

Denosumab is Effective for Controlling Serum Calcium Levels in Patients with Humoral Hypercalcemia of Malignancy Syndrome: A Case Report on Parathyroid Hormone-related Protein-producing Cholangiocarcinoma

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Abstract

Hypercalcemia resulting in the elevation of serum parathyroid hormone-related protein (PTHrP) and suppression of serum PTH was observed in a patient with advanced cholangiocarcinoma (CCC) and multiple lymph node metastases. We confirmed humoral hypercalcemia of malignancy based on PTHrP-producing CCC. Chemotherapy with gemcitabine and cisplatin could not control the patient's serum PTHrP levels and the patient was affected with bisphosphonate-refractory hypercalcemia. We administered a single dose of denosumab, an anti-receptor activator of nuclear factor-kappaB ligand monoclonal antibody, and the patient's serum calcium levels remained close to the normal range for approximately 3 weeks without additional treatment.

Key words: humoral hypercalcemia of malignancy (HHM), denosumab, anti-receptor activator of nuclear factor-kappaB ligand (RANKL) monoclonal antibody, cholangiocarcinoma

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Introduction

Several cases of humoral hypercalcemia of malignancy (HHM) syndrome caused by parathyroid hormone-related protein (PTHrP)-producing cholangiocarcinoma (CCC) have previously been reported (1-6). According to these reports, it is difficult to control the serum calcium levels when a reduction in the volume of the PTHrP-producing tumor is unattainable through therapies such as operation, radiation, or chemotherapy.

We herein report a case of refractory HHM syndrome caused by advanced PTHrP-producing CCC with serum calcium levels that were effectively controlled by the administration of denosumab.

Case Report

A 63-year-old man was referred to our hospital in June 2015 because of right-sided chest pain and hypercalcemia. He had a history of hypertension without serious illness. The laboratory data on admission are summarized in Table. His serum calcium level was 13.6 mg/dL and serum inorganic phosphorus level was 2.3 mg/dL. The whole PTH level was 7 pg/mL (normal range: 9-39 pg/mL). The intact PTHrP level was 49.2 pmol/L (normal range: <1.1 pmol/L). The carcinoembryonic antigen (CEA) level was 31.3 ng/mL (normal range: <5.0 ng/mL), carbohydrate antigen (CA) 19-9 level was 2,139.4 U/mL (normal range: <37.0 U/mL), and α -fetoprotein (AFP) level was 55.5 ng/mL (normal range: <10 ng/mL). A computed tomography (CT) scan indicated that he had multiple tumors, including one measuring ap-

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Table. Laboratory Findings on Admission.

		Normal Range
Total Protein (g/dL)	7.1	6.8-8.3
Albumin (g/dL)	3.7	3.8-5.3
Total bilirubin (mg/dL)	0.9	0.3-1.2
AST (IU/L)	51	12-37
ALT (IU/L)	26	7-45
LDH (IU/L)	239	114-220
ALP (IU/L)	381	124-367
γ -GTP (IU/L)	110	8-50
Ca (mg/dL)	13.6	8.6-10.1
IP (mg/dL)	2.3	2.2-4.1
Whole PTH (pg/mL)	7	9-39
Intact PTHrP (pmol/L)	49.2	0-1.1
BUN (mg/dL)	22.2	9-22
Creatinine (mg/dL)	1.01	0.6-1.1
CEA (ng/mL)	31.3	0-5.0
CA19-9 (U/mL)	2,139.4	0-37.0
AFP (ng/mL)	55.5	0-10
1,25(OH) ₂ D ₃ (pg/mL)	116	20-60

