XRF). methods. The compounds were incubated with target cells for 72 h. At the end of this incubation period, antiproliferative activity in vitro was determined by the MTT assay. In this work we have analysed the cell cycle phase distribution of compounds treated (2IC50) and untreated malignant cells, 24h, 48h, and 72h, after the treatment. After cell staining with propidium iodide cells were analyzed using a Becton Dickinson FAC-Scan flow cytometer.

Results: Results showed that the ligand as well as the complexes demonstrated excellent antiproliferative activity (IC50 values in range of nM, respectively) against all cell lines tested. Flow cytometric analysis showed that apoptosis was mostly induced in K562, HeLa, and MDA-MB-453 cells; MDA-MB-361 cells were with moderate sensitivity to the apoptotic action of compounds but, with marked increase in the percent of these cells in G1 phase.

Conclusion: This examination clearly indicated that thiosemicarbazones and their Zinc(II) complexes have a pronounced antitumor activity on the four neoplastic cell lines.

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Synthesis and primary cytotoxicity evaluation of non-heterocyclic thiosemicarbazones and their metal complexes

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Background: A new series of thiosemicarbazones (TSCs) and metal complexes were synthesized as potential antitumor agents. The aim of this work was to test the cytotoxicity of this compounds on different human tumour cell lines.

Methods: The compounds have been identified by elementar analysis and spectroscopic techniques (1H-NMR, 13C-NMR, IR, MS and XRF). In vitro cytotoxicity of the thiosemicarbazones and complexes was tested by MTT assay against the target cells: human cervix carcinoma (HeLa), chronic myelogenous leukemia (K562), breast carcinoma (MDA-MB-453) and breast adenocarcinoma (MDA-MB-361). Compounds solusions were added to neoplastic cells grown in 96 flat bottomed wells, 20h after cell seeding. Cell survival was determined 72h after the continuous agent action.

Results: Compouds H2SalpipF, H2AppipF and its zinc(II) complexes were evaluated and all the compounds demonstrated the most marked effects towards used target cell lines.

H2AppipF exhibited potent cytotoxic activity against chronic myelogenous leukemia (K562), (IC50=0.76 microM) and cervix carcinoma (HeLa), (IC50=0.89 microM). The complexes of TSCs showed significant improvement in cytotoxic activity against all cell lines (compound Zn(HAppipF)2; IC50=0.24-0.8 μM), and breast carcinoma (MDA-MB-453), (compound Zn(HSalpipF)2; IC50=0.79 μM).

Conclusion: This new group of compounds may offer novel exploratory derivatives for future investigations in the treatment of cancer.

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Nuclear c-Met and p-p38 at the invasive front of oral squamous cell carcinomas

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Background: The c-Met receptor tyrosine kinase is the receptor for hepatocyte growth factor. C-Met promotes cell motility, invasion and survival. Some studies report of nuclear c-Met expression, but the signalling outcome of the c-Met location is unknown. In the present study, we wanted to evaluate the nuclear expression of c-Met at the invasive front of oral squamous cell carcinomas (OSCCs). Furthermore, in an initial attempt to understand how other molecules e.g. activated p38 MAP kinase (p-p38), can influence the nuclear expression of c-Met in oral cancer, a 3D-co-culture organotypic oral cancer (OTOC) model was constructed.

Material and methods: By immunohistochemistry (IHC), the presence of nuclear c-Met and p-p38 was studied at the invasive front of 51 T1-T2 OSCCs. Furthermore, the nuclear expression of c-Met and p-p38 was studied in an OTOC model. In addition, the nuclear expression of c-Met was evaluated by IHC in OTOC models treated with a p38 inhibitor (SB203580).

