□ CASE REPORT □

Non-Islet Cell Tumor Hypoglycemia Is Caused by Big IGF-II in a Patient with a Carcinosarcoma of the Uterus

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Abstract

We report a 72-year-old Japanese woman with severe hypoglycemia. The laboratory data, which revealed the suppression of serum insulin, suggested the existence of non-islet cell tumor hypoglycemia (NICTH). Abdominal computed tomography demonstrated the presence of a huge uterine tumor. The patient was treated with a continuous infusion of glucose, but died of sepsis on day 46. An autopsy revealed the pathological diagnosis to be a carcinosarcoma of the uterus. Interestingly, an immunohistochemical study discovered the expression of insulin-like growth factor (IGF)-II in both the carcinoma and sarcoma cells. In addition, an immunoblot analysis of blood samples revealed the presence of circulating big IGF-II. Therefore, this is a novel case of NICTH that was caused by a uterine carcinosarcoma.

Key words: non-islet cell tumor hypoglycemia, carcinosarcoma, insulin-like growth factor-II

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Introduction

Non-islet cell tumor hypoglycemia (NICTH) is a very rare disorder caused by benign and malignant tumors. It is characterized by a remarkable increase in the circulating big insulin-like growth factor (IGF)-II, which is a precursor of IGF-II and is secreted by the tumor (1). Tumors that cause NICTH primarily have mesenchymal cell origins in the thorax and the retroperitoneum (2). NICTH caused by carcinosarcoma is, however, very rare (3). Herein, we report a novel case of NICTH that is caused by a carcinosarcoma of the uterus. Circulating big IGF-II and the expression of IGF-II in the tumor were both demonstrated.

Case Report

A 72-year-old woman visited the emergency room of our hospital for transient hemiparesis and loss of consciousness. She had been diagnosed as having carcinoma of the cervix

with pelvic invasion two days before this presentation, but had no previous history of neurologic abnormalities. A laboratory examination demonstrated a low blood glucose level (34 mg/dL), and her symptoms resolved with the intravenous administration of glucose. However, the discontinuation of glucose administration led to frequent recurrences of hypoglycemic episodes. Consequently, she was admitted to our hospital. Her height and weight were 152.0 cm and 48.0 kg, respectively. Her blood pressure on admission was 104/74 mmHg. She took a calcium antagonist, nitrendipine, for hypertension and pravastatin for hypercholesterolemia, but was on no other medications that would cause hypoglycemia. She had no past history linked to hypoglycemia. The serum insulin levels were consistently suppressed during the hypoglycemic episodes. Therefore, we excluded the possibility of iatrogenic hypoglycemia.

A physical examination revealed a large mass in the lower abdomen with no tenderness. An endocrinological examination showed the suppression of serum insulin and C-peptide. On the other hand, the levels of thyroid hormone, adreno-

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Table. Laboratory Data for the Patient.

CBC (Day 1)		Hormone		
WBC	15,710/mm ³	Day 2		(Reference range)
RBC	400×10 ⁴ /mm ³	TSH	0.43 μU/mL	0.41 - 4.13
Hb	8.8 g/dL	FT ₄	1.37 ng/dL	0.95 - 1.74
Plt	72×10 ⁴ /mm ³	GH	1.82 ng/mL	0.28 - 1.64
Biochemistry (Day 1)		IGF-I	19 ng/mL	38 - 207
Alb	2.7 g/dL	ACTH	15.2 pg/mL	7.2 - 63.3
AST	34 U/L	Cortisol	21.1 μg/dL	4.0 - 18.3
ALT	19 U/L	IRI	0.50 μU/mL	3.0 - 16.7
ALP	275 U/L	C-peptide	0.09 ng/mL	0.61 - 2.09
BUN	8.7 mg/dL	Glucagon	47 pg/mL	50.0 - 150.0
Cre	0.24 mg/dL	Aldosterone	20.6 pg/mL	35.7 - 240
Na	137 mEq/L	Day 14		
K	2.9 mEq/L	GH	0.15 ng/mL	0.28 - 1.64
Cl	94 mEq/L	IGF-I	27 ng/mL	38 - 207
Fe	12 μg/dL	IGFBP-3	1.15 μg/mL	1.99 - 3.19
UIBC	267 μg/dL	IRI	< 0.3 μU/mL	3.0 - 16.7
CRP	9.7 mg/dL	C-peptide	0.07 ng/mL	0.61 - 2.09
Glucose	34 mg/dL			
HbA1c	4.7%			
IRI	< 0.2 μU/mL	Immunology (Day 1)		
Blood gas (Day 1)		Anti-insulin antibody	(–)	
pH	7.526	Anti-insulin receptor antibody	(–)	
pCO ₂	35.4 mmHg			
pO ₂	111.9 mmHg			
HCO ₃	28.7 mmol/L			
BaseExcess	5.6 mmol/L			

