

Myeloma and Bone Disease

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Abstract

Purpose of Review Bone disease is a defining characteristic of multiple myeloma (MM) and the major cause of morbidity. It manifests as lytic lesions or osteopenia and is often associated with severe pain, pathological fracture, spinal cord compression, vertebral collapse, and hypercalcemia. Here, we have reviewed recent data on understanding its biology and treatment.

Recent Findings The imbalance between bone regeneration and bone resorption underlies the pathogenesis of osteolytic bone disease. Increased osteoclast proliferation and activity accompanied by inhibition of bone-forming osteoblasts leads to progressive bone loss and lytic lesions. Although tremendous progress has been made, MM remains an incurable disease. Novel agents targeting bone disease are under investigation with the goal of not only preventing bone loss and improving bone quality but also harnessing MM tumor growth.

Summary Current data illustrate that the interactions between MM cells and the tumor-bone microenvironment contribute to the bone disease and continued MM progression. A better understanding of this microenvironment is critical for novel therapeutic treatments of both MM and associated bone disease.

Keywords Multiple myeloma · Osteolytic bone disease · Tumor microenvironment · Bone marrow stromal cells · Bisphosphonates

Introduction

With more than 30,300 new cases diagnosed in 2016, multiple myeloma (MM) is the second most common hematological cancer and is the 14th leading cause of cancer death in the USA (<https://seer.cancer.gov/statfacts/html/mulmy.html>). MM is a B cell malignancy characterized by clonal expansion of terminally differentiated, immunoglobulin-producing, transformed plasma cells in the bone marrow (BM) [1, 2]. The evolution of the disease is characterized by an asymptomatic premalignant state, monoclonal gammopathy of undetermined significance (MGUS), and smoldering MM [2]. Smoldering MM is an asymptomatic, precancerous form of myeloma with increased level of plasma cells in bone marrow and/or monoclonal protein but without evidence of end organ involvement. Clinically, MM manifests with hypercalcemia, renal failure, anemia, and/or bone disease (also known as the CRAB criteria). The progression of the disease from a premalignant state into active MM is characterized by the development of osteolytic bone disease (OBD) [3] as one of its disease-defining features. At least 85% of MM patients show some degree of osteopenia [4] at diagnosis, and the severity of bone destruction typically correlates with tumor burden and prognosis [2, 5].

OBD is a consequence of bone homeostasis perturbation. Bone homeostasis is physiologically maintained by the balance between bone formation and bone resorption which is mainly regulated by osteoblasts and osteoclasts, respectively. MM cells are known to secrete factors that enhance osteoclast proliferation and activity while inhibiting osteoblast bone

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formation [6]. Additionally, the myeloma-driven deregulation of the bone compartment creates a permissive microenvironment for MM cell expansion [7]. Since MM burden and progression tightly depend on the interactions of MM cells with the bone microenvironment, therapies aimed at restoring bone homeostasis targeting osteoblasts, osteoclasts, or both hold promise for the treatment of MM. Animal studies have shown that inhibition of osteoclast function or stimulation of bone formation can reduce tumor burden and prevent OBD [8, 9]. Here, we review bone metabolism in the context of MM disease progression and treatment strategies to mitigate the impact of OBD.

Defining the Cellular Interplay in Myeloma-Derived Bone Disease

Bone Marrow Mesenchymal Stromal Cells

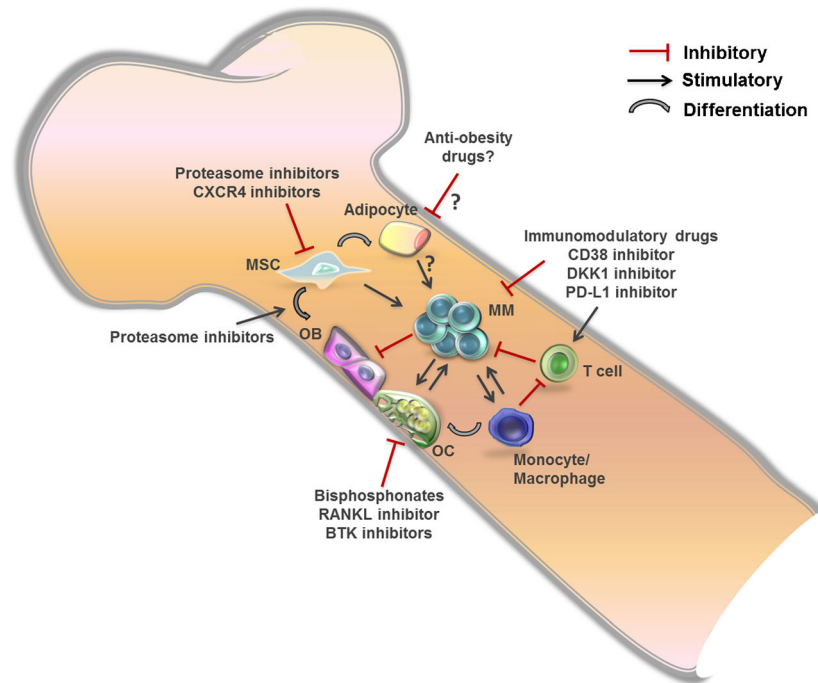
An understanding of the cellular interplay between tumor and the microenvironmental compartment is critical to developing novel therapies for MM (Fig. 1). Bone marrow stromal cells (BMSCs) comprise of cells of hematopoietic (red and white blood cells) and mesenchymal origin (osteoprogenitors, osteoblasts, adipocytes, chondrocytes, endothelial cells, and fibroblasts) [10]. BMSCs indirectly participate in hematopoiesis by secreting cytokines, chemokines, and growth factors such as CXCL12, IL7, LIF, G-CSF, GM-CSF, and IL-6. These factors play a critical role in the maintenance, proliferation, and differentiation of hematopoietic stem and progenitor cells [10–13]. Mesenchymal cells originate from a mesenchymal stem cell (MSC) [14] via differentiation into lineage-specific cells, which is strictly regulated by physical, chemical, and spatial stimuli. B lymphocytes originate and differentiate in the BM, and studies in mice have reported the crucial role of bone cells during B lymphopoiesis and plasma cell recirculation [13]. For example, induced *in vivo* cellular deficiency of osteoprogenitors and preosteoblasts depletes the common lymphoid precursor cells and overall B cell populations in the BM [15], while deletion of the α subunit in the G protein arrests B cell differentiation at the pro-B cell stage [16]. Interestingly, deletion of the parathyroid hormone (PTH) receptor, a G-protein-coupled receptor in osteoprogenitors, arrests the differentiation of the pro-B cell into a mature B cell. It also impairs egress of immature B cells from the BM, resulting in increased accumulation of maturing B cells in the BM. This accumulation occurs specifically along the bone surface in part due to overexpression of the vascular cell adhesion molecule 1 (VCAM1) by stromal and bone cells [17].

