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Central Administration of Leptin Inhibits Food Intake and Activates the Sympathetic Nervous System in Rhesus Macaques*

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ABSTRACT

The present study was performed to determine the effects of central administration of leptin on food intake and sympathetic nervous system activity in a nonrodent species, the rhesus monkey. Peripheral administration of leptin at doses (1 and 3 μg/kg, sc) that produced increments of circulating leptin concentrations within a physiological range did not inhibit food intake over the subsequent 3 days. In contrast, leptin (1 μg/kg, intracerebroventricularly) had no acute effect on food intake, but caused a significant and sustained suppression (40–50%) of food intake during the entire following day ($P < 0.01$). In addition, circulating norepinephrine levels increased by 55 ± 16% ($P < 0.02$) 1 h after intracerebroventricular leptin administration, but did not increase after artificial cerebrospinal fluid administration. These results indicate that leptin can provide a signal to the central nervous system that decreases food intake in primates and in addition acutely activates the sympathetic nervous system. However, the results showing an acute increase in circulating leptin concentrations after peripheral administration of human leptin suggest that in primates, increases in circulating leptin within the physiological range do not acutely regulate food intake. Leptin may be more important in regulating food intake when there are sustained changes in circulating concentrations of leptin (*e.g.* with obesity, prolonged energy restriction, or diabetes). (*J Clin Endocrinol Metab* **84:** 711–717, 1999)

LEPTIN, a hormone produced by adipocytes, is believed to play an important role in the regulation of body adiposity by acting at the level of the central nervous system to modulate both food intake and energy expenditure (1, 2). The original evidence for these actions of leptin came from studies in which exogenous hormone was administered to *ob/ob* mice, a strain with a defect in the gene that encodes for leptin (3–5). The *ob/ob* mice are markedly obese and hyperphagic. Administration of exogenous leptin to *ob/ob* mice reduces food intake, increases energy expenditure, and results in dramatic decreases in body weight (3–5). Congenital leptin deficiency or defects in its receptor leading to extreme early onset obesity have recently been reported in humans (6, 7). Although circulating leptin concentrations are closely related to body adiposity (1, 8–10), a dynamic role for leptin in the regulation of food intake and energy expenditure has been suggested by the finding that circulating leptin concentrations decrease during fasting (9, 10) or energy restriction (11) and increase after refeeding (9, 12). In addition, circulating leptin concentrations exhibit a diurnal pattern (13–15) with a nocturnal peak that is entrained by meal timing (16). Furthermore, insulin infusions that produce physiological increments in plasma insulin increase circulating leptin concentrations in human subjects (17). Lastly, 24-h leptin concentrations are increased in subjects consuming high carbohydrate meals, which enhance insulin secretion (18), an effect that may be mediated by increased adipocyte glucose metabolism (19).

Administration of exogenous leptin or activation of the gene that encodes for leptin in normal mice and rats has been shown to effectively decrease food intake, increase energy expenditure, and result in body weight loss (3, 4, 20–22). However, despite the wealth of data collected over the past 2 yr on these effects of leptin in rodents, several critical issues remain unresolved. First, very few of the studies administering leptin have measured the plasma levels created by exogenous hormone administration, and those that have indicate that most published studies have examined the effects of pharmacological doses of this hormone. Thus, we do not know whether the physiological changes in circulating leptin levels that occur with meal consumption and brief periods of fasting have a role in regulating food intake and energy expenditure. Second, there have been no reports to date of regulation of food intake and energy expenditure by leptin in nonrodent species; thus, the applicability of findings in rodents to the control of food intake and energy expenditure in nonrodent species has not been established.

Several lines of evidence indicate that in rodents, leptin appears to exert its actions on food intake and energy expenditure by acting at the level of the central nervous system.

