NOT TO BE MISSED

Clinical and Basic Research Papers – November 2008

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Bone Modeling, Remodeling and Repair


There are well-known differences in hip fracture risk between patients with OA of the hip and osteoporosis. From human biopsies taken at the time of arthroplasty, this study reveals that in addition to cortical thinning, loss of trabecular bone mass and connectivity plays a role in the skeletal fragility associated with hip fracture.—DGL


Sixteen-week-old female A/J, B6, and C3H inbred mouse femora were analyzed using Fourier transform infrared imaging. A/J femora had an increased mineral-to-matrix ratio compared to B6. The C3H mineral-to-matrix ratio was intermediate. C3H had reduced acid phosphate and carbonate levels and an increased collagen cross-link ratio compared to A/J and B6. A/J were the most stiff, B6 being the least, and C3H intermediate. In addition, highly mineralized and brittle A/J had the least amount of work-to-failure.—ES


In tissue engineering it seems desirable to induce vasculature and recruit cells as well as to have a specific osteogenic stimulus to promote differentiation and bone production. In this cranial defect study, VEGF alone, BMP-2 alone and the combination of the two were compared to control defects. VEGF alone increased vessel formation but did not provide a specific bone stimulus, so healing did not occur. VEGF added to BMP-2 increased bone formation at 4 weeks over BMP-2 alone but not at 12 weeks. It remains unclear if the pro-angiogenic and cell recruitment properties of BMP-2 are enough alone or whether augmenting with VEGF is needed.—DGL

Biomechanical techniques for characterizing bone strength are well-documented. This review/tutorial shifts the focus to measuring fracture toughness, i.e., bone’s resistance to fracture, with respect to whole bone testing in small animal studies. Optimizing the variability of measurements means smaller numbers of animals need to be tested to provide meaningful information. —DGL


Non-enzymatic glycation results in the formation of advanced glycation end-products (AGEs). Vehicle (VEH), alendronate (ALN 0.20, 1.00 mg/kg) or risedronate (RIS 0.10, 0.50 mg/kg) was administered using a canine animal model. Accumulation of AGEs was produced at high treatment doses (+49 to +86%; p < 0.001), compared to vehicle. Postyield work-to-fracture was reduced at high doses (-28% to -51%; p < 0.001). AGE accumulation inversely correlated with postyield work-to-fracture ($r^2 = 0.45$; $p < 0.001$). Bisphosphonates given at high doses result in accumulation of AGEs and a reduction in energy absorption of cortical bone. —ES


This editorial on small animal bone mechanical testing updates us on efforts being made to reduce variations in mechanical testing of small animal bones. Under the correct conditions, notched specimens can provide a comprehensive set of evaluations of the material, and not just geometric, properties of small animal bones. —DGL

Cancer and Bone


Pursuing their seminal work in this area, these authors show here that a bisphosphonate with very low affinity for bone reduces skeletal tumor growth in a mouse model of human breast cancer bone metastasis much more effectively than the high affinity bisphosphonate risedronate. Hence, preventing osteolysis by tumor-activated osteoclasts and preventing development of skeletal metastasis may both be achieved by bisphosphonates but may require quite different chemical structures. —SF

Clinical Studies and Drug Effects


Few data are available to determine whether any treatment reduces the absolute risk for fracture – i.e., that fracture rates during treatment in year 3 or beyond are lower than in the first or second year of treatment. Nor is it clear whether the relative risk reduction is maintained (in which case the absolute risk for fracture can go up or down but to different degrees in the groups). In about 1500 women receiving placebo or teriparatide 20 mg or 40 mg, risk for nonvertebral fractures and/or backpain decreased with time. The data suggest that the apparent rising risk for fracture is prevented, producing stable risk reduction, but it is not clear whether fracture rates in the latter months were actually lower than in earlier months; this would be consistent with reversal of fragility. —ES

Recent data indicate that intermittent PTH has greater effects on BMD and fractures compared to alendronate in glucocorticoid-induced osteoporosis. However, the mechanisms by which these drugs improve BMD and bone strength in response to corticosteroids remain poorly understood. This study uses a rodent model of bone loss and alteration of microstructure induced by prednisolone to unravel the tissue and molecular effects of PTH and risedronate treatment therein. Results indicate that both drugs improve bone mass, degree of bone mineralization, and bone strength in glucocorticoid-treated mice. PTH increases bone formation while risedronate reverses the deterioration of bone mineralization. —SF