MM cells influence the behavior of BMSCs through direct cell-cell contact and via secretion of paracrine factors. Adhesion of the myeloma tumor cells to BMSCs stimulates secretion of BMSC-derived factors (such as BAFF) that, in turn, upregulate expression of anti-apoptotic proteins (e.g.,

MCL-1 and BCL-2) and cell cycle regulating proteins (e.g., serine/threonine kinase Pim-2), which confer chemoresistance to MM cells [18–21]. Cell-cell interactions between BMSCs and MM cells are mediated via adhesion molecules, such as inter-cellular adhesion molecule 1 (ICAM-1) and VCAM-1 on BMSCs and very late antigen 4 (VLA-4) on MM cells. These interactions promote retention of MM cells within the BM and contribute to increased osteolytic bone lesions [22, 23, 24]. In addition to the MM-BMSC cell-cell interactions, secreted factors such as interleukin 6 (IL-6) are also critical promoters of MM progression [25]. Both MM and BMSCs secrete IL-6 upon stimulation of different signaling pathways including NF- κ B and Notch [26–29]. They also secrete other inflammatory cytokines like TNF- α and IL-1 β which can lead to OBD by activating osteoclasts, inhibiting osteoblasts, and increased IL-6 expression [30, 31]. Other factors secreted by both MM cells and BMSCs include vascular endothelial growth factor (VEGF), insulin-like growth factor (IGF-1), IL-1, TGF- β , angiopoietin-1 (Ang-1), platelet-derived growth factor (PDGF), basic-fibroblast growth factor (bFGF), and hepatocyte growth factor (HGF) [32, 33]. These growth factors have been shown to be associated with increased angiogenesis, osteoclastogenesis, and tumor growth in MM [34]. MM patients with advanced bone disease also show higher levels of activin A in the blood [35, 36]. Activin A is an osteoclast-activating factor and inhibitor of osteoblast differentiation but is not secreted by MM cells [36]. Interestingly, MSCs from MM patients, but not healthy donors, were found to secrete activin A, suggesting a possible intrinsic genetic defect in BMSCs of MM patients [36]. Preclinical models showed that targeting activin A reversed osteoblast inhibition and improved MM bone disease. Furthermore, lenalidomide, an immunomodulatory drug commonly used in the treatment of MM, increases activin A secretion from BMSCs, leading to inhibition of osteoblastogenesis. The combination of lenalidomide with activin A-neutralizing antibody could represent a new strategy in managing MM bone disease [37]. BMSCs of MM patients were found to have abnormal gene expression with decreased osteoblastic differentiation potential and aberrantly secrete growth differentiation factor 15 (GDF15), which supports myeloma stem cell survival and self-renewal [38–41]. Together, these findings highlight the crucial role of the BM microenvironment in the establishment and progression of MM [39, 40, 42, 43].

Drugs currently used in the clinic, like the proteasome inhibitors, can target the cross talk between MM and BMSCs. The proteasome inhibitor bortezomib inhibits MM cell growth by impairing MM cell adhesion to BMSCs and by reducing the secretion of cytokines necessary for MM survival [44]. Bortezomib acts on MSCs directly to increase osteoblast differentiation [45]. Another strategy to decrease MM cell adhesion to stroma involves the inhibition of CXCL-12/CXCR-4 axis, which is reinforced by VLA-4/VCAM-1 interaction, by

Fig. 1 Cellular interplay between tumor and the microenvironmental compartment critical to developing novel therapies for MM



using CXCR4 inhibitor AMD3100. Mobilization [36] of MM cells was found to enhance their sensitivity to chemotherapy [46].

