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Editorial Comment

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Treatment of immobilization-related hypercalcaemia with denosumab

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Hypercalcaemia has been reported to occur in ∼1–4% of the general adult population, and in 0.5–3% of hospitalized adult patients [1]. Hypercalcaemia results when the entry of calcium (Ca) into the extracellular fluid exceeds excretion in the urine or deposition in bone. This occurs when there is accelerated bone resorption, excessive gastrointestinal absorption or decreased renal excretion of Ca. Under most pathologic conditions, hypercalcaemia results from increased skeletal resorption or intestinal absorption with normal or decreased renal excretion. In some disorders more than one mechanism may be involved. As an example, in primary hyperparathyroidism, elevated parathyroid hormone (PTH) levels increase bone resorption, tubular Ca reabsorption and, indirectly, intestinal Ca absorption (by increasing renal synthesis of calcitriol). Among all causes of hypercalcaemia, primary hyperparathyroidism and malignancy are the most common, accounting for >90% of cases [2].

The differential diagnosis of hypercalcaemia may be broadly divided into PTH-mediated and non-PTH-mediated hypercalcaemia [3] (Table 1). PTH-mediated hypercalcaemia is associated most frequently with primary hyperparathyroidism. The most common cause of non-PTH-mediated hypercalcaemia is that of malignancy. Hypercalcaemia in patients with cancer is primarily due to increased bone resorption and the release of Ca from bone. There are three major mechanisms by which this can occur: osteolytic metastases with local release of cytokines which stimulates the differentiation of osteoclast precursors into mature osteoclasts and activates bone resorption (mainly breast cancer, myeloma and lymphomas), tumour secretion of PTH-related protein (PTHrP) (mainly solid tumours, such as squamous cell carcinomas and renal carcinomas) and tumour production of calcitriol (mainly lymphomas). In the latter condition, hypercalcaemia results from the increase of either bone resorption or intestinal absorption. Ectopic PTH secretion has been documented in single cases of small cell and squamous cell lung cancer, ovarian carcinoma, papillary thyroid carcinoma, hepatocellular carcinoma and undifferentiated neuroendocrine neoplasia. Non-PTH-mediated hypercalcaemia may be caused also by different endocrine disorders such as thyrotoxicosis and adrenal insufficiency. In hyperthyroidism, hypercalcaemia mainly results from increased bone resorption. Hypercalcaemia is a rare complication of pheochromocytoma. It can be due to concurrent hyperparathyroidism (in MEN, Type II) or to the pheochromocytoma itself due to tumoural production of PTHrp. Granulomatous diseases (such as sarcoidosis, and focal or disseminated tuberculosis) can cause hypercalcaemia by overproduction of 1,25-dihydroxyvitamin D. Immobilization is a rare cause of hypercalcaemia. For the diagnosis of immobilization-related hypercalcaemia, all the other causes of PTH- and vitamin D-dependent hypercalcaemia should be carefully excluded (Table 1). In this issue of *CKJ*, de Beus and Boer [4] report the case of a patient with chronic renal failure and immobilization-related hypercalcaemia successfully treated with denosumab, a monoclonal antibody with affinity for the receptor activator of nuclear factor-kB (RANK) ligand (RANKL). Prolonged immobilization uncouples bone remodelling because of the lack of mechanical stress. The greater deceleration in bone formation than in bone resorption results in a net efflux of Ca from bone that induces hypercalciuria and suppression of the parathyroid-1,25-vitamin D axis [5]. Hypercalcaemia develops when the efflux of Ca from bone exceeds the capacity of the kidney to excrete Ca. Immobilized patients with pre-existing states of high bone turnover (e.g. adolescents and patients with Paget's disease, thyrotoxycosis or primary hyperparathyroidism), and/or reduced renal function are at particular risk of developing severe hypercalcaemia [6–8]

The role of RANK and RANKL in the control of bone remodelling

The activity of osteoclasts and osteoblasts is intricately coordinated to continually remodel the adult skeleton. There is a well-balanced remodelling sequence in normal bone: bone is first resorbed by osteoclasts and then osteoblasts form bone at the same site. An imbalance in the bone remodelling process with excessive osteoclastic bone resorption exceeding the rate of osteoblastic bone formation results in a net release of Ca from bone, bone mass loss, hypercalciuria and hypercalcaemia. The RANK/RANKL interaction plays a key role in controlling bone remodelling by inducing osteoclast formation [9–10]. RANKL is a potent inducer of osteoclast formation.

