There is much excitement surrounding osteoprotegerin (OPG), its osteoprotegerin ligand (OPGL), and their role in bone metabolism. Bone growth and remodeling is a dynamic process, balancing bone matrix synthesis by osteoblasts and resorption of bone by multinucleated osteoclasts. Osteoprotegerin ligand, a member of the tumor necrosis factor receptor family of molecules, is a key regulator of bone remodeling through its key role in osteoclastogenesis.

OPGL was discovered by several groups, and was given different names depending on the reason behind its discovery: TRANCE [tumor necrosis factor (TNF) related activation induced cytokine], RANKL [receptor activator of nuclear factor-κB (RANK) ligand], osteoclast differentiation factor, and OPGL ligand for OPG. In addition to its essential role in the development and activation of osteoclasts, OPGL has been identified as an important costimulation molecule involved in T cell–dendritic cell communication and in dendritic cell survival. RANK is the downstream signaling receptor for OPGL. OPG is a soluble decoy receptor for OPGL, neutralizing its ability to bind with RANK and induce a signal. Due to its expression on activated T cells and its role in inflammation and bone loss, OPG has generated much interest in autoimmune disease research, in particular in inflammatory arthritis.

In this issue of The Journal, Masi and colleagues investigate the role of the OPG/OPGL system in modulating bone injury in children with juvenile idiopathic arthritis (JIA). They report an increased OPG/OPGL ratio in the peripheral blood of children with JIA compared to controls, as well as the association of the TT polymorphism of the OPG gene in children with JIA with lower bone mineral density. The authors suggest that increased OPG production in affected children may reflect a compensatory mechanism for overproduction of OPGL.

THE OPG/OPGL PATHWAY
OPGL is made by activated T cells and its expression is upregulated by many soluble factors affecting bone resorption, including the proinflammatory cytokines, interleukin-1 and TNF-α. T cells express a cell-surface membrane-bound OPGL that is cleaved by metalloproteinases into a soluble form. There may be some functional differences between membrane-bound and soluble OPGL, with cell-bound OPGL being more effective mediators of osteoclastogenesis when measured by in vitro assays. The presence of both soluble and membrane-bound forms of OPGL and their functional differences may explain the conflicting data from different groups. The method of quantitation of OPGL and the specimen used may also help address these differences. Investigators using ELISA assays on serum can only measure the soluble form, as in the report by Masi, et al. Those researchers using flow cytometry on single cell suspensions will only measure cell-bound forms of OPGL, and those using reverse transcription polymerase chain reaction (RT-PCR) to quantitate messenger RNA production will measure message precursors for both soluble and membrane-bound variants.

In the article presented in this issue, Masi, et al report that serum levels of OPGL protein, as measured by ELISA, in children with JIA are lower than those for healthy children. This would point to decreased osteoclastic activity in children with JIA compared to healthy children, a counterintuitive notion, given the body of literature demonstrating the increased risk and incidence of osteopenia in children with JIA. Data from Varsani, et al and our own data differ from those reported by Masi, in finding an increased level of OPGL in the peripheral circulation of children with JIA compared to healthy children. The critical difference is the method of quantitation of OPGL. In Varsani’s case, RT-PCR was used to measure OPGL mRNA, which represents precursors of both soluble and membrane-bound forms. The increase in OPGL in children with JIA is also seen when OPGL is assayed by flow cytometry, which measures cell-surface, membrane-bound forms of OPGL. These differences serve to highlight the need to further

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understand the biologic activity of membrane-bound versus soluble forms of OPGL to accurately interpret data within the context of disease and normal physiologic states.

Another important factor in the physiologic equation is the interactions of OPGL with its natural decoy receptor/neutralizer, OPG. Bone loss mediated by OPGL can be blocked by administration of OPG. OPG functions as a decoy receptor for OPGL, competing with RANK for binding with OPGL, effectively inhibiting osteoclastogenesis both in vitro and in vivo. Thus evaluation of OPG levels must go hand in hand with OPG levels, as the balance of the 2 will determine whether osteoclastic or osteoblastic activity dominates.

However, this evaluation is not a simple one-to-one calculation; OPG, although a member of the TNF receptor super family, is secreted as a 110 kDa homodimer, not a molecular trimer, as are others in this molecular family including its ligand OPGL. The report by Masi and colleagues shows that the ratio of OPG/OPGL is lower in the peripheral blood of children with JIA compared with healthy controls, again opposite to what one would expect in an osteopenic scenario.