Results: At the invasive front, 26 of the tumours showed nuclear c-Met expression while 25 tumours did not demonstrate nuclear c-Met. Thirty-five tumours demonstrated nuclear p-p38 staining at the invasive front while 16 tumours did not show nuclear p-p38. At the invasive front we found that when nuclear c-met was expressed in a high number of cells, the nuclear expression of p-p38 was much lower and vice versa (p=0.004). Inhibition of p38 in the OTOC model resulted in an increased number of cells with nuclear c-Met compared to the model without inhibition.

Conclusion: Nuclear c-Met and nuclear p-p38 was present in OSCCs. Either the expression of nuclear c-Met or nuclear p-p38 dominated at the invasive front. Inhibition of p38 increased the number of cancer cells expressing nuclear c-Met in the OTOC model. Our results suggest an association between c-Met and p-p38.

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p75NTR and pattern of invasion predict poor prognosis in oral squamous cell carcinomas

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Background: The high rate of treatment failure for patients with oral squamous cell carcinomas (OSCCs) indicates the need for better prognostic markers to identify tumours not responding to today's treatment. Thus, studies of the biological characteristics in OSCCs are of importance, also with regard to development of new treatment approaches.

Material and methods: In 53 T1-T2 OSCCs we evaluated the prognostic value of p75 neurotrophin receptor ($p75^{NTR}$) known to induce both cell survival and cell death. The results were related to conventional prognostic systems (TNM staging and WHO grading) and invasive front grading/IFG. Immunohistochemically, we assessed the expression of $p75^{NTR}$ both in central/superficial tumour parts and at the invasive front in OSCCs. Hematoxylin and eosin stained sections were graded according to WHO and IFG. Endpoint was disease-free survival.

Results: All tumours expressed $p75^{NTR}$. $p75^{NTR}$ both in central/superficial tumour areas and at the invasive front was associated with poor prognosis (p=0.03, and p=0.02 respectively). Tumours with marked cell dissociation (IFG parameter) was associated with poor prognosis (p=0.03). In tumours showing both $p75^{NTR}$ at the invasive front and marked tumour cell dissociation, the average risk of recurrence was increased about 17 times compared to tumours with low $p75^{NTR}$ expression and collective invasion (p=0.01). Conventional prognostic systems were not of prognostic significance.

Conclusion: $p75^{NTR}$ was expressed in all OSCCs. The $p75^{NTR}$ expression and the pattern of invasion were significantly associated with a poor prognosis, and both were better prognostic factors than conventional prognostic systems. The combination of $p75^{NTR}$ expression and the pattern of invasion strongly increased the identification of patients with low disease-free survival.

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Cold stress induces apoptosis in a multidrug resistant leukaemic cell line by a caspases-dependent mechanism

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The acquisition of a multidrug-resistant (MDR) phenotype that renders tumoural cells insensitive to anti-neoplasics is one of the main causes of failures on chemotherapy of human malignancies. Thus, the discovery of new stimuli able to induce cell death on drug resistant cells are fundamental on the design of new approaches to eliminate drug-resistant tumours. We have found that leukaemic cells with MDR phenotype are sensitive to hypothermia-induced cell death only when those cells are exposed to temperatures below 4 ºC for a long period of time (up to 12 hrs). We have analyzed cell surface exposure of phosphatidylserine by using Annexin-V-FITC and Propidium Iodide (PI) staining, and found that cold exposure significantly induced early apoptosis (as defined by Annexin V-positive/PI-negative) on MDR cells in comparison to their sensitive counterparts. Furthermore, we have determined the role of individual caspases such as the effector caspase-3 and the initiators caspase-8 (extrinsic pathway) and caspase-9 (intrinsic pathway) by using selective inhibitors of each one of these ptoreins and our results showed that the cold-induced cell death mechanism is caspases-dependent.

Together, these findings indicate that acquisition of MDR phenotype by leukaemic cells is accompanied by pleiotropic changes that result on reduced tumour capacity to survive under stress conditions such as hypothermia. Hence, these studies on MDR-tumours may assist in the design of specific therapeutic strategies that could complement current chemotherapy treatments.