IRI: immunoreactive insulin

Figure 1. Clinical imaging. A: Abdominal computed tomography using radiocontrast material shows a pelvic tumor with low intensity regions. The tumor has no contrast enhancement. B: Magnetic resonance imaging of the abdomen and pelvis.

corticotrophic hormone or cortisol were within the normal range (Table). Neither the anti-insulin antibody nor the anti-insulin receptor antibody was detected. The serum IGF-I and IGF binding protein (BP)-3 levels were both low. The serum potassium level was low on admission, but the level gradually increased to a normal range without specific treatment. She also had mild anemia and thrombocytosis, presumably due to iron deficiency. Abdominal computed tomography showed a large mass in the uterine body (Fig. 1A), whereas there were no abnormal lesions in the pancreas or the liver. Magnetic resonance imaging also demonstrated a large mass lesion in the pelvis with heterogeneous signal intensity

(Fig. 1B). We considered the possibility of NICTH, and treated her with an intravenous continuous infusion of glucose, as well as dietary glucose loading. A few weeks after admission, her blood culture test was positive for *Staphylococcus aureus*, indicating a septic state. The focus of the infection was not definitively determined, but we clinically suspected the formation of an abscess in the pelvic tumor and/or a central catheter infection. Due to the advanced stage of the uterine tumor, intensive treatment for this tumor, such as surgical resection, radiation and chemotherapy, was not conducted. She died of sepsis 46 days after admission.

Figure 2. The histopathological features of the tumor. A, B: Hematoxylin and Eosin staining specimens. A: Characteristic sarcoma pathology, i.e., proliferation of spindle-shaped cells with scattered giant cells, was demonstrated in the tumor tissue. B: Carcinoma tissue was identified in sarcoma tissue. There is no transition between carcinoma and sarcoma tissues. C, D: An immunohistochemical study of IGF-II. Staining was positive for IGF-II (appearing brown) in sarcoma cells (C). Similarly, IGF-II expression was observed in both carcinomatous and sarcomatous cells (D). The scales are indicated as yellow bars.

Pathological and immunohistochemical examinations

An autopsy revealed that the uterine tumor (measuring 26×12×12 cm in size and weighing 1.8 kg) occupied a large part of the pelvic cavity. This giant tumor showed necrotic changes, and had infiltrated the gall-bladder. There was neither the presence of a lymph node nor distant metastasis. Microabscess formation was observed in the right lung and right kidney, which is consistent with the clinical diagnosis of sepsis. A histopathological study revealed the proliferation of the carcinoma in the uterine body (Fig. 2B), as well as an endometrial stromal sarcoma (Fig. 2A, B). No transition between the carcinoma and sarcoma was identified. Therefore, the diagnosis was carcinosarcoma of the uterine body. An immunohistochemical examination, using the rabbit polyclonal antibody ab9754 (purchased from Abcam.co.jp, Tokyo, Japan), demonstrated positive staining for IGF-II in both the carcinoma (Fig. 2D) and sarcoma tissues (Fig. 2C, D).

The immunoblot analysis

An immunoblot analysis of the patient's serum using the anti-IGF-II antibody identified a broad band with a higher molecular weight (approximately 14-15 kDa, Fig. 3, lane 3) than the control, human recombinant IGF-II (7.5 kDa, Fig. 3, lane 1). Similar patterns showing a high molecular band for IGF-II were also observed in the cases previously diagnosed as having NICTH with a circulating big IGF-II (Fig. 3, lanes 2 and 6). This finding was consistent with big IGF-II harboring part of the E domain of proIGF-II.