**Genetics**


Gerodermia osteodysplastica (OMIM 231070) is a rare recessive progeria-like disorder of prematurely aged skin and severe osteoporosis. By performing a genome-wide linkage scan in 13 affected families of Mennonite origin, this study identifies a locus on 1q24 and subsequently a number of mutations in a gene that codes for a Golgi protein that is a partner of Rab6 and is thus involved in vesicular trafficking. How these gene mutations lead to osteoporosis and skin aging, however, remains to be fully elucidated, as does the potential role of this protein in common osteoporosis. —SF


Peak BMD obtained during young adulthood is a major determinant of osteoporotic fractures. This paper describes a fine-mapping study for chromosome 1q, which showed significant linkage (LOD = 4.3) with variation in spinal areal BMD in healthy premenopausal white women in a previously reported study by the same group. Through a two-step strategy, this research demonstrates that genetic factor(s) in a 230-kb LD block in chromosome 1q play an important role in peak spinal BMD in healthy premenopausal white women. —HWD


An autosomal genome-wide scan for BMD at the lumbar spine and femoral neck was conducted in a total of 103 pedigrees ascertained through a male relative with low BMD values at either the lumbar spine or femoral neck. The identified genomic regions largely overlapped with previous reported ones important to BMD or other bone-related traits.

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These regions also encompass several putative genes for osteoporosis such as COL1A1, SOST, and LRP5. —HWD


Polymorphisms of the CYP1B1 gene, a member of the CYP450 superfamily, were tested for association with estrogen metabolism and BMD in 468 postmenopausal Caucasian women. The authors' findings suggest that through its effect on the rate of estrogen catabolism, the Val432Leu polymorphism of the CYP1B1 gene represents a possible genetic risk factor for osteoporosis in American women. —HWD


Recent genome-wide association studies (GWAS) for osteoporosis have focused on single nucleotide polymorphisms (SNPs) associated with BMD and/or fracture. However, a substantial proportion of genetic variation is represented by copy-number variation (CNV) (see review below by Altshuler and colleagues). In this GWAS for hip fractures vs controls in Chinese individuals, the authors extracted information on CNVs from the Affymetrix 500k gene chip, found association of some CNVs with fracture risk and then identified a deletion variant in the gene UGT2B17, encoding an enzyme for steroid metabolism. As with all good association studies, the findings were replicated in an independent cohort of Chinese individuals with and without hip fractures. —SF

Molecular and Cell Biology


Biomechanical stimulation of bone formation is now known to be at least partly mediated by an inhibition of sclerostin and to engage the Wnt/LRP5/β-catenin pathway. This in vitro work on osteoblasts indicates that strain may directly induce translocation of β-catenin to the nucleus, i.e., act downstream of the receptor complex. —SF


Strontium (Sr\textsuperscript{2+}) dose-dependently stimulates the apoptosis of mature osteoclasts through the Ca\textsuperscript{2+}\textsubscript{o}-sensing receptor (CaR), which results in stimulation of a PLC-dependent signaling pathway and nuclear translocation of NF-kB. Unlike Ca\textsuperscript{2+}\textsubscript{o}, Sr\textsuperscript{2+}\textsubscript{o}-induced osteoclast apoptosis depends on PKCII activation and is independent of IP3 action. Sr\textsuperscript{2+}\textsubscript{o} and Ca\textsuperscript{2+}\textsubscript{o} in combination exert a greater effect on apoptosis than either by itself. —ES

The authors demonstrate that genetic ablation of TRPV4, a Ca\textsuperscript{2+}-permeable channel of the TRP family, increases bone mass by impairing bone resorption in mice. TRPV4 mediates basolateral Ca\textsuperscript{2+} influx specifically in large osteoclasts when Ca\textsuperscript{2+} oscillations decline. Thus, TRPV4-mediated Ca\textsuperscript{2+} influx secures intracellular Ca\textsuperscript{2+} concentrations to ensure NFATc1-regulated gene transcription, and regulates the terminal differentiation and activity of osteoclasts. TRPV4 can become a therapeutic target for bone diseases. —TM


