The brains of the bones: how osteocytes use WNT1 to control bone formation

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WNT proteins drive the development and maintenance of many tissues, including bone. It is less clear which of the many WNT proteins act on bone or where these WNTs act in the skeleton; however, loss-of-function mutations in *WNT1* cause bone fragility in children and adults. In this issue of the *JCI*, Joeng and colleagues demonstrate that bone formation is under the control of WNT1 produced by osteocytes, the cells that reside deep in the bone matrix and form dendritic networks. The implication of WNT1 in the control of bone formation identifies a potential new target for the treatment of low bone mass disorders, such as osteoporosis.

Which WNT, where?

A role for WNT signaling in bone development and maintenance was first discovered through the study of osteoporosis pseudoglioma syndrome, an ultra-rare disease that is characterized by blindness, low bone mass, and frequent fractures in children (1). This disorder is caused by homozygous loss-of-function mutations in the gene encoding LDL receptor–related protein 5 (LRP5), which acts as a coreceptor for WNT ligands on the surface of osteoblasts, the cells responsible for bone formation. When WNT ligands bind to LRP5, bone formation is stimulated; therefore, mutations that inactivate LRP5 lead to slow bone formation and low bone mass.

This basic machinery was discovered more than 15 years ago, but it is still unclear which of the many WNT proteins play an important role in human bone. Studies in children with another rare bone fragility disorder provided an answer. Specifically, homozygous inactivating mutations in *WNT1* lead to a severe form of recessive osteogenesis imperfecta that is characterized by frequent fractures in early childhood and, in some patients, structural brain abnormalities (2–5). Approximately 20 individuals with homozygous

WNT1 mutations have been reported to date. Bone and brain phenotypes similar to those seen in these children are also present in the swaying mouse, which has a naturally occurring *Wnt1* mutation (6). Heterozygous *WNT1* mutations give rise to less severe bone fragility and manifest as early-onset osteoporosis in adolescents or young adults (2, 4).

Mouse studies have indicated that the WNT1-expressing bone cells are osteocytes, rather than bone-forming osteoblasts or bone-resorbing osteoclasts (2). Osteocytes are former osteoblasts that have become embedded in mineralized bone matrix (7), where they maintain contact with each other, and with cells on the bone surface, via canalicular canals through which they project their dendrites. This canalicular system also allows for a remarkably close contact between osteocytes and bone matrix: the combined length of canaliculi in a cubic centimeter of mineralized bone amounts to 74 km, with 80% of the bone matrix located within 3 μm of the nearest canaliculus (8). It is thought that osteocytes monitor the deformation of bone matrix that results from the exposure of the skeleton to mechanical forces and thus play an essential role

in the biological adaptation of bones to mechanical stimuli (7). More generally, the osteocyte system is ideally positioned for integrating input from local and systemic factors and for orchestrating teams of effector cells (osteoblasts and osteoclasts) that are recruited to carry out the resulting action plan.

WNT1 in osteocytes

In this issue, Joeng et al. set out to explore the mechanism whereby WNT1 determines bone mass (9). Previous observations from this group established that global loss of WNT1 causes low bone mass and fractures, but left open the question as to whether this phenotype was explained by the absence of functional WNT1 specifically in osteocytes. As WNT1 is expressed in many cell types and tissues, such as B lymphocytes and brain, it was still possible that the observed effects on the skeleton were a secondary consequence of hematologic or neurologic abnormalities. Joeng and colleagues have now shown that targeted inactivation of *Wnt1* in osteocytes replicates the skeletal phenotype of animals with global WNT1 deficiency, including impaired bone formation, extremely low bone mass, and spontaneous fractures. In contrast, WNT1 overexpression in osteocytes markedly increased bone formation and resulted in more trabecular and cortical bone. Neither WNT1 inactivation nor overexpression seemed to have a major effect on bone resorption. Together, these experiments indicated that WNT1 production by osteocytes is essential for the control of bone formation and bone mass.

