A Decade of Bisphosphonate Bone Complications: What It Has Taught Us About Bone Physiology

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While the AIDS epidemic of the 1980s taught the medical and dental professions much about immune cells and the immune system’s cellular relationships, the bisphosphonate-induced osteonecrosis epidemic of the past decade has taught these same professions much about bone turnover, bone cell cross talk, the response and functional relationship of bone cells to loading, and drug effects on cellular dynamic relationships. The present article explores the literature as well as both evidence- and experience-based data to discuss known bone pathologies and physiologic mechanisms as well as uncover new findings: (1) bone remodeling is the mechanism by which bone adapts to loading stresses, termed either bone modeling or Wolff’s law, and it is also the mechanism for bone renewal; (2) osteoclastic bone resorption triggers bone renewal at a rate of about 0.7%/day by its release of growth factors; (3) bisphosphonates prevent the renewal of old and injured bone, thus making it brittle and more likely to fracture over time; (4) bisphosphonates have a half-life in bone of 11 years because of their irreversible binding to bone via their central carbon atom; (5) when administered intravenously, bisphosphonate loads bone and accumulates in bone 142.8 times faster than when administered orally; (6) osteoclastic resorption of bisphosphonate-loaded bone results in osteoclast death in which the cell bursts, releasing the bisphosphonate molecules to reenter the local bone or bone marrow in a re-dosing effect; (7) endosteal osteoblasts are dependent on the osteoclastic resorption/growth factor release/new bone formation mechanism of bone renewal, whereas periosteal osteoblasts are not; and (8) it is likely that endosteal osteoblasts and periosteal osteoblasts have different cell membrane receptors and arise from separate embryologic niches. ORAL CRANIOFAC TISSUE ENG 2012;2:309–320. doi: 10.11607/octe.0058

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The emergence of the AIDS epidemic in 1980 caught the fields of medical science, immunology, and medical and dental education unaware and unprepared. At that time, the immune system was simply understood as a two-class system consisting of phagocytes (eg, neutrophils, macrophages, etc) and secretory cells (eg, lymphocytes and plasma cells). The secretory cells were further categorized as T cells from thymus dependency—producing lymphokines and B cells from Peyer’s patches, lymph nodes, and other unknown sites related to the bursa of Fabricius in birds (hence the name B cells). It was taught that B cells produce antibodies, and they were further identified as producing various classes of antibodies (eg, immunoglobulins M, G, A, E). However, the identification of a virus with a specific predilection to a certain component of the immune system stimulated a frenetic search and study of immune cells and how they work, leading to the recognition of T-helper cells, the specific CD4 cell, chemokine receptor 5 and CXC chemokine receptor 4, natural killer T cells, antigen-presenting cells, etc. Knowledge of immune cells and the immune system grew rapidly within a short time and led to focused treatments for human immunodeficiency virus infection/AIDS, as well as an improved knowledge and treatment of other immune-based pathologies.
Now, in a similar sense, bisphosphonates have caused osteonecrosis of the jaws (ONJ), spawning a renewed interest and intense research in the last 10 years into how bone remodels and renews itself as well as how it ages, becomes injured, and fractures.

Although cases of ONJ occurred in drug company–sponsored trials in the 1990s, they were dismissed and not reported.\(^5\)–\(^7\) Therefore, the initial identification of ONJ caused by bisphosphonates to the scientific community occurred in a 2002 textbook.\(^8\) This was followed by four publications\(^9\)–\(^12\) in the latter half of 2003 and first half of 2004 and has since been followed by more than 1,300 publications, which have reported more than 15,000 cases, confirming the epidemic predicted in 2003.\(^9\) However, the majority of these publications have focused on prevalence, the clinical entity of exposed bone, and prevention and treatment recommendations.\(^13\)–\(^17\) Few have focused on the actual bone cells involved and what their newly discovered expanded role is in ONJ or even in normal skeletal homeostasis.