Discussion

The hypoglycemia observed in NICTH is presumed to be an effect of circulating big IGF-II, and successful treatment of the tumor diminishes its level, thereby ameliorating the hypoglycemia, which provides evidence for the role of big IGF-II in NICTH (1, 2). A patient with a carcinosarcoma of the ovary who had hypoglycemia and an increased serum IGF-II/IGF-I ratio was recently reported (3), which is con-

Figure 3. An immunoblot analysis using the antibody against IGF-II. Lane 1, recombinant human IGF-II (1-67) (control); lanes 2 and 6, samples from other patients with NICTH; lane 3, present patient; lane 4, a patient with hypoglycemia of unknown origin; and lane 5, a normal subject. The band corresponding to big IGF-II is indicated by a black arrow. In comparison to those represented in lanes 1 and 2, the present patient had almost exclusively big IGF-II.

sistent with our findings and strongly suggests a diagnosis of NICTH (2). In that report, however, neither the expression of IGF-II in tissue specimens nor the existence of big IGF-II was evaluated. Therefore, to the best of our knowledge, the present case is the first in which not only circulating big IGF-II, but also the expression of IGF-II in both carcinoma and sarcoma cells, was demonstrated in a carcinosarcoma. The low level of serum IGF-I in the present case was consistent with the findings in prior reports, although the serum IGF-II concentration was not evaluated and the IGF-II/IGF-I ratio was not determined in the present study. Growth hormone (GH) is usually suppressed through the central effect of circulating IGF-II (2), such that serum IGF-I is also suppressed. In the present case, IGFBP-3, which is a GH-dependent peptide, was also low.

Mature IGF-II is derived by the processing of its precursor, preproIGF-II, which contains 180 amino acids. A 24 amino acid-containing signal peptide is removed and pro IGF-II is thereby generated as a peptide that consists of 156 amino acids. The proteolytic cleavage sites of pro IGF-II are presumed to be Arg-68, Lys-88, and Arg-104, which ultimately results in a bioactive mature IGF-II that contains 1-67 amino acids (4). The removed peptide (68-156) is called the E-domain with O-glycosylation sites, and big IGF-II identified in NICTH patients has the part of the E domain that is glycosylated (5). Big IGF-II is known to exist in two isoforms; one isoform contains amino acids 1-87 and the other contains amino acids 1-104. Although we did not determine the isoform of big IGF-II in the present case, a recent study suggested that patients with NICTH commonly had circulating big IGF-II (1-87) (6).

Even if the total serum concentration of IGF-II is comparable to that of normal subjects, NICTH patients actually exhibit hypoglycemia (7). At this point, the difference in bioavailability between mature IGF-II and big IGF-II is known to be important. Mature IGF-II (1-67) forms the 150 kDa ternary complex with IGFBP-3 and the acid labile unit (8, 9). In contrast, big IGF-II exists preferentially as the 50 kDa binary complex (IGF-II and IGFBP), rather than the

ternary complex. Because of its lower molecular size, the binary complex is presumed to transverse the capillary membrane more easily (7, 10). A recent report clearly demonstrated that in a patient with NICTH, the binary complex dominated over the ternary complex in the preoperative period, while this was reversed in the postoperative period (11), suggesting that these complexes play a critical role in the clinical course of NICTH.

The hypoglycemic effect of IGF-IIs is mediated by the insulin receptor and/or the IGF-I receptor (7). A recent *in vitro* study characterized the two aforementioned isoforms of big IGF-II (1-87 and 1-104) (12). The ability to activate the IGF-I receptor and insulin receptor did not differ substantially between these two big IGF-IIs. More importantly, glycosylated IGF-IIs (1-87 and 1-104), but not unglycosylated IGF-II (1-104), showed a severely impaired ability to form the ternary complex (80-90% reduction). Therefore, glycosylation of the E-domain appears to be crucial for increasing the bioavailability of big IGF-IIs.

Carcinosarcoma of the uterus is known to show a very rapid tumor progression and a poor prognosis. The present case apparently had a typical uterine carcinosarcoma course because the tumor was huge and was accompanied by a large necrotic area. There are two theories as to the origin of the uterine carcinosarcoma. Since IGF-II expression was demonstrated in both the carcinoma and sarcoma tissues, the present case is likely to support the combination tumor theory, which involves the two tumor types originating from the same stem cell (13). However, it remains to be determined whether or not both of these tissues in the present case actually synthesized big IGF-II, because the antibody used for the immunostaining was not specific for big IGF-II. An immunoblot analysis indicated that the circulating IGF-II in the present case was almost exclusively big IGF-II (Fig. 3), and it is likely that both the carcinoma and sarcoma cells secreted this big IGF-II.

In summary, we herein described a novel case of NICTH that was caused by a huge uterine carcinosarcoma in which both the carcinoma and the sarcoma expressed IGF-II. Furthermore, we identified circulating big IGF-II using an immunoblot analysis, and it is highly likely that it was glycosylated. This observation is consistent with the recently established concept of NICTH.

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

Hiroyuki Honma and Mizue Matsui contributed equally to this work.

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