Osteolineage Cells

Cells of the osteoblast lineage are the main actors in mineral bone apposition and thus primarily affected in MM bone disease. Differentiation of MSCs into bone cells is regulated by activation of two major transcription factors: runt-related transcription factor 2 (RUNX2) and osterix (OSX) [47]. These transcription factors, essential for the maturation of osteoblasts and ossification, rely on the canonical Wnt signaling pathway. The binding of Wnt proteins to frizzled (FZ) family receptors and lipoprotein receptor-related protein (LRP)-5/6 coreceptors forces the release of β -catenin from the cytosolic destruction complex and is internalized in the nucleus, where it acts as coactivator of transcription factors that belong to the TCF/LEF family [48••]. However, Wnt signaling is inhibited by a variety of Wnt antagonists such as dickkopf-1 (DKK1), sclerostin (SOST), and secreted frizzled-related proteins (sFRPs) that compete for the same Wnt coreceptors [48••]. MM cells produce a variety of Wnt antagonists including DKK1, sRFP2, and sRFP3 [49–51]. High levels of DKK1 have been found in the BM and blood of MM patients and have been correlated with the presence of focal bone lesions [49]. In vitro and in vivo studies have shown that blocking DKK1 and increasing Wnt signaling restored osteoblast number and trabecular bone while decreasing tumor burden [9, 52–54]. A phase IB trial of an anti-DKK1 antibody, BHQ880, in relapsed/refractory MM showed a general trend

towards increased BMD, though interpretation is limited given that BHQ880 was given with zoledronic acid [55].

MM cells have been reported to also secrete SOST [56]. Although we and others were not able to detect SOST secretion from MM patient cells or MM cell lines [57], the level of circulating SOST in MM patients is significantly higher and correlates with advanced MM disease stage and fractures [58, 43]. Osteocytes are responsible for SOST secretion but the number of osteocytes in MM patients is drastically reduced because of osteolytic lesions. We have recently reported that MSCs of MM patients aberrantly secrete SOST, and this may, at least in part, account for the increased circulating SOST levels in MM patients [43, 57]. In vivo treatment of MM-bearing mice with SOST neutralizing antibody showed reduced bone loss and osteolytic lesions, although no effect was seen on tumor burden [59•]. However, the source of SOST secretion and its effects on MM and tumor microenvironment remain under investigation. Together, these results indicate that rescuing osteoblast activity and increasing bone formation in MM patients can be a potential tool to inhibit MM tumor growth and improve MM bone disease.

Adipocytes

Although BM adipocytes have not been directly implicated in MM bone disease, emerging evidence show that adipocytes support MM growth and survival which negatively correlate with bone mass. Adipocyte and osteoblast lineage commitment, arising from common progenitor MSCs, are tightly regulated events in BM. Wnt signaling is a major promoter of osteoblast lineage while suppressing adipocyte lineage

commitment regulated by PPAR γ signaling pathway [60, 61]. Moreover, marrow adiposity increases with age, from 30% of the BM volume in young adults to 70% in elderly people [62••]. Conversely, bone formation rates decrease with aging.

Obesity is positively correlated with increased risk of developing MM and is associated with higher levels of BM adiposity [63–65]. Compared to patients with MGUS, patients with MM had higher abdominal fat cross-sectional area and higher fat metabolic activity as demonstrated by FDG-PET/computed tomography (CT) [66]. A recent *in vivo* study demonstrated that diet-induced obesity promoted a MM-like syndrome in mice, suggesting that even an unhealthy diet may be implicated in the initiation of the tumor [67•]. *In vitro*, BM adipocytes have been shown to support MM growth and survival by secreting adipocyte-specific cytokines (adipokines) (leptin, insulin, adiponin, resistin, etc.) and growth factors [IL-1 β , IL-6, IL-10, IL-12, TNF α , monocyte chemoattractant protein 1 (MCP-1), IGF-1, VEGF, SCF, bFGF] [68–72] and protecting MM cells against chemotherapy-induced apoptosis through autophagy activation [73].

BM adipocytes also secrete lipids in the form of free fatty acids that can be used as energy for tumor cell proliferation [74]. Increased levels of free fatty acids were found in the serum of MM patients when compared to healthy donor serum [75]. Although some specific fatty acids resulted in anti-myeloma effects [76, 77], a recent study implicated lysolipids in the origin of monoclonal gammopathies, including MGUS and MM [78••]. They reported that lysolipids act as antigens for plasma cell-derived antibodies and reactivity to this category of lipids accounts for 33% of sporadic human monoclonal gammopathies. The results suggest that extensive immune activation due to lysolipids could potentially represent an initiation factor in sporadic monoclonal gammopathies and could represent novel biomarkers and targets to prevent disease progression. Many lipids also act as ligands for the nuclear receptors involved in PPAR γ signaling pathway. Therefore, increased levels of free fatty acids in MM patients likely lead to upregulation of PPAR γ signaling, which in turn enhances BM adipogenesis and MM survival while inhibiting osteoblast differentiation. A recent *in vitro* study demonstrated the direct lipotoxic effect of saturated fatty acids, stearate and palmitate, on human osteoblasts [79, 80]. Data also demonstrated that augmented adipogenesis during obesity and aging impaired bone regeneration [81]. Taken together, BM fat is emerging as an important regulator of MM development and bone disease and, therefore, may represent a new promising therapeutic target.

