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Maternal and infant vitamin D status during the first nine months of infant life — A cohort study

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Abstract: Objective: To assess vitamin D status and possible consequences of low plasma 25-hydroxyvitamin D (25OHD) levels in a population of healthy mothers and their infants. Furthermore, we wanted to examine the influence of vitamin D intakes on plasma 25OHD and the associations between parathyroid hormone (PTH) and 25OHD. **Material and methods:** A total of 107 women aged 24–41 years, gave birth to 108 infants. They were followed for three visits during nine months, for determination of 25OHD and PTH concentrations, in cord and venous blood samples at birth, four and nine months post partum. **Results:** Cord blood level (43.3 ± 20.4 nmol/l) was on averages 62 ± 16% below maternal levels (73.3 ± 30.7 nmol/l), as measured 1–2 weeks postpartum. Cord blood 25OHD correlated positively with maternal 25OHD levels *($r = 0.83$, $p < 0.001$)* and with maternal use of vitamin D supplementation *($r = 0.25$, $p < 0.02$)*. At birth 23% of mothers and 61% of infants had 25OHD<50 nmol/l. Vitamin D deficiency (P-25OHD<25 nmol/l) was present in 66% of the children born by mothers with 25OHD levels below 50 nmol/l ($p < 0.01$), whereas only one child was born with deficiency among mothers with 25OHD>50 nmol/l. Cord blood 25OHD levels correlated with the absolute weight of the nine month old infants *($r = 0.24$, $p = 0.025$)* and with changes in the infants length (growth in cm) between birth and month nine *($r = 0.29$, $p < 0.01$)*. Cord blood PTH levels were very low (median 0.21; inter quartile range 0.11–0.33 pmol/l) with increasing levels ($p < 0.01$) reaching 3.08 (2.67–3.92) pmol/l at the last visit. **Conclusion:** Vitamin D deficiency is widespread in newborn. Maternal 25OHD status at birth is the major determinant of infant 25OHD status even nine months after birth. Maternal 25OHD levels above 50 nmol/l are needed to assure that almost no children are born with deficiency. Low 25OHD levels at birth are associated with an impaired growth during the first nine months of life.

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Sclerostin serum levels are increased in prostate cancer patients

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Abstract: Sclerostin serum levels are increased in prostate cancer patients. **Introduction:** Androgen deprivation therapy (ADT) has clearly shown to improve survival of prostate cancer (PCa) patients when given in combination with radiotherapy for intermediate and high risk localized prostate cancer, but it is associated with an increased risk of bone loss and fractures. Sclerostin is a glycoprotein expressed by osteocytes that acts as a negative regulator of bone formation. Recent studies have evaluated serum sclerostin levels in bone diseases as osteoporosis, but there are no data on PCa patients. **Objectives:** The aim of this study was to compare serum levels of sclerostin in PCa patients and healthy controls, and to evaluate the relationship between sclerostin and bone metabolism in PCa patients. **Methods:** We performed a cross-sectional study including 20 patients with untreated PCa, 20 patients with PCa treated with ADT and 20 healthy controls. We measured serum sclerostin levels, sex steroid levels (total, free and bioavailable testosterone and estradiol) and bone turnover markers (CTX, OC and TRAP). **Results:** Serum sclerostin levels were significantly higher in PCa patients compared to control subjects: ADT: 73.7 ± 25.3 pmol/L; non ADT: 56.14 ± 16.45 pmol/L; healthy controls: 45.52 ± 11.98 pmol/L, $p < 0.05$ for both comparisons. Furthermore, in PCa group, ADT treated patients had significantly higher sclerostin levels than PCa patients without treatment ($p < 0.05$). In healthy controls, serum sclerostin was positively correlated with age ($r = 0.431$, $p = 0.045$). However, there was no correlation with age in PCa patients (ADT: $r = 0.096$, $p = 0.686$; non ADT: $r = 0.149$, $p = 0.497$). In PCa patients treated with ADT, there was an inverse relationship between serum sclerostin and sex steroids: (total T: $r = -0.587$, $p = 0.007$; free T: $r = -0.480$, $p = 0.032$; bioavailable T: $r = -0.480$, $p = 0.032$; total estradiol: $r = -0.498$, $p = 0.025$). There was no correlation between sclerostin and bone turnover markers in any group. **Conclusion:** Serum sclerostin levels are increased in patients with prostate cancer compared to healthy control. Moreover, ADT treated patients had higher sclerostin concentrations compared to patients without treatment. The inverse relationship between serum sclerostin and sex steroids in ADT-treated patients suggests that this protein may be involved in ADT-related bone loss.