**SPECIFIC BONE SCIENCE LESSONS LEARNED FROM BISPHOSPHONATES**

**Bone Remodeling Rates and Cellular Life Spans**

The fundamental effect of bisphosphonates is the impairment and apoptosis (cellular death) of not only adult osteoclasts but osteoclast precursors in the bone marrow (Figs 1a and 1b).\(^18\),\(^19\) Osteoclasts arise from hematopoietic stem cells in bone marrow and mature as mononuclear cells through the influence of transcription factors c-Fos and PU.1 (Fig 2).\(^20\) They are secreted into the general circulation, still as a mononuclear cell, which then fuses into a multinucleated cell through the effects of interleukin-1 and interleukin-6 as well as receptor activator of nuclear factor kappa-B ligand (RANKL). However, this mature osteoclast is a terminal cell with a life span of only 14 days.\(^21\) This is in contrast to osteoblasts, which become osteocytes, at which point they have a life span of about 180 days. Osteocytes are not mere quiescent cells but are tasked with the responsibility of acting as mechanoreceptors through their pseudopodia within their canaliculi as they traverse the mineral matrix. Osteocytes sense tension and pressure to regulate the resorption of their own mineral matrix through the secretion of osteoprotegerin (OP).\(^22\) That is, osteocytes sense tension and pressure, which in the jaws is highest in the lamina dura about the teeth, the edentulous alveolar ridge in denture wearers, and the posterior lingual cortex in the region of the mandibular molars—the areas of highest incidence of ONJ. This is a result of the balance between the prorresorption effects of RANKL and the inhibition of remodeling provided by OP (Fig 3). In areas of greater stress, the osteoblasts/osteocytes reduce their secretion of OP and increase their secretion of RANKL so that the area can resorb and re-form to accommodate this greater stress, a classic adaptive response.\(^23\),\(^24\) If the now-stimulated osteoclast attempts to resorb this stressed bone that
contains a bisphosphonate, it begins to do so but dies off before completing the resorption, thus disrupting the resorption-renewal cycle. The osteocytes then succumb to the stress and die before their 180-day life span, or they live out their life span but are not replaced. The bone may first become overmineralized (sclerotic) (Fig 4), but then it often becomes necrotic and exposed (Fig 5).

How do osteoclasts know when to resorb bone during normal skeletal remodeling? Osteoclasts self-secrete RANKL as an autocrine function and will resorb any bone that is vulnerable to be resorbed.23–25 This vulnerability is determined by the age and health of the bone itself. That is, young undamaged osteocytes secrete sufficient amounts of OP to inhibit osteoclastic bone resorption. As osteocytes age, become fewer in number, or are injured, however, their OP secretion drops, allowing osteoclasts to overcome this inhibition and resorb the mineral matrix.25 Therefore, osteoclasts are able to recognize not just necrotic bone and sequester it, but they recognize aged and injured bone as well, which keeps the skeleton in a dynamic renewal of young elastic bone capable of responding to the stresses placed upon it (Fig 6).

**The Coupling of Bone Resorption and Bone Remodeling (Renewal)**

The effects of bisphosphonates on the jaws help to clarify the too often interchangeable and incorrectly used terms *bone modeling, bone remodeling,* and
Bone modeling is the morphologic change in bone (e.g., thickening or thinning) of a bone in response to adaptive stress. The lamina dura is a good example of this. Here, within a marrow space, is an unexpected rim of dense cortical bone induced by Sharpey fibers from the periodontal ligament arising from root cementum and inserting into it. This transmits occlusal forces to the bone, which responds by laying down denser bone that is capable of withstanding this stress, which becomes the lamina dura. When a tooth is extracted, the lamina dura is seen to fade away radiographically (Fig 7). Before tooth extraction, there is resorption and new bone apposition, with the net result of a gain in bone density because of dental occlusal loading. After tooth extraction, the resorption followed by new apposition of bone results in a net loss of bone density to a degree equal to that of the surrounding trabecular bone in the marrow space because of the loss of occlusal loading stress. Therefore, the lamina dura remodels to the density of marrow space trabecular bone and becomes indistinct on radiographs. This action of osteoclast-mediated bone resorption followed by new bone apposition is referred to as bone remodeling. Therefore, bone remodeling is the mechanism by which bone modeling occurs. In a similar sense, old and dying bone that has lived out its average life span of 180 days undergoes renewal by bone resorption coupled with new bone apposition by this very same bone remodeling activity. Therefore, bone remodeling is also the mechanism by which bone renewal is achieved.

