348 POSTER

Targeting pancreatic cancers with a G-quadruplex binding small molecule

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We have previously reported on a novel series of tetraaminoalkyl substituted naphthalene diimide compounds that bind to G-quadruplex nucleic acids and have high potency against a panel of human cancer cell lines.
We now report on a further subset of highly water soluble derivatives with methyl piperazine end groups, which show a high affinity and selectivity for human telomeric G-quadruplexes over duplex DNA. These compounds are similarly potent in the cancer cell line panel, and also show potency against several pancreatic cancer cell lines, with typical IC_{50} values in the dimension of 0.1 μM. The compounds have >40-fold selectivity over a normal fibroblast cell line. Treatment with the compounds at sub cytotoxic concentrations over several weeks lead to a decrease in growth of pancreatic cancer cells, and they stained positive for senescence. *In vitro* experiments show that the compounds inhibit the binding of hPOT1 and TopoisomeraseIIα to G-quadruplex DNA. The fluorescent compounds were visualised inside pancreatic cancer cells by confocal microscopy. In a flow cytometry experiment the phosphorylated Histone H2A.X (Ser139), which occurs in response to DNA damage, was detected in MIA-Pa-Ca-2 cells incubated with the compounds for 14 h. Cell cycle analysis with Propidium Iodide showed an increase of cells in G2/M phase. One of the compounds was evaluated in an *in vivo* study using the pancreatic cancer MIA-Pa-Ca-2 xenograft model. It was well-tolerated when dosed 3 mg/kg every 48 h by intraperitoneal injection. The anti cancer activity of the compound is displayed in form of growth delay of the tumors in the treated animals. The results of the preliminary evaluation of the reported compounds including investigations in their mechanism of action and the *in vivo* study suggest that they are good candidates for G-quadruplex binding anti cancer drugs.

349 POSTER

Monocarboxylate transporter 1 as a potential therapeutic target in glioblastomas

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Background: Glioblastoma is the brain tumor with the highest prevalence and lethality worldwide, having a very low overall survival. Therefore, it is imperative to study new molecular therapeutic targets that could improve the life time of these patients. Dependence of aerobic glycolytic metabolism is a characteristic of malignant tumours, including glioblastomas. To allow continuous glycolysis and prevent cell death from increased intracellular acid accumulation, tumour cells upregulate pH regulators, such as monocarboxylate transporters (MCTs). The efflux of acid lactic produced by glycolysis is dependent of MCTs, which facilitate the co-transport of short-chain fatty acids such as lactic acid, coupled with a proton (proton symport). Expression of MCTs has been described in same tumours, however studies in brain tumours are scarce. Therefore, we aimed to characterize the expression of MCT1, MCT4 and their chaperone protein (CD147) in glioblastoma human samples, as well as in tumour cell lines. Further, we aimed to assess the sensitivity of glioma cell lines to MCT inhibitors.
Methodology: Expression of MCT1 and MCT4, as well as their chaperone protein CD147 was evaluated by immunohystochemistry in 78 cases of glioblastoma samples and in 8 different glioblastoma cell lines by immunohystochemistry and western blot. Sensitivity of these cell lines to MCT inhibitors was assessed using viability/proliferation assays.
Results: MCT1 and CD147 expressions were increased in glioblastomas tissues, compared to the non-neoplastic tissue. A variety of glioma cell lines expressed both MCT1 and MCT4 isoforms, although with different cellular localizations. In most glioma cell lines, both MCT1 and CD147 were expressed in the plasma membrane, while MCT4 expression was only detected in the cytoplasm. Further, MCT classical inhibitors significantly decreased the viability/proliferation of glioma cell lines.
Conclusions: These data provide evidence that MCT1 might play an important role in glioblastoma survival. Thus, exploitation of MCT inhibitors may represent promising strategy in glioblastoma therapy.