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Intermittent dosing with sunitinib (Pf-02783926) and docetaxel provides effective therapy against lytic bone metastasis of human breast cancer

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Abstract: Bone metastases are a frequent complication of many common malignancies including breast, prostate and lung carcinomas. Currently, there are no effective therapies to treat bone metastases of any tumor type. Breast carcinoma causes deterioration of bone associated with clinical symptoms including, pain, pathologic fractures, nerve compression and hypercalcemia. With recent advances in cancer management, patients with metastatic bone disease are likely to have a prolonged clinical course and therefore the early identification and initiation of treatment of patients at high risk for developing bone metastases may help curtail a complex and costly clinical problem and preserve the quality of life of those patients. The mechanism of bone metastases involves the interactions between tumor cells and bone cells, in particular osteoclasts. The primary goal of our study was to assess the therapeutic potential of combination therapy using cytotoxic agent Docetaxel and VEGF inhibitor sunitinib in preclinical models of bone metastases deemed to be predictive of the clinical outcomes. While Docetaxel is drug of choice for bone metastases, sutent (PF-02783926), an oral tyrosine kinase inhibitor of VEGFRs was chosen because bone tumors are known to be highly vascularized and could be susceptible to treatment with anti-angiogenic compounds. By using imaging (X-ray, m-CT, IVIS), serum biomarkers and histochemical methods we demonstrated that intermittent dosing with sunitinib helps normalization of tumor vasculature and significantly improves efficacy of cytotoxic therapy. The novel dosing paradigm applied in this study should guide clinicians not only to enhance design of clinical trials when VEGF inhibitors are used in combination with cytotoxic agents but also to consider targeting breast cancer patients with established bone metastasis using sunitinib and Docetaxel combination.

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Cytotoxicity of marine cyanobacteria extracts on osteosarcoma cells

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Abstract: Marine cyanobacteria have been identified as a rich source of secondary metabolites with potential pharmacological applications. Anti-inflammatory, antibacterial and anticancer activities are some examples of properties described for cyanobacteria compounds, being the cytoxicity against cancer cell lines one of the most documented. The Laboratory of Ecotoxicology Genomics and Evolution (LEGE) —

CIIMAR, Porto, Portugal, possesses a large collection of cyanobacteria strains isolated from the Portuguese coast. In order to investigate the interest of genera such as *Cyanobium*, *Synechocystis*, *Synechococcus*, *Leptolyngbya* and *Pseudoanabaena* which are genera that have been largely overlooked in terms of bioactivity, we have been screening their ability to induce cytotoxicity on human cancer cell lines. Assays have been conducted with a crude extract obtained by a dichloromethane and methanol extraction of freeze dried biomass and three fractions obtained using Si column chromatography with a gradient from 100% hexane, to 100% ethyl acetate to 100% methanol. The cytotoxicity of cyanobacteria crude extract and fractions has been evaluated by the MTT assay at 24, 48 and 72 h. Here we present the results concerning the cytotoxicity of 24 cyanobacteria strains on the osteosarcoma cell line MG63. The results show both inhibitory and stimulatory effects on cell growth within the same cyanobacteria strain. However, five cyanobacteria strains were found to induce a decrease in cell viability that reached the 80% within the ethyl acetate fraction, which makes this fraction interesting for the isolation and characterization of new bioactive compounds.

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Paracrine-induced osteoclastogenesis by human breast cancer cell lines

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Abstract: Paracrine communications between breast cancer cells and osteoclasts might have an important role in the development of osteolytic metastasis. The aim of this work is to evaluate the osteoclastogenic paracrine effects of four human breast cancer cell lines on osteoclast precursors and to characterize the underlying intracellular mechanisms.

Osteoclastic precursors were isolated from human peripheral blood. Cell cultures were performed in the presence of 10% or 20% of conditioned media (CM) from different breast cell lines (SK-BR-3, MCF-7, Hs-578T and T47D), collected at different culture periods. Cell cultures performed with recombinant M-CSF and RANKL were used as positive control. When indicated, cell cultures were also treated with MEK and NFkB pathway inhibitors and a PGE2 synthesis blocker. Cells were analyzed at days 7, 14 and 21 for several osteoclast-related parameters.

All the breast cancer cell lines displayed a high paracrine osteoclastogenic potential. T47D promoted osteoclast differentiation at a higher extent, followed by SK-BR-3, Hs-578T and MCF-7. The observed effects varied with the CM concentration and the culture period. Regarding the involved signaling pathways, NFkB pathway was essential in the osteoclastogenic response mediated by the CM from the four tested cell lines. PGE2 production was also relevant, particularly for the cell line T47D. Although important, the MEK pathway seemed to be less activated in the case of Hs-578T.