Table 1. Causes of hypercalcaemia

PTH-mediated
Primary hyperparathyroidism (sporadic)
Familial: multiple endocrine neoplasia (MEN-I and -IIa), familial hypocalciuric hypercalcaemia
Tertiary hyperparathyroidism (renal failure)
PTH-independent
Hypercalcaemia of malignancy (production of PTHrp, activation of extrarenal 1 alpha-hydroxylase with increased calcitriol production, osteolytic bone metastases and local cytokines production)
Vitamin D intoxication
Chronic granulomatous disorders (activation of extrarenal 1 alpha-hydroxylase with increased calcitriol production)
Medications (thiazide diuretics, lithium, teriparatide, excessive vitamin A, theophylline toxicity)
Miscellaneous
Hyperthyroidism
Acromegaly
Pheochromocytoma
Adrenal insufficiency
Immobilization
Parenteral nutrition
Milk alkali syndrome

Adapted from: Khairallah *et al*. [3].

Osteoclasts arise from precursor cells in the monocyte–macrophage lineage. Both systemic factors and locally acting factors induce the formation and activity of osteoclasts. Systemic hormones such as PTH, 1,25-dihydroxyvitamin D3, thyroxine and prostaglandins (such as PGE2) stimulate the formation of osteoclasts by inducing the expression of RANKL on marrow stromal cells and osteoblasts rather than by acting directly on osteoclast precursors [10, 11]. RANKL can also be released in a soluble form by T cells in inflammatory states. RANKL binds the RANK receptor on osteoclast precursors and signals through the nuclear factor kB (NF-kB) and Jun *N*-terminal kinase pathways to induce the activation, migration, differentiation and fusion of haematopoietic cells of the osteoclast lineage to begin the process of bone resorption [10]. In addition, the formation and activation of the osteoclasts can be stimulated by the release of interleukin-6, interleukin-1, prostaglandins and colony-stimulating factors (CSFs) by the osteoblasts [11] (Figure 1). Osteoblasts arise from mesenchymal stem cells, which form osteoblasts, adipocytes and muscle cells [12]. A transcription factor that is critical for the differentiation of osteoblasts is Runx-2, or core-binding factor alfa-1 (CBFA1). CBFA1 drives the expression of most genes associated with osteoblast differentiation [13]. Bone does not develop in mice that lack the CBFA1 gene [14]. Both systemic factors and locally acting factors can enhance the proliferation and differentiation of osteoblasts (Figure 1). These include PTH, prostaglandins and cytokines as well as growth factors such as platelet-derived growth factor (PDGF) produced by lymphocytes. In addition, bone matrix is a major source of growth factors, which can enhance the proliferation and differentiation of osteoblasts. These include the bone morphogenetic proteins (BMPs), transforming growth factor β (TGF-β), insulin-like growth factors (IGFs) and fibroblast growth factors (FGFs) [9, 15]. Osteoprotegerin (OPG) is a soluble 'decoy receptor' that is expressed by osteoblasts and binds to RANKL with high affinity [16]. Because OPG directly competes with RANK for the binding sites of RANKL, OPG inhibits osteoclastogenesis and subsequent bone resorption [17]. The ratio of RANKL to OPG determines the level of osteoclastogenesis. Overproduction of OPG in transgenic mice causes severe osteopetrosis, whereas the absence of OPG