Osteoclasts

The pathogenesis of MM-induced bone disease underlies primarily in upregulation of osteoclast differentiation and activity that results in unbalanced bone resorption giving rise to

characteristic osteolytic lesions [82]. Positive correlations were found among osteoclast numbers, resorptive surface, and disease progression [5, 83, 84]. Several factors mediate osteoclast activation in MM patients. MM cells directly secrete cytokines with osteoclastogenic potential, including IL-1, IL-3, IL-6, TNF α , MIP-1 α , MIP-1 β , and decoy receptor 3 (DcR3) [85–90].

MSCs and bone cells are also a source of osteoclast-activating cytokines. In addition to BAFF, activin A, CXCL12, IL-6, and VEGF, two proteins produced by bone cells play a central role in osteoclast differentiation: the receptor activator of nuclear factor kappa-B ligand (RANKL) and osteoprotegerin (OPG) [91–93]. Adhesion of MM cells to BMSCs stimulates RANKL expression by osteoblasts which promote osteoclast differentiation through stimulation of NF- κ B and JunN-terminal kinase pathways [94]. In addition, RANKL is able to inhibit osteoclast apoptosis. Denosumab is a human monoclonal antibody that binds to RANKL with high affinity resulting in rapid and extensive bone resorption inhibition but also MM burden modulation [95]. Denosumab is approved for the treatment of bone metastases due to solid tumors (see below). OPG is significantly decreased and correlates with advanced bone disease in MM patients [96]. A high RANKL/OPG ratio is associated with a worse prognosis [5]. Treatments aimed to normalize the RANKL/OPG ratio by increasing OPG and/or reducing RANKL were effective in arresting bone destruction and MM growth *in vivo* [97–100].

High levels of the pro-inflammatory cytokine MIP-1 α , mainly secreted by MM cells and osteoclasts, correlate with bone disease and poor prognosis [101]. MIP-1 α promotes MM cell survival and migration through activation of ERK and AKT signaling, stimulates osteoclastogenesis, and inhibits osteoblastic differentiation by downregulating osterix [102–104]. Similar to MIP-1 α , high expression of Bruton's tyrosine kinase (BTK) in MM cells and osteoclasts, fundamental to B-lymphocyte development as well as osteoclast differentiation, has been implicated in MM cell growth and increased bone resorption [105, 106]. The BTK inhibitor ibrutinib demonstrated cytotoxicity on MM cells and was effective in reducing osteolysis through inhibition of the NF- κ B pathway [107, 108]. The combination of the Btk inhibitor, CC-292, with the proteasome inhibitor carfilzomib *in vivo* showed a synergistic effect in reducing the tumor burden and increasing bone volume [109]. In addition to their fundamental role in bone turnover, osteoclasts are implicated in a variety of functions [110, 111]. Osteoclasts are terminally differentiated cells of the monocyte/macrophage lineage and as all cells of the innate immune system, share similar immune receptors and activities. They interact directly with T lymphocytes and activate them in the context of autoimmune disease [112]. Moreover, while T cells produce potent osteoclastogenic cytokines (RANKL and IL-1 β), osteoclasts are able to suppress T lymphocyte proliferation in order to maintain a balance in bone remodeling [113].

Cells of the myeloid lineage, including macrophages, dendritic cells, and other myeloid-derived suppressor cells (MDSCs), have been implicated in the establishment of a protective environment for MM cells by suppressing killer T cells and preventing chemotherapy-induced apoptosis [114–116]. A recent study demonstrated that osteoclasts protect MM cells against T cell-mediated cytotoxicity via direct inhibition of proliferating T cells [117]. Because defective T cell-mediated response is a key mechanism for tumor escape from immunological surveillance [118], the immunosuppressive function of osteoclasts and their crucial role in osteolytic disease need to be considered primary targets in the treatment of MM.

Imaging of Multiple Myeloma

Traditionally, imaging using plain films, i.e., the skeletal survey, has been used for the initial assessment of bone disease [119]. Conventional radiography has the advantage of cost and widespread availability. However, a significant limitation is that 30–50% of bone can be destroyed before it can be detected by this modality [120]. Cross-sectional imaging by CT, MRI, or PET-CT have increased sensitivity and are increasingly used for assessing bone disease with the aim of detecting disease at an earlier time point [121].

Whole-Body Low-Dose CT

CT overall has a 4–33% higher detection rate for picking up lytic lesions compared to conventional radiography [122]. To take advantage of the increased sensitivity of CT and to minimize radiation exposure, whole-body low-dose CT (WBLDCT) protocols have been developed [123]. The radiation exposure from WBLDCT is minimal, about double that of the skeletal survey (4.1 versus 2.4 mSV) [124] versus an estimated 21 mSV for conventional chest, abdomen, and pelvic CT [125]. WBLDCT is more sensitive than conventional radiography for detecting lesions in the spine, pelvis, and thoracic cage, and in one study, led to change in treatment in 18.2% patients [126] and in another study, detected lytic lesions in an additional 23% of patients with plasma cell disorders [127]. In a recent retrospective study by the International Myeloma Working Group, 21.4% of patients who were considered to have smoldering multiple myeloma by skeletal survey were upstaged to active multiple myeloma through WBLDCT [128]. In addition to increased sensitivity, WBLDCT has the advantages of speed and convenience and has replaced the skeletal survey at some European institutions [124].

