Turkish Journal of Biology

Volume 38 | Number 2 Article 4

1-1-2014

Serum leptin profiles, food intake, and body weight in melatonin-implanted Syrian hamsters (Mesocricetus auratus) exposed to long and short photoperiods

BÜLENT GÜNDÜZ

TÜBİTAK

Turkish Journal of Biology

http://journals.tubitak.gov.tr/biology/

Research Article

Turk J Biol
(2014) 38: 185-192
© TÜBİTAK
doi:10.3906/biy-1308-40

Serum leptin profiles, food intake, and body weight in melatonin-implanted Syrian hamsters (*Mesocricetus auratus*) exposed to long and short photoperiods

Bülent GÜNDÜZ*
Department of Biology, Faculty of Arts and Sciences, Çanakkale Onsekiz Mart University, Çanakkale, Turkey

Received: 16.08.2013 • **Accepted:** 31.10.2013 • **Published Online:** 28.03.2014 • **Printed:** 28.04.2014

Abstract: We have shown that circadian changes in leptin concentrations are inversely linked to circulating melatonin levels in the Syrian hamster. The present study aimed to assess the effects of subcutaneous melatonin implants under different photoperiodic conditions: did the implants affect serum leptin levels, body weight, food consumption, and/or testicular weight? Male hamsters were exposed to long and short photoperiods for 10 weeks and received subcutaneous melatonin implants (1 mg in 24 mg beeswax) every 2 weeks. Blood samples were collected every week at midday (1200 hours) or midnight (0000 hours) to assess leptin and melatonin concentrations. Testes were measured to calculate weight. Body weight and food intake were determined every week. Melatonin implants blocked the testicular regression produced by short photoperiods. No differences in body weight were observed among any of the groups. Food consumption increased only with the melatonin implant in the short photoperiod. Serum leptin levels in both photoperiods remained constant. Differences were apparent between noon and night leptin profiles. Noon leptin levels were high (16–18 ng/mL) compared to night leptin levels (8–11 ng/mL) in untreated controls. On the other hand, serum leptin concentrations declined in melatonin-implanted hamsters in both photoperiods. The effects of melatonin on leptin hormone profiles are very pronounced, and melatonin seems to have both physiologically and pharmacologically suppressive effects on leptin production by direct or indirect mechanisms.

Key words: Melatonin, leptin, hamster, implant, photoperiod, pineal gland

1. Introduction

For many animals, successful reproduction and survival in seasonal environments depends upon an ability to predict changes in environmental conditions and make anticipatory adjustments in physiological and behavioral factors (Dark et al., 1983; Bronson and Heideman, 1994). Such adjustments include a change in body mass and energy utilization, a molt to a summer or winter pelage, and cessation or initiation of reproductive activity (Steinlechner and Niklowitz, 1992). The predictive cue used by many animals to anticipate seasonal transitions is day length. Therefore, in Syrian hamsters short day exposure, for example, increases the body mass. This increase is primarily associated with an increase in fat mass (Hoffman et al., 1982; Davis, 1989) which offers us the opportunity to study the effects of a photoperiod-induced regulation of adipose tissue metabolism. The pineal gland is primarily involved with conveying the photoperiodic message along the neuroendocrine axis. The release of melatonin from the pineal gland shows a circadian rhythm; blood levels of melatonin are high during darkness and low during the day (Reiter, 1991).

The activity of the reproductive axis is sensitive to the adequacy of nutrition and stores of metabolic reserves. Mainly, the adipocyte-derived hormone leptin is involved in the regulation of food intake and body weight and serves as a metabolic gate to the reproductive system (Wade et al., 1996; Ziylan et al., 2009) and to cellular immunity (Baltaci and Mogulkoc, 2012). Genetically obese ob/ob mice are infertile, and leptin treatment has been shown to restore fertility (Chehab et al., 1996). Severely food-restricted animals have reduced circulating levels of leptin, which are associated with markedly reduced secretion from the gonadotropins (Matthew et al., 1999; Karakas et al., 2005). Treatment of food-restricted mice, rats, sheep, and monkeys with exogenous leptin reverses the diet-induced inhibition of gonadotropin secretion. Although leptin would appear to play a role in relaying metabolic information to the reproductive axis, the mechanism(s) by which this is accomplished are not presently known. Exposure to a short photoperiod decreases leptin gene expression and hormone release in adipose tissue (Klingenspor et al., 1996, 2000), suggesting that leptin might be involved in photoperiod-mediated

* Correspondence: bgunduzbio@comu.edu.tr

seasonal adaptations of mammals independent of food deprivation or overfeeding. In addition, circulating concentrations of leptin exhibit pulsatility and circadian rhythmicity. The levels of plasma leptin vary directly with body mass and body fat. Moreover, circadian changes in leptin concentrations are inversely linked to circulating melatonin levels in the Syrian hamster (Gündüz, 2002).

