Clinical and Basic Research Papers – October 2009

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Bone Modeling, Remodeling, and Repair


In dogs, clinical or high-dose treatment for 3 years with alendronate does not adversely affect the material properties of femoral cortical bone. Intrinsic (material) properties (ultimate stress, elastic modulus, and toughness) were estimated using standard formulae. Would fatigue testing rather than loading to failure give different results? —DGL


This article is not just for orthopedics. It discusses the impact of change management on practice. —DGL


Lin et al. report that unloading decreases Wnt/beta-catenin signaling accompanied by upregulation of Sost. Sclerostin inhibited Wnt/beta-catenin in vivo and suppressed the activity of osteoblasts and the viability of osteoblasts and osteocytes. Sost(-/-) mice were resistant to unloading-induced bone loss and unaccompanied by reduced bone formation or decreased Wnt/beta-catenin signaling. Sclerostin is a target for preventing disuse osteoporosis. —ES


◆ McGee-Lawrence ME, Wojda SJ, Barlow LN, Drummer TD, Bunnell K, Auger J, Black HL, Donahue SW. Six months of disuse during hibernation does not increase intracortical porosity or decrease cortical bone geometry, strength, or mineralization in black bear (Ursus americanus) femurs. J Biomech. 2009 Jul 22;42(10):1378-83. [Abstract]

The important concept here is that hibernating bears possess a mechanism by which
they do not experience bone loss (either trabecular or cortical) associated with disuse. There was no increase in cortical porosity, and no difference in trabecular bone mass after hibernation compared to before. Also, trabecular bone showed no effect on eroded surface or osteoid surface, suggesting that the bone remodeling rate was not altered during hibernation. Studies of hibernation could lead to an expanded understanding of how to reduce the negative bone balance associated with disuse or age-related bone loss. —Matthew R. Allen


Overexpression of OPG reduced osteoclasts and turnover, and increased peak load in vertebrae. Femurs from OPG-Transgenic (Tg) rats were of normal length, but had osteopetrotic changes, reduced periosteal perimeter (-6%) and reduced bending strength. —ES


This is an interesting follow-up paper showing that Hif activation and VEGF production result from inhibition of the prolyl hydroxylase enzyme (PHD) with simple small molecules such as desferrioxamine (DFO). This and other inhibitors were applied by direct injection at the fracture site in a stabilized murine femur fracture model. PHD inhibition increased vascularity at 14 days and increased callus size as assessed by microCT at 28 days. No significant differences in mechanical properties were noted, but this may require a larger animal model to produce. —DGL


The investigators report a reduction in bones’ resistance to crack formation as glycation increases. This may be an important cause of bone fragility during aging and a problem with treatments that reduce bone remodeling as suppression of remodeling, at least with bisphosphonates, is a well-documented cause of increased glycation in animal studies. —ES

Tsuji K, Cox K, Gamer L, Graf D, Economides A, Rosen V. Conditional deletion of BMP7 from the limb skeleton does not affect bone formation or fracture repair. J Orthop Res. 2009 Sep 24. [Epub ahead of print] [Abstract]

This is a follow-up article to work showing that conditional deletion of BMP-2 in the limbs leads to spontaneous fractures that do not heal. Here, conditional deletion of BMP-7 from the embryonic limb prior to the onset of skeletogenesis has no effect on postnatal limb growth, articular cartilage formation, maintenance of bone mass, or fracture healing. Other BMPs present in adult bone seem sufficient to compensate for the absence of BMP7. —DGL

Clinical Studies and Drug Effects

Cawthon PM, Ewing SK, McCulloch CE, Ensrud KE, Cauley JA, Cummings SR, Orwoll ES;

Bone loss accelerates as age advances because the same or a higher intensity of remodeling removes the same or an increasing volume of bone from an ever decreasing volume. Among 4720 community-dwelling men followed over 4.6 years, femoral neck (FN) BMD loss was 1.7% and accelerated with age. FN BMD loss in men 85 years of age was 2.5 times greater than for men 65 years of age, increasing the risk of hip fracture by 25%. Men with lower BMD at baseline lost the most BMD over follow-up. —ES


Finkelstein et al. report a blunting of the response to teriparatide after retreatment in terms of BMD and remodeling markers. —ES


In 704 postmenopausal osteoporotic patients randomized to daily oral 1 mg minodronate (n = 359) or placebo (n = 345) for 24 months, treatment reduced the risk of vertebral fractures by 59% (95% CI, 36.6-73.3%). —ES

