BoneKEy-Osteovision. 2007 December;4(12):322-326
http://www.bonekey-ibms.org/cgi/content/full/ibmske;4/12/322
DOI: 10.1138/20070283

MEETING REPORT

Meeting Report from the 29th Annual Meeting of the American Society for Bone and Mineral Research

September 16-19, 2007 in Honolulu, Hawaii, USA

GENETICS OF HUMAN BONE DISEASES

Serge Ferrari

Division of Bone Diseases, WHO Collaborating Center for Osteoporosis Prevention, Department of Rehabilitation and Geriatrics, Geneva University Hospital, Geneva, Switzerland

The sessions dedicated to the genetics of bone diseases started with an outstanding symposium entitled "Novel Insights into Bone Metabolism through Genetics.**"** M. Whyte first reviewed disorders associated with mutations in the RANKL/OPG-RANK system. Juvenile Paget's Disease, JPD (also known as familial hyperphosphatasia, OMIM No #239000), is a recessive disorder caused by OPG (*TNFRSF11B* gene) null mutations. The skeletal deformities and high bone turnover characteristic of this disorder may be complicated by vascular calcifications, as seen in the OPG null mouse, manifest as retinal exudate and bleeding. As an alternative to bisphosphonates, recombinant OPG is a promising new therapy for these patients. In a few JPD cases, however, OPG mutations were not found. RANK (*TNFRSF11A* gene) mutations that affect the signal peptide region, resulting in increased signaling (activating mutations), cause familial expansive osteolysis, FEO (OMIM No #174810). Other constitutive RANK mutations were found in some rare cases of early-onset Paget's disease, PDB2 (OMIM No #602080). Most recently, Sobacchi *et al*. described RANKL (*TNFSF11* gene) loss-of-function mutations in an osteoclast-poor form of autosomal recessive osteopetrosis, OPTB2 (OMIM No #259710, see Not To Be Missed, BoneKEy, 2007 Aug;4(8):204-8). Of note, an abstract at the ASBMR meeting (1) reported no differences in RANKL gene polymorphisms between subjects with/without sporadic Paget's disease, whereas OPG polymorphisms were reported to be associated with Paget's in this group.

E. Shore reviewed her own findings concerning the causes of fibrodysplasia ossificans progressiva, FOP (OMIM No #135100), and progressive osseous heteroplasia, POH (OMIM No #166350). These two disorders are characterized by new bone formation independent of the skeleton, *i.e.*, new bone at the wrong place and at the wrong time: endochondral ossification in FOP, intramembranous ossification that occurs in the skin in POH. They are autosomal dominant disorders with a frequency of about 1/million individuals that in the case of FOP can be inherited from either the father or the mother, whereas POH is inherited exclusively from the father. From a phenotypic point of view, these two disorders differ by the presence of big toe malformation in FOP but not in POH. Linkage approaches revealed that FOP is due to mutations in the type 1 BMP receptor (*ACVR1* gene, see S. Ferrari, BoneKEy, 2006 December;3(12):11-29). In contrast, linkage approaches have been difficult in POH due to the small number of known cases (about 50). The responsible gene has nevertheless been identified by a candidate gene approach based on the phenotypic similarities that may exist between POH, Albright's hereditary osteodystrophy, AHO, and pseudohypoparathyroidism PHP1a. In two-thirds of POH cases, this revealed heterozygous mutations of the G protein α s (*GNAS*) allele that is paternally-derived (XL-

α-s protein). However, how this mutation and/or paternal imprinting explains the restricted expression of the mutant protein in the skin remains unclear.