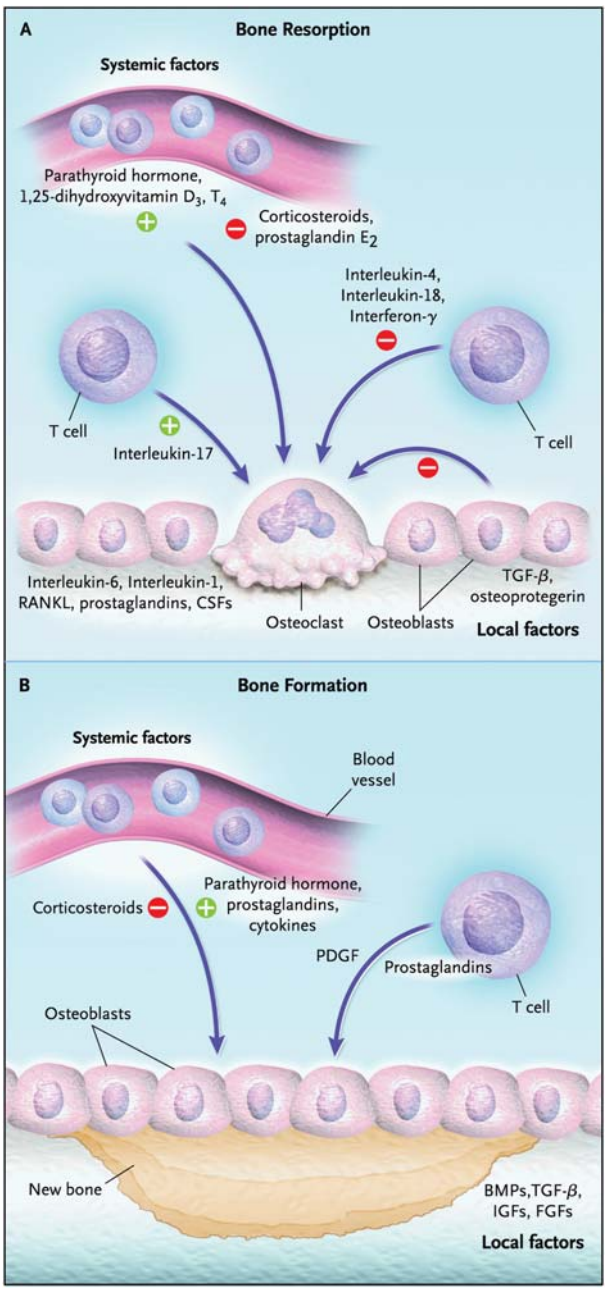

Fig. 1. Factors implicated in bone resorption (**A**) and formation (**B**). Both systemic factors and locally acting factors induce the formation and activity of osteoclasts (Panel A). Systemic hormones such as PTH, 1,25-dihydroxyvitamin D_3 and thyroxine (T_4) stimulate the formation of osteoclasts by inducing the expression of receptor activator of nuclear factor-κB ligand (RANKL) on marrow stromal cells and osteoblasts. In addition, osteoblasts can produce interleukin-6, interleukin-1, prostaglandins and CSFs, which induce the formation of osteoclasts. Osteoblast can also produce factors, which inhibit the formation of osteoclasts, such as TGF-β, and osteoprotogerin. Helper cells such as T cells can produce cytokines that can inhibit the formation of osteoclasts, such as interleukin-4, interleukin-18 and interferon-γ. Both systemic factors and locally acting factors can enhance the proliferation and differentiation of osteoblasts (Panel B). These include PTH, prostaglandins and cytokines as well as growth factors such as PDGF produced by lymphocytes. In addition, bone matrix is a major source of growth factors, which can enhance the proliferation and differentiation of osteoblasts. These include the BMPs, TGF-β, IGFs and FGFs. Corticosteroids can induce apoptosis of osteoblasts and block bone formation. Plus signs indicate stimulation, and minus signs inhibition. Reproduced with permission from Roodman GD [11].

