

NOT TO BE MISSED

Clinical and Basic Research Papers – January 2010

Serge Ferrari, Editor-in-Chief

Ego Seeman, Clinical Editor

David G. Little, Associate Editor

Toshio Matsumoto, Associate Editor

Bone Modeling, Remodeling, and Repair

◆Allen MR, Erickson AM, Wang X, Burr DB, Martin RB, Hazelwood SJ. Morphological assessment of basic multicellular unit resorption parameters in dogs shows additional mechanisms of bisphosphonate effects on bone. *Calcif Tissue Int.* 2009 Dec 2. [Epub ahead of print] [[Abstract](#)]

Bisphosphonates reduce the rate of bone remodeling in part by reducing the initiation of new basic multicellular units (BMUs) as reflected by a reduction in the proportion of the endosteal envelope undergoing bone resorption. This reduction in tissue level remodeling is well-documented but whether these drugs reduce the volume of bone resorbed at the cellular level by each BMU is not known. Indeed there is no evidence for this except perhaps for estrogen therapy. The authors report resorption depth (Rs.De), area (Rs.Ar), and width (Rs.Wi) in vertebral trabecular bone of beagles after 1 year of vehicle (VEH), alendronate (ALN; 0.10, 0.20, or 1.00 mg/kg/day), or risedronate (RIS; 0.05, 0.10, or 0.50 mg/kg/day). ALN or RIS at doses used in osteoporosis (0.20 and 0.10 mg/kg/day, respectively) had lower Rs.Ar (-27%) and Rs.Wi (-17%), with no difference in Rs.De, compared to VEH. Low doses of ALN and RIS did not affect any parameters. There were no differences between RIS and ALN. —ES

◆Emerton KB, Hu B, Woo AA, Sinofsky A, Hernandez C, Majeska RJ, Jepsen KJ, Schaffler MB. Osteocyte apoptosis and control of bone resorption following ovariectomy in mice. *Bone.* 2009 Nov 17. [Epub ahead of print] [[Info](#)]

Bone matrix remodeling is initiated from signals within the matrix that must communicate with the marrow environment. This is achieved by osteocytes that die by apoptosis and communicate the location of damage to the endosteal lining cells that form the roof of a bone remodeling compartment within which precursors of the osteoclast and osteoblast lineages are recruited and differentiated to then target, remove and replace damaged bone; no apoptosis, no remodeling. The authors report co-localization of osteocyte apoptosis and endocortical remodeling following ovariectomy predominately in the posterior diaphyseal cortex in rats, but apoptosis preceded resorption by several days and when suppressed bone resorption was suppressed as well. Elegant work. —ES

◆Habermann B, Kafchitsas K, Olender G, Augat P, Kurth A. Strontium ranelate enhances callus strength more than PTH 1-34 in an osteoporotic rat model of fracture healing. *Calcif Tissue Int.* 2009 Dec 4. [Epub ahead of print] [[Abstract](#)]

This paper compares strontium ranelate (SR) and PTH(1-34) administration in OVX fracture healing. SR was given at 600 mg/kg/day, which gave serum levels similar to a

clinical dose. The PTH dose given was 20 µg 3 x per week. At 28 days only treatment with SR led to a significant increase in callus resistance compared to the OVX control rats, whereas both PTH and SR increased the bone volume/tissue volume ratio of the callus. The callus tissue volume increase induced by SR was significant, contrary to the changes induced by PTH. Callus in SR-treated animals was more resistant to torsion compared with OVX control rats. Unfortunately we will have to wait for another report for mechanistic information. Were the SR effects anabolic? Was the PTH-treated group more remodeled and therefore the callus was already smaller, or was there an increase in cartilaginous callus via PTH? —DGL

- ◆Kolios L, Hoerster AK, Sehmisch S, Malcherek MC, Rack T, Tezval M, Seidlova-Wuttke D, Wuttke W, Stuermer KM, Stuermer EK. Do estrogen and alendronate improve metaphyseal fracture healing when applied as osteoporosis prophylaxis? *Calcif Tissue Int.* 2009 Dec 1. [Epub ahead of print] [\[Abstract\]](#)

