markers, including Beclin 1 and ATG5, as well as a relocalization of LC3B protein, was observed, suggesting an enhanced propensity of the cells to undergo autophagic cell death as a determinant of their higher sensitivity to cisplatin. When we assessed the susceptibility of PCa cells to other chemotherapeutic agents, we observed an increased resistance of DU145/miR-205 cells to the mTOR inhibitor RAD001, whereas a comparable sensitivity to paclitaxel was observed for the two cell lines. Overall, these findings suggest that modulation of EMT in PCa cells may result in a different response as a function of the tested drug and that, only for selected agents, combination treatments including EMT-modulators, such as miR-205, can be envisaged to improve cell response.

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Newcastle disease virus Iraqi local isolate as a therapy for murine mammary adenocarcinoma: In vitro and in vivo study

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The avian paramyxovirus, Newcastle disease virus (NDV), is considered to be very promising. Since cytolytic strains of NDV possess a number of desirable properties in this regard. To evaluate the effectiveness of NDV Iraqi isolate (NDV-Iraqi Ahmed Nahi − IAN) as a tumor cytolytic agent, we have performed in vitro and in vivo experiments. In vitro tests studied oncolytic activity on different tumor cell lines by light and electron microscope. In vivo experiment using murine mammary adenocarcinoma allograft grown in mice. We compared antitumor activity of intratumoral injection of NDV-IAN to systemic intraperitonial treatment. In vitro results revealed necrosis and apoptosis induction. While in vivo results showed intratumoral treatment caused average of 92% growth inhibition ($p < 0.0001$), while intraperitonial treatment show 79% growth inhibition at the end of the experiment ($p < 0.0001$) compared to control group. Furthermore treatment groups showed prolong surviving. Histopathological pictures showed massive area of necrosis with infiltration of inflammatory cells mainly lymphocyte. Ultrastructural study showed budding of the virus from the treated tumor cells. Our results suggest that NDV Iraqi isolate (NDV-IAN) as a promising antitumor agent.

Genetics and epigenetics

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Potent in vitro and in vivo anti-tumor activity of ITF2357 by modulation of c-myc related miRNA signature in human Burkitt's lymphoma

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Background and Objectives: Recent studies support the existence of a c-*myc*–microRNA (miRNA) interaction within the genesis and the maintenance of Burkitt's lymphoma (BL). Myc oncoproteins have been found to inhibit the transcription of tumor suppressor genes by recruiting histone deacetylase (HDAC) proteins to target genes. We studied the in vitro and in vivo anti-tumor activity of a novel hydroxamate HDAC pan-inhibitor ITF2357 (Givinostat®, Italfarmaco S.p.A.) on BL cell lines with respect to its ability to modulate BL miRNA expression profile and c-myc target genes.
Methods: Standard MTT assay was used to define the half maximal cell-growth inhibitory concentration (IC_{50}) of ITF2357. Apoptosis and cell cycle phase distribution of treated and untreated cells were analysed by flow cytometry. MiRNA modulation was investigated by array analysis and real time PCR. Cell signalling proteins affected by ITF2357 treatment were analyzed by western blot, immunohistochemistry and confocal microscopy. In vivo anti-tumor activity of ITF2357 alone or in combination of cyclophosphamide (CTX) was studied in subcutaneous Raji xenografted SCID mice.
Results: Namalwa and Raji cell lines treated with 200 nM ITF2357 (48h IC_{50}) showed late and early apoptosis, with subG1 peak formation and G1 arrest respectively. To identify the molecular pathways affected by ITF2357, we investigated c-myc expression and NF-kB activation before and after treatment. Noteworthy, c-myc protein expression was reduced in treated BL cells while its mRNA levels did not change or even increased. As a possible mechanism impairing c-myc translation, we investigated the modulation of miRNA expression profile after treatment with ITF2357. Interestingly, in treated BL cell lines, let-7a and miR-26a that can negatively affect c-myc translation were up-regulated. According to recent evidences about the pro-apoptotic effects of NF-kB activation in human BL, we found that ITF2357 increased the acetylation of NF-kB subunit RelA and NF-kB nuclear localization in BL treated cell lines. The administration of 50 mg/kg ITF2357 to Raji xenografted SCID mice significantly reduced tumor growth compared to untreated control mice. Results from molecular analyses of the in vivo treated tumors were consistent with those obtained in in-vitro experiments. Finally, the combination of ITF2357 and CTX resulted more effective compared to CTX alone in completely eradicating the tumor in vivo.
Conclusion: The in vitro and in vivo anti-tumor effects of ITF2357 against BL cell lines were found associated with the reversion of crucial events in the *c-myc* driven lymphomagenesis, including the restoration of NF-kB activity and let-7a and miR-26a expression. The potent in vivo anti-tumor effects provided by the combined administration of ITF2357 and CTX might be translated in a novel and more effective therapeutic option for BL patients.