results in marked osteopenia [18, 19]. The importance of RANKL in the formation of osteoclasts has been also documented by the knockout mouse models in which the RANKL or RANK gene has been deleted. These animals lack osteoclasts, and as a result, severe osteopetrosis develops [20]. During physiological bone remodelling, the ratio of RANKL to OPG is balanced. An excess of RANKL is found in many clinical conditions such as oestrogen deficiency [21], systemic glucocorticoid exposure [22], active inflammatory process in rheumatoid arthritis [23], skeletal malignancies such as multiple myeloma [24] and bone metastases [25], resulting in exacerbated bone loss. The imbalance of the RANKL/OPG system seems to play a pivotal role also in the pathogenesis of immobilization-related hypercalcaemia [26, 27]. Mechanical strain applied to murine primary stromal cells decreased RANKL mRNA levels by ∼40% which was paralleled by a 50% reduction of osteoclast formation [28]. OPG administration by inhibiting osteoclast activity ameliorates the decrease in both bone mineral density and bone strength in immobilized rats [29]. Increased RANKL production by osteocytes plays a pivotal role in the bone loss associated with unloading [27]. Thus, mechanical strain enhances the RANKL-to-OPG ratio, and lack of mechanical strain during the periods of immobilization may lead to an imbalance in this ratio resulting in increased bone resorption [26, 27].

Bisphosphonates and denosumab for treatment of bone resorption-induced hypercalcaemia: safety, tolerability and precautions in patients with renal failure

The administration of bisphosphonates was until recently the only way to reduce bone resorption and control hypercalcaemia when osteoclastic hyperactivity was the major mechanism of hypercalcaemia, and systemic factors, such as increased production of PTH or calcitriol, were not involved in the osteoclastic stimulation. Now, denosumab, a monoclonal antibody with affinity for the RANKL, has become available on the market. Denosumab was approved by the United States Food and Drug Administration for the treatment of osteoporosis in postmenopausal women, and for the prevention of skeletal-related events (SREs: fracture, spinal cord compression, bone pain requiring surgery/radiation therapy and hypercalcaemia of malignancy) in patients with bone metastasis from solid tumours. Denosumab blocks the binding of RANKL to RANK and thereby reduces the formation, function and survival of osteoclasts, which results in the decreased bone resorption and increased bone density in osteoporosis [30, 31]. In solid tumours with bony metastases, RANKL inhibition decreases osteoclastic activity leading to decreased SRE and tumour-induced bone destruction [31, 32]. Thus, the mechanism of action of denosumab is quite different from that of bisphosphonates. Pharmacokinetics and pharmacodynamics studies, and recent clinical trials suggest that denosumab might be a promising alternative to bisphosphonates for the treatment of resorption-related hypercalcaemia in patients with renal insufficiency, for a more rapid and sustained effect on bone resorption, and a better tolerability compared with bisphosphonates.

The bisphosphonates are non-hydrolysable analogues of inorganic pyrophosphate which adsorb to the surface of bone hydroxyapatite and inhibit calcium release by interfering with osteoclast-mediated bone resorption. Bisphosphonates also reduce osteoclast activity by decreasing osteoclast progenitor development and recruitment, and by promoting osteoclast apoptosis [33]. They are effective in treating hypercalcaemia resulting from excessive bone resorption of any cause. All of the bisphosphonates are relatively non-toxic compounds and they are more potent than calcitonin and saline for patients with moderate or severe hypercalcaemia [34, 35]. As a result, they have become the preferred agents for management of hypercalcaemia due to excessive bone resorption from a variety of causes, including malignancy-related hypercalcaemia [34, 35]. Intravenous zoledronic acid (4–8 mg) and pamidronate (60–90 mg) are generally the bisphosphonates of choice. Zoledronic acid is favoured by some because it is more potent than pamidronate [36]. Their maximum effect occurs in 2 to 4 days, so that they are usually given in conjunction with saline and/or calcitonin, which reduce serum Ca concentration more rapidly. Bisphosphonates are excreted by the kidney. In subjects with normal renal function, about half of the administered dose binds to the bone and the rest is excreted within several hours by the kidney. Bisphosphonates are cleared rapidly from the plasma (half-life is ∼1h), but may persist in bone for the patient's lifetime [37]. Zoledronic acid and pamidronate have been associated with both acute and chronic renal failure [36, 38, 39]. Although acute renal failure may be clinically reversible, varying degrees of irreversible impairment may persist and eventually lead to chronic renal failure. In addition, pamidronate has been associated with nephrotic syndrome, tubulointerstitial nephritis and Fanconi syndrome [39].The risk of renal damage is directly related to the drug infusion time and dosage. The incidence of renal failure associated with zoledronic acid varies by patients' underlying diseases, from ∼10–20% [39, 40]. Previous treatments with bisphosphonates, advanced age and the presence of chronic kidney disease increase the risk of developing renal insufficiency [39–41]. Thus, intravenous bisphosphonates should be used with caution in patients with impaired renal function. Adequate hydratation with saline, dose reduction and a slower infusion rate may minimize the risk of renal damage in patients with chronic renal insufficiency [39].

