Leptin induces elongation of cardiac myocytes and causes eccentric left ventricular dilatation with compensation

Yukiko Abe,1 Koh Ono,2 Teruhisa Kawamura,1 Hiromichi Wada,1 Toru Kita,2 Akira Shimatsu,3 and Koji Hasegawa4

1Division of Translational Research and 2Clinical Research Institute, Kyoto Medical Center, National Hospital Organization, Kyoto; and 3Department of Cardiovascular Medicine, Graduate School of Medicine, Kyoto University, Kyoto, Japan

Submitted 3 June 2006; accepted in final form 9 January 2007

Leptin induces elongation of cardiac myocytes and causes eccentric left ventricular dilatation with compensation. Am J Physiol Heart Circ Physiol 292: H2387–H2396, 2007. First published January 12, 2007; doi:10.1152/ajpheart.00579.2006.—One of the major manifestations of obesity is an increased production of the adipocyte-derived 16-kDa peptide leptin, which acts mainly on hypothalamic leptin receptors. Leptin receptors are widely distributed in various tissues, including the heart. Whereas increased plasma leptin levels have been reported in patients with congestive heart failure, systemic alterations induced by obesity can affect cardiac hypertrophy, and the direct effects of leptin on cardiac structure and function still remain to be determined. We first exposed primary cardiac myocytes from neonatal rats to leptin for 48 h. This resulted in a significant increase in myocyte long-axis length (P < 0.05 at 50 ng/ml) but not in the short-axis width. Leptin induced the rapid phosphorylation of STAT3 and its DNA binding in cardiac myocytes. Administration of a JAK2 inhibitor, AG-490, completely inhibited all of these effects by leptin. Furthermore, we examined the effect of continuous infusion of leptin for 4 wk following myocardial infarction in mice. Echocardiography demonstrated that left ventricular fractional shortening in the leptin-infused group (28.4 ± 2.8%) was significantly higher than that in the PBS-infused group (18.4 ± 2.2%) following myocardial infarction. Interestingly, left ventricular diastolic dimension in the leptin-infused group (4.56 ± 0.12 mm) was also higher than that in the PBS-infused group (4.13 ± 0.09 mm). These results demonstrate that leptin induces the elongation of cardiac myocytes via a JAK/STAT pathway and chronic leptin infusion causes eccentric dilatation with augmented systolic function after myocardial infarction.

obesity; hypertrophy; heart failure; signal transduction

A GROWING EPIDEMIC OF OVERWEIGHT and obesity is affecting many countries worldwide. Obesity, even when uncomplicated by hypertension or diabetes, is frequently associated with left ventricular (LV) hypertrophy as assessed through ECG criteria or echocardiography (6, 7, 11, 12). Previous reports have strongly suggested that LV hypertrophy associated with obesity is eccentric in nature (3, 5, 30). In moderate to severe cases of obesity, this may lead to LV diastolic dysfunction. LV systolic dysfunction may also occur if wall stress remains high because of inadequate hypertrophy (2). Therefore, identifying the key mechanism for the development of cardiac hypertrophy in obese patients may offer unique insights into the understanding of obesity-induced changes in cardiovascular function.

One of the major manifestations of obesity is an increased production of the adipocyte-derived 16-kDa peptide leptin. Leptin regulates cellular homeostasis and glycemic control (31). While leptin was initially described as an adipocyte-derived protein with expression and secretion restricted to adipose tissue, recent reports have demonstrated that it is also locally expressed in other tissues (8, 15). Leptin receptor isoforms have been shown to be expressed in the myocardium and in isolated cardiac myocytes (19). This suggests that leptin has specific effects on the heart. Several recent studies have supplied evidence that leptin directly induces hypertrophy in cardiomyocytes in culture (13, 20, 26, 29). The observation that elevated plasma leptin levels are found in patients with LV hypertrophy or congestive heart failure suggests that leptin may play a role in human cardiac hypertrophy (17, 18, 21). On the other hand, one report suggested that leptin mediates “antihypertrophic” effects (4). Therefore, the direct effects of leptin on myocardial cell morphology and cardiac function still remain to be determined.

In this report we provide evidence for a potential link between leptin and LV eccentric hypertrophy by demonstrating that leptin induces cardiac myocyte elongation in vitro and eccentric dilatation with augmented systolic function after myocardial infarction (MI) in vivo.

