Regular Article

Incidence and Risk Factors of Osteonecrosis of the Jaw in Advanced Cancer Patients after Treatment with Zoledronic Acid or Denosumab: A Retrospective Cohort Study

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Zoledronic acid and denosumab are two antiresorptive drugs currently in use for treating osteoporosis. They have different mechanisms of action, but both have been shown to delay the onset of skeletal-related events in patients with advanced cancer. However, medication-related osteonecrosis of the jaw (MRONJ) has been reported in cancer patients treated with zoledronic acid or denosumab. We studied 155 patients with several types of advanced cancer who were treated with zoledronic acid or denosumab in our hospital during the period from April 2010 through March 2013. Thirteen of these 155 patients (8.4%) developed MRONJ. MRONJ development was significantly associated with the number of zoledronic acid or denosumab infusions ($p<0.001$) and the duration of zoledronic acid or denosumab therapy ($p<0.001$). Logistic regression analysis showed that diabetes [odds ratio (OR)=6.699, 95% confidence interval (CI), 1.435–31.277, $p=0.016$], anemia [OR=14.559, 95% CI, 2.161–98.069, $p=0.006$], and pus discharge [OR=6.491, 95% CI, 1.514–27.835, $p=0.012$] significantly increased the risk of developing MRONJ. However, the risk of MRONJ was significantly lower [OR=0.137, 95% CI, 0.020–0.944, $p=0.043$] when patients received periodical dentistry maintenance. Diabetes, anemia, and pus discharge may also play roles in its development. These findings suggest that the active inclusion of dentistry maintenance in bisphosphonate or denosumab treatment of cancer patients can reduce MRONJ development.

Key words medication-related osteonecrosis of the jaw (MRONJ); zoledronic acid; denosumab; periodical dentistry maintenance

Antiresorptive agents that target osteoclasts, thereby inhibiting bone resorption and subsequent bone loss, are currently considered the cornerstone of osteoporosis prevention and treatment. Bisphosphonates currently represent the first-line antiresorptive agents for the management of postmenopausal osteoporosis, with zoledronic acid being considered the most potent.[1)] Recently, denosumab has been launched for the treatment of postmenopausal osteoporosis. The use of either zoledronic acid or denosumab decreases bone turnover markers, improves bone mineral density, and decreases the risk of vertebral, nonvertebral, and hip fractures.[2,3)]

Bisphosphonates are a class of drugs that prevent the loss of bone mass. Intravenous bisphosphonates are primarily used and effective in the treatment and management of cancer-related conditions. Bisphosphonates are known inhibitors of bone resorption[4)] and angiogenesis.[5,6)] They are used in the treatment of various medical conditions, including osteoporosis, multiple myeloma, and breast cancer with skeletal metastasis, because they reduce bone pain, hypercalcemia, and the risk of pathologic fractures.[7–11)] The use of bisphosphonates to prevent skeletal-related events (SREs) in patients with bone metastatic cancer has been associated with the occurrence of medication-related osteonecrosis of the jaw (MRONJ), due to excessive inhibition of bone turnover.[12)] Zoledronic acid has been considered the standard of care and has been shown to prolong the time to first SRE and reduce the number of SREs; however, many patients with bone metastases continue to experience SREs. In a pivotal phase 3 trial, zoledronic acid demonstrated efficacy in preventing skeletal complications in patients with bone metastases secondary to solid tumors other than breast cancer.[13)]

Denosumab is a subcutaneously dosed monoclonal antibody against receptor activator of nuclear factor-κB ligand (RANKL) for the prevention of SREs in patients with solid tumors metastatic to bone.[14)] Three international, randomized, double-blind, double-dummy phase 3 studies have evaluated denosumab *versus* zoledronic acid for the delay and treatment of SREs in breast and prostate cancers, and in combined solid tumors and multiple myeloma.[15–17)] Denosumab's superior efficacy over zoledronic acid was demonstrated in the studies of patients with advanced breast or prostate cancer, as well as in a prespecified integrated analysis of all patients enrolled across the three studies.[15,16,18)] Findings from both preclinical studies and trials of patients with bone metastases suggest that tumor-induced bone destruction is largely caused by the activation of bone-resorbing osteoclasts.[19)]

