The study of gene expression as a tool for predicting response to chemotherapy in pancreatic cancer.

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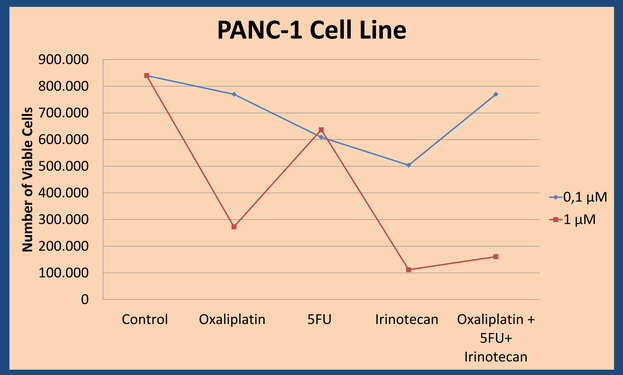
Background: Pancreatic cancer is a malignant neoplasm arising from malignant cells which form the pancreas. The combination of 5-Fluorouracil, irinotecan and oxaliplatin is mostly used in treatment of pancreatic cancer. In the present study a group of genes, which are correlated with basic signal transduction pathways, were tested in order to be determined whether any of these could be an indicator of response to the above chemotherapeutics.

Methods: The experiments were performed by using a human Caucasian pancreatic cell line (PANC-1) and a human primary pancreatic adenocarcinoma cell line (BxPC-3) provided by the European Collection of Cell Cultures (ECACC). Each cell line was treated with different concentrations, both in combination of the above drugs, as well as in monotherapy. The cMET, AKT-1, AKT-2, IGFR-1, IGFR-2, PDGFRα, PDGFRβ, EGFR, ERK1, ERK2, MEK1, MEK2, FGFR2, VEGFR3 and mTOR genes were tested with RT-qPCR in both cases (pre- and post- treatment). The 18SrRNA was used as reference gene and the analysis was performed according to Livak method ($2^{-\Delta\Delta Ct}$) normalized to a normal PBMC's cell line. Finally, the cell viability was measured with propidium iodide staining.



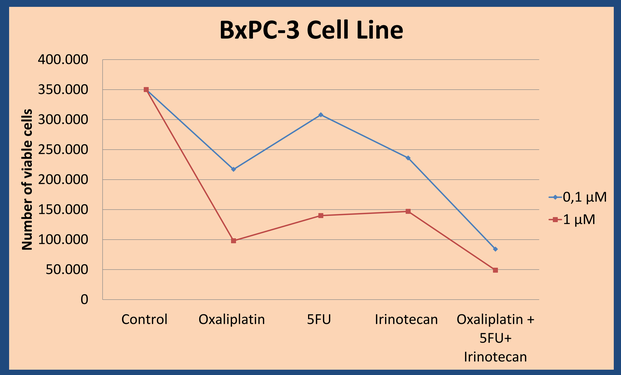
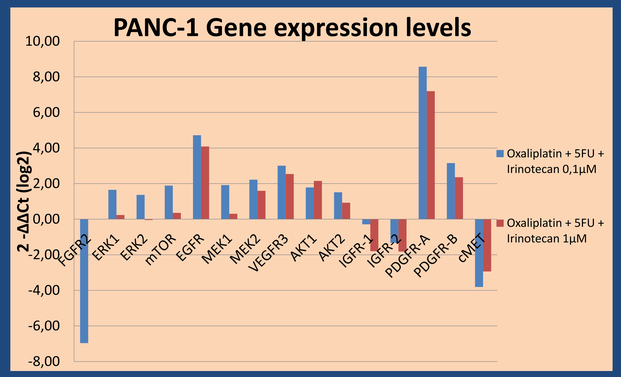


Figure 1: Viability of human pancreatic cancer cells, after 48h of incubation

Conclusions: The resistance to chemotherapy is one of the major problems in cancer treatment. The resistance is either natively-existed (primary), or developed by the cancer cells (secondary). The study of the above panel of genes or individual could be very useful in determining the response to chemotherapeutics. The method is easily implemented, with minimum time and cost requirements. It is essential to perform further studies in other pancreatic cancer cell lines and in more samples, so as to be used at clinical level. However the first results may be quite encouraging.

Results: The cellular-based assays have shown that the combination of drugs was, in most of cases more effective compared to monotherapy. In PANC-1 cell line, the cells that were treated with 0.1μM of each drug, showed lower mortality rate than the others.
The molecular biology assays showed that gene expression levels were different between the two cell lines pre- and post- treatment. However, the greater difference was observed for FGFR2, EGFR, VEGFR3, PDGFRα and cMET genes.



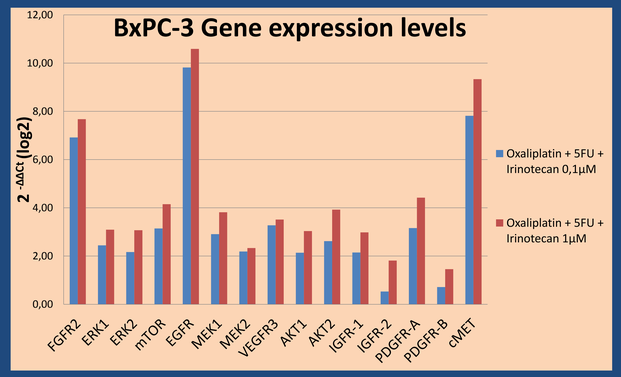


Figure 2: Relative gene expression among PANC-1, BxPC-3 and normal PBMC cell lines.

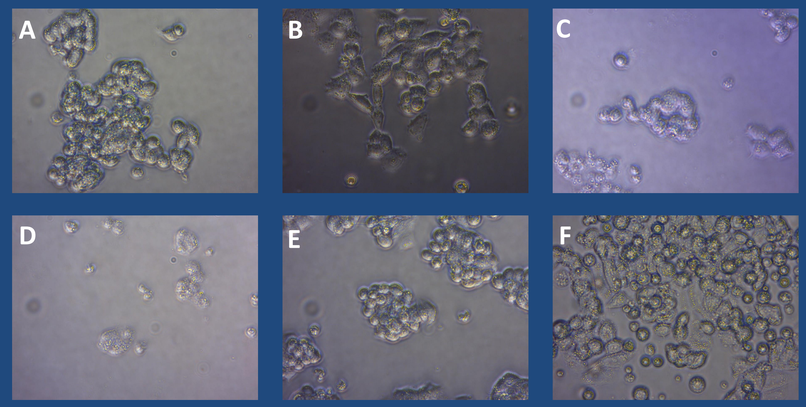


Figure 3: PANC-1 cell line pre- and post- treatment.
A: Untreated, **B**: Oxaliplatin 1μM, **C**: 5FU 1μM, **D**: Irinotecan 1μM,
E: 0.1μM 5FU+ 0.1μM Oxaliplatin+0.1μM Irinotecan,
F: 1μM 5FU+ 1μM Oxaliplatin+1μM Irinotecan.

Selected Reference:

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•Mondo EL, Noel MS, Katz AW, Schoeniger LO, Hezel AF. Unresectable locally advanced pancreatic cancer: treatment with neoadjuvant leucovorin, fluorouracil, irinotecan, and oxaliplatin and assessment of surgical resectability. J Clin Oncol. 2013 Jan 20;31(3):e37-9.