J. Marini reviewed autosomal recessive, mostly lethal forms of osteogenesis imperfecta (OI) characterized by white (normal) sclerae, small head circumference, and a round face. In 10 to 15% of OI cases, *Col1A1* and *Col1A2* mutations are not found. In OI type VII and IIB, for instance, (OMIM No #610682 and #610854), homozygous and compound mutations were found in a cartilage-associated protein (*CRTAP* gene) that shares 53% homology and forms a complex with prolyl 3-hydroxylase 1 (*P3H1* gene, also known as leprecan, *LEPRE1* gene). This complex is responsible for hydroxylation of the pro986 residue of Col1A1 necessary for the assembly of the triple helix. A mouse model of the *CTRAP* null mouse published by Morello *et al*. (2) demonstrates that this protein is expressed in chondrocytes at the chondro-osseous junction. Interestingly hypomorphic (non-lethal) mutations do exist in both *CRTAP* and *LEPRE1*, the latter found in families of African origin with OI type VIII.

OI cases with unusual presentations were reported by a group in Sweden (3), the most interesting being three girls with multiple fractures but high BMD (+3SD) by DXA ("dense OI" according to the authors) and normal bone turnover markers. In all three cases, novel *Col1A1* or *Col1A2* mutations were found, in particular at the splice site of the C-terminal peptide. The major limitation of this work was the fact that the pathogenesis of the identified mutations was not directly demonstrated whereas concomitant (HBM) mutations in other genes were not ruled out. Also related to OI was a very important communication (4) reviewing experience with IV pamidonate in children with OI in relation to the potential risk of osteonecrosis of the jaw (ONJ). As a reminder, dentinogenesis imperfecta is also often present in these children who therefore require a number of dental procedures during growth. Despite follow-ups of up to 10 years in some children, extraction sockets were never found to be complicated by ONJ.

A group in France (5) reported its experience with the highly variable response to treatment with active vitamin D (1α vitamin D, 1-2 microg/d) and phosphorus in a large series of patients (n=81) with X-linked hypophosphatemic rickets (XLH). Their results suggested that genetic variation in the vitamin D receptor (*VDR*), in particular a haplotype that they identified as Hap1, influenced the height and Ca-Pi metabolic response to treatment. Dr. M. Whyte, however, pointed out that the outcome of XLH is primarily influenced by the type of *PHEX* mutations that are present, *i.e.*, deactivating vs. non-deactivating mutations.

Moving from monogenic disorders to osteoporosis genetics, *LRP5* gene polymorphisms previously described to be associated with BMD and/or fractures in several independent cohorts were analyzed in relation to these phenotypes in an extraordinarily large collection of participants (n=37,000) from both European and US cohorts (6). This prospective meta-analysis at the participant level confirmed previous findings in that *LRP5* missense SNPs in exons 9 and 18 were associated with spine and femur aBMD and fracture risk, with a maximal 26% increase in risk of vertebral fracture per 667M allele. Another abstract (7) reminded us that heterozygous carriers of *LRP5* OPPG mutations have a lower BMD and a higher prevalence of vertebral fractures than controls, further suggesting that heterozygous *LRP5* mutations should be suspected whenever osteoporosis is detected in middle-aged subjects. This study also indicated a higher prevalence of diabetes and hypercholesterolemia in these subjects, thereby providing a possible molecular clue to explain the association of these disorders with osteoporosis.

An interesting approach to predict osteoporotic fractures using *Col1A1* polymorphisms was taken by Australian investigators (8). They developed models that indicated the TT genotype of *Col1A1* (5% of the population) to more than double

the risk of any fracture at all ages independently of femur neck BMD, so that homozygozity for the T allele would be equivalent to +20 yrs of age or -1SD of BMD. A new candidate gene to be associated with BMD is *FNLB*, filamin B, mutations in which cause osteochondrodysplasia (9). This gene is in a region previously mapped for linkage with osteoporosis (3p14) by one group in England. Now the authors report association of *FNLB* SNPs with BMD in two independent cohorts, *i.e.*, they were able to replicate the association they found. Large-scale genotyping methods to simultaneously analyze thousands of SNPs were presented. By analyzing 113 SNPs in 54 selected genes with microarray-based techniques, sex-specific associations with BMD were reported, such as *LRP5* and *ESR2* in men, and *IL6* and *ESR1* in women (10). A genome-wide association study (GWA) using the 100k SNP gene chip method found numerous SNPs associated with BMD and bone geometry at the hip (11). However virtually no SNP shared association with BMD and bone geometric indices, such as femoral neck width, suggesting that these traits are determined by separate genes. A region-wide association study based on 200 tag SNPs covering 6Mb on chromosome 3p21 identified a voltage-dependent calcium channel subunit, CACNA2D2, to be associated with BMD in Chinese subjects (12).

