PB1846

BORTEZOMIB INHIBITS OSTEOCLASTOGENESIS AND MODULATE CHIT1 AND YKL40 EXPRESSION

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Background: Osteolytic bone disease is a common manifestation of multiple myeloma (MM) that leads to progressive skeleton destruction and is the most severe cause of morbidity in MM patients. It results from increased osteolytic activity and decrease osteoblastic function. Activation of mammalian chitinases CHIT1 and YKL40 is associated with osteoclast (OCs) differentiation and bone digestion.
Aims: In the current study, we investigated the effect of two Bortezomib's concentration (BO) (2.5 nM and 5nM) on osteoclastogenesis by analyzing regulation of chitinase expression.
Methods: In order to obtained the OCs, the conditioned medium was supplemented with 25 ng/mL soluble rhRANK ligand (Peproteck, BDA, Italy) and 20 ng/mL rhM-CSF (Peproteck, BDA, Italy), for 21 days w/o Bortezomib (2.5nM or 5nM). The medium was replaced every 3 days. Cells and supernatants were harvested every 3 days for enzymatic assay, qRT-PCR, immunofluorescence and Western blotting. The supernatants were stored at − 20 °C. To confirm that macrophages achieved OCs differentiation, suitable markers were analyzed by qRT-PCR. Finally, in order to evaluate the ability of MM cell lines (U266) to digest bone, dentine discs were added to the wells before cell seeding. U266 cultured with conditioned medium (without BO) for 24 h were used as a control.
Results: OCs exposition to BO was able to inhibit the expression of different OCs markers such as RANK, CTSK, TRAP and MMP9. In addition BO-treatment reduced CHIT1 enzymatic activity and both CHIT1 and YKL40 mRNA expression levels and cytoplasmatic and secreted protein. Moreover,, immunofluorescence evaluation of mature OCs showed that BO was able to translocate YKL40 into the nucleus, while CHIT1 remained into the cytoplasm. Since MM

cell lines such as U266, SKM-M1 and MM1 showed high levels of CHIT1 activity, we analyzed bone resorption ability of U266 using dentin disc assay. After 3 days of incubation, we observed that U266 cells were able to form resorption pits on a dentin disc. Silencing the chitinase proteins in U266 cell line with specific siRNAs, resulted in pits number reduction on dentine discs
Summary and Conclusions: In conclusion we showed that BO decreases osteoclastogenesis and reduces bone resorption in OCs and U266 cell line by modulating the chitinases CHIT1 and YKL40. These results indicate that chitinases may be a therapeutic target for bone disease in MM patients.

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POLYMORPHISM WITHIN THE BFGF PROMOTER REGION IS ASSOCIATED WITH DISEASE PROGRESSION AND RESPONSE TO CHEMOTHERAPY

increase of chromosomal aberrations detection in patients with PC neoplasms (75%) and also allows iFISH study

PB1849

THE ROLE OF FLT3-LIGANT IN THE PROGRESSION OF MULTIPLE MYELOMA: CORRELATION WITH ANGIOGENIC CYTOKINES.

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Background: Background. Multiple myeloma (MM) is a malignant proliferation of monoclonal plasma cells, resulting in a variety of clinical manifestations including osteolytic bone lesions, anemia, hypercalcemia and renal failure.