Denosumab is not excreted by the kidney and renal function does not have a significant effect on denosumab pharmacokinetics or pharmacodynamics [42]. These findings suggest that dose adjustment based on the glomerular filtration rate is not required. Denosumab elimination is thought to occur through the immunoglobulin clearance pathway via the reticuloendothelial system, similar to that of other monoclonal antibodies and is thus thought to be independent of renal or hepatic function [43]. Pharmacokinetic studies showed a rapid and prolonged absorption of denosumab after subcutaneous injection, starting 1 h post-dose and reaching maximum serum levels as late as 21 days later [44, 45]. Effects on bone resorption start within 12 h, as documented by a rapid dose-dependent decrease in urinary and serum *N*-telopeptide levels. In patients treated with denosumab (doses ranging from 0.1 to 3 mg/kg), *N*-telopeptide levels decreased within 1 day and this effect lasted through Day 84 in the higher dose levels, whereas the effect of pamidronate (90 mg iv) reached a maximum at 3 days and lasted about 28 days [44]. The mean half-life of denosumab ranges between 25 and 40 days in relation to the dosages [44, 45].

RANKL-inhibitor agents have been shown to induce greater suppression of bone resorption and hypercalcaemia compared with bisphosphonates in murine models of humoral hypercalcaemia of malignancy [46]. Denosumab decreases bone resorption more rapidly and the effect is longer compared with pamidronate, in patients with multiple myeloma or bone metastases [32]. Denosumab significantly prolonged the time to a first SRE compared with zoledronic acid in a Phase III trial conducted in patients with bone metastases from breast cancer [47]. In another Phase III trial, denosumab was not significantly inferior to zoledronic acid in delaying time to first SRE in 1776 patients with multiple myeloma or bone metastases from a solid tumour other than breast or prostate cancer [48]. The recommended dose and schedule for denosumab for the prevention of SREs is 120 mg administered subcutaneously every 4 weeks.

Data on the safety of denosumab come mainly from the results of the large clinical trials in osteoporotic patients. In the osteoporosis clinical trials, denosumab (60 mg every 6 months) was generally well tolerated, and the incidence of serious adverse events was not different from that observed with placebo [49, 50]. However, eczema, cellulitis and flatulence were more common in women assigned to denosumab than placebo [49]. Two cases of osteonecrosis of the jaw were observed in the sixth year of extension of the FREEDOM trial [50]. In a *post-hoc* analysis of FREEDOM trial data, stratifying the patients with post-menopausal osteoporosis by levels of kidney function, denosumab was effective in reducing the risk of fracture independently of renal function and the incidence of adverse events did not differ by levels of kidney function [51]. Thus, denosumab for its efficacy and tolerability represents a valid alternative to bisphosphonates for patients with renal insufficiency affected by post-menopausal osteoporosis or hypercalcaemia of malignancy [51, 52]. The use of denosumab for treatment of other causes of resorption-related hypercalcaemia has not been reported so far.