AST: aspartate aminotransferase, ALT: alanine aminotransferase, LDH: lactate dehydrogenase, ALP: alkaline phosphatase γ -GTP: gamma-glutamyl transpeptidase, BUN: blood urea nitrogen, Ca: calcium, IP: inorganic phosphorus, CEA: carcinoembryonic antigen, CA19-9: carbohydrate antigen 19-9, AFP: α -fetoprotein, 1,25(OH)₂D₃: 1,25-dihydroxycholecalciferol

proximately 8 cm in the liver, and multiple lymph node metastases (Fig. 1A and B). These tumors were only enhanced at the peripheries, but not on the inside. Parathyroid swelling and other primary tumors (except for the liver) were not detected in either the CT scan or on an echogram. The chest CT scan revealed osteolytic lesions in the thoracic vertebrae (Fig. 1C), but whole body bone scintigraphy was negative. Endoscopic examinations of the upper and lower gastrointestinal tracts were normal. The histological diagnosis of a biopsy specimen from the left neck metastatic lymph node confirmed adenocarcinoma (Fig. 2A). An immunohistochemical examination showed supportive patterns for CCC because cytokeratin (CK) 7, CK19 (Fig. 2B), CK20, and CA19-9 (Fig. 2C) were positive, and thyroid transcription factor-1, napsin A, and hepatocyte were negative. CA19-9 positivity indicated that the elevation of serum CA19-9 levels was caused by tumor cell production. In addition, tumor cells from the metastatic lymph node showed positive staining for PTHrP (Fig. 2D). According to the results of imaging and immunohistochemical studies, particularly those pertaining to CK19, CA19-9, and PTHrP positivity, we diagnosed the patient as having PTHrP-producing CCC, which caused HHM syndrome. Because his CCC was inoperable, he received chemotherapy with gemcitabine and cisplatin combined with repeated zoledronic acid hydrate after hydration to improve hypercalcemia during hospitalization (Fig. 3). The multiple liver tumors and metastatic lymph nodes had increased in size and number (Fig. 1D), and his intact PTHrP levels elevated to 87.2 pmol/L even after chemotherapy. His serum calcium levels were only transiently

reduced by combination therapy of zoledronic acid hydrate, elcatonin (Fig. 3), furosemide, and betamethasone. Despite the administration of zoledronic acid hydrate, his serum calcium levels increased within a short duration.

Consequently, we administered a single dose of anti-receptor activator of nuclear factor-kappaB ligand (RANKL) monoclonal antibody, a subcutaneous injection of 120 mg denosumab, after obtaining his informed consent, although we thought HHM syndrome in this case was caused by PTHrP-producing CCC rather than local osteolytic hypercalcemia (LOH). After the administration of denosumab, his serum calcium levels were controlled at a level almost below 10.5 mg/dL for 3 weeks, which was achieved without the additional administration of bisphosphonates, although his serum inorganic phosphorus levels were consistently decreasing. He was administered 10 mmol sodium phosphate as 310 mg inorganic phosphorus daily (Fig. 3).

The patient eventually died of disease progression on the 89th day after admission.

Discussion

The most common features of HHM syndrome are hypercalcemia associated with the overexpression of PTHrP accompanied by suppressed PTH secretion. HHM associated with PTHrP may occur in patients with malignancies originating from various tissues (7). A CT scan revealed multiple liver tumors and lymph node metastases, but no other primary tumor in this case. The pattern of enhancement of liver tumors was quite different from hepatocellular carcinoma