Melatonin modulates fat metabolism in some mammalian species (Rasmussen et al., 1999; Wolden-Hansen et al., 2000; Baltaci and Mogulkoc, 2007). As for the Syrian hamster, no previous study has tested the effects of continuous release of melatonin on circulating leptin concentrations. The present study was designed to determine whether a high dose of continuous-release melatonin administered to adult male Syrian hamsters has an effect on serum leptin rhythmicity (both in total and in day/night variation) and to test the hypothesis that photoperiod-dependent changes in reproductive function are mediated by leptin.

2. Materials and methods

2.1. Animals

Forty male Syrian hamsters (*Mesocricetus auratus*) were obtained from the breeding colony (14L:10D) maintained at Çanakkale Onsekiz Mart University. Adult hamsters (weighing ~70 g) were exposed to either 14L (14 h light, 10 h darkness, lights off at 2000 hours) (n = 20) or 10L (10 h of light, 14 h of darkness, lights off at 2000 hours) (n = 20) photoperiods. Animals were housed in plastic cages (16 × 31 × 42 cm) with pine shavings used as bedding. The procedures used in this study were carried out in accordance with the university's animal scientific procedures and approved by the Çanakkale Onsekiz Mart University Institutional Animal Care and Use Committee. All lighting was provided by cool-white fluorescent tubes controlled by automatic programmable timers. Ambient temperatures in the animal facilities were held constant at 22 ± 1 °C in air-ventilated rooms. Tap water was accessible ad libitum. The food supplied was standard Purina rodent pellets containing 22% protein, 5% fat, and 5% crude fiber. Food consumption was calculated for each week. Since there were 2 animals per cage, food consumed was necessarily based upon group consumption rather than upon the individuals therein. Food remaining in the wire mesh food hopper was weighed and a weighed amount of fresh food was added. In all cases, obvious food particles that had dropped through the mesh hopper were removed and weighed as well. The difference between these 2 measurements was calculated as a measure of food consumed per 2-animal cage. Throughout the study, all animals had free access to food and water at all times.

2.2. Surgical procedures

Before surgery, hamsters were anesthetized subcutaneously with ketamine (20 mg/kg BW, Sigma Chemical Company, MO, USA) and intraperitoneally with pentobarbital (32.5 mg/kg BW). Depth of anesthesia was monitored by frequent testing for the presence of leg flexion reflexes and active muscle tonus. Implants were prepared according to the methods of Horton et al. (1992). Crystalline melatonin (Sigma) was dissolved in melted beeswax (1 mg melatonin/24 mg beeswax). The mixture was then aspirated into 15-cm lengths of PE 320 tubing (2.69 mm i.d. × 3.5 mm o.d.; Intramedic, Clay Adams, Parsippany, NJ, USA). When the beeswax had cooled to room temperature and hardened, the tubing was cut into 10-mm capsules. Control capsules were prepared in a similar way but filled with beeswax only. The melatonin implants were open-ended and released approximately 10 µg of melatonin per day (Stetson et al., 1983). Every 2 weeks, melatonin-containing capsules were removed and replaced with new ones. Implants were inserted subcutaneously in animals under ketamine and pentobarbital anesthesia through a small dorsal skin incision in a shaved area on the back. The wound was closed with a steel wound clip.

2.3. Testes measurements

Reproductive condition was assessed in lightly anesthetized hamsters by measuring the width and length of the left testis with analog calipers. The product of testis length × width^2 provided an estimated testis volume, which is highly correlated with testis weight and function in rodents (Watson-Whitmyre and Stetson, 1985). Testis volume was then converted to paired testes weight using a predetermined linear regression formula. All data from in situ testicular measurements are reported in the form of paired testes weights derived by this method.