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Receptors for leptin have been localized within several brain areas, importantly in hypothalamic regions including the paraventricular nucleus and the arcuate nucleus, that are known to be areas important in the control of food intake (23–26). Moreover, administration of leptin systemically has been shown to activate neurons in these regions, as indicated by increased expression of Fos (27). Leptin receptors have been colocalized in neuropeptide Y (NPY) neurons in rodents (23) as well as in nonhuman primates (28). Leptin decreases the synthesis and release of NPY, a hypothalamic neuropeptide known to be a potent stimulator of food intake (29–31). Leptin administration intracerebroventricularly (icv) can decrease food intake and increase energy expenditure in rats (20, 25, 31–33). Perhaps most convincingly, several studies have shown that microinjection of leptin into the ventromedial hypothalamus (34) and the arcuate nucleus (35) can potently decrease food intake in the rat. A report by Collins *et al.* (36) showing that norepinephrine turnover in brown fat is increased soon after leptin administration, before an effect of leptin on food intake, suggests that leptin-induced increases in energy expenditure may at least in part reflect an activation of the sympathetic nervous system. This has been supported by the finding that leptin increases the firing rates of sympathetic nerves in rats (37).

The first goal the of the present study was to determine whether administering leptin at doses that produce acute increases in circulating leptin concentrations within the physiological range decreases food intake in rhesus monkeys. The second goal was to determine whether leptin administered directly into the central nervous system (icv into the third ventricle) is effective in regulating food intake and sympathetic nervous system activity in primates.

Materials and Methods

Animals

Sixteen adult male rhesus monkeys, weighing 9.6–13.5 kg, were used for these studies. Monkeys were housed in individual cages in a temperature-controlled room (24 ± 2 C) with lights on 12 h each day (0700–1900 h). Monkeys were maintained on a standard *ad libitum* feeding regimen, with two meals each day at 0900 and 1600 h, consisting of 1100 Cal (20 biscuits) of Purina High Protein Monkey Diet, jumbo size (no. 5047, Ralston Purina, St. Louis MO) and one quarter piece of fresh fruit at the morning meal. Before the initiation of experiments, monkeys had been adapted for at least 6 weeks to this feeding schedule, and their food intake was stable. Water was available at all times *ad libitum*. The studies were reviewed and approved by the institutional animal care and use committee of the Oregon Regional Primate Research Center.

Catheterization procedure

Five monkeys had a subclavian venous catheter and a third cerebroventricular cannula implanted at least 2 months before these experiments. The venous catheter was implanted using standard sterile surgical procedures, and monkeys were maintained on jacket/tether/swivel systems, as described previously (38).

Before placement of the third ventricular cannula, monkeys were anesthetized [with ~20 mg/kg Nembutal (Abbott Laboratories, North Chicago, IL)] and placed in a stereotaxic frame. The ventricular system was visualized by radiograph after injection of 300 μL of a radioopaque solution (Omnipaque, Winthrop Pharmaceuticals, New York, NY) into the lateral ventricle. Based on this radiograph, the third ventricular cannula was lowered into position through a second hole in the skull, so that the tip of the cannula was in the lower third of the third ventricle, above the median eminence. Placement of the cannula was confirmed by radiograph subsequent to an injection of 50 μL Omnipaque. The top portion of the cannula was adhered to the skull with dental acrylic. A catheter of polyethylene tubing (PE 50, Becton Dickinson and Co., Parsippany NJ), filled with artificial cerebrospinal fluid (aCSF; composed of 0.166 g/L $CaCl_2$, 7.014 g/L NaCl, 0.298 g/L KCl, 0.203 g/L $MgCl_2 \cdot 6H_2O$, and 2.10 g/L $NaHCO_3$; Life Technologies, Grand Island, NY) was attached to the cannula and tunneled sc to exit with the venous catheter between the scapulae. The ventricular cannula was passed through the same tether as the venous catheter and was kept patent with a constant infusion of aCSF at a rate of approximately 600 μL/24 h.

Blood-sampling procedures

Blood samples were collected via indwelling venous catheters into sterile heparinized syringes. Samples for measurement of leptin were transferred into sterile glass tubes and kept cold until centrifugation. Samples for measurement of catecholamines were transferred into ice-cold glass tubes containing 2.25 mg ethyleneglycol-bis-(β-aminoethyl ether)-*N*,*N*,*N*′,*N*′-tetraacetic acid and 1.5 mg reduced glutathione and centrifuged immediately. Both samples were centrifuged at 2400 rpm for 10 min. The red blood cells from the leptin samples were resuspended in sterile saline and reinfused through the indwelling venous catheters. The plasma from the catecholamine samples was placed in Eppendorf vials immediately after centrifugation, snap-frozen, and stored at −80 C.