350 POSTER

Target therapy and endoarterial chemotherapy with ozonotherapy in combined treatment of metastatic colorectal cancer

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Background: Improvement of quality and increasing of longevity of metastatic colorectal cancer we used endoarterial infusion combined with target therapy, immunotherapy and ozonotherapy.
Materials and Methods: We have taken randomized study on 68 patients in period during 2006–2010 year from them in 1st group (23 patients) included patients that have taken Bevacizumab (5 mg/kg intravenous infusion every 7 days), immunomodulator by intramuscular injection of Transfer Albuminatus Factor (TAF) and intraarterial infusion of ozonized liquor with chemotherapy (CT) by scheme FOLFOX-4 (Oxaliplatin-100 mg/m^2 on 1st day, Leukovorin-200 mg/m^2 on 1st day, 5 Fluorouracil (5FU)-400 mg/m^2 intraarterial intermittent administration on 1st day, then 5FU − 2.4–3.0 g/m^2 48 hours flat continuous infusion) with interval 3 weeks. 2nd group (20 patients) taking CT by similar scheme of intravenous infusion withoutimmunotherapy and target therapy, with interval 3 weeks.
Results: All patients analyzed for toxicity. Main G 2−3 side-effects were: neutropenia on I group − 4.3%, II group − 50%, diarrhea − 4.3; 20%, stomatitis − 4.3%; 10%, neurotoxity − 0; 5%, deep venous thrombosis − 0%; 5%, hypertension − 4.3%; 15%, cardiac ischemia − 0; 5%.No toxic deaths have occurred. All patients have been evaluated for response and we observed 7 cases in I group (1 in II group) complete and 13 (15 in II) partial response rate and 3 stable disease (4 in II group). Up to now 18 patients in I group and 6 in II underwent post-CT surgical resection of metastases with curative intent and 14; 2 − R0 resection have been performed. At median follow-up of 16.3 and 11.8 months, 9 and 18 patients have progressed and median progression-free survival (PFS) is 13.1; 9.2 months with an actual PFS at 10 months of 72%; 43%. To date 7; 16 patients have died and median overall survival (OS) has not yet been reached.
Conclusion: Using endoarterial chemotherapy can be safely combined with ozonotherapy, immunotherapy and target therapy without increasing toxicities no causing unforeseen adverse events. Preliminary data in terms of RR, secondary resection of metastases and PFS are promising increase effect of endoarterial CT.

351 POSTER

Inhibition of monocarboxylate transporter 1 in cervical cancer cells: effect of alpha-cyano-4-hydroxycinnamate and siRNA

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Background: Up-regulation of glycolytic metabolism, even in the presence of oxygen (Warburg effect), has been described as a possible adaptive mechanism to overcome intermittent hypoxia in pre-malignant lesions. In this context, monocarboxylate transporters (MCTs) emerge as important players due to their dual function, as lactate exporters, allowing continuous glycolysis, and tumour intracellular pH regulators, by co-transporting lactate and a proton, inducing extracellular acidosis. We have recently described the up-regulation of monocarboxylate transporter 1 (MCT1) along the progression towards invasive cervical carcinoma. Therefore, we aimed to evaluate the effect of MCT1 inhibition in cervical cancer cells.
Material and Methods: The metabolic behaviour of the human cervical cancer cell lines SiHa, CaSki, HeLa, Sw756, C33 and HaCat, with different high-risk HPV status, was characterised. Extracellular lactate and glucose were quantified using commercial kits. MCT1 activity was inhibited in HeLa cells using the classical MCT inhibitor alpha-cyano-4-hydroxycinnamate (CHC) while MCT1 expression was silenced using siRNA. The inhibitory effects were estimated using the Sulforhodamine B assay. Statistical analysis was performed using the SPSS statistical software.
Results: Glucose consumption and lactate production varied among cell lines, with CaSki, HeLa and HaCat being the most glycolytic cell lines and SiHa showing the lowest rates of glucose consumption and lactate production. Hence, HeLa was further used to perform MCT1 inhibition studies. When exposing cells to CHC, we found a significant decrease in total cell biomass, with an IC_{50} value of 7.29 mM. Importantly, this inhibition was accompanied by a significant decrease in extracellular lactate content. siRNA inhibition of MCT1 expression also showed a significant decrease in total cell biomass.