The example of the lamina dura represents a steady state of bone modeling. However, it is well known that both hyperocclusion and bisphosphonates independently cause a thickening of the lamina dura and together cause an even more prominent thickening of the lamina dura (Fig 8). Hyperocclusion transmits increased forces to the lamina dura through the periodontal ligament. The lamina dura thickens in response up to a point beyond which it may break down, resulting in periodontal bone loss. Bisphosphonates prevent the resorption phase of lamina dura remodeling, resulting in hypermineralization of the lamina dura with old unremodeled bone. If the bisphosphonate dose accumulation over time in the lamina dura becomes too great, the bone dies and becomes exposed. This is why the observed bone exposure of bisphosphonate-induced osteonecrosis always begins in the alveolar bone or over the remodeling surface of a torus. This is the case even when it is first seen as a spontaneous exposure of the lingual cortex in the mandibular molar region; this occurs because axial loading of the mandibular molars is actually exerted on the lingual cortex rather than the inferior border (Fig 9). It also explains why the exposed bone in this area is always above the mylohyoid ridge, which anatomically is alveolar bone (Fig 10).

The Biochemistry of Bone
Bone is type 1 collagen wrapped as a left-handed triple helix in which crystals of hydroxyapatite are interspersed. However, upon formation of the mineral matrix, osteoblasts insert trace quantities of the growth factors bone morphogenetic protein (BMP) and insulin-like growth factors 1 and 2 (ILG-1 and ILG-2). When an osteoclast begins normal resorption of old or injured bone, it seals itself to the bone surface and establishes a closed compartment that will become the Haversian lacuna. It then secretes protons (acid pH < 1) to dissolve the hydroxyapatite crystals and collagenase to break down the bone collagen. However, in doing so it liberates the acid-insoluble BMP, ILG-1, and ILG-2, which in turn causes a proliferation and differentiation of local resident stem cells and osteoprogenitor cells to re-form the bone with younger and more elastic tissue (Fig 11). This coupling of bone resorption followed by new bone formation (bone remodeling) is not only the basis of bone renewal but the fundamental physiology of
Osteopenia and osteoporosis for which the oral bisphosphonates alendronate, resldronate, and ibandronate are prescribed (and more recently the intravenous [IV] form of zoledronate known as Reclast and the RANKL inhibitor denosumab subcutaneously), ie, bone renewal throughout the entire skeleton as well as the focal areas of interest for osteopenia and osteoporosis. The lumbar spine and femoral head remodel with a net gain in bone mass until an individual’s early 20s. After the early to mid-20s, a slow but steady decline in bone mass takes place in both men and women. This is because less than 100% of the bone that is resorbed about every 180 days is replaced by newly forming bone. Over the years, this slight shortfall in bone renewal adds up to a significant loss of bone mass. It is heightened in women after menopause because estrogen is one of the requirements for osteoblast differentiation from bone marrow osteoprogenitor cells. Therefore, postmenopausal women undergo a more rapid and significant decline in bone mass (crudely measured as bone mineral density by the dual energy x-ray absorptiometry scan). Thus, the logical strategy but also the fallacy of treating osteopenia/osteoporosis with bisphosphonates comes to light.

**The Strategy and Fallacy of Bisphosphonates for Osteopenia/Osteoporosis**

Bisphosphonates inhibit osteoclast-mediated bone resorption and renewal. Therefore, similar to their effect on the lamina dura, they retain existing bone; under normal function, this increases mineralization, resulting in an increase in bone mineral density and
theoretically an increased resistance to osteoporosis-related fractures.\textsuperscript{28,31} However, preventing osteoclast-mediated bone resorption/renewal results only in the retention of old bone, which lives out its natural life span and becomes brittle and thus even more prone to fracture.\textsuperscript{32,34} This is why the more recent literature is reporting an increasing number of femur fractures actually caused by alendronate rather than prevented by it (Fig 12)\textsuperscript{35–38} and why independent investigators have not been able to duplicate the prevention of osteoporosis fractures first published by the drug companies.\textsuperscript{39}