2.4. Experimental procedures

Each photoperiod contained control and melatonin-implanted animal groups. Each group had 10 adult hamsters. Testes weights were calculated and body weights were taken every week throughout the 10-week experimental period. Blood (approximately 1.0 mL) was taken every week at midday (between 1200 and 1300 hours) and midnight (between 0000 and 0100 hours) from the orbital sinus of each animal under light ether anesthesia to determine leptin levels. Samples taken during the dark phase were taken under dim red light. To prevent the loss of circulating plasma volume, 0.9% NaCl was injected intraperitoneally immediately after each blood collection in the same volume as drawn. NaCl replacement solution was sterilized and warmed to body temperature prior to replacement. Blood samples were centrifuged at 4 °C for 30 min at 1000 × *g*. Serum aliquots were aspirated and frozen at –20 °C. Hormones were measured by commercial ELISA kits according to the manufacturer's instructions (ICN, Costa Mesa, CA, USA).

Serum melatonin levels were measured in duplicate using 96-well microtiter plates coated with captured antibody goat anti-rabbit Ig. Each microtiter plate was filled with 50 mL of blanking reagent; zero calibrators; standard solutions containing 5, 10, 20, 50, 500, and 1000 pg/mL of melatonin; controls of human serum with specific amount of melatonin; or extracted samples. Optical densities were determined at 450 nm in an automatic microplate reader. Serum melatonin concentrations were expressed as pg/mL. The sensitivity of the melatonin assay was 3.0 pg/mL. Serum concentrations of hamster leptin were measured in duplicate with a lower detection limit of 0.5 ng/mL. Both the intra- and interassay coefficients of variation were less than 10% in the 2 assays.

2.5. Statistics

Data were analyzed using SPSS 10.0 (SPSS Inc., Chicago, IL, USA). Testes, body weights, and food intake were analyzed using a repeated-measure 2-way or 3-way analysis of variance (ANOVA) to view the effects of photoperiod, treatment, and timing of the sampling. Data are expressed as means ± SEM for serum leptin and melatonin for each sampling point. Hormone levels were analyzed by 3-way ANOVA (effect of implant, photoperiod, and timing of sampling as parameters) followed by Duncan's multiple range test. Differences were considered statistically significant at $P < 0.05$.

3. Results

No differences in body weights were observed ($P > 0.05$; Figure 1A). Melatonin implants blocked testicular regression produced by short photoperiods at week 10 ($P < 0.001$ versus control; Figure 1B). There were no significant changes in testes weights of 14L animals; they retained their large testes size.

Serum leptin levels in both short (10L) and long (14L) photoperiods remained constant. Differences were apparent between noon and night leptin levels ($P < 0.001$). Noon leptin levels were high (16–18 ng/mL) compared to night leptin levels (8–11 ng/mL) in untreated controls under both photoperiods (Figures 2A and 2B). On the other hand, serum leptin concentrations declined in melatonin-implanted hamsters in both photoperiods throughout the experiment (Figures 2C and 2D). No differences were detected between noon and night leptin levels. The differences between control and melatonin-implanted hamsters in both photoperiods were apparent ($P < 0.001$). The implants suppressed the leptin concentrations equally in both photoperiods (Figure 2).

Daytime plasma levels of melatonin were low for both short and long photoperiods (control, 9–11 pg/mL). Melatonin treatment resulted in significantly ($P < 0.001$) increased levels (28–30 pg/mL) in both photoperiods (Figure 3A). The levels of plasma melatonin during the night in both short and long photoperiods (30–35 pg/mL) significantly increased with the melatonin implant (48–53 pg/mL) (Figure 3B).

Food consumption in the control and melatonin-implanted groups was relatively consistent throughout the experiment (~7 g/day) in the long photoperiod (Figure 4A). However, food consumption in both groups was higher in the short photoperiod (Figure 4B). Moreover, incremental increases in consumption in the melatonin-implanted group took place from week 7 and increased above 10 g/day.