A nice model of stabilized metaphyseal fractures in OVX rats. This study examined bone formation indices throughout healing, as well as a radiographic architectural technique (µCT would be better) and mechanical testing. In summary, OVX resulted in poorer results than sham. Estrogen supplementation reversed the effects of OVX (as one would expect?) but alendronate treatment before and after did not show great improvements at 5 weeks. Other studies have shown improvements with bisphosphonates but this model may be more relevant. A longer follow-up would undoubtedly show the enlarged calluses seen in other studies. No negative effects on healing were seen with alendronate. Most metaphyseal fractures heal in clinical practice. An interesting aside would be to see if long-term high dose alendronate treatment did interfere with this fracture model, or if adding steroids also leads to a decrement in healing as reported by Odvina et al. —DGL

- ◆Li YF, Luo E, Feng G, Zhu SS, Li JH, Hu J. Systemic treatment with strontium ranelate promotes tibial fracture healing in ovariectomized rats. *Osteoporos Int.* 2009 Dec 3. [Epub ahead of print] [\[Abstract\]](#)

Metaphyseal osteotomies in OVX rats were treated with SR or nothing. The ultimate load was increased by 211.0% and 61.4% ($p < 0.01$), and the total bone volume of callus by 74.8% and 79.3% ($p < 0.01$) at 4 and 8 weeks post-fracture, respectively. SR treatment also promoted healing progress with increased osteogenesis at 4 weeks. If these results can be replicated in larger animals SR could be a candidate for bone healing. The results presented are strikingly large. —DGL

- ◆Roberts MD, Santner TJ, Hart RT. Local bone formation due to combined mechanical loading and intermittent hPTH-(1-34) treatment and its correlation to mechanical signal distributions. *J Biomech.* 2009 Nov 13;42(15):2431-8. [\[Abstract\]](#)

Another paper looking at interactions of PTH treatment and mechanical loading. Previously PTH has been shown to act at least partially through SOST, implicating a mechanosensory apparatus effect. In this careful study, objective comparison of bone apposition patterns was made to a library of candidate mechanical signals. Candidate mechanical signals were generated with the assistance of finite element analysis. Four-point bending treatment plus or minus PTH(1-34) treatment were compared to look for synergy by examining the magnitude and distribution of bone formation at the endocortical surface. PTH and bending results were found to be additive, but not synergistic. This article speaks somewhat against PTH exerting its effects solely via a mechanosensitive apparatus. —DGL

Clinical Studies and Drug Effects

- ◆Imaz I, Zegarra P, González-Enríquez J, Rubio B, Alcazar R, Amate JM. Poor bisphosphonate adherence for treatment of osteoporosis increases fracture risk: systematic review and meta-analysis. *Osteoporos Int*. 2009 Dec 5. [Epub ahead of print] [\[Abstract\]](#)

The main result of this meta-analysis based on 5-6 informative studies totaling more than 200,000 patients treated with bisphosphonates and followed for one year or more confirms that in clinical practice adherence is suboptimal, with a pooled persistence mean of half a year, a medication possession ratio (MPR) mean of 67%, and a 46% increased risk of fractures, particularly clinical vertebral fractures, in non-compliant (<80% MPR) vs. compliant patients. —SF

Genetics

- ◆Choi JW, Sutor SL, Lindquist L, Evans GL, Madden BJ, Bergen HR 3rd, Hefferan TE, Yaszemski MJ, Bram RJ. Severe osteogenesis imperfecta in cyclophilin B-deficient mice. *PLoS Genet*. 2009 Dec;5(12):e1000750. [\[Abstract\]](#)

It is now clear that osteogenesis imperfecta (OI) can be caused not only by mutations in the collagen 1 alpha 1 and 2 chain genes, but also in the enzymatic complex that is necessary to process collagen into fully mature fibrils. Hence mutations in LEPRE1 and CRTAP in mice and/or humans also cause OI. This study now shows that mice lacking cyclophilin B, a third partner of the collagen hydroxylation enzyme complex, also exhibit phenotypic features of OI. —SF