Simply stated, osteoclasts are a normal cell in skeletal homeostasis. Removing them and their useful function by toxic drugs can be expected to result in, and has resulted in, the same complications seen in the genetic loss of osteoclast function in the disease known as osteopetrosis,\textsuperscript{40} ie, exposed bone in the maxilla and/or mandible and leg fractures. With bisphosphonates, it is not a matter of genetic expression but one of dose and accumulation in bone over time. This is why, in September 2011, the U.S. Food and Drug Administration (FDA) encouraged physicians to reassess osteopenia/osteoporosis patients treated with oral bisphosphonates after 3 years of treatment and discouraged treatment beyond 5 years.\textsuperscript{41}

**The Biochemistry of Bisphosphonates Related to Bone**

Bisphosphonates are a synthetic of pyrophosphates, which are diphosphonates,\textsuperscript{42} as exemplified by the common bone scan technetium-99 methylene diphosphonate. Such scans are used routinely to show areas of increased activity in bone; the technetium is water soluble and therefore rapidly eliminated through the kidneys. Any view of a total-body technetium bone scan will identify a generally greater uptake in the jaws, indicative of the greater turnover rate of jawbone (Fig 13). That, together with occlusal loading and denture wearing, is why the jaws are more vulnerable to bisphosphonates and why exposed-bone ONJ only occurs in the jaws.

In the synthesis of bisphosphonates, the central-backbone oxygen atom of the pyrophosphates is replaced by a carbon atom, which imparts irreversible binding to the hydroxyapatite in bone (Fig 14).\textsuperscript{53,44} The result is a documented half-life in bone of 11 years and an accumulation to toxic effects with repeated doses that can result in ONJ or femur fractures.\textsuperscript{45} This explains the greater numbers and greater severity of ONJ cases with longer-term use and the fact that the vast majority of femur fractures occur only after 6 or more years of alendronate use (Fig 12).

While the central-backbone carbon atom imparts irreversible binding to bone, the R-group attached to the central carbon atom confers potency (Fig 14). Nitrogen within this R-group side chain increases potency sufficiently such that non–nitrogen-containing bisphosphonates, eg, etidronate and tiludronate, are not known to cause ONJ. Variations in the configuration of the nitrogen-containing side chains confer different degrees of potency, with zolendronate being the most potent of the IV bisphosphonates and therefore causing the majority of IV ONJ cases\textsuperscript{46} and alendronate being the most potent of the oral bisphosphonates and therefore causing the majority of oral ONJ cases.\textsuperscript{47–49}

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**Fig 12** Subtrochanter midshaft fracture of the femur caused by long-term alendronate (Fosamax) use.

**Fig 13** Technetium-99 methylene diphosphonate bone scans show a generalized increased uptake in the jaws, which is indicative of a higher turnover rate.
The Pharmacodynamics of Bisphosphonates and Bone

It has been widely recognized that ONJ cases caused by IV bisphosphonates occur after fewer doses, are more severe, require surgical intervention more often and of a greater magnitude, and are less responsive to drug holidays.\(^47,48\) This is a result of the immediate uptake and binding of bisphosphonates to bone via the IV route, as compared to the oral route, where less than 0.7% is absorbed through the gastrointestinal mucosa before it circulates and binds to bone.\(^7,47,50,51\) Therefore, for each milligram of the equally potent IV bisphosphonate pamidronate (Aredia) vs the oral bisphosphonate alendronate, there is a 1/0.007 bone absorption ratio, meaning that bone absorption and accumulation is 142.8 times higher for an IV than an oral bisphosphonate. This explains not only the difference in severity and the degree of exposed bone but why as few as four doses of an IV bisphosphonate can place patients at a significant risk of ONJ, while it may take 100 or more weekly doses (2 years) of an oral bisphosphonate to create the same risk level.

Because all bisphosphonates affect the osteoclast precursors in bone marrow as well as those osteoclasts remodeling mineralized bone in the general skeleton,\(^19,52\) IV doses affect the bone marrow precursors quickly and more severely. This reduces the ability of the bone marrow to replace lost osteoclasts that attempt to resorb bisphosphonate-loaded bone in the jaws at other skeletal units. By contrast, the poor gastrointestinal absorption of oral bisphosphonates represents a trickle effect into the bone marrow and affects the precursor cells more weakly, allowing them to keep pace with lost osteoclasts during resorption of bisphosphonate-loaded bone in the jaws and elsewhere. This retained ability to replace a population of dying osteoclasts explains why drug holidays of 9 months or longer are effective in reducing the risk of ONJ in an oral bisphosphonate individual but usually are not in a patient that has been treated with an IV bisphosphonate.