MATERIALS AND METHODS

Immunocytochemistry and measurement of cell size. The investigation conformed to the Guide for the Care and Use of Laboratory Animals, published by the National Institutes of Health (NIH Publication No. 85-23, Revised 1996), and was approved by the Institutional Animal Research Committee of Kyoto University. Primary neonatal rat ventricular cardiac myocytes were prepared from Sprague-Dawley rats as previously described (9). The cardiac myocytes were grown on flask-style chambers with glass slides (Nalgene Nunc, Naperville, IL) and then stimulated with saline or 5–500 ng/ml of leptin in serum-free medium for 48 h. The cells were then fixed with 3% formaldehyde in phosphate-buffered saline (PBS) for 15 min at room temperature. Immunocytochemical staining for β-myosin heavy chain (β-MHC) was performed using the indirect immunoperoxidase method. The anticardiac β-MHC polyclonal antibody (Novo-Castra, Newcastle, UK) was used at a dilution of 1:50. A total of 50 myocardial fibers were selected randomly from cardiac myocytes stained with anti-β-MHC antibody, and the surface areas of these cells were measured semiautomatically with the aid of an image analyzer (Luzex 3U; Nikon, Tokyo, Japan) as previously described (10). Cell long-axis length was defined as the maximum longitudinal extension

Address for reprints requests and other correspondence: K. Ono, Dept. of Cardiovascular Medicine, Kyoto Univ., 54 Shogoin-Kawahara Cho, Sakyo-ku, Kyoto, 606-8507, Japan (e-mail: kohono@kuhp.kyoto-u.ac.jp).

The costs of publication of this article were defrayed in part by the payment of page charges. The article must therefore be hereby marked “advertisement” in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.
of individual cells. Maximum short-axis width was measured perpendicular to the axis that defined cell length. Immunocytochemical staining for STAT3 was performed using the indirect immunofluorescence method. The cells were fixed with Bouin’s solution for 10 min at room temperature and subsequently autoclaved for 10 min at 121°C in 10 mM citrate, pH 6.0. The cells were incubated with anti-STAT3 monoclonal antibody (Santa Cruz Biotechnology) at a dilution of 1:100. STAT3 signals were detected with anti-mouse FITC-conjugated secondary antibody at a dilution of 1:75 for 45 min.

Analysis of the phosphorylation state of STAT3, ERK1/2, p38, and JNKS. Cells were lysed with 2× Laemmli loading buffer (2% SDS, 20% glycerol, 0.04 mg/ml bromophenol blue, 0.12 M Tris·HCl, pH 6.8, and 0.28 M β-mercaptoethanol) (150 μl/35-mm dish). Filters were probed with antiphosphospecific STAT3, ERK1/2, p38, and JNK signals with antiphosphospecific STAT3, ERK1/2, p38, and JNK signals.

Fig. 1. Leptin induces the elongation of cardiac myocytes. Neonatal rat ventricular cardiac myocytes were isolated, treated with saline or leptin (50 ng/ml) for 48 h, and subjected to immunocytochemistry. The primary antibody to β-myosin heavy chain (β-MHC) was stained with a secondary antibody conjugated with peroxidase (brown signals). A: representative photomicrographs. B: myocardial cell surface area and short- and long-axis lengths were measured as described in MATERIALS AND METHODS. Values are means ± SE. The data are from 40 cells in each group. C: mRNA expression levels of atrial natriuretic factor (ANF) and GAPDH were measured by real-time PCR after leptin treatment. Mean ANF/GAPDH value was set at 1.0. Values are means ± SE.
antibody (New England Biolabs, Beverly, MA). To normalize for protein loading after immunoprecipitation, blots were stripped by incubation in 6.25 mM Tris-HCl, pH 6.8, 100 mM β-mercaptoethanol, and 2% SDS for 30 min at 50°C, washed twice with PBS and 0.05% Tween, and then probed with an antibody that recognizes both phosphorylated and nonphosphorylated forms of STAT3, ERK1/2, p38, and JNK (New England Biolabs). The mean of the integrated density obtained from three independent files was used as a representative value for each band.