Molecular and Cell Biology

- ◆Hikiji H, Ishii S, Yokomizo T, Takato T, Shimizu T. A distinctive role of the leukotriene B4 receptor BLT1 in osteoclastic activity during bone loss. *Proc Natl Acad Sci U S A*. 2009 Dec 15;106(50):21294-9. [\[Abstract\]](#) [\[Full Text\]](#)

Leukotriene B4 (LTB4) is a potent mediator of inflammation. Both LTB4 and its high-affinity receptor BLT1 were produced by osteoclasts. Trabecular BV/TV at the distal femur was similar in mice deficient for BLT1 and control mice, however, BLT1 KO mice were resistant to the decrease of BV/TV following systemic inflammation (induced by LPS) or ovariectomy. The latter observation adds to the evidence that inflammatory/immune mediators are involved in estrogen deficiency-induced bone loss. —SF

- ◆Koizumi K, Saitoh Y, Minami T, Takeno N, Tsuneyama K, Miyahara T, Nakayama T, Sakurai H, Takano Y, Nishimura M, Imai T, Yoshie O, Saiki I. Role of CX3CL1/fractalkine in osteoclast differentiation and bone resorption. *J Immunol*. 2009 Dec 15;183(12):7825-31. [\[Abstract\]](#)

Beyond the overwhelming role of the OPG/RANKL/RANK system, the search for coupling factors between osteoblasts and osteoclasts continues. This study demonstrates that osteoblasts express the chemokine CX3CL1 in close contact with osteoclast precursors, and induces migration and adhesion of osteoclast precursors expressing the CX3CR1 receptor. Blocking antibodies against CX3CL1 inhibited osteoclast differentiation in vitro, and reduced osteoclast and erosion surfaces in vivo, providing a rationale for a new therapeutic approach. —SF

- ◆Li H, Xie H, Liu W, Hu R, Huang B, Tan YF, Liao EY, Xu K, Sheng ZF, Zhou HD, Wu XP, Luo XH. A novel microRNA targeting HDAC5 regulates osteoblast differentiation in mice and contributes to primary osteoporosis in humans. *J Clin Invest*. 2009 Dec;119(12):3666-77. [\[Abstract\]](#)

This could be the most brilliant and innovative study of the year in the bone field. Cell development and functions are not only regulated by transcriptional activators and repressors, but also at the translational level, i.e., by repressing the translation of specific mRNAs into proteins. The outstanding experiments shown here indicate that a microRNA induced in osteoblasts by BMP2 in turn represses the synthesis of a factor (HDAC5) that normally promotes degradation of the osteoblast transcription factor RUNX2. Hence silencing this microRNA in mice increases HDAC5 and decreases RUNX2 protein, leading to an inhibition of bone formation and bone mass. Most impressively, this study reports on 2 subjects with juvenile osteoporosis caused by an inactivating mutation of this microRNA. —SF

- ◆Rached MT, Kode A, Silva BC, Jung DY, Gray S, Ong H, Paik JH, DePinho RA, Kim JK, Karsenty G, Kousteni S. FoxO1 expression in osteoblasts regulates glucose homeostasis through regulation of osteocalcin in mice. *J Clin Invest*. 2010 Jan;120(1):357-68.