In 2014, the term MRONJ was introduced, replacing the common expression bisphosphonate-related osteonecrosis of the jaw (BRONJ), as this condition can also result from treatment with other antiresorptive (denosumab) and antiangiogenic therapies. MRONJ was first reported in patients with a broad range of cancers receiving chemotherapy and intra-

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venous bisphosphonates in the early 2000s. In retrospective studies, various incidences of osteonecrosis of the jaw (ONJ) have been reported.[20–24] A recent updated position paper (from 2014) issued by the American Academy of Oral and Maxillofacial Surgeons (AAOMS) defined medication-related ONJ as cases in which all of the following 3 characteristics are present: 1) current or previous treatment with antiresorptive or antiangiogenic agents, 2) exposed bone or bone that can be probed through an intraoral or extraoral fistula in the maxillofacial region that has persisted for longer than 8 weeks, and 3) no history of radiation therapy to the jaws or obvious metastatic disease to the jaws.[25] A recent meta-analysis reported a mean incidence of MRONJ of 6.1% in these patients.[26] However, this proportion can be reduced with a screening dental visit before the start of therapy and the avoidance of invasive procedures, such as dental extractions.[27,28] MRONJ is a well-recognized complication associated with bisphosphonates or monoclonal antibody of the immunoglobulin G_2 (IgG_2) subtype targeting RANKL therapy.[20,29–32] It presents as exposed bone in the maxillofacial area showing delayed signs of healing.

MRONJ results from bone exposure in the oral cavity with subsequent necrosis, often following dental procedures or traumatic injuries.[33] However, some patients develop MRONJ without invasive dental procedures during treatment with bisphosphonates or denosumab. Therefore, the true incidence, etiology, and risk factors that contribute to MRONJ pathogenesis are unknown in cancer patients. The resulting knowledge gap has impaired the determination of individual susceptibility and the development of preventative measures against MRONJ. The primary objectives of this study were thus to estimate the frequency of MRONJ in patients treated with zoledronic acid or denosumab, to understand better the clinical presentation of MRONJ, and to identify risk factors associated with the development of osteonecrosis in these patients. We conducted a retrospective study to determine the frequency and risk factors of MRONJ in patients with cancer treated with bisphosphonates or anti-RANKL antibody.

PATIENTS AND METHODS

Patients Firstly, we extracted information of patients who were administered zoledronic acid (Zometa®; Novartis Pharmaceuticals Corporation, East Hanover, NJ, U.S.A.) or denosumab (Ranmark®; Daiichi Sankyo, Tokyo, Japan) between April 2010 and March 2013 at Okayama University Hospital. A total of 164 patients were found, and we investigated all administration records of these drugs retrospectively (not limited to 2010). On the basis of this investigation, a total of 155 consecutive patients who had received zoledronic acid or denosumab for cancer management at least 2 times were enrolled in this study, and a retrospective cohort study was performed. This study was approved by Okayama University Graduate School of Medicine, Dentistry, and Pharmaceutical Sciences Ethics Committee (No. 883).

Zoledronic Acid or Denosumab Administration Zoledronic acid was administered at 4 mg every 3 to 6 weeks over 15 min, and denosumab was administered at 120 mg every 4 to 5 weeks, by subcutaneous injection. Zoledronic acid dose adjustment was carried out based on the patient's calcium level and/or renal function.

ONJ Diagnosis The MRONJ diagnosis was determined from the clinical and radiographic findings. MRONJ was diagnosed by dentists in Okayama University Hospital according to the description in the AAOMS position paper.[25] Details are described in Introduction. Suspicious MRONJ lesions were identified through medical records containing documentation of exposed bone, osteomyelitis, or delayed healing in the jaw bones.

Evaluated Risk Factors We evaluated risk factors as follows: anti-cancer drugs (in use/not in use), smoking (current or former smoker *versus* non-smoker), tooth mobility, complaint of toothache, oral dryness, pus discharge from gingiva, dental care, stomatitis, diabetes, anemia (Hb <10 g/dL), obesity (body mass index $\geqq 25$), and zoledronic acid or denosumab infusions before the treatment of zoledronic acid or denosumab.