The mediators of osteoblast recruitment by osteoclasts are still poorly known. Platelet-derived growth factor (PDGF) has been suggested to be one of them. Using Boyden co-culture chambers, this study further shows that RANKL-differentiated osteoclasts producing PDGF-bb stimulate recruitment of osteoblast precursors in vitro and that either inhibition of PDGF gene expression in osteoclasts or the PDGF receptor gene in osteoblasts reduces chemoattraction of osteoblasts by osteoclasts. —SF


BMP2 induced Msx2 and Osterix expression and promoted osteoblastic differentiation in mesenchymal cells from Runx2-deficient mice. Overexpression of Smad1 and Smad4 upregulated Osterix expression, and Smad6 suppressed BMP2-induced Osterix expression in Runx2-deficient cells. Overexpression of Msx2 enhanced and knockdown of Msx2 inhibited Osterix expression enhanced by BMP2 in Runx2-deficient cells. In addition, Osterix and Runx2 regulated the expression of distinct proteins. Thus, Osterix is regulated not only by Runx2-dependent but also Msx2-dependent mechanisms, and Osterix and Runx2 control osteoblast differentiation by regulating distinct gene expression. —TM


Deletion of the C/EBP\beta gene from mice resulted in suppression of osteoblast differentiation and delayed bone formation with concurrent suppression of chondrocyte maturation. In osteoblasts, C/EBP\beta heterodimerized with ATF4, and this complex transactivated OSE1 of the osteocalcin promoter. C/EBP\beta also enhanced the synergistic effect of ATF4 and Runx2 on osteocalcin promoter transactivation by enhancing their interaction. These results provide evidence that C/EBP\beta is a crucial co-factor in the promotion of osteoblast maturation by Runx2 and ATF4. —TM

Osteoimmunology is not only about modulation of bone turnover by immune and inflammatory cells, but also about the importance of the bone niche for hematopoietic/immune cells. Here, deletion of Gsα (a subunit from the heterotrimeric G protein complex responsible for cAMP signaling) in osteoblast progenitors – using an Osterix promoter Cre – not only resulted in dramatically reduced osteoblast surfaces and low trabecular bone volume, but also in decreased numbers of B cells, whereas T cells were preserved. Low B cell numbers may be explained by a marked reduction of IL-7 expression by osteoblasts without cAMP signaling. —SF

Physiology and Metabolism


Activation of the renin-angiotensin system (RAS) increases bone turnover. Transgenic mice expressing the human renin gene were normotensive and had a low bone mass. Angiotensin II (AngII) acted on osteoblasts and increased RANKL and VEGF, thus stimulating the formation of osteoclasts. Knockdown of the AT2 receptor inhibited AngII activity, while silencing of the AT1 receptor enhanced it, suggesting a functional interaction between the 2 AngII receptors on the osteoblastic cell surface. Finally, treatment of THM mice with an ACE inhibitor, enalapril, improved osteoporosis as well as hypertension, whereas treatment with losartan, an ARB specific for AT1, resulted in exacerbation of the low bone mass phenotype. —ES


Structure-function studies of PTH ligand and receptor have led here to the development of PTH analogs with prolonged binding and cAMP signaling in vitro. In vivo testing of these compounds indicates increased bone resorption, hypercalcemia and hypophosphatemia, together with improved trabecular bone volume. These results are similar to previous observations with a long-acting PTH-Fc fusion molecule, which therefore reinforces the new paradigm that bone anabolism by long-lasting PTH analogs can be obtained simultaneously with catabolism and hypercalcemia. —SF

Public Health


Of 23,146 patients who had a hip fracture, 6% received treatment. Bisphosphonate treatment was dispensed to 2.6% and 3.6% of the patients within six months and one year after the occurrence of the hip fracture, respectively. —ES

Reviews, Perspectives and Editorials


Other Studies of Potential Interest


Lewis J. From signals to patterns: space, time, and mathematics in developmental biology. Science. 2008 Oct 17;322(5900):399-403. [Abstract] [Full Text]


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