The transcription factor FoxO1 is a major target for insulin signaling, which in turn negatively regulates insulin sensitivity in many cells and suppresses pancreatic β cell proliferation and function. On the opposite side, uncarboxylated osteocalcin has been shown to favor β cell proliferation, insulin secretion and sensitivity. This study shows that mice lacking the transcription factor FoxO1 in osteoblasts have higher osteocalcin levels and reduced osteocalcin carboxylation (by ESP). In turn these mice show hyperinsulinemia and increased insulin sensitivity, i.e., a phenotype opposite to that of osteocalcin KO mice. These experiments further delineate the osteoblast as a cell involved in glucose homeostasis. —SF

- ◆Sunters A, Armstrong VJ, Zaman G, Kypta RM, Kawano Y, Lanyon LE, Price JS. Mechano-transduction in osteoblastic cells involves strain-regulated, estrogen receptor alpha-mediated, control of IGF-1R sensitivity to ambient IGF, leading to PI3-K/AKT dependent, Wnt/LRP5 receptor-independent activation of beta-catenin signaling. *J Biol Chem*. 2009 Dec 30. [Epub ahead of print]

Some in vivo experiments have suggested that the Wnt-LRP5 signaling pathway is essential to the mechanotransduction response of the skeleton. This in vitro study further examines the signaling pathways involved in the osteoblastic response to a single strain. It confirms previous work of this group indicating that estrogen receptor α is implicated in this response by co-activating the IGF-1 receptor, and shows that β -catenin activation by strain is not inhibited by Dkk1, and hence is probably not mediated by Wnt-LRP5/Fzd in these cells, but rather directly mediated by activation of the PI3K/AKT pathway downstream of the IGF-1 receptor. —SF

Public Health

- ◆Berry SD, Kiel DP, Donaldson MG, Cummings SR, Kanis JA, Johansson H, Samelson EJ. Application of the National Osteoporosis Foundation Guidelines to postmenopausal women and men: the Framingham Osteoporosis Study. *Osteoporos Int*. 2010 Jan;21(1):53-60. [\[Abstract\]](#)

- ◆Dawson-Hughes B, Looker AC, Tosteson AN, Johansson H, Kanis JA, Melton LJ 3rd. The potential impact of new National Osteoporosis Foundation guidance on treatment patterns. *Osteoporos Int*. 2010 Jan;21(1):41-52. [\[Abstract\]](#)

These two studies evaluate the proportion of women and men in the US who would be candidates for osteoporosis treatment based on the 2008 NOF guidelines incorporating the FRAX[®] tool to estimate fracture probability. Hence subjects with a FRAX[®] probability of fractures $\geq 20\%$ ($\geq 3\%$ for hip fractures only) should be considered for treatment, in addition to those with previous fragility fractures and those with a BMD < -2.5 T-score at the spine or hip. In the Framingham cohort and the NHANES population, 40 to 50% of white Caucasian women and 17 to 20% of men above 50, and up to 86% of women and 50% of men above 75, would be considered for treatment according to these guidelines. —SF

- ◆ Shi N, Foley K, Lenhart G, Badamgarav E. Direct healthcare costs of hip, vertebral, and non-hip, non-vertebral fractures. *Bone*. 2009 Dec;45(6):1084-90. [\[Abstract\]](#)

The word osteoporosis triggers images of woman, a dowager's hump, vertebral fractures and trabecular bone loss. This perception was okay in the last century, not this one. The authors report the following burden in patients 50-64 years: non-hip, non-vertebral (NHNV, n=27,424), vertebral (n=3386) and hip (n=2423); ≥ 65 years: NHNV (n=40,960), vertebral (n=11,751) and hip (n=21,504). Adjusted mean first-year costs associated with hip, vertebral, and NHNV fractures were \$26,545, \$14,977, and \$9183 for the 50-64 age cohort, and \$15,196, \$6701, and \$6106 for patients ≥ 65 years. Taking prevalence into account, the proportion of the total fracture costs accounted for by NHNV, hip, and vertebral fractures was 66%, 21% and 13% for the 50-64 age cohort, and 36%, 52% and 12% for the ≥ 65 age cohort. This is no joke. There is little evidence for anti-fracture efficacy against NHNV fractures. Few studies have these fractures as a primary endpoint, most do not show efficacy at all and when reported the risk reduction is about 20-30 percent, about half that observed for vertebral fracture risk reduction. We are looking in the wrong place in the 21st century. —ES