**RT-PCR.** RT-PCR was used to analyze the expression of rat isoform-specific Ob-Ra and Ob-Rb as described previously (23). For real-time PCR, the reaction was performed with a SYBR Green PCR master mix (Applied Biosystems), and the products were analyzed with a thermal cycler (ABI Prism 7900HT sequence detection system). Levels of GAPDH transcript were used to normalize cDNA levels. Gene-specific primers were as follows: rat Ob-Ra sense, 5′ TGGCCCATGAGTAAAGTGAATGCTG-3′; rat Ob-Ra antisense, 5′ TCAAAGAGTGTCCGCTCTTTTGG-3′; rat Ob-Rb sense, 5′ TGGCCCATGAGTAAAGTGAATGCTG-3′; rat Ob-Rb antisense, 5′ TCTTCTGAAACTGGTTCAGGCTCCA-3′; atrial natriuretic factor (ANF) sense, 5′ CGTGCCCCGACCCACGCCAGCATGGGCTCC-3′; ANF antisense, 5′ GGCTCCGAGGGCGGACGAGCGAGCCCTCA-3′; mouse Ob-Ra and Ob-Rb sense, 5′ TGGTGGGAGCATGTTCTCCA-3′; mouse Ob-Ra antisense, 5′ GCTTGGTAAAAGATGCTCATG-3′; GAPDH sense, 5′ TTGCTCATGGATGACCTTGGC-3′.

**Electrophoretic mobility shift assays.** We used double-stranded oligonucleotides (purchased from Santa Cruz Biotechnology) that contained STAT3-binding sites and mutant STAT3-binding motif for the assay. We also used oligonucleotides with a promoter-specific transcription factor (SP1) binding site as a control probe. Electrophoretic mobility shift assays (EMSA) were carried out at 4°C for 20 min in 15-μl reaction mixtures containing 10 μg of nuclear extract, 0.25 ng (20,000 cpm) of radiolabeled double-stranded oligonucleotide, 500 ng of poly(dI-dC), 5 mM Tris-HCl (pH 7.5), 1 mM EDTA, 0.5 mM dithiothreitol, 37.5 mM KCl, and 4% Ficoll 400. For cold competition experiments, a 100-fold excess of unlabeled competitor oligonucleotide was included in the binding reaction mixture. Protein-DNA complexes were separated by...
electrophoresis on 4% nondenaturing polyacrylamide gels in 0.25× TBE (1× TBE is 100 mM Tris, 100 mM boric acid, and 2 mM EDTA) at 4°C.

Experimental MI, implantation of micro-osmotic pumps, and trans-thoracic echocardiography. C57/BL6 mice were anesthetized with 1.0 to 1.5% isoflurane, and open-chest coronary artery ligation was performed. MI was induced by ligating the left anterior descending coronary artery. During this procedure, micro-osmotic pumps (Du-rect) were implanted subcutaneously in the intrascapular region of each mouse. The pumps infused solutions at a rate of 2.5 μl/h for 28 days. The reservoir of each pump was preloaded with 2,000 μl of either sterile PBS or recombinant mouse leptin (R&D Systems) (1.54 μg/μl), to give a leptin infusion rate of ~0.32 μg·g⁻¹·day⁻¹. The dose of leptin was chosen to not increase blood pressure. The cardiac functions of mice were evaluated noninvasively by echocardiography after 4 wk. The animals were anesthetized with ketamine (50 μg/g body wt) and xylazine (2.5 mg/g). Transthoracic echocardiography was performed with a cardiac ultrasound recorder (Toshiba Power Vision, Tokyo, Japan) using a 7.5-MHz transducer. After the acquisition of high-quality two-dimensional images, M-mode images of the LV were recorded. LV end-diastolic (LVDD) and end-systolic (LVED) internal dimensions were measured according to the leading edge-to-leading edge convention adopted by the American Society of Echocardiography. Percent fractional shortening (%FS) was calculated as follows:

\[
%FS = \left[\frac{LVDD - LVED}{LVDD}\right] \times 100.
\]

At least three independent M-mode measurements were obtained per animal by an examiner who was blinded to the genotype of the animal. Plasma leptin concentration was measured by a mouse leptin immunoassay kit (R&D Systems).

Fig. 3. Leptin induces the nuclear translocation of STAT3 in cardiomyocytes. Immunocytochemical staining for STAT3 was performed using the indirect immunofluorescence method. The cells were incubated with anti-STAT3 monoclonal antibody, and STAT3 signals were detected with anti-mouse FITC-conjugated secondary antibody. β-MHC staining was performed using rabbit anti β-MHC polyclonal antibody, and the signals were detected with anti-rabbit rhodamine-conjugated secondary antibody. 4′,6-Diamidino-2-phenylindole dihydrochloride (DAPI) staining indicates the nucleus of the cells.
Statistical analysis. Data are presented as means ± SE. Statistical comparisons were performed using unpaired two-tailed Student’s t-tests or an analysis of variance with Scheffe’s test where appropriate, with a probability value of <0.05 taken to indicate significance.