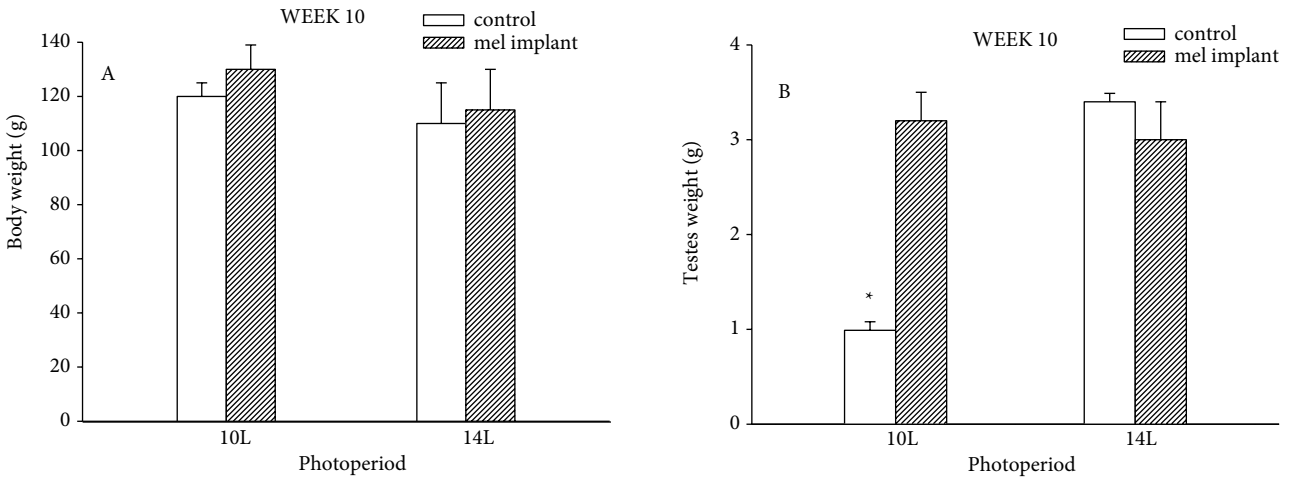

Figure 1. Body weights of male Syrian hamsters determined over 10 weeks of short and long photoperiod exposure. Unfilled bar represents control groups, and shaded bar represents melatonin-implanted animals at week 10 (A). Testes weights in male Syrian hamsters determined throughout 10 weeks of short and long photoperiod exposure. Testes weights were determined by laparotomy and are expressed as means ± SEM of 10 hamsters in each group (B). Asterisk indicates significant difference ($P < 0.05$).