MRI

MRI plays a key role in the imaging of MM due to its sensitivity for detecting BM involvement in contrast to conventional radiography or CT, which are better suited for detecting

lytic lesions [129]. Several patterns of bone marrow involvement have been described, including a normal appearance, focal involvement, diffuse infiltration, and a variegated pattern [130]. MRI is also more sensitive than plain films for detecting lesions; for example, MRI detected lesions in 74% of MM patients compared to 56% by skeletal survey. In the spine, MRI revealed more lesions than the skeletal survey by conventional radiography, 78 versus 16% [131]. This is clinically relevant, as identification of more than one focal lesion by MRI in patients traditionally classified as smoldering MM is associated with a high risk of progression to active disease: 70% progression in 2 years [132]. Furthermore, MRI findings change in response to treatment; there was a strong relationship between response to treatment and changes in diffuse and focal patterns in one study [133]. Due to the prognostic relevance of findings on MRI, the definition for active multiple myeloma has been recently updated to include more than one focal lesion (≥ 5 mm) seen on MRI [134].

PET/CT

Functional imaging using 18 fluorodeoxyglucose (18F-FDG) PET/CT has also emerged as a valuable tool for assessing bone disease, particularly assessing the burden of disease and for distinguishing between active and inactive lesions. It already is a standard component for diagnosis and assessment of response in patients with lymphoma and solid tumors. The IMWG has recently described guidelines for its use [135]. An advantage of PET/CT over MRI is the ability to evaluate disease outside the field of view of MRI as well as evaluating extramedullary sites of disease. PET/CT plays an important role in staging plasma cell dyscrasias, for example, confirming a solitary site of disease in solitary plasmacytoma versus active multiple myeloma. Recently, the Intergroupe Francophone du Myélome (IFM) compared PET/CT and MRI in a prospective randomized study in newly diagnosed MM patients [136]. This study found that normalization of PET/CT following induction treatment was associated with significant improvement in progression-free survival. In contrast, normalization of MRI did not have prognostic value. This suggests that PET/CT may play a larger role in the future for assessing response to treatment and be a potential prognostic factor.

Bisphosphonates

Bisphosphonates are a cornerstone of treating and preventing bone disease in MM [137]. Bisphosphonates are related to pyrophosphate and are characterized by a P-C-P backbone. They deposit in bone and reduce osteoclast activity by inhibiting farnesyl pyrophosphate synthase [138]. In MM, the development of higher-potency bisphosphonates such as pamidronate and zoledronic acid, which are given intravenously, was a

significant advance for relieving pain due to bone disease and preventing skeletal-related events (SREs) such as pathologic fracture, irradiation or surgery on bone, or spinal cord compression [139–141]. Pamidronate was the first bisphosphonate to show a benefit in MM. In patients with at least one osteolytic lesion, pamidronate given every 4 weeks significantly reduced SREs compared to placebo, 24 versus 41% ($p < 0.001$) after 9 cycles, respectively [139] (Table 1). Importantly, pamidronate also significantly improved quality of life, with decreases in pain scores seen within a month. Zoledronic acid is another bisphosphonate that is also given monthly and is more potent than pamidronate. For treatment of hypercalcemia of malignancy, zoledronic acid was superior to pamidronate [142]. In MM, the efficacy of zoledronic acid in preventing SREs was comparable to pamidronate [143], though zoledronic acid has the main practical advantage of a shorter infusion time compared to pamidronate.

In addition to playing an important supportive role, bisphosphonates may have an anti-MM effect. The MRC Myeloma IX trial is a key trial that compared zoledronic acid to oral clodronate in patients with newly diagnosed MM [144••]. This study found that zoledronic acid reduced mortality by 16% and had superior overall survival, 50 versus 44.5 months with clodronate ($p = 0.04$). There also was a lower incidence of SREs with zoledronic acid, 27 versus 35% ($p = 0.0004$) compared to clodronate [145]. Moreover, patients who did not have bone lesions at baseline also had fewer SREs with zoledronic acid [145], though the benefit in survival was seen only in patients with bone disease on study entry [146].

Based on these findings, the National Comprehensive Cancer Network [147] and the International Myeloma Working Group [148] recommend bisphosphonates (pamidronate or zoledronic acid) for all patients with symptomatic MM. The IMWG guidelines note that the optimal duration of bisphosphonate treatment has not been determined for patients who achieve at least a very good partial response; in these patients, bisphosphonates should be given for at least 12 months and up to 24 months. For patients who achieve less than a VGPR, the group recommended treatment until disease progression and further continued at relapse.

Osteonecrosis of the Jaw

Osteonecrosis of the jaw (ONJ) is one of the most serious adverse events of bisphosphonates [149, 150]. ONJ is traditionally defined as exposed, necrotic bone in the jaw that does not heal after 8 weeks, and it is generally painful. The highest risk of ONJ occurs with zoledronic acid due to its increased potency, and initial studies showed an incidence of 4–11% which increases with duration of exposure [151, 152]. In the Myeloma IX trial, the cumulative incidence of ONJ was 3–4% at a median follow-up of 3.7 years [153].

A major risk factor for ONJ is dental extraction [152, 154]. Attention to dental hygiene and minimizing invasive procedures (such as tooth extractions or dental implants) may reduce the risk of ONJ [155]. IMWG guidelines recommend holding bisphosphonate therapy before and after invasive dental procedures for 90 days (though routine dental cleanings and procedures, including root canals, may occur) [148]. Treatment of ONJ is supportive. After healing has occurred, the IMWG recommends resuming bisphosphonate treatment.