RESULTS

Effect of leptin on primary cardiomyocytes. We exposed primary cardiac myocytes from neonatal rats to leptin at concentrations of 0, 5, 50, and 500 ng/ml for 48 h. As shown in Fig. 1A, brown signals, which indicated the presence of β-MHC, were observed in both saline- and leptin-stimulated cardiac myocytes. Cardiac myocytes that had been stimulated with leptin at a concentration of 50 ng/ml, which is usually observed in obese patients (14), showed increases in cell size and myofibrillar organization when compared with saline-stimulated cells. As shown in Fig. 1B, leptin stimulation (50 ng/ml) resulted in a significant increase in the myocyte surface area (left) and long-axis length (center) but not in the short-axis length (right). An increase in the leptin concentration from 50 ng/ml to 500 ng/ml resulted in a slight but not significant increase in the myocyte cell surface area. Thus a physiological concentration of leptin (50 ng/ml) was sufficient to induce the elongation of cardiac myocytes in culture. We also measured the mRNA levels of ANF, which is closely associated with cardiomyocyte hypertrophy, and GAPDH after leptin treatment. As shown in Fig. 1C, leptin stimulation (50 ng/ml and 500 ng/ml) resulted in a significant increase in the relative expression level of ANF/GAPDH.

Leptin induces STAT3 activation in cardiac myocytes. To investigate the intracellular pathway for leptin signaling in cardiac myocytes, we first examined the expression of two leptin receptor isoforms, short (Ob-Ra) and long (Ob-Rb) forms, by RT-PCR. A previous study in neonatal rat ventricular myocytes demonstrated the predominant expression of Ob-Ra and leptin-induced activation of ERK1/2 and p38 at 5 min after stimulation (20). In contrast with that study, we observed...
mRNA expression of Ob-Rb, which is linked to JAK/STAT signaling, in addition to that of Ob-Ra under our experimental conditions (Fig. 2A). Next, we examined the phosphorylation (activation) of MAP kinases (ERK1/2, p38, and JNKs) and STAT3 in cardiac myocytes by Western blotting. We did not recognize the significant activation of ERK1/2, p38, or JNKs at 10 min after treatment with 50 ng/ml of leptin (Fig. 2B). As shown in Fig. 2C, the phosphorylated form of STAT3 was increased in leptin-stimulated cardiac myocytes (lanes 2–4) when compared with saline-stimulated cells (lane 1), compatible with the expression of Ob-Rb. Activation was evident as early as 5 min after stimulation (lane 2). These findings suggest that leptin stimulation predominantly activated the JAK/STAT pathway in cardiac myocytes.

**Leptin induces nuclear translocation of STAT3 in cardiac myocytes.** To further examine the activation of STAT3 by leptin in cardiac myocytes, we performed an immunofluorescence microscopic analysis. As shown in Fig. 3, STAT3, which is indicated by the green signal of FITC, was detected in the cytoplasm of nearly all saline-stimulated cardiac myocytes (top left). However, stimulation of cardiac myocytes with leptin for 15 min markedly changed this localization and caused the nuclear translocation of STAT3 (top right). By double staining with an antibody against cardiac β-MHC, we confirmed that these cells are myocytes (middle). These findings provide further evidence for the leptin-induced activation of the JAK/STAT pathway in cardiac myocytes.

**Leptin increases the DNA-binding activity of STAT3.** To determine whether leptin modulates the DNA-binding activity of STAT3 in cardiac myocytes, EMSAs were performed. Nuclear extracts were prepared from neonatal cardiac myocytes that had been stimulated with leptin in the absence or presence of AG-490 (1 µM), a specific inhibitor of JAK2, or that had been treated with saline as a control. These extracts were probed with a radiolabeled double-stranded oligonucleotide containing the consensus STAT3 site. As shown in Fig. 4A, competition EMSAs revealed that a retarded band represented specific binding, as evidenced by the fact that it was competed out by an excess of unlabeled STAT3 oligonucleotide (lane 4), but not by the same amount of an oligonucleotide containing the STAT3 site with a mutation (lane 5). The amount of the specific complex containing STAT3 was markedly increased in nuclear extracts from leptin-stimulated myocytes (lane 2) compared with those from saline-treated cells (lane 1). Notably, the leptin-stimulated increase in STAT3-binding activity was almost completely blocked by 1 µM AG-490 (Fig. 4A, lane 3, and Fig. 4C). In contrast, SP1-binding activities were not altered by either leptin or leptin plus AG-490 (Fig. 4B).