In this issue, de Beus and Boer [4] report the case of a patient with chronic renal failure and immobilization-related hypercalcaemia successfully treated with denosumab after a partial and transient response to pamidronate. The case suggests that denosumab might be a promising alternative to bisphosphonates also for the treatment of resorption-related hypercalcaemia in patients with renal insufficiency. However, particular attention should be paid to ensuring that patients are supplemented with calcium and vitamin D prior to starting therapy. In the denosumab trials, all women with osteoporosis were supplemented with daily calcium (1000 mg) and vitamin D (400 to 800 Units) and the incidence of hypercalcaemia was negligible [49]. Thus, in patients with normal renal function, adequately supplemented with calcium and vitamin D, hypercalcaemia typically is not a concern. However, in patients with conditions that predispose to hypercalcaemia, such as chronic kidney disease, malabsorption syndromes or hypoparathyroidism, symptomatic hypercalcaemia may occur. In a study of 55 patients with varying degrees of chronic kidney disease, the proportion of patients with serum calcium <7.5 mg/dL (1.9 mmol/L) or symptomatic hypercalcaemia was higher, occurring in 10 and 29% of subjects with creatinine clearance of 50–80 and <30 mL/min, respectively [42]. Thus, in patients with chronic renal failure, calcium and vitamin D supplementation is recommended and serum calcium, phosphorus and magnesium should be closely monitored during therapy.

Conflict of interest statement. None declared.

(See related article by E. de Beus and W.H. Boer. Denosumab for treatment of immobilization-related hypercalcaemia in a patient with advanced renal failure. *Clin Kidney J* 2012; 5: 566–571)

References

1. Frolich A. Prevalence of hypercalcemia in normal and hospitalized populations. *Dan Med Bull* 1998; 45: 436–439
2. Lafferty FW. Differential diagnosis of hypercalcemia. *J Bone Miner Res* 1991; 6(Suppl 2): S51–S59
3. Khairallah W, Fawaz A, Brown EM *et al*. Hypercalcemia and diabetes insipidus in a patient previously treated with lithium. *Nat Clin Pract Nephrol* 2007; 3: 397–404
4. de Beus E, Boer WH. Denosumab for treatment of immobilization-related hypercalcemia in a patient with advanced renal failure. *Clin Kidney J* 2012; 5: 566–571
5. Stewart AF, Adler M, Byers CM *et al*. Calcium homeostasis in immobilization: an example of resorptive hypercalciuria. *N Engl J Med* 1982; 306: 1136–1140
6. Meythaler JM, Korkor AB, Nanda T *et al*. Immobilization hypercalcemia associated with Landry-Guillain-Barré syndrome. Successful therapy with combined calcitonin and etidronate. *Arch Intern Med* 1986; 146: 1567–1571
7. Massagli TL, Cardenas DD. Immobilization hypercalcemia treatment with pamidronate disodium after spinal cord injury. *Arch Phys Med Rehabil* 1999; 80: 998–1000
8. Gopal H, Sklar AH, Sherrard DJ. Symptomatic hypercalcemia of immobilization in a patient with end-stage renal disease. *Am J Kidney Dis* 2000; 35: 969–972
9. Kodama H, Nose M, Niida S *et al*. Essential role of macrophage colony-stimulating factor in the osteoclast differentiation supported by stromal cells. *J Exp Med* 1991; 173: 1291–1294
10. Hofbauer LC, Heufelder AE. Role of receptor activator of nuclear factor-kB ligand and osteoprotegerin in bone cell biology. *J Mol Med* 2001; 79: 243–253
11. Roodman GD. Mechanism of bone metastasis. *N Engl J Med* 2004; 350: 1655–1664
12. Aubin JE. Bone stem cells. *J Cell Biochem Suppl* 1998; 30–31: 73–82
13. Yang X, Karsenty G. Transcription factors in bone: developmental and pathological aspects. *Trends Mol Med* 2002; 8: 340–345
14. Komori T, Yagi H, Nomura S *et al*. Targeted disruption of Cbfa1 results in a complete lack of bone formation owing to maturational arrest of osteoblasts. *Cell* 1997; 89: 755–764
15. Mundy GR, Chen D, Zhao M *et al*. Growth regulatory factors and bone. *Rev Endocr Metab Disord* 2001; 2: 105–115
16. Simonet WS, Lacey DL, Dunstan CR *et al*. Osteoprotegerin: a novel secreted protein involved in the regulation of bone density. *Cell* 1997; 89: 309–319
17. Burges TL, Qian Y, Kaufman S *et al*. The ligand for osteoprotegerin (OPGL) directly activates mature osteoclasts. *J. Cell Biol* 1999; 145: 527–538
18. Min H, Morony S, Sarosi I *et al*. Osteoprotegerin reverses osteoporosis by inhibiting endosteal osteoclasts and prevents vascular calcification by blocking a process resembling osteoclastogenesis. *J Exp Med* 2000; 192: 463–474
19. Mizuno A, Amizuka N, Irie K *et al*. Severe osteoporosis in mice lacking osteoclastogenesis inhibitory factor/osteoprotegerin. *Biochem Biophys Res Commun* 1998; 247: 610–615
20. Dougall WC, Glaccum M, Charrier K *et al*. RANK is essential for osteoclast and lymph node development. *Genes Dev* 1999; 13: 2412–1224
21. Eghbali-Fatourechi G, Khosla S, Sanyal A *et al*. Role of RANK ligand in mediating increased bone resorption in early postmenopausal women. *J Clin Invest* 2003; 111: 1221–1230