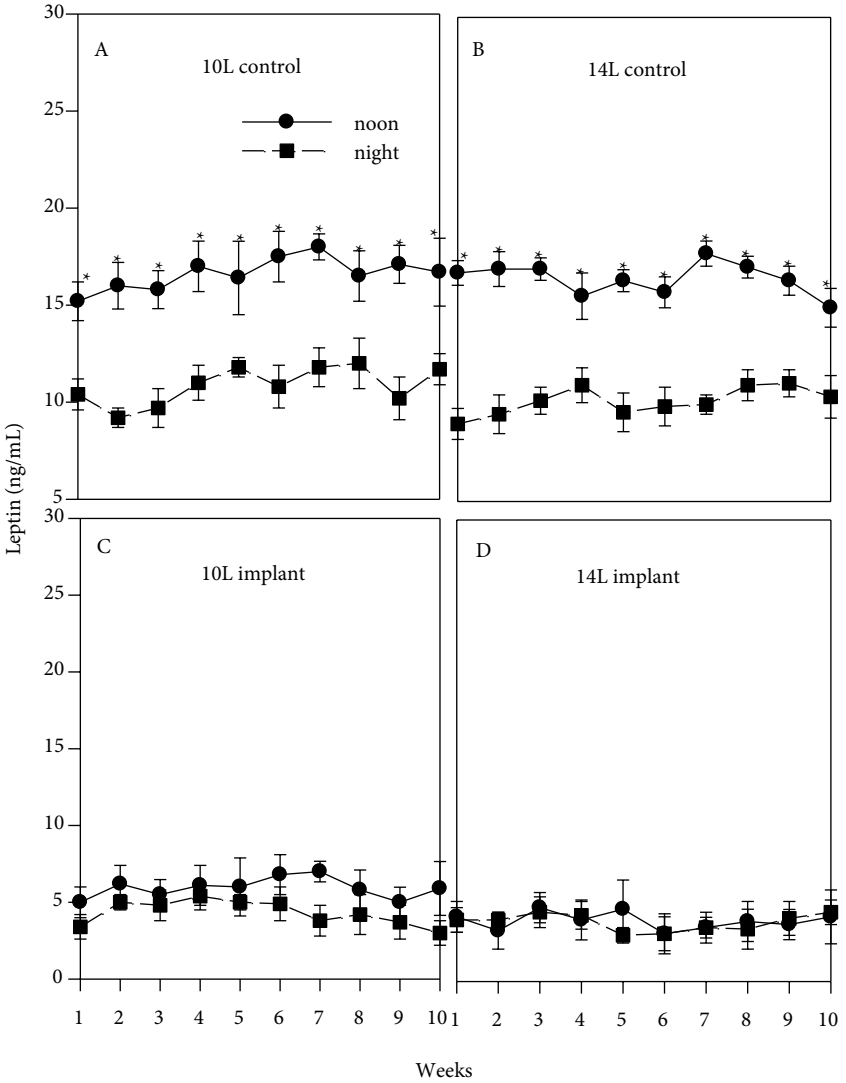

Figure 2. Syrian hamster mean serum leptin concentrations. (A) Leptin concentrations of control animals in short photoperiod (10L). (B) Leptin concentrations of control animals in long photoperiod (14L). (C) Leptin concentrations of melatonin-implanted animals in short photoperiod (10L). (D) Leptin concentrations of melatonin-implanted animals in long photoperiod (14L). Circular symbol represents serum leptin levels taken at noon and square symbol represents serum leptin levels taken at night. Blood samples were collected every week from the same animals per group (n = 10). Asterisk indicates significant difference ($P < 0.05$).

4. Discussion

The results show that melatonin implants have no effect on body weight when male Syrian hamsters are maintained in 14L or 10L photoperiods. Animals given constant-release melatonin implants did not differ from animals given empty implants over the course of the 10-week treatment period. These data are similar to those obtained in other studies (Hoffman, 1983; Bartness and Wade, 1984). This shows that the body weight response to photoperiod change may be somewhat independent of the perception of an endogenous melatonin rhythm.

Melatonin implants block gonadal regression in short-photoperiod–exposed animals. This result is consistent with previously obtained results (Turek, 1979; Reiter, 1980), indicating that implants leave the animals in a preoperative mode, presumably via a melatonin receptor system. It has been suggested that continuous exposure of the melatonin receptor to melatonin, by means of a

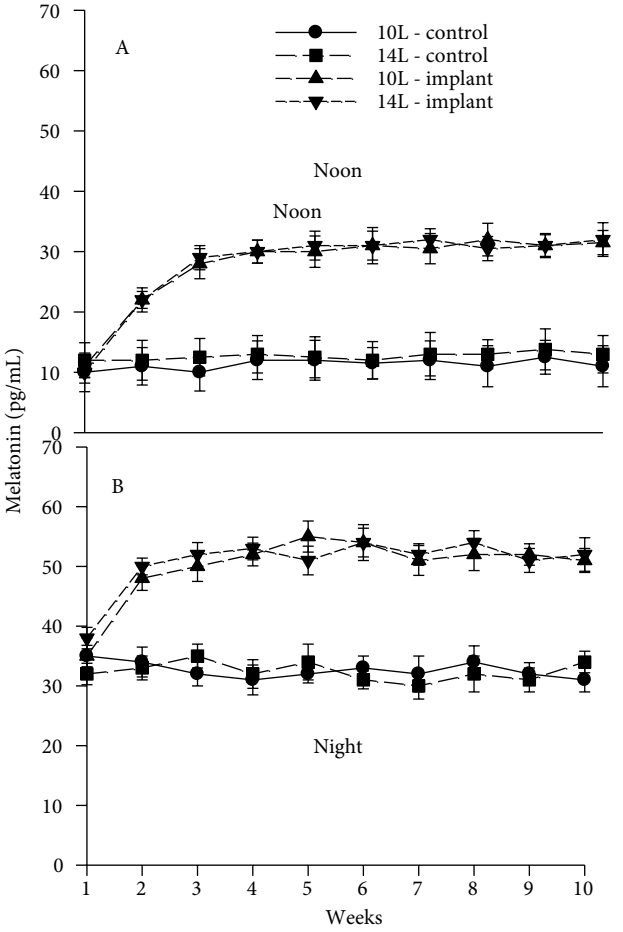

Figure 3. Syrian hamster mean serum melatonin concentrations. (A) Melatonin concentrations of control and melatonin-implanted animals (blood taken at noon in both photoperiods). (B) Melatonin concentrations of control and melatonin-implanted animals, blood taken at night in both photoperiods. Blood samples were collected every week from the same animals per group (n = 10).