The interaction of OPGL and OPG at the molecular level is not known. One way to assist in data interpretation is to link the determinations of OPGL and OPG with biologic activity, in this case osteoclastogenesis leading to bone loss, a biologically relevant measure of activity in the OPG/OPGL pathway. To that end, investigators have quantitated the presence of osteoclast precursors and their functional ability to resorb bone as a correlate to OPGL levels and the balance of activities between OPGL and OPG. Proof of principle would include neutralization of this OPGL-mediated biologic activity by exogenous OPG.

**OPGL, T CELLS, AND BONE LOSS**

T cells appear to be the link between inflammation and bone loss. Increasingly, the role of OPGL as a critical second signal involved in optimal T cell activation is recognized. T cell-derived OPGL can also regulate the development and activation of osteoclasts resulting in bone loss. This mechanism is true both for systemic and local activation of T cells. In Lewis rats, adjuvant arthritis is a T cell-dependent disease leading to joint inflammation and bone erosions. T cells from inflamed joints express OPGL. Although T cells appear to be important regulators of bone metabolism, mutant mice that lack T cells do not have abnormal bones or dentition. But under conditions of chronic systemic activation of T cells as in viral infections and multisystem autoimmune disease, or chronic local inflammation involving bone and joints such as in arthritis, bone remodeling via production of OPGL by T cells plays a more prominent role. IL-1β, IL-6, IL-11, IL-17, and TNF-α, proinflammatory cytokines that stimulate osteoclastogenesis, increase the expression of OPGL but decrease OPG expression.

In contrast, cytokines inhibiting osteoclastogenesis, such as IL-13, interferon-γ, and transforming growth factor-β1 suppress the expression of OPGL but enhance expression of its inhibitor OPG. Interestingly, glucocorticoids, widely used to treat multisystem autoimmune disease and arthritis, strongly induce OPGL expression and decrease expression of its decoy receptor, OPG. One of the well documented morbidities of corticosteroid use is bone loss.

In people with inflammatory arthritis, OPGL has also emerged as an important factor regulating bone loss. Synovial tissue from patients with rheumatoid arthritis and osteoarthritis have been shown to express high levels of OPGL. Similarly, patients with spondyloarthropathies have also been found to express high levels of OPGL in affected synovial tissue. There remains debate whether cartilage destruction occurs independently or is dependent on subchondral bone loss. In the adjuvant-induced rat arthritis model, both cartilage and bone are protected by administration of OPG, but in collagen-induced arthritis in rats, the protective effects of OPG were less in cartilage than in bone. In patients with erosive psoriatic arthritis, increased numbers of osteoclast precursors (OCP) were found in the peripheral blood. OCP numbers were decreased by administration of OPG and anti-TNF.

Although administration of OPG in animal models of arthritis is bone protective, inflammation and its systemic effects are not altered. Many cell types, both immune and non-immune, have now been found to express OPGL including lymphocytes, monocytes, macrophages, dendritic cells, synovial fibroblasts, and mammary gland epithelial cells of lactating females. Additionally, the downstream effects of inflammatory cytokines on the OPG/OPGL pathway may be the temporal answer to the apparent “disconnect” between inflammation and bone loss in arthritis in some situations. This may in part explain the lack of correlation between serum levels of members of the OPG/OPGL family and classic markers of inflammation in patients with arthritis. As reported by Masi, et al, there is no correlation between OPG or OPGL and common laboratory markers of inflammation such as erythrocyte sedimentation rate or C-reactive protein, but genetic control of OPGL expression may be related to local bone loss and generalized osteopenia.

The OPG/OPGL pathway is a key regulator of bone metabolism through its effect on development and activation of osteoclasts. In addition to its role in osteoclastogenesis, OPGL is a critical factor in the immune system, from regulating development of lymph nodes and Peyer’s patches to serving as an important costimulation molecule in optimal T cell activation and mediating dendritic cell survival. Beyond its role in inflammation and bone loss, OPGL appears to regulate skeletal calcium release and is crucial in the morphogenesis of the lactating mammary gland. This aspect of OPGL biology is only beginning to be understood in the context of disease and normal physiologic states.
elucidated and may have implications in the hormonal regulation of osteoporosis. These multifaceted functions serve to highlight the need for further work in understanding the molecular mechanisms responsible for the biologic effects of this intriguing family of molecules.

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