22. von Tirpitz C, Epp S, Klaus J *et al*. Effect of systemic glucocorticoid therapy on bone metabolism and the osteoprotegerin system in patients with active Crohn's disease. *Eur J Gastroenterol* 2003; 15: 1165–1170
23. Mori H, Kitazawa R, Mizuki S *et al*. RANK ligand, RANK, and OPG expression in type II collagen-induced arthritis mouse. *Histochem Cell Biol* 2002; 117: 283–292
24. Standal T, Seidel C, Hjertner Ø *et al*. Osteoprotegerin is bound, internalized, and degraded by multiple myeloma cells. *Blood* 2002; 100: 3002–3007
25. Michigami T, Ihara-Watanabe M, Yamazaki M *et al*. Receptor activator of nuclear factor kappaB ligand (RANKL) is a key molecule of osteoclast formation for bone metastasis in a newly developed model of human neuroblastoma. *Cancer Res* 2001; 61: 1637–1644
26. Hofbauer LC, Kuhne CA, Viereck V. The OPG/RANKL/RANK system in metabolic bone diseases. *J Musculoskelet Neuronal Interact* 2004; 4: 268–275
27. Xiong J, Onal M, Jilka RL *et al*. Matrix-embedded cells control osteoclast formation. *Nat Med* 2011; 17: 1235–1241
28. Rubin J, Murphy T, Nanes MS *et al*. Mechanical strain inhibits expression of osteoclast differentiation factor by murine stromal cells. *Am J Physiol Cell Physiol* 2000; 278: C1126–C1132
29. Mochizuki S, Fujise N, Higashio K *et al*. Osteoclastogenesis inhibitory factor/osteoprotegerin ameliorates the decrease in both bone mineral density and bone strength in immobilized rats. *J Bone Miner Metab* 2002; 20: 14–20
30. Tsourdi E, Rachner TD, Rauner M *et al*. Denosumab for bone disease: translating bone biology into target therapy. *Eur J Endocrinol* 2011; 165: 833–840
31. Bone HG, Bolognese MA, Yuen CK *et al*. Effects of denosumab treatment and discontinuation on bone mineral density and bone turnover markers in postmenopausal women with low bone mass. *J Clin Endocrinol Metab* 2011, 96: 972–980
32. Body JJ, Lipton A, Gralow J *et al*. Effects of denosumab in patients with bone metastases with and without previous bisphosphonate exposure. *J Bone Miner Res* 2010; 25: 440–446
33. Rodan GA, Fleisch HA. Bisphosphonates: mechanisms of action. *J Clin Invest.* 1996; 97: 2692–2696
34. Stewart AF. Clinical practice. Hypercalcemia associated with cancer. *N Engl J Med* 2005; 352: 373–379
35. Berenson JR. Treatment of hypercalcemia of malignancy with bisphosphonates. *Semin Oncol* 2002; 29: S12–S18
36. Major P, Lortholary A, Hon J *et al*. Zoledronic acid is superior to pamidronate in the treatment of hypercalcemia of malignancy: a pooled analysis of two randomized, controlled clinical trials. *J Clin Oncol* 2001; 19: 558–567
37. Lin JH. Bisphosphonates: a review of their pharmacokinetic properties. *Bone* 1996; 18: 75–85
38. Markowitz GS, Fine PL, Stack JI *et al*. Toxic acute tubular necrosis following treatment with zoledronate (Zometa). *Kidney Int* 2003; 64: 281–289