Atypical Femur Fracture

Atypical femur fractures have been better appreciated in patients undergoing oral bisphosphonate therapy for osteoporosis, but are now being seen in MM patients treated with pamidronate or zoledronic acid [156]. These represent isolated cases, as the incidence of atypical femur fracture remains to be determined, as these femur fractures are typically assumed to be pathologic.

Renal Toxicity

Nephrotoxicity is another limiting factor in bisphosphonate use [157]. Bisphosphonates are excreted unchanged in the urine, and impaired renal function can lead to accumulation of the drug. With zoledronic acid, the risk of kidney injury (in this case defined as a rise in creatinine of 0.5 mg/dL) was appreciated in the initial phase III trials, prompting a dose reduction from 8 to 4 mg and increase in the duration of the infusion from 5 to 15 min [140]. Acute tubular necrosis is the main pathology observed. The American Society of Clinical Oncology provides guidelines for the dosing of zoledronic acid in patients with renal impairment [158]. Notably, zoledronic acid is not recommended in patients with severe renal impairment (creatinine clearance <30 mL/min). In the Myeloma IX trial, the rate of acute renal failure was 5–7% in the zoledronic acid arm, similar to 6% in the clodronate arm [144••].

Effect of Myeloma Therapy on Bone Disease

Another key component in managing the bone disease is an effective treatment of the underlying disease. The landscape of MM treatment has changed dramatically in the past decade, with the introduction of two highly effective classes of drugs, immunomodulatory drugs (e.g., lenalidomide, pomalidomide) and proteasome inhibitors (e.g., bortezomib, carfilzomib), along with the recent approval of drugs with novel mechanisms of action, such as the monoclonal antibody targeting CD38 (daratumumab) [159]. The use of these novel agents has been responsible for significant improvements in overall survival [160, 161], and patients diagnosed more recently in

Table 1 Randomized trials of bone-directed agents in multiple myeloma

	Eligibility	Treatment	N	Median time to first SRE	ONJ	Renal toxicity	SRE incidence
Berenson et al. [139]	At least one osteolytic bone lesion; newly diagnosed and relapsed	Pamidronate	196				24% ^a
		Placebo	181				41% ^a
Rosen et al. [140]	At least one osteolytic bone lesion; newly diagnosed and relapsed	Zoledronic ^b acid	129	12.5 months			
		Pamidronate	65	9.4 months			
Gimsing et al. [190]	Newly diagnosed patients	Pamidronate 30 mg	252	10.2 months	0.8%		33.7%
		Pamidronate 90 mg	252	9.2 months	3.2%		35.2%
Morgan et al. [144••, 145]	Newly diagnosed patients	Zoledronic acid	981		4%		27%
		Clodronate	979		<1%		35%
Himelstein et al. [195]	Newly diagnosed patients with at least one bone lesion ^c	Zoledronic acid q12 weeks	139		1% ^c	15.5% ^c	55%
		Zoledronic acid q4 weeks	139		2% ^c	19.9% ^c	60%
Raje et al. [181••]	Newly diagnosed patients with at least one bone lesion	Denosumab	859	22.8 months	4.1%	10%	43.8%
		Zoledronic acid	859	24 months	2.8%	17.1%	44.6%

^a After 9 cycles of treatment

^b Includes cohorts treated with zoledronic acid 4 and 8 mg with subsequent dose reduction to 4 mg for renal safety

^c Multiple myeloma cohort in study that included patients with breast cancer and prostate cancer; ONJ in overall cohort and renal toxicity in overall cohort, defined as increase in creatinine 0.5 mg/dL if baseline 1.4 mg/dL or less and 1 mg/dL if baseline was greater than 1.4 mg/dL

2012 were 25% more likely to survive to 2 years than those diagnosed only 6 years previously [162].

Improvements in treatment have also correlated with improvements in bone-related outcomes. For example, in a subset analysis of the previously described Myeloma IX trial (which randomized patients to zoledronic acid versus pamidronate), the frequency of SREs was lower by 26% in patients randomized to the more effective treatment arm with cyclophosphamide, thalidomide, and dexamethasone compared to the melphalan and prednisone arm [146]. Additionally, the depth of response may influence the impact of bisphosphonate therapy. In a retrospective analysis of the Myeloma IX trial, in patients who have undergone autologous stem cell transplant, the benefit in SRE and overall survival of zoledronic acid versus clodronate was mainly seen in patients who did not achieve a complete response [163].

Moreover, the proteasome inhibitor class has been shown to have bone anabolic effects. Preclinical studies show that proteasome inhibitors such as bortezomib and carfilzomib stimulate osteoblast growth and differentiation and inhibit osteoclast activity [164, 165]. An earlier study showed that patients receiving the combination of bortezomib and dexamethasone (and who also received zoledronic acid) showed an improvement in lumbar spine bone mineral density [166]. In the phase III VISTA study comparing the combination of bortezomib (Velcade) with melphalan and prednisone (VMP) versus melphalan and prednisone (MP) in newly diagnosed transplant-ineligible patients, bone disease and need for radiation therapy was less frequent in the bortezomib arm

[167]. There was also radiological evidence of bone healing in some of the patients treated with VMP (versus none of the patients treated with MP) where data was available. Similarly, consolidation with bortezomib, thalidomide, and dexamethasone following autologous stem cell transplant was shown to reduce bone resorption and was associated with a very low incidence of SREs; notably, bisphosphonates were not used [168]. However, in a recent phase II study specifically investigating the effect of consolidation with bortezomib versus observation on bone disease as a primary endpoint in patients who had a partial response or better after autologous stem cell transplant did not see an effect on bone mineral density [169]. Interpretation of these findings is limited as a substantial proportion of patients in both arms previously received bortezomib upfront, and zoledronic acid use was significantly higher in the control arm.