Drug Holidays and Bisphosphonates

A drug holiday is a temporary discontinuation of a bisphosphonate to allow bone marrow recovery and the formation of new osteoclasts to repopulate the population that has been depleted by the bisphosphonate. As stated, because IV bisphosphonates confer 142.8 times the bone loading dose of oral bisphosphonates, the depletion of osteoclast precursors and the accumulation of the bisphosphonate in bone are too great to be predictably reversed by a drug holiday. However, because an oral bisphosphonate’s systemic absorption is so low, its effects on the osteoclast bone marrow precursors are less pronounced, allowing the bone marrow to keep pace with the loss. Therefore, a drug holiday becomes effective because a residual population of marrow stem cells and osteoclast progenitor cells repopulates the osteoclast pool to return bone remodeling/renewal to levels that remain depressed but are still consistent with bone healing (Figs 15 and 16).

Arbitrary drug holidays that have been recommended by various associations are not based on evidence.\(^53-55\) This is because the length of a drug holiday depends on the duration and dose of the various
bisphosphonates used. For instance, alendronate is recommended and marketed for 70 mg per week, as compared to residronate, at 35 mg per week, and ibandronate, at 150 mg per month (35 mg/week). Perhaps this is why alendronate has accounted for more than 96% of ONJ cases caused by an oral bisphosphonate and why it is the only oral bisphosphonate known to cause fractures of the femur.38,48

The length of a drug holiday from an oral bisphosphonate or even the very need for a drug holiday may be guided by a serum morning fasting c-terminal telopeptide (CTX) test. Although the clinical utility of this blood test remains controversial, with several studies showing data to attest to its clinical correlation48,56–59 and others denying its usefulness,53,60 it is frequently used as the primary biochemical marker of bone turnover. In fact, it has been used in nearly every study of the therapeutic effectiveness of oral bisphosphonates and those of many of the IV bisphosphonates that were presented to the FDA.5,6,30,33,61

The CTX measures an eight–amino-acid sequence of collagen released by osteoclasts during bone resorption.56 However, since most cancers break down collagen during their invasion of normal tissue, the CTX results are unreliably higher in cancer patients, making it a useless study in this group. Therefore, the clinical utility of the CTX is limited to noncancer patients on oral bisphosphonates. In addition, in patients who have taken or are taking methotrexate or prednisone, the CTX is unreliably low and is also not recommended. These patients are usually those with autoimmune diseases such as rheumatoid arthritis or systemic lupus erythematosus. Because methotrexate reduces the bone marrow stem cell population, fewer osteoclasts are produced, resulting in lower bone turnover and therefore a lower CTX level. Because prednisone
inhibits collagen synthesis, including type 1 collagen in bone, the release of collagen fragments during bone turnover is reduced, resulting in a lower CTX level.\textsuperscript{56,59} Normal CTX level tests in bisphosphonate-naïve adults range between 350 and 500 pg/mL.\textsuperscript{48,50,56} CTX levels in patients taking an oral bisphosphonate may range from 20 to 55 pg/mL, depending on the specific drug, the dose, and the number of doses taken. Studies have shown that a CTX value of 150 pg/mL is sufficient for healing in alveolar bone after surgery and represents a goal for a drug holiday.\textsuperscript{48,56} Upon beginning a drug holiday in an uncomplicated osteopenia/osteoporosis patient, a low CTX is usually seen to gradually rise to a level of 150 pg/mL within 9 months. However, one notable exception highlights an important lesson to be learned about osteoclasts and bisphosphonates. That is, in patients who have taken alendronate steadily for 7 years or more, the CTX is usually very low (30 to 60 pg/mL). Upon beginning a drug holiday, the CTX is seen to rise, as one would expect from bone marrow recovery. However, once the CTX rises to levels around 100 pg/mL, CTX levels paradoxically fall again to an intermediate level of 60 to 90 pg/mL. This seems to be the result of the drug holiday allowing bone marrow recovery and the regeneration of new osteoclasts; these new osteoclasts begin resorbing heavily bisphosphonate-loaded bone. In doing so, they die and burst (apoptosis), releasing the metabolically stable bisphosphonate into the local environment, where some of it becomes bound once again to adjacent bone; most is taken up in the circulation, effectively re-dosing the body with the bisphosphonate, thereby suppressing the bone marrow osteoclast precursors once again and lowering the CTX value.