Statistical Analysis All analyses were performed using SPSS statistical software (SPSS for Windows, version 21; SPSS Inc., Chicago, IL, U.S.A.). The χ^2 test was used for comparisons of proportions across levels of categorical variables. For continuous variables, the *t*-test or Wilcoxon rank sum test was used as appropriate. To analyze possible associations between clinical and demographic characteristics with the occurrence of MRONJ, we used a two-tailed Fisher's exact test. To adjust the analysis for possible confounding factors, we used a logistic regression model that only included variables that presented an association with the occurrence of MRONJ. This was done by univariate statistical analysis with $p \leqq 0.20$. Logistic regression was used to investigate multivariate associations with the development of MRONJ. Throughout the analysis, a level of 5% was used to denote statistical significance.

RESULTS

Patients The baseline characteristics of the 155 patients included in our analysis are listed in Table 1. Other diagnoses included cancer of the colon ($n=13$), sarcoma ($n=8$), kidney ($n=7$), stomach ($n=3$), esophagus ($n=1$), liver ($n=1$), ovary ($n=1$), and unknown origin ($n=3$). One hundred and thirteen patients were treated with zoledronic acid, 16 patients were treated with denosumab, and 26 patients received both zoledronic acid and denosumab. There were no significant differences in age ($p=0.161$) or sex ($p=0.071$) of the patients with or without MRONJ. A total of 13 MRONJ cases (8.4%) were identified. MRONJ was noted in 9 of the 59 patients (15.3%) with breast cancer, 2 of the 47 patients (4.3%) with lung cancer, 1 of the 11 patients (9.1%) with prostate cancer, and 1 of the 38 patients (2.6%) with other malignancies.

Bisphosphonate and Anti-RANKL Antibody Exposure The median number of administrations of zoledronic acid or denosumab infusions to the whole population was 13 (range, 2 to 99) and the median duration of exposure was 13.0 months (range, 2.1 to 92.6 months). MRONJ development was significantly associated with the number of administrations of zoledronic acid or denosumab infusions ($p<0.001$; Table 2) and the duration of zoledronic acid or denosumab therapy ($p<0.001$; Table 3). Patients with MRONJ had significantly more administrations of zoledronic acid or denosumab infusions (median: 38.5 (range, 4 to 17) *vs.* median: 12.0 (range, 2 to 99), $p<0.001$) (Table 2) and significantly longer duration of zoledronic acid or denosumab therapy (median: 38.4 (range,

Table 1. Patient Characteristics (n=155)

	Osteonecrosis of jaw				p
	+		−		
	No. of patients	%	No. of patients	%	
Sex					0.071
Male	3	23.1	43	30.3	
Female	10	76.9	99	69.7	
Age					0.161
Average	60.2		65.2		
Standard deviation	12.7		6.4		
Chemotherapy					0.387
Yes	10		124		
Cancer species					0.093
Breast cancer	9	69.2	50	35.2	
Lung cancer	2	15.4	45	31.7	
Prostate cancer	1	7.7	10	7.0	
Other	1	7.7	37	26.1	
Systemic condition					
Diabetes	3	23.1	14	9.9	0.156
Anemia	3	23.1	12	8.5	0.116
Obesity	3	23.1	11	7.7	0.098
Stomatitis	4	30.8	51	35.9	1.000
Habit					
Smoking	1	7.7	9	6.3	0.595
Oral condition					
Moving tooth	7	53.8	40	28.2	0.065
Oral drying	6	46.2	64	45.1	1.000
Toothache	6	46.2	42	29.6	0.224
Pus discharge	7	53.8	38	26.8	0.055
Periodical dentistry maintenance	4	30.8	78	54.9	0.145

*p<0.05.

Table 2. Incidence of Osteonecrosis Related to Number of Infusions

	Osteonecrosis of jaw				p
	Yes*		No		
Total No. of infusions					<.001
Median	38.5		12.0		
Range	4–71		2–99		
	No. of patients	%	No. of patients	%	
No. of infusions					
2–5	1	7.7	39	27.5	
6–12	0	0	35	24.6	
13–24	2	15.3	36	25.4	
25–36	3	23.1	19	13.4	
37–48	3	23.1	5	3.5	
49–60	3	23.1	5	3.5	
>60	1	7.7	3	2.1	

*Number of administrations times until ONJ occurred (not counted after the onset of ONJ). p Values were calculated by a Wilcoxon rank sum test.