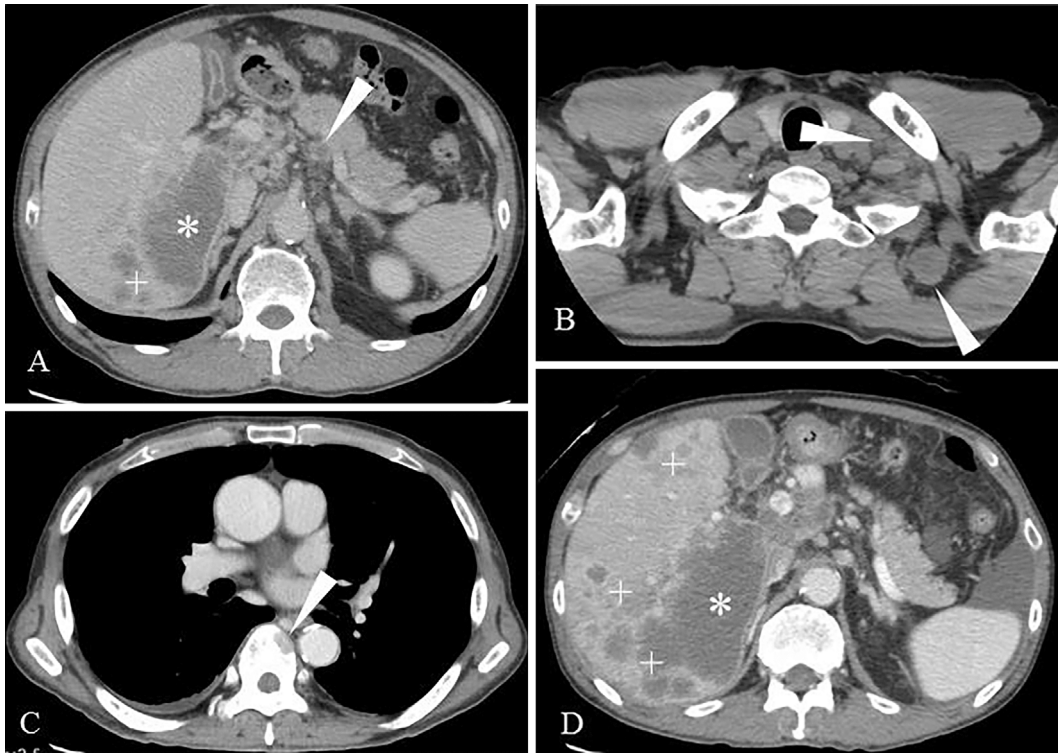


Figure 1. CT scans taken on admission and after chemotherapy. An abdominal CT scan taken at admission (A) revealed multiple liver tumors and lymph node metastases (arrows). A neck CT scan (B) revealed multiple lymph node metastases (arrows). A chest CT scan (C) revealed osteolytic changes in the thoracic vertebrae (arrow). An abdominal CT scan taken after chemotherapy (D). Liver tumors increased in size (*) and number (+) even after chemotherapy.