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Id1 enhances RING1b E3 ubiquitin ligase activity through the Mel-18/Bmi-1 polycomb group complex

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The helix–loop–helix inhibitor of differentiation and DNA binding (Id1) is well-known as an oncogene in various tumors. Although it has been reported that Id-1 promotes several oncogenic processes, it is still unclear whether Id1 functions through epigenetic transcriptional regulation. In this study, we examined the effect of Id1 on polycomb group (PcG) proteins, which are crucial epigenetic gene silencers, and found that Id1 regulated the expression of Mel-18 and Bmi-1, both of which belong to PRC1. We also confirmed that Id1 induced Mel-18 downregulation, which was mediated by the Akt pathway, and consequently upregulated the transcription of its target gene, c-Myc. Using a promoter-reporter, we demonstrated that Id1 regulated Bmi-1 transcription through c-Myc binding to its E-box in the promoter. Finally, we examined the activity of E3 ligase RING1b whose catalytic activity is increased by binding with the RING finger protein Bmi-1, and found that Id1 over-expression enhanced RING1b E3 ligase activity leading to accumulation of H2A ubiquitination and ubiquitin/proteasome-mediated degradation of geminin. Taken together, our study provided a novel link between Id1 and PcG proteins and suggested that Id1 may contribute to tumor development through PcG-mediated epigenetic regulation.

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Hydroxamate-tethered short chain fatty acid designer cancer prevention molecule

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Among various classes of histone deacetylase (HDAC) inhibitors, short-chain fatty acids exhibit the least potency, with IC(50) in the millimolar range. We rationalized that this weak potency was, in part, attributable to their inability to access the zinc cation in the HDAC active-site pocket, which is pivotal to the deacetylation catalysis. Based on the knowledge that the acetylation status of core histones plays a pivotal role in regulating gene transcription through the modulation of nucleosomal packaging of DNA. In a hypoacetylated state, nucleosomes are tightly compacted, resulting in transcriptional repression due to restricted access of transcriptional factors to their targeted DNA. Conversely, histone acetylation leads to relaxed nucleosomal structures, giving rise to a transcriptionally permissive chromatin state. The level of this posttranslational modification is maintained by a dynamic balance between the activities of histone acetyltransferases (HATs) and histone deacetylases (HDACs), both of which are recruited to target genes in complexes with sequence-specific transcription activators. Aberrant regulation of this epigenetic marking system has been shown to cause inappropriate gene expression, a key event in the pathogenesis of many forms of cancer. For cancer prevention safety and low toxicity are of high importance for drug development. Hence, starting from butyric acid (present in dietary sources) however, concentrations are in milimolar range for this to be meaningful. Here we report an Hydroxamate-tethering approach wheer we explored the structural optimization of valproate, butyrate, phenylacetate, and phenylbutyrate by coupling them with Zn(2+)-chelating motifs (hydroxamic acid and o-phenylene diamine) through aromatic omega-amino acid linkers. This strategy has led to a novel class of Zn(2+)-chelating, motif-tethered, short-chain fatty acids that exhibited varying degrees of HDAC inhibitory potency. One hydroxamate-tethered