Experimental protocols

Exp 1. To determine whether normal physiological variations in plasma leptin concentrations play a role in acutely regulating food intake in rhesus monkeys, leptin (Linco Research, Inc., St. Charles, MO) or saline was administered via sc injection to 10 noncatheterized monkeys, and food intake was measured for the subsequent 3 days. Before this study, a pilot study was performed with five catheterized monkeys to determine the time course and magnitude of fluctuations in leptin secretion across the 24-h day on the same *ad libitum* feeding regimen. We found that maximal plasma leptin concentrations occurred between 2300–0400 h and were about 2-fold greater than the minimum daily plasma leptin concentrations. Thus, to examine the effects of systemic leptin administration on food intake, we chose to enhance the magnitude of the endogenous nighttime rise in plasma leptin concentrations by injecting a dose of 3 μg/kg leptin at 0130 h and monitoring food intake for 3 days. The leptin dose was based on the results from another study in our laboratory in which adult male rhesus monkeys were given sc injections of leptin at 1 and 3 μg/kg at 0130 h (Cameron, J. L., and P. J. Havel, unpublished data). Lastly, we repeated this study, injecting 3 μg/kg leptin, sc, to monkeys at 1600 h and monitoring food intake for 3 days.

Exp 2. The second experiment was performed to examine the effects of icv injection of leptin on food intake and plasma catecholamines. Five monkeys with indwelling icv and venous catheters were studied twice, once after the administration of leptin (1 μg/kg in 20 μL aCSF, icv) and once after the administration of aCSF (icv). For this experiment, recombinant human leptin was purchased from Linco Research, Inc. Each of the two studies was conducted over a 3-day period. On day 1, a bolus icv injection (leptin or aCSF) was given at 1600 h, at the same time that the afternoon meal was provided. Food intake was monitored by counting remaining biscuits at 30, 60, 90, 120, and 240 min after the meal was provided and the following morning before the morning meal. Food intake continued to be monitored in a similar fashion after both the morning and afternoon meals on the second and third days of the study. Blood samples were collected at 15-min intervals for the first 12 h after the meal and at 30-min intervals for the following 2 h; one more blood sample was collected the following morning.

Assays

Plasma leptin concentrations were measured with a sensitive RIA for primate leptin with reagents supplied by Linco Research, Inc. Plasma norepinephrine and epinephrine were measured in duplicate with a highly sensitive and specific radioenzymatic assay (39). The intra- and interassay coefficients of variation for the plasma catecholamine assay were 6% and 12%, respectively.

Data analyses

In each experiment, monkeys were studied twice, once after leptin administration and once after administration of vehicle (aCSF or saline). Food intake and plasma hormone and neurotransmitter responses were compared at each time point using paired one-tailed Student's t test. $P \leq 0.05$ was considered significant.

Results

Effect of systemic leptin administration on food intake

In our pilot study to determine the time course of leptin secretion in male monkeys across the 24-h day, we found that maximal plasma leptin concentrations occurred between 2300–0400 h and were about 2-fold greater than the minimum daily plasma leptin concentrations (Fig. 1). To augment the nocturnal rise in plasma leptin levels that occurs over the course of a normal day, we chose to administer a 3 μg/kg dose of leptin, sc. This dose was based on the results from a pilot study in our laboratory in which four adult male rhesus monkeys were given sc injections of leptin at doses of 1 and 3 μg/kg (Cameron, J. L., and P. J. Havel, unpublished data). Figure 2 shows the increase in circulating leptin that occurred after injection of leptin at doses of 1 and 3 μg/kg leptin given as an sc injection at 0130 h (the time at which peak levels in circulating leptin normally occur during the 24-h day). Leptin administration at 1 and 3 μg/kg induced increases in plasma leptin more than 2- and 3-fold, respectively, over baseline fasting concentrations in all four monkeys used in the pilot experiment. Increased plasma leptin concentrations lasted for more than 8 h after leptin administration. However, in Exp 1 we found that leptin administered at a dose of 3 μg/kg at 0130 h had no effect on food intake in the 10 monkeys studied either acutely or during the succeeding 3 days (Fig. 3). Lastly, to examine the effect of systemic leptin administration on food intake at a time when leptin concentrations are normally low, we administered leptin (3 μg/kg, sc) to 10 monkeys at 1600 h, but similar to leptin administration at 0130 h, there was no effect on food intake either acutely or for the succeeding 3 days (data not shown).