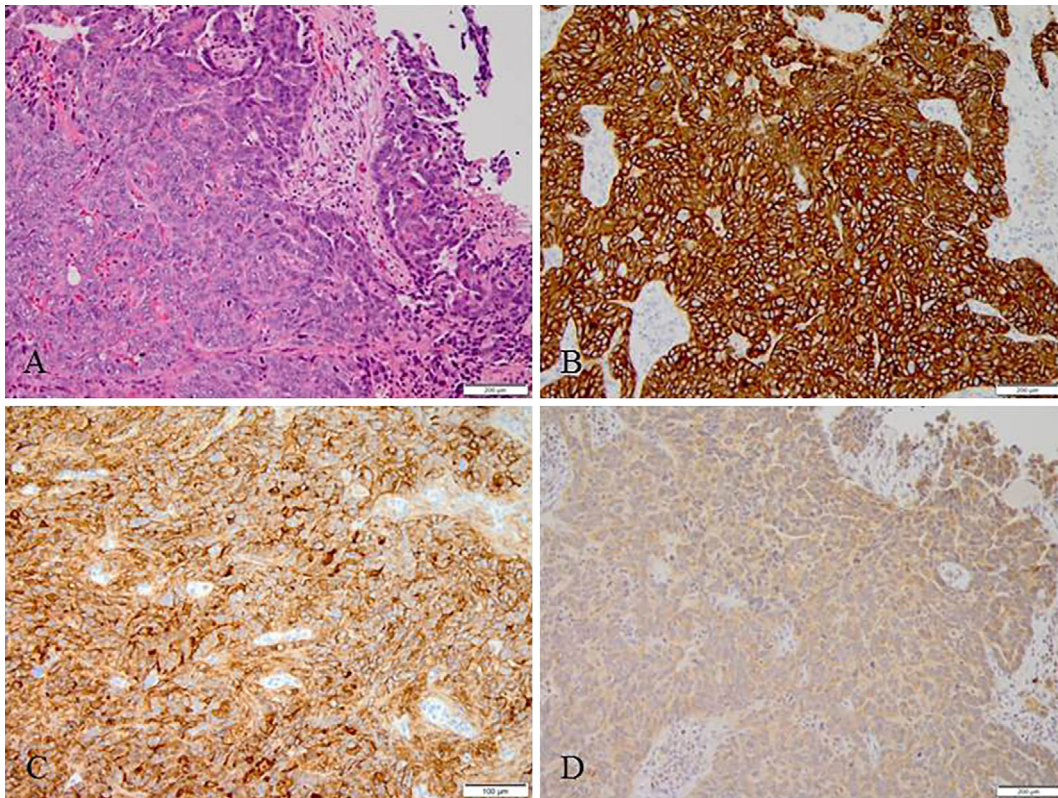


Figure 2. Histological examination of the metastatic neck lymph node. Moderately differentiated adenocarcinoma showing a papillotubular structure (A). Immunostaining for CK19 (B) and CA19-9 (C) was positive. Furthermore, tumor cells were positive for PTHrP (D). Magnification, 200 \times .