---

**Fig. 5.** AG-490 suppresses the leptin-induced phosphorylation of STAT3 and inhibits cardiac myocyte hypertrophy. A: total cell lysates were prepared from neonatal cardiac myocytes that had been stimulated with leptin in the absence or presence of AG-490 (1 µM) or treated with saline as a control. Western blotting analysis with antiphosphospecific and total STAT3 was performed, and their signals were quantified as in Fig. 2. The relative level in cardiac myocytes without treatment was set at 1.0 in each experiment. Values are means ± SE from three independent experiments. B: cell surface area was measured as in Fig. 1. Values are means ± SE (in µm² of myocardial cell surface area). The data are from 40 cells in each group.
AG-490 inhibits cardiomyocyte hypertrophy induced by leptin. We tested whether AG-490 affects leptin-induced myocardial cell hypertrophy. Neonatal rat ventricular myocytes were preincubated with or without 1 μM AG-490 for 1 h. Activation of STAT3 was evaluated by Western blot analysis as described above. As shown in Fig. 5A, AG-490 inhibited leptin-induced activation of STAT3 (compare lanes 1, 3, and 4) but did not affect the phosphorylation status in the basal state (compare lanes 1 and 2). As shown in Fig. 5B, the same dose of AG-490 completely inhibited the increase in the myocardial cell surface area induced by leptin. AG-490 alone did not affect the area in saline-stimulated cardiac myocytes. These data demonstrate that AG-490 can selectively suppress the hypertrophic response induced by leptin.

Effect of chronic leptin infusion on cardiac structure and function following MI. To determine the effect of chronic leptin infusion on cardiac structure and function following MI, 12-wk-old mice were subjected to MI or sham operation. Each mouse was then subcutaneously fitted with an Alzet miniosmotic pump that delivered either PBS or 400 ng/h (0.32 μg·g⁻¹·day⁻¹) of leptin for 4 wk. The plasma leptin concentration significantly reduced after MI in mice infused with PBS. Chronic infusion of leptin significantly increased the plasma leptin level in both sham-operated and MI mice (Fig. 6A). At this...
Fig. 7. Effect of chronic leptin infusion on cardiac structure and function following MI. 

A: heart rate and blood pressure data at 4 wk after sham or MI operation. SBP and DBP, systolic and diastolic blood pressure. 

B: echocardiography data at 4 wk after sham or MI operation. LVEDD, left ventricular chamber diameter in end diastole; LVESD, LV chamber diameter in end systole; FS, fractional shortening. 

C: mRNA expression levels of mouse Ob-Ra, Ob-Rb, and GAPDH in the heart were measured by real-time PCR after MI. The mean ANF/GAPDH value was set at 1.0. Values are means ± SE.
LEPTIN INDUCES ELONGATION OF CARDIOMYOCYTES

concentration, leptin infusion did not affect food intake or body weight during examination. The heart weight-to-body weight (HW/BW) ratio and the relative expression level of ANF were significantly enhanced after MI. The relative expression level of ANF/GAPDH in infarcted hearts in leptin-infused mice was significantly higher than that in PBS-infused mice following MI (Fig. 6C). Blood pressure and heart rate were similar between the PBS- and leptin-infused groups (Fig. 7A). As shown in Fig. 7B, echocardiography demonstrated that there were no differences in LV dimensions or systolic functions between leptin- and PBS-infused mice with sham operation (open bars). In MI mice (solid bars), however, both LV chamber diameter in end diastole (LVEDD; Fig. 7B, left) and FS (Fig. 7B, right) were significantly higher in the leptin-infused group when compared with the PBS-infused group. Thus chronic leptin infusion resulted in eccentric dilatation with augmented systolic function following MI. To identify a possible link between leptin infusion and LV dilatation, we measured the expression levels of Ob-Ra and Ob-Rb in these mouse hearts. Interestingly, whereas Ob-Rb expression was significantly enhanced after MI (Fig. 7C), there was no change in the level of Ob-Ra.

DISCUSSION

Leptin is an adipocyte-derived peptide, the production of which is increased in patients with obesity. In addition to its action as a neuropeptide, it has become clear that leptin plays many roles as a growth factor in several cell types, including cardiac myocytes (19, 20, 26, 29). Clinical evidence has implicated an increased plasma leptin level as a potential independent risk factor for coronary heart disease, and leptin has been associated with cardiac hypertrophy and heart failure (17, 18, 21, 27). Therefore, it has become increasingly important to understand the effect of obesity-associated hyperleptinemia on cardiovascular systems.