**Endosteum and Periosteum**

The mandible and maxilla, like most bones, have an outer rim of cortical bone covered by a thin periosteum on their outer cortical surface. On the inner cortical surface is a layer of endosteum, which unlike the periosteum is not a defined membrane but a series of connected cells that line the surface of a network of interconnected trabecular bone. This trabecular bone network extends from the buccal cortex to the lingual cortex, which is lined by endosteal cells throughout. Between these bone trabeculae are various marrow spaces, which in the adult contain a dwindling number of true hematopoietic and mesenchymal stem cells as well as age-related fibrofatty marrow.

In resected specimens of both IV and oral bisphosphonate–induced ONJ, there is a noted formation of red viable periosteal new bone that can be separated from the underlying necrotic bone in many cases (Figs 17a to 17c). Additionally, new periosteal bone can be seen radiographically to form months and years after resection (Fig 18). This indicates that the network of trabecular bone in the marrow space and the inner cortical surface are dependent on the
osteoclast-mediated coupling of bone renewal and homeostasis that is induced by bone resorption/release of growth factors/new bone formation, whereas the outer cortical surface and periosteal cells do not depend on this coupling.\textsuperscript{62–64} This further implies a significant genetic difference between periosteal and endosteal cells and likely a different embryology as well. Moreover, it has also been noted that when recombinant human BMP is used in contact with both endosteum and periosteum, the regenerated bone arises solely from the endosteum, implying that periosteal cells lack the receptors for BMP (Fig 19).\textsuperscript{65} This has already been translated to the clinical arena by sheer experience, as surgeons do not rely on the periosteum to regenerate bone when using recombinant human BMP-2 in an absorbable collagen sponge as a graft but instead increase its contact with a greater area of endosteal bone surface.\textsuperscript{66}

CONCLUSIONS

1. The toxic effects of bisphosphonates on bone and the resulting cases of osteonecrosis of the jaw, similar to human immunodeficiency virus/AIDS, have reinforced the age-old axiom that “disease teaches us much about normal.”
2. Bone remodeling is the mechanism by which bone renews itself and by which it adapts to functional stress, often termed Wolff’s定律 or bone modeling.
3. Osteoclastic bone resorption is coupled to bone remodeling and therefore bone modeling and bone renewal by the growth factors bone morphogenetic protein and insulin-like growth factors 1 and 2.
4. The inhibition of bone remodeling by bisphosphonates results in the retention of old hypermineralized brittle bone, which may appear as a thickened lamina dura or an area of generalized sclerosis in the jaws, may die off to result in osteonecrosis of the jaw, or may result in subtrochanteric fractures of the femur.
5. Bisphosphonates are irreversibly bound to bone and can be released from bone only by osteoclastic acid–based resorption. This irreversible binding is caused by the backbone carbon atom in this relatively small molecule. Potency is determined by the nitrogen-containing side chain on the backbone carbon atom.
6. Osteonecrosis caused by intravenous bisphosphonates is more prevalent, more severe, less responsive to drug holidays, and less responsive to debridement surgeries than osteonecrosis induced by oral bisphosphonates, because intravenous bisphosphonates load and accumulate in bone at a rate 142.8 times higher than oral bisphosphonates.
7. Drug holidays are useful only in cases of oral bisphosphonate use uncomplicated by cancer, methotrexate, or prednisone because the 0.7% gastrointestinal absorption of oral bisphosphonates induces only a gradual toxicity to the bone marrow, allowing the marrow to keep pace, recover in response to a drug holiday, and repopulate the lost osteoclasts.
8. Osteoclasts that resorb bone during a drug holiday after several years of bisphosphonate use release the bisphosphonate, which is then taken up in the circulation to act in a re-dosing effect on the bone marrow as well as bone formation. This is reflected by lower c-terminal telopeptide values.
9. While the endosteum is dependent on the cascade of bone resorption/release of growth factors/new bone formation, periosteum is not and is therefore resistant to bisphosphonate toxicity.
10. Periosteal and endosteal cells are likely genetically and embryologically different, as they respond in opposite fashions to bisphosphonates and bone morphogenetic protein.

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