Negative reports are sometimes also useful. By studying genetic variation in the PTH system (i.e., *PTH*, *PTHrP*, *PTH1R* and *PTH2R* genes) in a prospective study of 1000 elderly women, OPRA investigators found no association with BMD, fractures and/or PTH levels, except between some *PTH* haplotypes and fracture risk (13).

A major component of skeletal strength, besides bone mineral density, is the microarchitecture of cortical and trabecular bone. Using high-resolution pQCT, the heritability (h2, %) of human bone microstructure was evaluated at the distal radius and tibia in more than 100 mother-daughter pairs (14). H2 was generally high and similar to h2 for aBMD. However, some heritability estimates decreased sharply past the menopause, suggesting that intense bone remodeling may overcome additive genetic effects on peak bone microstructure. Also, interestingly, the mean heritability of trabecular and cortical microarchitecture was near 50% once adjusted for body size and BMD at the same site, adding to the evidence that genetic effects may specifically affect bone structure.

Although reviewing the plethora of abstracts on mouse genetics is beyond the scope of this summary, it is worth mentioning an interesting approach that used the Haplotype Association Mapping (HAM) program to extract information from SNPs and BMD in 18-week female mice from 30 mouse strains (15). This technique identified a positional candidate gene, *CER1*, in which a non-synonymous SNP was later found to be associated with low BMD in 1000 Chinese premenopausal women as well. Finally, one of the most interesting abstracts in this field (16) identified a highly polymorphic region in the 3'-UTR of the *Ppar*γ gene by comparing sequences from B6 and C3H mice. Studies in congenic mice revealed that this region regulates the *Ppar*γ transcription level in response to a high fat diet. Moreover, SNPs in the human synthenic region of mouse *Ppar* 3'-UTR were also reported to interact with dietary fat in association with BMD.

Conflict of Interest: The author reports receiving consulting and speaker fees, as well as research grants, from Amgen, and consulting and speaker fees from Merck, the Alliance for Better Bone Health, and Eli Lilly Switzerland**.**

References

1. Beyens G, de Freitas F, Vanhoenacker F, Verbruggen L, Zmierczak HG, Westhovens R, Van Offel J, Devogelaer J, Van Hul W. No role for polymorphisms in the gene encoding RANKL in the development of sporadic Paget's disease of bone in contrast to some OPG polymorphisms. *J Bone Miner Res*. 2007 Sep;22(Suppl 1):S287. [Abstract]

2. Morello R, Bertin TK, Chen Y, Hicks J, Tonachini L, Monticone M, Castagnola P, Rauch F, Glorieux FH, Vranka J, Bächinger HP, Pace JM, Schwarze U, Byers PH, Weis M, Fernandes RJ, Eyre DR, Yao Z, Boyce BF, Lee B. CRTAP is required for prolyl 3- hydroxylation and mutations cause recessive osteogenesis imperfecta. *Cell*. 2006 Oct 20;127(2):291-304.

3. Lindahl K., Brändström H, Kindmark A, Ljunggren Ö. Sequencing of collagen I in three patients with multiple fractures of unknown aetiology reveals novel mutations causing osteogenesis imperfecta with normal to high BMD. *J Bone Miner Res*. 2007 Sep;22(Suppl 1):S55. [Abstract]

4. Chahine CJ, Cheung M, Head TW, Schwartz S, Rauch F, Glorieux FH. Extraction socket healing in pediatric patients treated with pamidronate: a retrospective chart review. *J Bone Miner Res*. 2007 Sep;22(Suppl 1):S56. [Abstract]