Table 3. Incidence of Osteonecrosis Related to Time of Exposure

	Osteonecrosis of jaw				p
	Yes*		No		
Total months of exposure					<.001
Median	38.4		11.5		
Range	5.2–62.3		2.1–92.6		
	No. of patients	%	No. of patients	%	
Time of exposure, months					
0–3	0	0	34	23.9	
4–12	1	7.7	42	29.6	
13–24	3	23.2	29	20.4	
25–36	2	15.3	20	14.1	
37–48	2	15.3	11	7.8	
49–60	3	23.2	3	2.1	
>60	2	15.3	3	2.1	

*Administration times until ONJ occurred (not counted after the onset of ONJ). p Values were calculated by a Wilcoxon rank sum test.

Table 4. Risk Factors for Developing MRONJ

Parameter	Odds ratio	95% CI	p
Sex	0.132	0.017–1.008	0.051
Age	1.072	0.995–1.155	0.066
Diabetes	6.699	1.435–31.277	0.016*
Anemia	14.559	2.161–98.069	0.006*
Obesity	2.762	0.511–14.934	0.238
Moving tooth	0.706	0.138–3.617	0.067
Pus discharge	6.491	1.514–27.835	0.012*
Periodical dentistry maintenance	0.137	0.020–0.944	0.043*

*p<0.05.

5.2 to 62.3) *vs.* median: 11.5 (range, 2.1 to 92.6), p<0.001) (Table 3) than patients without MRONJ.

Systemic and Oral Risk Factors Associated with MRONJ Development We examined several modeled clinical factors including sex, age, diabetes, anemia, obesity, moving tooth, pus discharge, and primary care dental treatment (Table 4). There were no significant differences in sex (p=0.051), age (p=0.066), obesity (p=0.238), and moving tooth (p=0.067). However, regression analysis showed diabetes [odds ratio (OR)=6.699, 95% confidence interval (CI), 1.435–31.277, p=0.016], anemia [OR=14.559, 95% CI, 2.161–98.069, p=0.006], and pus discharge [OR=6.491, 95% CI, 1.514–27.835, p=0.012] to be significantly associated with ONJ development. It is well known that diabetes and anemia are risk factors of MRONJ development. On the other hand, even with an increased number of infusions, significant reduction of risk was implied to be associated with periodical maintenance by dentists [OR=0.137, 95% CI, 0.020–0.944, p=0.043].

DISCUSSION

This study suggested that increased cumulative dose and long-term zoledronic acid or denosumab treatment are the most important risk factors for MRONJ development. Pus discharge must also play important roles in MRONJ development, from the findings in this study. Numerous reports have presented risk factors for MRONJ development.[20,32,34] Nitrogen-containing bisphosphonates (pamidronate, alendronate, risedronate, incadronate, zoledronate) target the intracellular enzyme farnesyl diphosphonate synthase, inhibiting the mevalonate pathway, resulting in disruption of post-translational

intracellular signaling proteins such as Ras.[35] This would alter cytoskeletal organization and cell motility, resulting in osteoclast apoptosis. Furthermore, bisphosphonates disrupt normal bone homeostasis, resulting in impaired healing, especially in bone exposed to constant trauma, which may result in necrosis.[36] Namely, MRONJ development may occur through endothelial proliferation *via* intraosseous circulation and bone blood flow.[37,38] In contrast, the effect of denosumab as an antibody is temporary. It has been shown that remodeling of the jaw bone resumes when denosumab is discontinued.[39] This is in line with the finding of the present study that there is an increase in bony turnover when denosumab is being administered. However, the mechanism of action of bisphosphonates and denosumab remains nuclear.[40]

MRONJ development was significantly associated with longer duration (median: 38.4 (range 5.2 to 62.3) *vs.* median: 11.5 (range 2.1 to 92.69), $p<0.001$) and an increased number of bisphosphonate infusions (median: 38.5 (range 4 to 17) *vs.* median: 12.0 (range 2 to 99), $p<0.001$) in this study (Tables 2, 3). In addition, the incidence of MRONJ was higher in patients with breast cancer than in those with lung cancer, prostate cancer, and other neoplasms. We considered that the risk of MRONJ in breast cancer may be linked to improved patient survival with the introduction of novel therapeutics such as trastuzumab. Therefore, the patients with breast cancer had the most prolonged exposure to bisphosphonates. Namely, it is possible that these results are consistent with earlier studies that noted that a prolonged duration of bisphosphonate exposure is a significant risk factor for the development of MRONJ.[20,30,32,34]

