INTRODUCTION

Leptin is one of anorexigenic peptides and suppresses food intake and body weight by binding leptin receptor of hypothalamus. Previously, it was reported that leptin regulates bone formation and bone resorption through the central nervous system. Recently, it was demonstrated that other neuropeptides such as Neuropeptide Y (NPY) and cocaine and amphetamine-regulated transcript (CART) also regulate bone metabolism centrally. Neuregulin 1 (NMU) is one of hypothalamic neuropeptide and plays an important role in the regulation of feeding behavior and energy metabolism. Hanada et al. reported that NMU knockout (NMUKO) mice show obesity and hyperphagia and that NMU suppresses food intake and body weight centrally. However, the role of NMU in bone metabolism is yet to be clarified. We took notice of the similarity between leptin and NMU and made a hypothesis that NMU also regulates bone metabolism through the central nervous system.

METHODS

Experimental Animals
Wild type (WT) and NMU knockout (NMUKO) mice in a C57BL/6J background were used for all experiments. These mice were generated from the mating of NMU heterozygous mice.

Histological and Histomorphometric Analysis and Micro-CT Analysis
Histological and histomorphometric analysis were performed on vertebral bodies of 3-month-old mice by using undecalcified sections. Cortical thickness and cortical cross sectional area in the center of femur were measured by micro-CT. The samples from at least 6-8 mice per group were examined.

Measurement of Bone Resorption Marker
Deoxypyridinoline cross-links were measured in morning urines of 3-month-old WT and NMUKO mice. Creatinine values were used for standardization between samples.

Intracerebroventricular Infusion
After anesthesia, a cannula was implanted into third ventricle and rat NMU was infused at 3nmol/day for 28 days using an osmotic pump.

RESULTS

NMUKO Mice Have High Bone Mass
To study the role of NMU in bone metabolism, histological analysis and micro-CT analysis were performed. BV/TV of vertebral body was increased significantly in male NMUKO mice compared with male WT mice. Cortical thickness and cortical cross sectional area were also significantly increased in NMUKO mice.

Increased Bone Formation and Normal Bone Resorption in NMUKO Mice
To reveal whether NMU promotes bone formation or inhibits bone resorption, histomorphometric analysis was performed. We found that mineral apposition rate (MAR) and bone formation rate (BFR) were higher in NMUKO mice than in WT mice. Osteoblast surface was also significantly increased in NMUKO mice. On the other hand, in osteoclast surface and osteoclast number, there was no significant difference between WT mice and NMUKO mice. Furthermore, in measurement of urine deoxypyridinoline cross-links, there was no significant difference between two groups.

NMU Regulates Bone Mass through the Central Nervous System Independent of Leptin Signaling.
To investigate whether NMU regulates bone metabolism through a central nervous system, central administration of rat NMU to 2-month-old female WT mice and female ob/ob mice for 28 days was performed. In WT mice, both BV/TV and BFR were decreased by central administration of NMU, but not significantly. In ob/ob mice, BV/TV was significantly decreased by central administration of NMU. Both cortical thickness and cortical cross sectional area of femur were also decreased. These findings indicate that NMU regulates bone mass through the central nervous system and acts independent of leptin signaling.

DISCUSSION

Both NMU and leptin have a specific receptor in the hypothalamus and suppress food intake and body weight centrally. The function of NMU is very similar to that of leptin. Recently it was reported that leptin regulates bone metabolism through the central nervous system. Therefore, we speculated that NMU also regulates bone metabolism through the central nervous system.

As the result of analysis of NMUKO mice, we found that NMUKO mice have high bone mass due to increased bone formation, not decreased bone resorption. Furthermore, reduction of bone mass in WT mice and ob/ob mice with central administration of NMU suggests that NMU regulates bone mass through a central nervous system independent of leptin signaling.

REFERENCES

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