In contrast, the beneficial effect of lenalidomide, a widely used immunomodulatory drug, on bone disease is less certain. A retrospective study of treatment with lenalidomide and dexamethasone in relapsed disease showed that reduction of bone resorption biomarker CTX (carboxy-telopeptide fragments of collagen type-I $\alpha 1$ chains) only occurred in patients responding to treatment, and the treatment did not have any effect on bone formation [170]. On the other hand, the addition of bortezomib to lenalidomide and dexamethasone was associated with increased bone formation markers. The lack of apparent benefit of lenalidomide on bone formation may be mediated by activin A, a member of the TGF- β superfamily that is most commonly associated with embryogenesis and

gonadal hormone signaling, but is also involved in bone remodeling and osteoclast formation. This has motivated the study of inhibiting activin A with sotatercept (ACE-011), an activin type IIA receptor fusion protein that binds to activin A, in combination with standard treatment with melphalan, prednisolone, and thalidomide [171] or lenalidomide or pomalidomide in patients with relapsed MM (NCT01562405) [172].

Denosumab

A newer agent for managing bone disease is denosumab. Denosumab is a fully human monoclonal antibody given subcutaneously that targets RANKL. As described above, RANKL is a cytokine produced by osteoblasts that activates the RANK receptor present on osteoclast precursors and osteoclasts and promotes the formation, function, and survival of osteoclasts [173]. Since it is a monoclonal antibody, unlike bisphosphonates, denosumab does not accumulate or persist in bone. It has a circulatory half-life of approximately 26 days, and like other monoclonal antibodies, the clearance of denosumab is through the reticuloendothelial system and importantly does not depend on renal clearance [174].

Like zoledronic acid, denosumab is approved for the prevention of SREs in patients with bone metastases due to solid tumors based on several phase III clinical trials [175]. In these studies, patients were randomized to denosumab 120 mg subcutaneously versus zoledronic acid 4 mg intravenously (or equivalent creatinine clearance adjusted dose) every 4 weeks. In two of the studies, denosumab was superior to zoledronic acid in delaying SREs [176, 177]. Similar to zoledronic acid, the risk of ONJ was seen in about 1.8% of patients [178].

However, in a third study, the 244 study, about 10% of the population were MM patients, and an ad hoc analysis showed that survival was unexpectedly worse in the MM cohort treated with denosumab compared to zoledronic acid [179]. Consequently, denosumab is not approved in MM (at the time of this writing). However, conclusions from the study about the MM cohort are limited [180]. Given the small number of MM patients, there were imbalances between the study arms that favored the zoledronic acid arm; the denosumab arm had more patients with renal dysfunction, and the zoledronic acid arm patients were treated more often with novel agents and autologous stem cell transplantation. In addition, there was unequal early withdrawal censoring that may have favored the zoledronic acid arm. These findings confounded the interpretation of outcomes in the MM cohort.

To address these limitations in the 244 study, a larger phase III study enrolling only patients with MM was recently conducted, and presented at the International Myeloma Workshop [181••]. This is the largest international study in MM, randomizing 1718 newly diagnosed patients with at least one bone lesion to denosumab versus zoledronic acid, both given

according to standard q4 week schedules. Patients were stratified by type of therapy, intent for autologous stem cell transplant, ISS stage, and region. The primary endpoint was time to SRE, defined as pathologic fracture (vertebral or non-vertebral), need for radiation therapy or bone surgery, or spinal cord compression. In this study, denosumab was non-inferior to zoledronic acid (22.83 versus 23.98 months); overall survival was similar in both arms. Interestingly, progression-free survival, an exploratory endpoint, was longer in the denosumab arm (46.09 months) compared to the zoledronic acid arm (35.38 months), $p = 0.036$. Adverse events of interest include ONJ, which was 4.1% in the denosumab arm compared to 2.8% in the zoledronic acid arm. Importantly, the incidence of renal toxicity was significantly lower with denosumab, 10 versus 17.1% ($p < 0.001$), especially in patients with renal insufficiency at baseline (creatinine clearance ≤ 60 mL/min), 12.9 versus 26.4%, respectively. Additionally, there were fewer acute phase reactions in the denosumab group, 5.4 versus 8.7%. Overall, these findings may change practice, given that kidney dysfunction is common in MM and often presents a major barrier to effective use of osteoclast-targeted therapy. Indeed, denosumab may be able to play a key role for managing and preventing bone disease in this challenging patient population. The improved PFS observed in the denosumab arm warrants further evaluation.

Vertebroplasty and Kyphoplasty

Vertebroplasty (injection of methyl methacrylate or bone cement) and kyphoplasty (use of an inflatable balloon followed by instillation of bone cement) are percutaneous procedures for treating compression fractures, which are common in MM [182, 183]. The Cancer Patient Fracture Evaluation study randomized 134 patients with malignancy and at least one vertebral compression fracture to balloon kyphoplasty versus non-surgical management [184]. Forty percent of the patients in the study had MM, and at 1 month, there was significant improvement in back pain and quality of life. A recent retrospective series showed durable improvement in disability which persisted at 1 year and also noted better outcomes in patients after treatment was initiated [185]. These procedures can play a key adjunctive role for managing bone-related pain.