Genetics


This is another large-scale genome-wide association study searching for genes underlying osteoporosis. The researchers identified 20 genetic regions that were associated with BMD of the lumbar spine and femoral neck, 13 of which had not been reported previously. This study enhances our understanding of the complex genetic architecture underlying osteoporosis. —HWD


Previous studies have suggested the importance of chromosome 3p14-p21 and ARHGEF3 gene polymorphisms in BMD determination. The product of the ARHGEF3 gene activates the RHOA GTPase, whose encoding gene is also located in chromosome 3p14-p21. This study reports significant associations of RHOA
polymorphisms with spine and hip BMD in a discovery cohort of 769 female sibs. The association was then confirmed in another cohort of 780 postmenopausal women with the same effect direction. In short, multiple lines of evidence highlight the involvement of the RhoGTPase-RhoGEF pathway in BMD determination. —HWD

**Molecular and Cell Biology**


Mice with type 1 cannabinoid receptor (CB1) deficiency (CB1(-/-)) show increased peak bone mass due to reduced bone resorption, but develop age-related osteoporosis with reduced bone formation and accumulation of adipocytes in the bone marrow. Marrow stromal cells from CB1(-/-) mice show, and pharmacological blockade of CB1 in stromal cells from wild-type mice produces, enhanced adipocyte differentiation and reduced osteoblast differentiation. CB1 expression is upregulated with age in marrow stromal cells, osteoblasts, adipocytes and osteoclasts. Thus, while the CB1 receptor acts negatively on peak bone mass through an effect on osteoclast activity, it protects against age-related bone loss by enhancing osteoblast differentiation. This study suggests that peripheral CB1 agonists may protect against age-related bone loss when used in the aged, and CB1 antagonists may increase peak bone mass when used in the young. —TM


Bcl-xL is a member of the B cell lymphoma 2 (Bcl-2) family, but its role in bone cells was unknown. Osteoclast-specific conditional deletion of Bcl-x reveals that it negatively regulates Src activation and thereby osteoclastic bone resorption. Hence Bcl-x cKO mice have osteopenia that worsens with age due to high bone resorption, despite normal osteoblast number and surface. —SF


These researchers have generated OASIS-deficient mice to find out that these animals have decreased bone mass and spontaneous fractures. A series of in vivo and in vitro studies elucidate the dual role of the basic leucine zipper transcription factor (TF), OASIS, in the direct transcriptional activation of the Col1a1 gene, its response to BMP2 and collagen processing through the endoplasmic reticulum (ER) for secretion. Hence these results identify a new TF downstream of Runx2 that appears to be essential for osteogenesis and bone formation. —SF

This study shows that, in response to intermittent PTH, TCRβ(-/-) mice devoid of αβ T cells display diminished Wnt signaling in preosteoblasts and blunted osteoblastic function with decreased bone anabolism. Among Wnt family ligands with bone anabolism, intermittent PTH increases only Wnt10b production in CD8+ T cells. Furthermore, intermittent PTH shows no bone anabolism in Wnt10b(-/-) mice, as well as in TCRβ(-/-) mice reconstituted with Wnt10b(-/-) T cells. Thus, activation of Wnt signaling in osteoblastic cells by T-cell-derived Wnt10b plays a key permissive role in the anabolic effect of intermittent PTH. These results, along with the observation that intermittent PTH decreases Wnt inhibitors in bone cells, contribute to the understanding of the mechanism of bone anabolism by intermittent PTH. However, because LRP5-mediated Wnt signaling is shown to stimulate bone formation by inhibiting serotonin synthesis in the duodenum, signaling molecules mediating the activation of the Wnt pathway in osteoblastic cells remain to be clarified. —TM


Using an elegant combination of conventional and tissue-specific gene deletions of tryptophan hydroxylase 1 (Tph1), Tph2, Adrβ2, Htr1a, Htr2b, Htr3c and ObRb in wild-type and ob/ob mice, this study convincingly demonstrates that, in order to regulate bone mass accrual, appetite and energy expenditure, leptin acts in Tph2-expressing neurons of the brainstem to inhibit serotonin synthesis. Brainstem-derived serotonin favors bone mass accrual by binding to Htr2c receptors on ventromedial hypothalamic neurons, and appetite via Htr1a and 2b receptors on arcuate neurons. Leptin inhibits bone formation and appetite, and increases energy expenditure because it reduces firing of serotonergic neurons and serotonin synthesis. These results modify the map of leptin signaling in the brain and indicate that the serotonergic neuronal circuitry exerts a fundamental influence on the common control of bone mass and energy metabolism. —TM