5. Jehan F, Nguyen M, Walrant-Debray O, Sinding C, Garabedian M. Growth and response to active vitamin D and phosphorus treatment is highly associated with haplotype of the vitamin D receptor promoter in X-linked hypophosphatemic rickets. *J Bone Miner Res*. 2007 Sep;22(Suppl 1):S55. [Abstract]

6. van Meurs JBJ, Trikalinos TA, Ralston SH, Rivadeneira F, Ioannidis JPA, Uitterlinden AG, The Genomos Study Group. Large-scale analysis of association between polymorphisms in the LRP-5 and -6 genes and bone mineral density and fracture. *J Bone Miner Res*. 2007 Sep;22(Suppl 1):S56. [Abstract]

7. Saarinen A, Saukkonen T, Somer M, Laine CM, Toiviainen-Salo S, Lehesjoki A, Mäkitie O. LRP5 mutations are associated with osteoporosis, impaired glucose tolerance and hypercholesterolemia. *J Bone Miner Res*. 2007 Sep;22(Suppl 1):S112.

8. Tran BH, Nguyen ND, Frost SA, Center JR, Eisman JA, Nguyen TV. Enhancement of prognosis of osteoporotic fractures by genetic marker: contribution of the collagen I alpha 1 gene. *J Bone Miner Res*. 2007 Sep;22(Suppl 1):S57. [Abstract]

9. Wilson SG, Mullin BH, Jones MR, Dick IM, Dudbridge F, Spector TD, Prince RL. Variation in the FLNB gene regulates bone density in two populations of Caucasian women. *J Bone Miner Res*. 2007 Sep;22(Suppl 1):S57. [Abstract]

10. Gennari L, Tejedor D, Merlotti D, De Paola V, Cadaval A, Martini G, Valleggi F, Franci B, Campagna S, Lucani B, Simon L, Martinez A, Nuti R. Identification of single nucleotide polymorphisms (SNPs) contributing to bone mineral density and fractures: a DNA microarray based study. *J Bone Miner Res*. 2007 Sep;22(Suppl 1):S111.

11. Karasik D, Cho YK, Cupples LA, Kiel DP, Demissie S. Genome-wide measure of pleiotropy among osteoporosis-related traits in the Framingham Study. *J Bone Miner Res*. 2007 Sep;22(Suppl 1):S111.

12. Huang Q, Cheung C, Fong P, Ng M, Mak W, Chan V, Sham P, Kung A. Identification of the CACNA2D2 gene on chromosome 3p21 as a novel susceptibility gene for osteoporosis. *J Bone Miner Res*. 2007 Sep;22(Suppl 1):S185. [Abstract]

13. Tenne M, Gerdhem P, Luthman H, Åkesson K, Mc Guigan F. Relationship between variation in the PTH, PTH related peptide and the PTH receptors 1 and 2 genes and BMD and fracture in elderly women. *J Bone Miner Res*. 2007 Sep;22(Suppl 1):S287. [Abstract]

14. Ferrari SL, Chevalley T, Bonjour JP, Rizzoli R. Heritability of bone

microstructure in women. *J Bone Miner Res*. 2007 Sep;22(Suppl 1):S68. [Abstract]

15. Cheung CL, Tang PL, Sham PC, McClurg P, Chan S, Smith DK, Su AI, Cheah KS, Kung AW, Song Y. Genome-wide haplotype association mapping (HAM) in mice leads to an identification of a genetic variant in CER1 associated with bone mineral density in premenopausal women. *J Bone Miner Res*. 2007 Sep;22(Suppl 1):S283. [Abstract]

16. Ackert-Bicknell CL, Graber J, Cho K, Demissie S, Ordovas J, Beamer WG, Kiel DP, Rosen CJ. An interaction between dietary fat and polymorphisms in a previously undetected, alternate 3' UTR of the PPARG gene influences bone mass in both mice and humans. *J Bone Miner Res*. 2007 Sep;22(Suppl 1):S186. [Abstract]