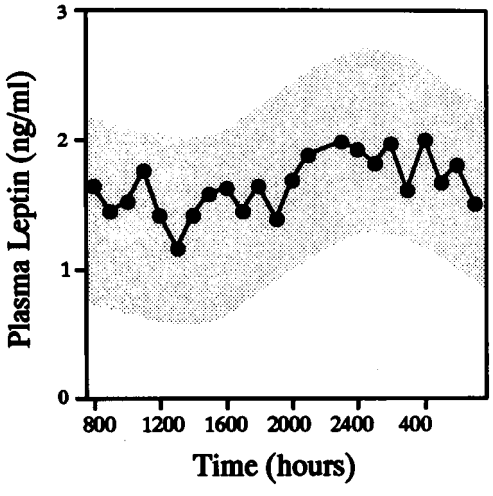

FIG. 1. Mean plasma leptin concentrations in five monkeys on a day of normal food intake (a morning meal at 0930 h and an afternoon meal at 1600 h). Blood samples were collected hourly. The *shaded area* depicts the SEM. Lights were on from 0700–1900 h, as indicated by the *black bar*.

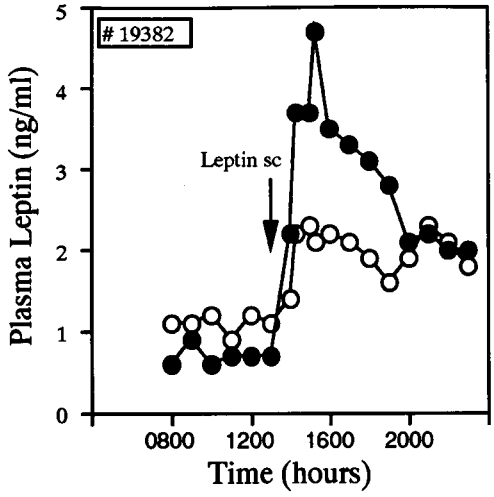

FIG. 2. Plasma leptin concentrations in a monkey fasted for 24 h and then given a sc injection of 1 μg/kg (*open circles*) or 3 μg/kg (*closed circles*) recombinant human leptin.

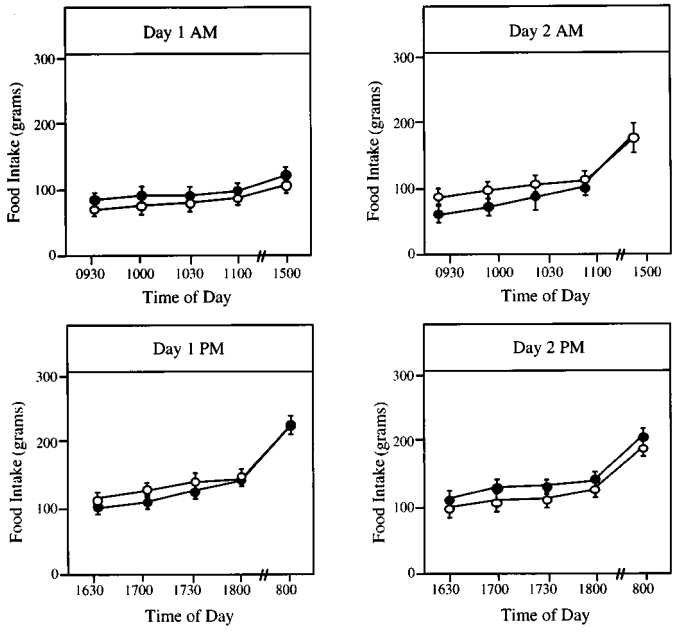

FIG. 3. Food intake over four meals in 10 monkeys after a single sc injection of either vehicle (*open circles*) or human leptin (3 μg/kg; *closed circles*) at 0130 h before day 1. Data are shown as the mean (SEM).