Pathophysiology


Eddleston et al. report that an antibody to sclerostin (Scl-AbI) prevents inflammation-induced bone loss in a model of colitis in animals. Scl-AbI-treated animals had higher femoral BMD, and a reverse in the decline of both intrinsic and extrinsic mechanical properties of the femur, when treatment was initiated after colitis-associated bone loss had occurred such that strength was no different to noncolitic controls. —ES


This study adds to our understanding of the pathophysiology of ONJ by following 30 cases (24 malignancies and 6 osteoporosis) over nearly 2 years and obtaining histological analyses of the necrotic bone in 26 samples. It shows prominent infiltration by
inflammatory cells and bacteria in necrotic and perinecrotic bone, whereas osteocytic lacunae were empty in necrotic bone (by definition). In perinecrotic bone, osteocyte number decreased and empty lacunae/apoptotic osteocytes increased with the degree of inflammation. Numerous TRAP⁺/CTR⁻ cells were observed in the inflammatory tissue around the necrosis, but bone formation was not detectable when inflammation was marked. The authors propose a chronological scheme of events starting with inflammation, leading to osteocyte apoptosis, bone necrosis – aseptic at first and then infected – and finally to ONJ. The role of BiPi and low bone turnover, if any, in this inflammatory-based process remains unclear. —SF


This exciting study shows that overexpression of C-type natriuretic peptide (CNP) in the liver or continuous infusion of CNP reverses the phenotype in a mouse model of achondroplasia (Ach). FGFR3 activation is known to act principally through the RAS/MAPK pathway. CNP inhibited this pathway and effectively widened the hypertrophic zone and restored longitudinal growth without any discovered side effects. Although the immediate clinical application seems limited, as a proof-of-principle, this work shows that skeletal dysplasias may be treatable. —DGL

Public Health


The prevalence of risk factors used in the new NOF FRAX®-based Clinician’s Guide was estimated using NHANES III and suggests that 20% of men and 37% of women are candidates for treatment. —ES


In 169,145 patients with a first hip fracture (HFx) followed a median of 3.8 years, 27,834 had a second HFx; a cumulative incidence of 9% after 1 year and 20% after 5 years. The RR of second HFx was 2.2 (95% CI: 2.0-2.5) at 1 year and normalized at 15 years. Mortality at 1 and 5 years after a second HFx was higher compared with the background population (men – 1 year: 27% versus 9%; 5 years: 64% versus 40%; women – 1 year: 21% versus 10%; 5 years: 58% versus 41%). —ES

Reviews, Perspectives and Editorials

Eliasson P, Jönsson JI. The hematopoietic stem cell niche: Low in oxygen but a nice place to be. J Cell Physiol. 2009 Sep 1. [Epub ahead of print] [Abstract]

Other Studies of Potential Interest


Matsushita T, Chan YY, Kawanami A, Balmes G, Landreth GE, Murakami S. ERK1 and ERK2 play essential roles in osteoblast differentiation and in supporting osteoclastogenesis. Mol Cell Biol. 2009 Sep 8. [Epub ahead of print]


Sugiyama T, Price JS, Lanyon LE. Functional adaptation to mechanical loading in both cortical and cancellous bone is controlled locally and is confined to the loaded bones. Bone. 2009 Sep 3. [Epub ahead of print] [Abstract]

Yang CM, Hsieh HL, Yao CC, Hsiao LD, Tseng CP, Wu CB. Protein kinase C-delta transactivates platelet-derived growth factor receptor-alpha in mechanical strain-induced collagenase 3 (matrix metalloproteinase-13) expression by osteoblast-like cells. J Biol Chem. 2009 Sep 18;284(38):26040-50. [Abstract] [Full Text]


Conflict of Interest: Dr. Ferrari reports that he receives research support from Amgen and is an advisory committee member and lectures occasionally at conference symposia for Merck Sharp & Dohme, the Alliance for Better Bone Health (sanofi aventis/P&G), Amgen, Eli Lilly (Switzerland), Servier (Switzerland), and Novartis (Switzerland). Dr. Little reports that he receives royalties, research funds and consultancy fees from Novartis Pharma, as well as research support from Stryker Biotech. Dr. Seeman reports that he is an advisory committee member for sanofi-aventis, Eli Lilly, Merck Sharp & Dohme, Novartis, and Servier, and that he lectures occasionally at conference symposia for those companies. Dr. Matsumoto and Dr. Deng report no conflicts of interest.