It is well known that MRONJ is a complication that is correlated with the long-term use of bisphosphonates. Recently, this complication of a long duration of bisphosphonate use has received much publicity and been associated with much controversy.[41,42] The American Society of Clinical Oncology (ASCO) guidelines for breast cancer suggest that bisphosphonates should be administered "until there is evidence of a substantial decline in the patient's general performance status." In the studies on which the current guidelines are based, bisphosphonates were usually administered for a maximum of 2 years.[8,43] Our study indicated that an increased number of infusions and prolonged administration of zoledronic acid or denosumab resulted in a higher risk of MRONJ. On the other hand, it was reported that, in patients with cancer exposed to zoledronate or denosumab, the incidence rates of developing MRONJ were, respectively, 0.6% or 0.5% at 1 year, 0.9% or 1.1% at 2 years, and 1.3% or 1.1% at 3 years, with the risk for MRONJ in denosumab-exposed patients plateauing between years 2 and 3.[25] However, there were some patients who did not develop MRONJ even with an increased number of infusions in this study. Our results suggest that there is a significant reduction of the risk of developing MRONJ in patients receiving periodical oral maintenance by dentists, even with an increased number of infusions. Patients should improve their oral hygiene, whereas oncologists and dentists should be aware of this complication and its management.

On the other hand, some clinical studies have reported that MRONJ is associated with intravenous bisphosphonate in cancer patients rather than oral administration for osteoporosis patients.[25] We noted an overall MRONJ frequency of 8.4% (15.3% with breast cancer, 4.3% with lung cancer, 9.1% with prostate cancer, 2.6% with other malignancies). These percentages are higher than in previous reports.[20,21,32,44] These reports indicated that intravenous bisphosphonate induced MRONJ at a frequency of approximately 5% (range, 0.72 to 7.4%) in patients with cancer. It is possible that the high incidence rates of MRONJ can be explained by the participation of dentists or dental hygienists in this study. Namely, dentists or dental hygienists detected MRONJ before its progression.

Recently, the dosing frequency of zoledronic acid was reported by Hortobagyi *et al.* at the 2014 ASCO Annual Meeting.[45] They reported that the dosing frequency of zoledronic acid can be reduced by 67% without compromising efficacy and safety in women with breast cancer and bone metastases, according to phase III data. Such reduced dosing can potentially lower the risk of developing MRONJ associated with zoledronic acid or denosumab. Namely, we have to consider an administration schedule of zoledronic acid or denosumab that is safer and has similar effects compared with the current schedule in the future.

Logistic regression analysis showed that pus discharge was significantly associated with the development of MRONJ (Table 4). This finding emphasizes the importance of assessing the oral health as well as physical condition of patients and implementing preventive dentistry if necessary before initiating therapy. It is known that MRONJ development is associated with the oral environment. It was reported that the incidence of MRONJ decreased after routine preventive dental measures were implemented.[27,28] Namely, MRONJ is a manageable and preventable condition. Our data confirm that MRONJ risks are reduced by periodical dentistry maintenance by avoiding detrimental conditions of patients' mouths, such as pus discharge (Table 4). However, it was not well known that the association of systemic risk factors with MRONJ was less pronounced than the association observed with oral factors. There was some association of MRONJ with the use of corticosteroids and a near doubling of cases of MRONJ in association with the use of antiangiogenic agents. On the other hand, there was no association with the use of chemotherapeutic agents. A recent analysis of MRONJ incidence in patients with breast cancer and bone metastases found no association between the development of MRONJ and exposure to these agents.[46] Further studies are in progress to clarify the association of oral conditions.

In conclusion, long-term administration of bisphosphonate or denosumab, as well as pus discharge, are risk factors for MRONJ development in cancer patients. By applying periodical dentistry maintenance, it was suggested that MRONJ risk could be reduced, even with an increased number of infusions. However, the retrospective nature of the present preliminary study might have had an inherent bias, which may be a limitation. Furthermore, the issue of reinitiating bisphosphonate and denosumab therapy in patients suffering from osteonecrosis has been debated and warrants additional investigation in further clinical study. Prospective randomized studies are needed to assess the incidence of MRONJ and the safety of bisphosphonates and denosumab in cancer patients.

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Conflict of Interest The authors declare no conflict of interest.

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