Effect of central leptin administration on food intake

Central administration of leptin (1 μg/kg) had no immediate effect on food intake (*i.e.* the day 1 afternoon meal; Fig. 4). However, leptin treatment reduced food intake by 40–50% throughout the period of morning meal consumption compared to that during the control study when aCSF alone was administered. None of the animals exhibited more than a 66% decrease in food intake after central leptin administration. This reduction of food intake lasted into the afternoon meal on day 2. On day 3, food intake was generally normal, and on day 4 it was completely normal. Plasma leptin concentrations were slightly, but not significantly, higher on the day of leptin *vs.* the day of aCSF treatment.

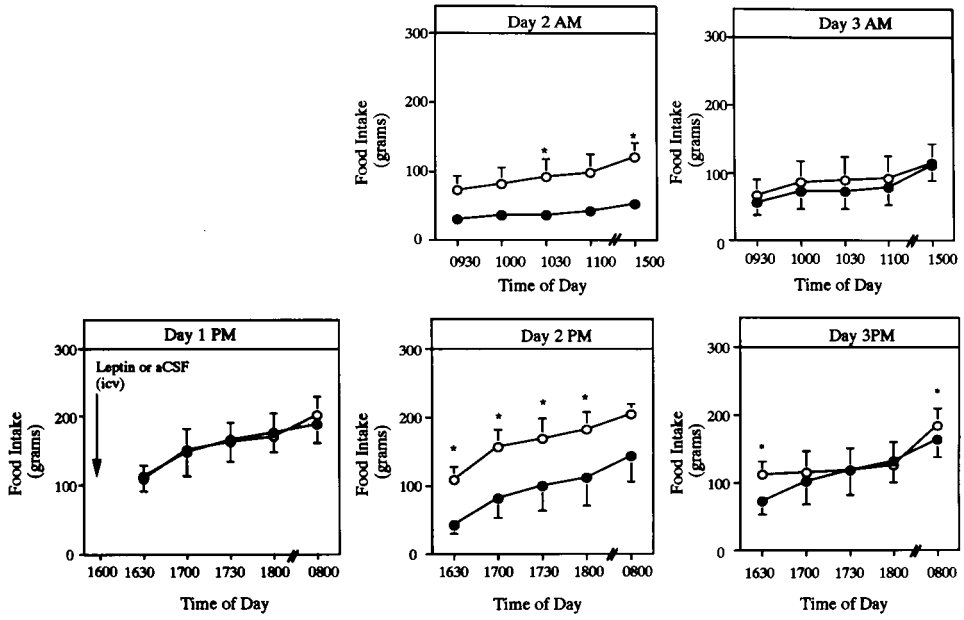

FIG. 4. Food intake (grams) over five meals in five monkeys after a single icv injection of either aCSF (*open circles*) or human leptin (1 μg/kg; *closed circles*) on day 1 at 1600 h. Data are shown as the mean ± SEM. *Asterisks* indicate a significant difference between vehicle and leptin trials ($P < 0.05$).

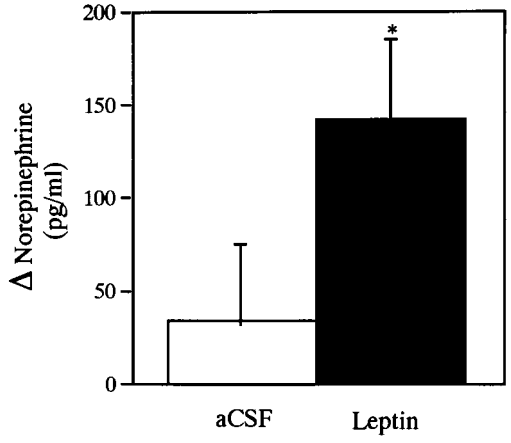

FIG. 5. The change in plasma norepinephrine (picograms per mL) concentrations in response to leptin or aCSF during the first hour postadministration. Data are shown as the mean (SEM). The *asterisk* indicates a significant difference between vehicle and leptin trials ($P < 0.02$).