Further mechanistic studies implicated an mTOR pathway in the control of osteoblasts through WNT1 (9). Overexpression of WNT1 in a stromal cell line accelerated osteoblast differentiation and increased mineralization in vitro, and this effect was diminished by rapamycin, which inhibits mTOR complex 1 (mTORC1) signaling. Accordingly, rapamycin treatment of mice overexpressing WNT1 in osteocytes allevi-

ated the high-bone-mass phenotype. Conversely, genetic stimulation of mTORC1 signaling in osteocytes of the swaying mouse increased bone mass and prevented spontaneous fractures. Thus, mTORC1 signaling at least in part mediates the effect of WNT1 signaling on osteoblasts.

Finally, Joeng et al. showed that anti-sclerostin antibody treatment of the swaying mouse increased bone mass, improved mechanical bone properties, and decreased the fracture rate (9). These observed improvements in the swaying mouse following anti-sclerostin administration are significant on both mechanistic and translational levels. Sclerostin is an osteocyte-secreted protein that decreases WNT signaling and thereby inhibits bone formation. It does so by interacting with LRP5 on the surface of osteoblasts, which prevents the binding of WNT ligands to LRP5. Anti-sclerostin antibody treatment therefore has bone anabolic properties. The fact that the anabolic effect of anti-sclerostin therapy is still seen in the absence of WNT1 indicates that other WNT ligands are available in the bone environment to signal through LRP5. Potential candidates for such a compensatory mechanism include WNT7b and WNT10b, which both enhance bone formation and interact with LRP5 (10, 11). Nevertheless, WNT1 seems to be the predominant WNT ligand in this context, as the results by Joeng et al. show that anti-sclerostin treatment is unable to completely restitute bone mass in WNT1-deficient mice (9).

From a translational perspective, the preclinical studies by Joeng et al. suggest that treatment with anti-sclerostin antibody is worth exploring as a potential therapeutic strategy for bone fragility caused by *WNT1* mutations. At present, antiresorptive therapy with bisphosphonates is the most widely used approach for treating bone fragility disorders. Unfortunately, this approach is not very effective in patients with *WNT1* mutations (4, 12). Anti-sclerostin antibody treatment may thus become a more effective treatment option in the presence of *WNT1* mutations. Another bone anabolic approach is

treatment with teriparatide (parathyroid hormone 1-34), which decreases sclerostin expression in osteocytes (7). Interestingly, recent pilot studies showed encouraging results with teriparatide treatment of adults who had osteoporosis caused by heterozygous *WNT1* mutations (13).

Future directions

In their series of elegant experiments, Joeng et al. have established that WNT1 production in osteocytes plays a key role in the control of bone formation and bone mass. This leads to the question of what drives WNT1 production in osteocytes under physiological conditions. Is WNT1 part of the mechanostat machinery, the putative system whereby osteocytes adapt bone strength to local mechanical requirements, or is osteocytic WNT1 under the control of systemic hormones?

The results by Joeng et al. also make WNT1 a potential player in bone disorders characterized by dysfunctional osteocytes and abnormal bone mass. For example, in X-linked hypophosphatemic rickets, which is caused by phosphate-regulating endopeptidase homolog, X-linked (*PHEX*) mutations, the bone matrix surrounding osteocytes is hypomineralized, but overall bone mass is paradoxically increased (14). In osteogenesis imperfecta, which is caused by mutations in collagen type I–encoding genes, osteocyte density is conspicuously increased, and bone mass is typically very low (15). Disordered WNT1 signaling might contribute to the bone mass abnormalities in these conditions.

Finally, the results by Joeng and colleagues offer new perspectives for the treatment of common bone mass disorders, such as osteoporosis. Pharmacological interventions to target WNT1 production specifically in osteocytes or to modify WNT1-dependent signaling pathways in osteoblasts could become promising therapeutic avenues. Studies of extremely rare disorders thus may help treat common ones.

Acknowledgments

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