In conclusion, all tested breast cancer cell lines induced a high degree of osteoclastogenic differentiation, involving, at least partially, different intracellular signaling pathways. These results might contribute to a better clarification of the osteolytic metastasis process associated to breast cancer.

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Effect of strontium ranelate in men with prostate cancer after chemotherapy

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Abstract: Effect of strontium ranelate in men with prostate after chemotherapy. **Background:** New therapeutic agents are significantly improving survival rates in patients with prostate cancer. Such therapy has improved outcome, but can provide bone complications—low bone mineral density (osteoporosis). The management of low bone mineral density remains a challenge. Our clinical survey has assessed the impact of strontium ranelate on bone mineral density. **Methods:** 20 men(mean age 52+/−3.6 years) with prostate cancer. All patients underwent chemotherapy. All patients performed bone scintigraphy for possible detection of bone metastasis. All the group of patients was reported no metastatic. Bone mineral density was measured by DEXA at spine and hip regions. All patients were diagnosed osteoporotic (T score > −2,5). For the management of osteoporosis all patients received strontium ranelate 2 gr. per day for 12 months. **Results:** All the regions measured by DEXA showed a good response to the therapeutic agent. DEXA results showed a significant increase of bone mineral density (+4.1 +/−0.4%). No side effects from other organs and systems were reported. **Conclusion:** The preliminary results of this study demonstrate that strontium ranelate with daily dose of 2 gr. is a potent therapeutic agent for increasing low bone mineral density induced by chemotherapy.

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PP388

Previous use of amino-bisphosphonates and circulating gamma/delta T cells

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Abstract: Circulating γδ T cell number is an important determinant of the occurrence of acute phase response (APR) after administration of amino-Bisphosphonates (N-BPs). Aim of this study was to explore whether the desensitization to the occurrence of APR in patients previously treated with N-BPs might be due to a long-lasting reduction of the number of circulating γδ T cells.

Seventy-six patients with postmenopausal or senile osteoporosis were enrolled in this study. The number of γδ T cells in the 24 who never received N-BPs treatment was 46 ± 25/ul, while in the 27 patients previously treated with oral N-BPs was 29 ± 17/ul ($p<0.01$ vs. never treated), and in the 25 patients who received intravenous (i.v.) zoledronic acid one year earlier the number of γδ T cells was 28 ± 22/ul ($p<0,02$ vs. never treated). The proportion of circulating γδ T cells were also significantly lower in previous users, both oral and i.v of N-BPs.

These results indicate that N-BPs treatment is associated with long-lasting decrease in circulating γδ T and this may explain the lower incidence of APR in patients previously exposed to N-BPs. The clinical meaning of this sustained effect of N-BPs remains obscure.

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PP389

Osteoblast differentiation stage determines the bidirectional cross-talk with bone metastatic prostate cancer cells

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Abstract: Metastases to the bone are often the most unfavorable and yet incurable final outcomes of tumor growth. The first contact of metastasizing tumor cells with the bone is at the endosteal surface in the bone marrow, consisting of osteoblasts in different stages of differentiation. The aim of our study is to unravel whether the interaction between prostate metastatic cells and osteoblasts depends on the differentiation stage of the osteoblast. We used an in vitro differentiating pre-osteoblast cell line (SV-HFO) and two prostate metastases cell lines, a bone (PC-3) and a lymph metastasis (LNCaP). Interaction was studied both after direct and indirect contact between tumor cells and osteoblasts during three differentiation stages of the osteoblasts (pre-mineralization, early and late mineralization). After 7 days of co-culture osteoblast growth and differentiation and tumor cell growth were assessed. For assessing effects on cell growth in direct contact studies stably GFP expressing PC-3 and LNCaP cells have been generated, to distinguish tumor cells from osteoblasts. Effects on tumor cell growth: Osteoblasts have a different effect on prostate metastasis cells growth depending on the stage of differentiation: pre-mineralization osteoblasts have stimulatory effects on tumor cell growth; early and late mineralization osteoblasts have inhibitory effects on tumor cell growth, both on PC-3 and LNCaP cells. The stimulatory effects are similar after direct or indirect contact; the inhibitory effects are much stronger after direct contact. This suggests the presence of stimulatory soluble factors produced by early stage osteoblasts and inhibitory factors present on the membrane or the mineralized matrix of late stage osteoblasts. Effects on osteoblast growth and differentiation: Only PC-3 cells inhibited alkaline phosphatase activity, especially during the pre-mineralization stage and strongest after direct contact. Both PC-3 and LNCaP cells had no clear effects on osteoblast growth, either after direct or indirect contact co-culture. Comparative gene expression profiling studies of the various osteoblast differentiation stages were performed and are