Plasma leptin did not increase after icv leptin administration and was not different during the 4-h postinjection period between the leptin and aCSF treatment days.

Effect of central leptin on plasma catecholamines

When monkeys received leptin (1 μg/kg, icv), there was an acute increase in circulating norepinephrine concentrations, with levels increasing from a baseline of 254 ± 50 to 396 ± 81 pg/mL [change (Δ), +142 ± 41 pg/mL; %Δ, +55 ± 16%; both $P < 0.02$] at 1 h after administration (Fig. 5). In contrast, plasma norepinephrine did not increase significantly after intracerebroventricular administration of aCSF, averaging 256 ± 53 pg/mL before and 290 ± 54 pg/mL 1 h after injection [Δ, +34 ± 20 pg/mL (P = NS); %Δ, +15 ± 9% (P = NS)]. By the following morning, circulating norepinephrine levels had decreased and were similar during the leptin and aCSF trials. Circulating plasma epinephrine concentrations were unaffected by either leptin or aCSF administration, averaging 95 ± 30 pg/mL before and 100 ± 28 pg/mL 1 h after leptin administration and 95 ± 13 pg/mL before and 92 ± 28 pg/mL 1 h after aCSF treatment.

Discussion

Subcutaneous administration at doses designed to produce increments in circulating plasma leptin concentrations within the normal physiological range did not suppress food intake either acutely or during several subsequent meals. In our first study, leptin was administered at 0130 h to enhance the magnitude of the normal diurnal peak in leptin that occurs in the middle of the night. A similar diurnal pattern, with peak levels occurring at night, has been reported in humans (13–16), and it has been hypothesized that increased levels of leptin occurring at night may prevent individuals from awakening from hunger and may serve to limit food intake during the morning meal (1). Our results do not support such a role in rhesus monkeys, in that the 3 μg/kg injection of leptin would have raised circulating leptin concentrations by 3-fold above normal nighttime levels for 6–9 h, yet there was no suppression of food intake during the morning meal.

We subsequently wondered whether an effect of leptin on food intake would be more readily apparent if leptin was administered at a time of day when endogenous leptin levels are low. Leptin was administered systemically at 1600 h, a time at which we found that icv leptin administration was effective in suppressing food intake during the following day. However, administration of leptin at this time did not inhibit subsequent food intake. Our results suggest that increases in leptin that occur over the course of a normal day do not play a role in acutely regulating the magnitude of daily food intake in rhesus monkeys. This is the first study to our knowledge to examine the effects of leptin, within the physiological range, on food intake in primates. Most of the studies published to date that have documented suppression

of food intake after systemic administration of leptin have used leptin-deficient *ob/ob* mice, animals that exhibit enhanced sensitivity to leptin (40) or have used very large doses of leptin in normal animals (22, 40). A previous study examined the effects of lower doses of leptin on neuroendocrine function in fasted mice at a time when circulating leptin levels resembled those in the fed state (41). However, these lower concentrations occurred 12 h after administering twice daily injections of leptin over 48 h, which acutely raised plasma leptin concentrations by up to 40-fold above normal levels for several hours.

Our study examined the effect of a single injection of leptin on food intake, and this leaves open the possibility that physiological shifts in circulating leptin concentrations may modulate food intake when they are sustained for a period of time. Sustained alterations in circulating leptin concentrations, associated with changes in adiposity and body fat distribution, occur with gender (42–44), with weight gain (12) and loss (8), and over normal development (43, 44). A recent report by Halaas *et al.* (45) supports the hypothesis that sustained physiological increases in plasma leptin decrease body weight and adiposity in normal mice. Further evidence for a physiological role for endogenous leptin in regulating food intake is provided by the report that icv administration of purified antileptin antiserum increases food intake in normal rats, but not in obese Zucker rats (46).