Reviews, Perspectives and Editorials

- ◆ Jones DC, Glimcher LH. Regulation of bone formation and immune cell development by schnurri proteins. *Adv Exp Med Biol*. 2010;658:117-22. [\[Abstract\]](#)
- ◆ Nakchbandi IA, van der Merwe SW. Current understanding of osteoporosis associated with liver disease. *Nat Rev Gastroenterol Hepatol*. 2009 Nov;6(11):660-70. [\[Abstract\]](#)
- ◆ Schneider MR, Sibilio M, Erben RG. The EGFR network in bone biology and pathology. *Trends Endocrinol Metab*. 2009 Dec;20(10):517-24. [\[Abstract\]](#)
- ◆ Takayanagi H. Osteoimmunology and the effects of the immune system on bone. *Nat Rev Rheumatol*. 2009 Dec;5(12):667-76. [\[Abstract\]](#)
- ◆ Takayanagi H. The unexpected link between osteoclasts and the immune system. *Adv Exp Med Biol*. 2010;658:61-8. [\[Abstract\]](#)
- ◆ Zanotti S, Canalis E. Notch and the skeleton. *Mol Cell Biol*. 2009 Dec 7. [Epub ahead of print]

Other Studies of Potential Interest

- ◆ Apel PJ, Crane D, Northam CN, Callahan M, Smith TL, Teasdall RD. Effect of selective sensory denervation on fracture-healing. An experimental study of rats. *J Bone Joint Surg Am*. 2009 Dec;91(12):2886-95. [\[Abstract\]](#) [\[Full Text\]](#)

◆Archer DF, Pinkerton JV, Utian WH, Menegoci JC, de Villiers TJ, Yuen CK, Levine AB, Chines AA, Constantine GD. Bazedoxifene, a selective estrogen receptor modulator: effects on the endometrium, ovaries, and breast from a randomized controlled trial in osteoporotic postmenopausal women. *Menopause*. 2009 Nov-Dec;16(6):1109-15. [\[Abstract\]](#)

- Also read other related papers on bazedoxifene effects on the reproductive tract and mammographic breast density in the same issue.

◆Brownstein CA, Zhang J, Stillman A, Ellis B, Troiano N, Adams DJ, Gundberg CM, Lifton RP, Carpenter TO. Increased bone volume and correction of HYP mouse hypophosphatemia in the *Klotho/HYP* mouse. *Endocrinology*. 2009 Dec 1. [Epub ahead of print] [\[Abstract\]](#)

◆Cruzoé-Souza M, Sasso-Cerri E, Cerri PS. Immunohistochemical detection of estrogen receptor beta in alveolar bone cells of estradiol-treated female rats: possible direct action of estrogen on osteoclast life span. *J Anat*. 2009 Dec;215(6):673-81. [\[Abstract\]](#)

◆Dobnig H, Stepan JJ, Burr DB, Li J, Michalská D, Sipos A, Petto H, Fahrleitner-Pammer A, Pavo I. Teriparatide reduces bone microdamage accumulation in postmenopausal women previously treated with alendronate. *J Bone Miner Res*. 2009 Dec;24(12):1998-2006. [\[Abstract\]](#)

◆Hikiji H, Ishii S, Yokomizo T, Takato T, Shimizu T. A distinctive role of the leukotriene B4 receptor BLT1 in osteoclastic activity during bone loss. *Proc Natl Acad Sci U S A*. 2009 Dec 15;106(50):21294-9. [\[Abstract\]](#) [\[Full Text\]](#)

◆Inose H, Ochi H, Kimura A, Fujita K, Xu R, Sato S, Iwasaki M, Sunamura S, Takeuchi Y, Fukumoto S, Saito K, Nakamura T, Siomi H, Ito H, Arai Y, Shinomiya KI, Takeda S. A microRNA regulatory mechanism of osteoblast differentiation. *Proc Natl Acad Sci U S A*. 2009 Dec 8;106(49):20794-99. [\[Abstract\]](#) [\[Full Text\]](#)

