclude that the type and location of C-KIT mutation may be the additional parameter for predicting prognosis and effectiveness of treatment for GISTs.

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Decreased xanthine oxidoreductase is a predictor of poor prognosis in early stage gastric cancer

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Background: Xanthine oxidoreductase (XOR) is a key enzyme in the degradation of DNA, RNA and high energy phosphates. Alterations in XOR expression have been reported in experimental tumorigenesis. We showed previously that breast **cancer** is accompanied by a decrease in XOR expression in about half of the cases, and loss of XOR independently identifies breast cancer patients with unfavorable prognosis. The purpose of the present study was to assess the clinical relevance of XOR in gastric **cancer**.
Materials and Methods: In this study we determined the XOR levels by immunohistochemistry in tissue microarray specimens of 337 patients with gastric **cancer** and assessed the relation between XOR expression and a series of clinicopathologic variables as well as disease specific survival.
Results: XOR expression was moderately decreased in 41% and undetectable in another 14% of the tumors as compared to the corresponding normal tissue. Decreased XOR was associated with advanced stage, deep tumor penetration, diffusely spread tumor location, positive lymph node status, large tumor size, non-curative disease, cellular aneuploidy, high S-phase fraction and high cyclooxygenase-2 expression, but not with p53 expession or Borrmann classification. Downregulation of XOR was associated with unfavourable outcome, and the cumulative five year gastric specific survival in patients with strong XOR expression was 47% compared to 22% in those with moderate-to-negative expression ($P < 0.0001$). This was also true in patients with stage I-II ($P = 0.0124$) and lymph node negative ($P = 0.018$) disease as well as in patients with smaller ($\leqslant 5$ cm) tumors ($P = 0.02$).
Conclusions: Our data suggest that XOR expression in gastric cancer might be a new marker for a more aggressive gastric cancer biology, similar to that as previously reported for breast cancer.