An additional question that, until recently, has not been addressed is whether decreases in plasma leptin concentrations within the physiological range serve as a signal to increase food intake, either acutely or chronically. Fasting (9, 10), energy restriction (11), and insulin-deficient diabetes (47) all produce marked and sustained decrements in circulating leptin concentrations that are probably the result of decreased adipocyte glucose metabolism (19). It has now been shown that replacing leptin by osmotic minipump infusion in streptozotocin diabetic rats, at a rate that prevents the marked fall in plasma leptin after induction of diabetes, also prevents the onset of diabetic hyperphagia (47, 48). In addition, there is a strong correlation between the magnitude of the decrease in circulating leptin concentration and the increase in hunger ratings reported during a prolonged moderate energy deficit in women (49). Together, the available data suggest a role for leptin as a long term regulator of energy balance, rather than as a short term determinant of appetite or satiety.

The significant and sustained suppression of food intake in rhesus monkeys we observed after intracerebroventricular administration of a 1 μg/kg dose of leptin indicates that, as has been previously documented in rodents (7, 12, 19, 20), leptin is able to act at the level of the central nervous system to suppress food intake in primates. Thus, it is likely that in the monkey, leptin acts in locations adjacent to the third ventricle, areas that also appear to be involved in leptin's action on food intake in rodents (21, 22). Although the icv dose of leptin employed, based on body weight, was substantially lower than the doses used in studies with icv leptin in rodents (20, 25, 32, 33), it may have been a pharmacological dose. Average concentrations of leptin in the CSF of healthy human subjects are approximately 0.2–0.3 ng/mL (50, 51). It is very likely that central administration of leptin at 1 μg/kg produced acute elevations of CSF leptin levels far above this concentration. However, because, like insulin (52), leptin is likely to gain entry to the CSF compartment subsequent to transport into the brain parenchyma, it is not possible to determine how concentrations of a plasma hormone reaching a specific brain region correlate with CSF levels of that hormone. Thus, we can conclude that, as in rodents, centrally administered leptin suppresses food intake in primates. However, it is not known whether the concentrations of leptin in the hypothalamic regions regulating food intake were within a physiological or a pharmacological range.

There was no immediate effect of leptin administered icv on food intake (*i.e.* no suppression of food intake during the 90 min after the afternoon meal provided at the same time as leptin was administered). Rather, suppression of food intake was first evident the following morning, 17.5 h after leptin administration. This finding differs from studies in the rat that have reported a suppression of food intake within 1 h of icv leptin administration (25, 33). However, as monkeys eat most of their meal within the first hour after meal presentation, it is possible that leptin inhibits food intake much earlier than the following morning, but given our meal feeding paradigm we were unable to detect such an earlier inhibition. Nevertheless, unlike in the rat, icv leptin administration did not suppress food intake within the first hour after injection.

The delay in the effect of leptin to suppress food intake may reflect the amount of time needed for leptin to transit from the third ventricle to receptors within the parenchyma of the brain. It may also reflect a multistep pathway of leptin action on appetite. In fact, there is growing evidence that leptin, at least in part, suppresses food intake by acting on hypothalamic NPY neurons. The *ob/ob* mice with knockout of the NPY gene are not as obese and are less hyperphagic than *ob/ob* mice with an intact NPY system (53). Leptin receptor messenger ribonucleic acid (mRNA) has been identified in NPY-expressing neurons (23, 28), and leptin administered icv to rats suppresses NPY synthesis (31). Leptin may also act on hypothalamic POMC neurons to modulate food intake. Recent evidence indicates that activation of melanocortin-3 and -4 receptors (which bind POMC-derived peptides) inhibits feeding in a number of rodent models of hyperphagia (54). POMC neurons in the arcuate nucleus have been shown to express leptin receptor mRNA in both rodents (55) and nonhuman primates (28). The *ob/ob* mice have been reported to have significantly reduced levels of POMC mRNA in the arcuate nucleus, and leptin treatment restores POMC mRNA to normal levels (56–58). Additionally, pharmacological blockade of melanocortin-4 receptors has been reported to prevent leptin-induced suppression of food intake in the rat (59).