◆Ji JD, Park-Min KH, Shen Z, Fajardo RJ, Goldring SR, McHugh KP, Ivashkiv LB. Inhibition of RANK expression and osteoclastogenesis by TLRs and IFN-gamma in human osteoclast precursors. *J Immunol*. 2009 Dec 1;183(11):7223-33. [\[Abstract\]](#)

◆Kendler DL, Ringe JD, Ste-Marie LG, Vrijens B, Taylor EB, Delmas PD. Risedronate dosing before breakfast compared with dosing later in the day in women with postmenopausal osteoporosis. *Osteoporos Int*. 2009 Nov;20(11):1895-902. [\[Abstract\]](#)

◆Kim MH, Ryu SY, Choi JS, Min YK, Kim SH. Saurolectam inhibits osteoclast differentiation and stimulates apoptosis of mature osteoclasts. *J Cell Physiol*. 2009 Dec;221(3):618-28. [\[Abstract\]](#)

◆Lee Y, Ha J, Kim HJ, Kim YS, Chang EJ, Song WJ, Kim HH. Negative feedback inhibition of NFATc1 by DYRK1A regulates bone homeostasis. *J Biol Chem*. 2009 Nov 27;284(48):33343-51. [\[Abstract\]](#) [\[Full Text\]](#)

◆Li H, Xie H, Liu W, Hu R, Huang B, Tan YF, Liao EY, Xu K, Sheng ZF, Zhou HD, Wu XP, Luo XH. A novel microRNA targeting HDAC5 regulates osteoblast differentiation in mice and contributes to primary osteoporosis in humans. *J Clin Invest*. 2009 Dec;119(12):3666-77. [\[Abstract\]](#)

◆Lu C, Xing Z, Yu YY, Colnot C, Miclau T, Marcucio RS. Recombinant human bone morphogenetic protein-7 enhances fracture healing in an ischemic environment. *J Orthop Res*. 2009 Nov 13. [Epub ahead of print] [\[Abstract\]](#)

- ◆Maass PG, Wirth J, Aydin A, Rump A, Stricker S, Tinschert S, Otero M, Tsuchimochi K, Goldring MB, Luft FC, Bähring S. A cis-regulatory site downregulates PTHLH in translocation t(8;12)(q13;p11.2) and leads to Brachydactyly Type E. *Hum Mol Genet.* 2010 Jan 4. [Epub ahead of print]
- ◆Potier E, Noailly J, Ito K. Directing bone marrow-derived stromal cell function with mechanics. *J Biomech.* 2009 Dec 2. [Epub ahead of print] [\[Abstract\]](#)
- ◆Sarahrudi K, Mousavi M, Thomas A, Eipeldauer S, Vécsei V, Pietschmann P, Aharinejad S. Elevated levels of macrophage colony-stimulating factor in human fracture healing. *J Orthop Res.* 2009 Nov 30. [Epub ahead of print] [\[Abstract\]](#)
- ◆Wang W, Lian N, Li L, Moss HE, Wang W, Perrien DS, Elefteriou F, Yang X. Atf4 regulates chondrocyte proliferation and differentiation during endochondral ossification by activating Ihh transcription. *Development.* 2009 Dec;136(24):4143-53. [\[Abstract\]](#)
- ◆Wilson SG, Jones MR, Mullin BH, Dick IM, Richards JB, Pastinen TM, Grundberg E, Ljunggren O, Surdulescu GL, Dudbridge F, Elliott KS, Cervino AC, Spector TD, Prince RL. Common sequence variation in FLNB regulates bone structure in women in the general population and FLNB mRNA expression in osteoblasts in vitro. *J Bone Miner Res.* 2009 Dec;24(12):1989-97. [\[Abstract\]](#)
- ◆Yu YY, Lieu S, Lu C, Miclau T, Marcucio RS, Colnot C. Immunolocalization of BMPs, BMP antagonists, receptors, and effectors during fracture repair. *Bone.* 2009 Nov 11. [Epub ahead of print] [\[Abstract\]](#)

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 reports no conflicts of interest.