Leptin exerts its effects through short (Ob-Ra) and long (Ob-Rb) receptor isoforms. It has been suggested that these two isoforms are coupled with distinct downstream signaling cascades; Ob-Ra is associated with MAP kinases, and Ob-Rb is coupled with JAK/STAT. A previous study demonstrated the predominant expression of Ob-Ra in neonatal rat cardiomyocytes and the leptin-induced activation of MAP kinases (20). Another study in human ventricular myocytes showed the leptin-induced activation of both MAP kinases and JAK/STAT as well as the expression of both Ob-Ra and Ob-Rb in these cells. The present study demonstrated that leptin induced activation of the JAK/STAT pathway in neonatal rat cardiac myocytes and that this pathway is required for elongation of these cells. Although we detected both Ob-Ra and Ob-Rb by RT-PCR, we did not detect the significant activation of MAP kinases after exposure to leptin for 10 min. Differences in the technique for preparing primary cardiac myocytes and their culture conditions might explain the discrepancy in the in vitro experimental results regarding receptor subtype expression and signaling cascades.

It has been shown that the gp130/leukemia inhibitory factor (LIF) receptor β-dependent cytokines cardiocrin-1 and LIF induce an increase in myocyte size characterized by a marked increase in cell length but little or no change in cell width (24, 28). Compatible with the fact that Ob-Rb is considerably similar to gp130 and LIF receptor, leptin had a more pronounced effect on cardiomyocyte length than on cardiomyocyte width. Myocytes from hearts with eccentric hypertrophy, which is often induced by volume overload, exhibit the assembly of sarcomeric units in series, whereas those from hearts with pressure overload-induced concentric hypertrophy show the parallel assembly of sarcomeric units (1, 16, 25). Our results in vitro also suggest that obesity-induced hyperleptinemia is, at least in part, attributable to eccentric hypertrophy. This finding prompted us to study the in vivo effect of leptin on cardiac morphology and function.

Previously, it was reported that infusion with leptin at 1 μg·kg⁻¹·min⁻¹ caused sustained increases in blood pressure (22). Since hypertension can induce cardiac hypertrophy by itself, we examined the effects of leptin on cardiac function at a lower concentration (400 ng/h = 0.32 μg·g⁻¹·day⁻¹) than that used in the previous report. Chronic infusion of leptin at this concentration did not cause any changes in food intake, blood pressure, or heart rate, suggesting that activation of the sympathetic nervous system did not occur under our conditions. We demonstrated here that, in MI mice, both LV diastolic dimension and fractional shortening in the leptin-infused group were significantly higher than those in the PBS-infused group. Therefore, leptin induces the enhancement of LV systolic function as well as LV eccentric dilatation after MI without increase in HW/BW ratio in vivo. In this experiment, we also demonstrated that the expression level of Ob-Rb is augmented in hypertrophied hearts after MI. Therefore, it is possible that the JAK/STAT pathway may be especially enhanced through Ob-Rb after MI.

A previous study suggested that leptin mediates antihypertrophic effects (4). In this study, infusion of leptin into ob/ob mice, which lack leptin and exhibit LV hypertrophy, reversed the increase in LV wall thickness. However, there is a possibility that the original hypertrophy in these mice may be the result of systemic alterations caused by a chronic lack of leptin. Moreover, the ability of leptin to reverse hypertrophy most likely reflects the correction of whole body physiological parameters that influence cardiac structure rather than a direct effect on myocytes. Therefore, on the basis of a consideration of our data together with those in previous works (13, 20, 26, 29), we believe that the acute direct effects of leptin on cardiac myocytes may be prohypertrophic.

Although the present study suggests that leptin may contribute to the development of eccentric LV dilation in obese patients, it is unlikely that a single cytokine or growth factor can explain all of the changes in cardiac morphology and function in these patients. However, clarifying the precise roles of leptin in the regulation of cardiac function may aid in the development of new therapeutic strategies for heart failure, especially in obese patients.

ACKNOWLEDGMENTS

The authors thank N. Sowa, N. Nagata, and M. Nishikawa for providing technical assistance.

GRANTS

This study was supported in part by grants (to T. Kita, K. Hasegawa, and K. Ono) from the Ministry of Education, Science, and Culture of Japan.
REFERENCES