39. Tanvetyanon T, Stiff PJ. Management of the adverse effects associated with intravenous bisphsphonates. *Ann Oncol* 2006; 17: 897–907
40. Berenson J, Hirschberg R. Safety and convenience of a 15-minute infusion of zoledronic acid. *The Oncologist* 2004; 9: 319–329
41. Guarneri V, Donati S, Nicolini M *et al*. Renal safety and efficacy of i.v. bisphosphonates in patients with skeletal metastases treated for up to 10 years. *Oncologist* 2005; 10: 842–848
42. Block GA, Bone HG, Fang L *et al*. A single-dose study of denosumab in patients with various degrees of renal impairment. *J Bone Miner Res* 2012; 27: 1471–1479
43. Tabrizi MA, Tseng CM, Roskos LE *et al*. Elimination mechanism of therapeutic monoclonal antibodies. *Drug Discov Today.* 2006; 11: 81–88
44. Body JJ, Facon T, Coleman RE *et al*. A study of the biological receptor activator of nuclear factor-kappa B ligand inhibitor, denosumab, in patients with multiple myeloma or bone metastases from breast cancer. *Clin Cancer Res* 2006; 12: 1221–1228
45. Yonemori K, Fujihiro Y, Minami H *et al*. Phase 1 trial of denosumab safety, pharmacokinetics, and pharmacodynamics in Japanese women with breast cancer-related bone metastases. *Cancer Sci* 2008; 99: 1237–1242
46. Morony S, Warmington K, Adamu S *et al*. The inhibition of RANKL causes greater suppression of bone resorption and hypercalcemia compared with bisphosphonates in two models of humoral hypercalcemia of malignancy. *Endocrinology* 2005; 146: 3235–3243
47. Stopeck AT, Lipton A, Body JJ *et al*. Denosumab compared with zoledronic acid for the treatment of bone metastases in patients with advanced breast cancer: a randomized, double-blind study. *J Clin Oncol.* 2010; 28: 5132–5139
48. Henry DH, Costa L, Goldwasser F *et al*. Randomized, double-blind study of denosumab versus zoledronic acid in the treatment of bone metastases in patients with advanced cancer (excluding breast and prostate cancer) or multiple myeloma. *J Clin Oncol.* 2011; 29: 1125–1132
49. Cummings SR, San Martin J, McClung MR *et al*. Denosumab for prevention of fractures in postmenopausal women with osteoporosis. *N Engl J Med* 2009; 361: 756–765
50. Papapoulos S, Chapurlat R, Libanati C *et al*. Five years of denosumab exposure in women with postmenopausal osteoporosis: results from the first two years of the FREEDOM extension. *J Bone Miner Res* 2011; 27: 694–701
51. Jamal SA, Ljunggren O, Stehman-Breen C *et al*. Effects of denosumab on fracture and bone mineral density by level of kidney function. *J Bone Miner Res* 2011; 26: 1829–1835
52. Brown-Glaberman U, Stopeck AT. Role of denosumab in the management of skeletal complications in patients with bone metastases from solid tumors. *Biologics* 2012; 6: 89–99

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