Palliative Radiation Therapy

Radiation also plays an important role for palliation of painful bony lesions in MM. An estimated 38% of patients are expected to receive radiation over the course of their illness [186], and this figure remains similar in the era of novel treatments [187]. The primary indication for the use of radiation therapy is treating bone pain, and other indications include impeding fracture, cord compression, or relief of symptoms associated with a mass (i.e., cranial nerve palsies or organ or joint dysfunction).

Minimal doses of radiation can be used, given the radiosensitivity of plasma cell lesion [188]. Doses of 20–35 Gy can be used, but it is essential to consider ability to retreat when designing treatment fields, particularly of the spine. In our experience, we have found that 20 Gy delivered in either five or ten fractions provide adequate symptom relief [189]. A retrospective series found that 76.4% of patients reporting complete relief of pain with palliative radiation with a median dose of 30 Gy [187].

Ongoing Areas of Study

An ongoing question is the amount of bisphosphonate necessary for improving bone disease. This is especially true given that current MM therapies are increasingly more effective than when the initial studies with bisphosphonates were conducted as well as concerns for adverse events such as ONJ with chronic exposure.

A lower dose of pamidronate was evaluated by the Nordic Myeloma Study group, which compared a 30 mg dose of pamidronate versus the standard dose of 90 mg in a double-blind, randomized study [190]. The rates of SREs were similar, and there was a trend towards less adverse events in the 30 mg dose group with fewer episodes of ONJ and nephrotoxicity.

With zoledronic acid, its monthly dosing schedule is based on suppression of urine N-telopeptide of type I collagen (NTX), a biomarker for bone resorption, for 4 to 8 weeks after a single dose of zoledronic acid [191, 192]. To further understand the duration of bone resorption suppression, a study measured urine NTX in 29 patients who achieved at least a partial response and who previously received bisphosphonate therapy. In these patients, urine NTX (uNTX) continued to be suppressed in nearly all these patients for 6 months after a single dose of zoledronic acid, and this correlated with freedom from SRE [193], suggesting that the zoledronic acid could be given less frequently.

These findings were further supported in the Z-MARK study, which evaluated less frequent zoledronic acid dosing based on urine NTX levels in patients who had already received at least 1 year of bisphosphonate therapy [194]. For patients where uNTX <50 nmol/mmol creatinine, zoledronic acid was given every 12 months; otherwise, it was given every 4 weeks. This study enrolled 121 patients, and nearly all patients, 117 patients, were assigned to the q12 week arm. The SRE rate was low, 5.8% in the first year and 4.9% in the second year, and the 2-year incidence of ONJ was 3.3%. These findings suggest that less frequent dosing is feasible with low risk of SRE.

Evaluation of less frequent dosing has been evaluated in a phase III study, CALGB 70604, which randomized bisphosphonate-naïve metastatic breast cancer and prostate cancer, and MM patients with at least one bone lesion to every 12

week dosing of zoledronic acid versus every 4 week dosing [195]. In the MM cohort, which comprised 15.3% of the study, the SRE rate was similar; 55% of patients had an SRE in the every 12 week arm versus 60% in the every 4 week arm. In the overall group, the rate of kidney dysfunction, defined as increase ≥ 0.5 mg/dL over baseline if baseline creatinine ≤ 1.4 or ≥ 1 mg/dL if baseline >1.4 mg/dL, was higher in the every 4 week arm, 19.9 versus 15.5% ($p = 0.02$). The ONJ rate in the overall cohort trended towards higher in the every 4 week arm, 2 versus 1% ($p = 0.08$). A limitation of the study is the high dropout rate due to patient withdrawal, with only 47% of patients with two or more years of treatment. Overall, this study adds support for every 12 week dosing, though the effect of this approach on overall survival remains to be determined.

Finally, patients with severe kidney dysfunction (creatinine clearance <30 mL/min) have an unmet need for managing bone disease, as current guidelines do not recommend use of zoledronic acid. Denosumab, given its lack of nephrotoxicity and lack of dependence on renal clearance, is currently being evaluated in a phase II study in this patient population (NCT02833610).

Conclusions

Here, we discuss the complexity of the cross talk between MM and the surrounding BM stromal microenvironment. Although the overall survival and quality of life of MM patients have significantly improved due to the introduction of novel therapeutic strategies, MM remains an incurable disease. MM progression and resistance tightly depend on the interactions with the stromal compartment; hence, treatments targeting this interplay are fundamental for the cure of MM and osteolytic bone disease. Osteoclast-targeted therapy with bisphosphonates has become an integral part of supportive therapy and plays a significant role in addressing the bone disease. Newer drugs, such as denosumab and other bone anabolic agents, are currently under investigation and may represent promising tools in the treatment of MM-derived bone disease. Further understanding of the MM stromal microenvironment and the development of novel agents targeting them are therefore needed.

Compliance with Ethical Standards

Conflict of Interest Cristina Panaroni and Andrew Yee declare no conflict of interest.

Noopur Raje reports payment for work as a consultant for Amgen and Novartis, outside the submitted work.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

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