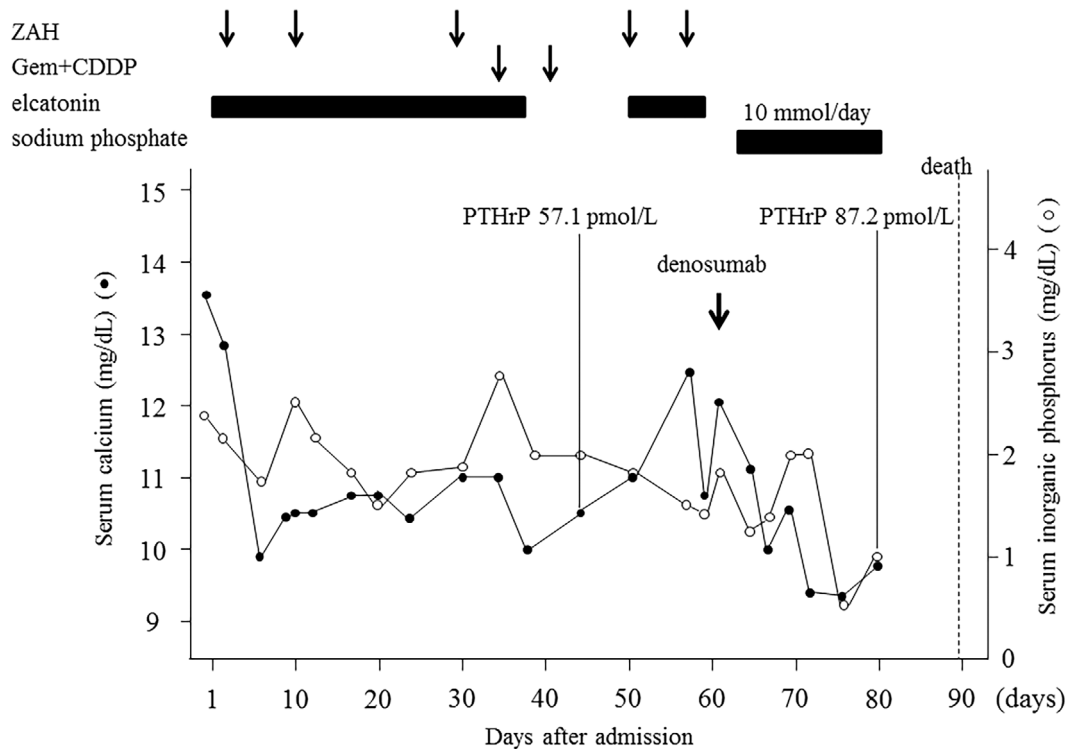


Figure 3. Clinical course of this case. We focused on the serum calcium levels (closed circles) and inorganic phosphorus levels (open circles). Arrows indicate the administration of zoledronic acid hydrate, gemcitabine, cisplatin, and denosumab. Closed squares indicate the durations for the administration of elcatonin or sodium phosphate. CDDP: cisplatin, Gem: gemcitabine, ZAH: zoledronic acid hydrate

(Fig. 1A). Roskams et al. reported that PTHrP is a useful marker for CCC, particularly in the differential diagnosis of hepatocellular carcinoma and metastatic adenocarcinoma (8). The tumor cells from metastatic lymph node had features typical of adenocarcinoma (Fig. 2A) and stained positive for CK7, CK19 (Fig. 2B), CA19-9 (Fig. 2C), and PTHrP (Fig. 2D), which suggested CCC. The CT scan also indicated that he had osteolytic lesions in the thoracic vertebrae (Fig. 1C), but whole body bone scintigraphy was negative. These data suggested that abnormal PTHrP secretion from tumor cells mainly caused HHM syndrome rather than LOH in this case. We concluded that HHM syndrome occurred because of PTHrP-producing CCC.

The prognosis of CCC is poor. The median survival time ranges from 2 to 15 months in trials using combination systemic chemotherapy and neoadjuvant chemoradiation in unresectable cases (9). HHM syndrome is one of the most clinically significant paraneoplastic syndromes because controlling the serum calcium levels are very difficult and an important matter in the quality of life for patients who have a poor prognosis. Most patients with HHM syndrome respond favorably to bisphosphonates when the drugs are preceded by saline hydration, however, a significant number of patients remain hypercalcemic or attain only transient effects after these treatments (10). Matsumoto et al. reported a case of combined hepatocellular-choleangiocarcinoma producing PTHrP that required palliative mass reduction surgery. The

aim is to reduce the serum PTHrP levels and control the serum calcium levels because hypercalcemia is medically difficult to manage (1). In our case, liver tumors and lymph node metastases increased in size and number (Fig. 1A and D), and the serum PTHrP levels elevated even further after chemotherapy (Fig. 3). On the other hand, there were no changes in the osteolytic lesions in his thoracic vertebrae on the CT scan.

In the bone, PTH or PTHrP acts on PTH/PTHrP receptors on osteoblast precursors to increase the production of macrophage colony-stimulating factor (MCS-F) and RANKL and decrease the production of osteoprotegerin (OPG). MCS-F and RANKL stimulate the production of osteoclasts and increase the activity of mature osteoclasts by binding to the receptor RANK. OPG blocks the interaction between RANKL and RANK receptor (11). Thus, in HHM syndrome, the inhibition of the increased interaction between RANKL and RANK receptor by anti-RANKL antibody, instead of a decreased OPG, may suppress bone resorption and hypercalcemia. The inhibition of RANKL has been reported to cause greater suppression of bone resorption and hypercalcemia compared with bisphosphonates in two models of HHM (10). Therefore, the administration of anti-RANKL antibody should thus be the first choice of treatment for HHM syndromes.

There have been three case reports on the administration of denosumab, an anti-RANKL antibody, for refractory hy-

percalcemia with HHM syndrome caused by PTHrP production in nonparathyroidal malignancy. In these reports, denosumab was indicated to be effective for bisphosphonate-refractory hypercalcemia, although hypocalcemia was one of the main adverse effects and careful monitoring of the calcium levels was required (12-14). In our case, the serum calcium level was only transiently controlled by bisphosphonates and the serum calcium levels increased within a short duration (Fig. 3). Because denosumab may suppress bone resorption and improve refractory hypercalcemia, we administered it, although we thought that HHM syndrome in our case was caused by PTHrP-producing CCC rather than LOH. After the administration of denosumab, the serum calcium levels of this patient were almost controlled to a normal range without the additional administration of bisphosphonates. The pathogenesis for the inhibition of phosphate transport via specific sodium-phosphate (NaPi) cotransporters on renal tubules by PTH or PTHrP is independent of RANKL (14). In this case, we had to supply phosphate even after the reduction in the serum calcium levels by the administration of denosumab because the serum inorganic phosphorus levels were consistently decreasing (Fig. 3).