The effect of leptin administered icv on food intake was apparent for more than 24 h. This finding is similar to results reported in rats in which a single injection of leptin icv suppressed food intake for 2–3 days, resulting in a loss of body weight that was sustained for at least 6 days (32). The prolonged effect of leptin administered icv on food intake may result from a slow rate of leptin degradation within the brain.

It is likely that the suppression of food intake subsequent

to central leptin administration results from a specific action of leptin, rather than from the induction of a general state of malaise. Monkeys administered leptin icv exhibited no outward signs of nausea, such as drooling, yawning, or a decrease in general activity. Moreover, the fact that leptin resulted in a delayed, rather than an acute, inhibition of food intake suggests that leptin was modulating food intake via a ligand-receptor interaction, not by a nonspecific induction of nausea or taste aversion. Additionally, leptin administration did not completely suppress feeding, but only reduced food intake by 50–60%. This conclusion is further supported by the work of Thiele and co-workers (33), who showed that leptin administered icv to rats (3 μg/rat) does not cause a conditioned taste aversion.

The finding that leptin, administered icv, led to a significant elevation in plasma norepinephrine levels provides strong evidence that central leptin administration activates the sympathetic nervous system. To our knowledge, this is the first report that leptin activates the sympathetic nervous system, as assessed by an increase in circulating norepinephrine levels. However, this finding supports the previously published work of Collins and co-workers (36) in mice, which showed that leptin increases norepinephrine turnover in brown adipose tissue, and the more recent demonstration in the rat that leptin activates sympathetic nerves innervating brown adipose tissue, kidneys, hindlimb, and adrenals (37). Moreover, peripheral administration of leptin at pharmacological doses in rhesus monkeys produces acute increases in circulating glucose and lactate that are blocked by combined α- and β-adrenergic blockade, demonstrating that the acute metabolic effects of leptin in the primate are mediated by sympathetic activation (60). It has been well documented in rodent models that there exists an inverse relationship between activation of the sympathetic nervous system and food intake (61, 62). To date, most substances that inhibit food intake cause an increased activity in the sympathetic nervous system (*i.e.* cholecystokinin, serotonin, *etc.*), and substances that increase food intake (*i.e.* NPY and norepinephrine) inhibit the sympathetic nervous system. Thus, it is likely that the increased sympathetic activity we have found in this study, contributes to both the inhibition of food intake and the leptin-induced increases in energy expenditure that have been previously reported in mice and rats (3, 5, 22).

Interestingly, the effect of leptin on plasma norepinephrine concentrations occurred soon after leptin administration, before leptin had any effect on food intake. Thus, the effect of leptin to activate the sympathetic nervous system appears to be mediated through a mechanism separate from that underlying leptin-induced changes in food intake. However, it is still possible that the effect of leptin on the sympathetic nervous system is mediated by a leptin-induced decrease in the activity of NPY-ergic neurons, in that NPY has been shown to potently reduce energy expenditure via a reduction in sympathetic output (63). Lastly, central leptin administration did not increase plasma epinephrine concentrations. Thus, the effect of leptin to activate sympathetic nervous system was specific for noradrenergic sympathetic and excluded those preganglionic sympathetic nerves innervating the adrenal medulla.

In summary, icv leptin administration causes a potent and sustained suppression of food intake and an acute increase in circulating norepinephrine concentrations in rhesus monkeys, indicating that leptin is capable of acting as a signal to the central nervous system to modulate feeding behavior and the level of sympathetic nervous system activity in primates. Nevertheless, our finding that acute increases in circulating leptin within the physiological range did not inhibit food intake in rhesus monkeys argues against a role for leptin in the short term control of daily fluctuations in food intake. Rather, our data support the hypothesis that leptin serves as a long term signal to modulate energy balance and is probably involved in regulating appetite when decreases (*e.g.* insulin-deficient diabetes or prolonged energy restriction), or increases (*e.g.* chronic overfeeding) of circulating leptin concentrations are sustained.

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