In conclusion, we herein reported a case of HHM syndrome caused by PTHrP-secreting CCC and reviewed the previously administered treatments, which inhibited RANKL in patients with HHM syndrome. The anti-RANKL monoclonal antibody, denosumab, should therefore be a first-line treatment after hydration for HHM syndrome caused by PTHrP, when reduction of PTHrP is unattainable through other anti-tumor therapies.

The authors state that they have no Conflict of Interest (COI).

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References

1. Matsumoto M, Wakiyama S, Shiba H, et al. Combined hepatocellular-cholangiocarcinoma producing parathyroid hormone-related protein: report of a case. *Surg Today* **44**: 1577-1583, 2014.

2. Lim S, Han J, Park KH, et al. Two cases of humoral hypercalcemia of malignancy in metastatic cholangiocarcinoma. *Cancer Res Treat* **45**: 145-149, 2013.
3. Yamada M, Shiroeda S, Sato K, Arisawa T, Tsutsumi M. Cholangiocarcinoma producing parathyroid hormone-related peptide treated with chemoradiation using gemcitabine and S-1. *Intern Med* **48**: 2097-2100, 2009.
4. Sohda T, Shiga H, Nakane H, Watanabe H, Takeshita M, Sakisaka S. Cholangiocellular carcinoma that produced both granulocyte-colony-stimulating factor and parathyroid hormone-related protein. *Int J Clin Oncol* **11**: 246-249, 2006.
5. Yamada S, Sanefuji H, Morimoto H, et al. Parathyroid hormone-related peptide producing cholangiocellular carcinoma with a marked psammoma formation. *J Gastroenterol Hepatol* **15**: 1442-1446, 2000.
6. Davis JM, Sadasivan R, Dwyer T, Van Veldhuizen P. Case report: cholangiocarcinoma and hypercalcemia. *Am J Med Sci* **307**: 350-352, 1994.
7. Burtis WJ, Brady TG, Orloff JJ, et al. Immunochemical characterization of circulating parathyroid hormone-related protein in patients with humoral hypercalcemia of cancer. *N Engl J Med* **322**: 1106-1112, 1990.
8. Roskams T, Willems M, Campos RV, Drucker DJ, Yap SH, Desmet VJ. Parathyroid hormone-related peptide expression in primary and metastatic liver tumors. *Histopathology* **23**: 519-525, 1993.
9. Olnes MJ, Erlich R. A review and update on cholangiocarcinoma. *Oncology* **66**: 167-179, 2004.
10. 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* **146**: 3235-3243, 2005.
11. Kronenberg HM, Melmed S, Polonsky KS, Reed Larsen P. *Williams Textbook of Endocrinology*. 11th ed. Saunders Elsevier, Philadelphia, PA, 2008: 1209-1212.
12. Adhikaree J, Newby Y, Sundar S. Denosumab should be the treatment of choice for bisphosphonate refractory hypercalcemia of malignancy. *BMJ Case Rep* in press.
13. Teng J, Abell S, Hicks RJ, et al. Protract hypocalcemia following a single dose of denosumab in humoral hypercalcemia of malignancy due to PTHrP-secreting neuroendocrine tumour. *Clin Endocrinol (Oxf)* **81**: 940-942, 2014.
14. Bikos SA, Hammers HJ. Denosumab for the treatment of bisphosphonate-refractory hypercalcemia. *J Clin Oncol* **30**: e299, 2012.

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