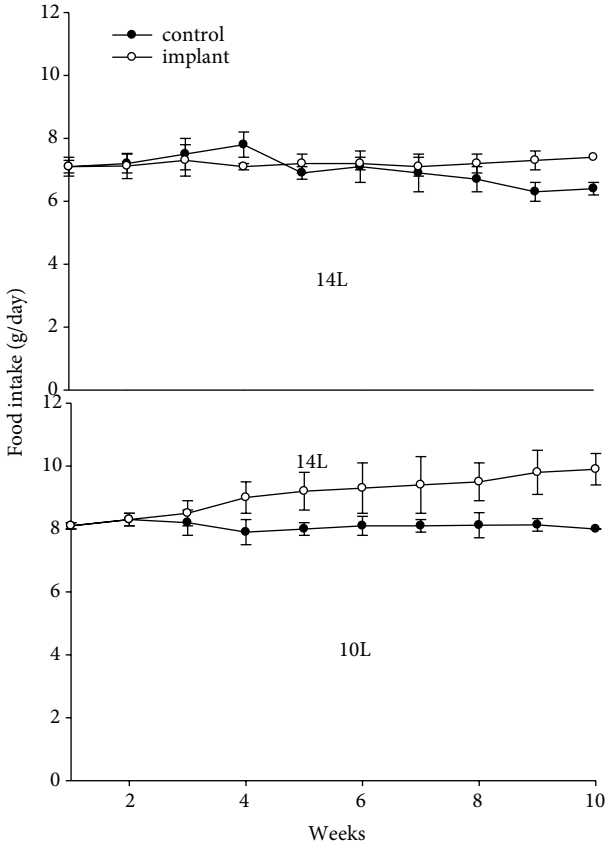

Figure 4. Food intake (g) in control and melatonin-implanted adult male hamsters in 14L (A) and 10L (B) over a 10-week period. Values are given as the mean ± SEM of 10 animals per group at each time point.

melatonin implant, prevents the receptor function and therefore blocks the action of short photoperiods.

The differences between noon and night leptin levels of the control groups in both photoperiods were significant. The leptin levels of the melatonin-treated animals were much lower. Furthermore, it has been suggested that melatonin may be the key regulator of leptin secretion in raccoon dogs and the Syrian hamster (Gündüz, 2002; Nieminen et al., 2002). There was a significant drop in plasma leptin levels of the melatonin-treated hamsters throughout the experiment, indicating that exogenous melatonin had lowered the leptin levels. The continuous-release melatonin capsules suppressed leptin concentrations in animals regardless of photoperiod; this suppression occurred by the first sample (within 1 week) and persisted for the duration of the study. Melatonin also abolished the diurnal pattern of variation in leptin. The suppression of leptin concentrations had no physiological correlates, as exemplified by changes in testicular weight or body weight, thus leading us to reject the hypothesis that photoperiod-dependent changes in reproductive function are mediated by leptin in Syrian hamsters.

The mechanism of the drop in plasma leptin concentrations by exogenous melatonin remains unclear. There might be a direct effect of melatonin on the expression of the ob mRNA. Zalatan et al. (2001) reported that physiological levels of melatonin could block fatty acid transport via a melatonin-receptor–mediated mechanism. Leptin is known to be produced in adipocytes in response to nutrient cycling (Wang et al., 1998). Thus, melatonin may decrease the release of leptin into the circulation by blocking fatty acid transport into the adipocytes. After implantation, leptin release into the circulation may decrease due to decreased fatty acid transport into adipocytes. It has been shown that melatonin given in

drinking water for 12 weeks suppresses plasma leptin levels independent of total body fat (Wolden-Hansen et al., 2000). In our study, melatonin was given subcutaneously for 10 weeks. During this time, changes in body weight were not seen; thus, chronic application of melatonin can be said to decrease leptin release without affecting body weight. The mechanism could also be the indirect stimulation of feeding efficiency and weight gain due to the shortened photoperiod. This leads to an increase in the amount of fat present in the body to secrete more leptin; however, in our model, this is not the case, because leptin levels drop (Figure 2C). This may be a signal to consume more food (Figure 4B) and gain more body weight in the presence of melatonin. The neuroendocrine response to fasting triggered by the fall of leptin levels in winter is a part of this survival mechanism (Ahima et al., 1996). The shortening photoperiod increases endogenous melatonin secretion, which is a signal for weight gain and fat storage in Syrian hamsters. The stored fat in the white adipose tissue informs the central nervous system via leptin about the amount of energy available for different purposes, such as wintering or reproduction. After the onset of wintering, food is scarce, and animals are forced to enter a phase of energy preservation. It is then reasonable to allow the neuroendocrine response to fasting, triggered by the fall of plasma leptin levels, to take place.

Since blood levels of leptin follow a diurnal rhythm in Syrian hamsters, and are linked to the melatonin diurnal rhythm, the genesis of this diurnal pattern and melatonin control over leptin release must be elucidated. In fact, it is not even clear whether, like many other hormonal rhythms, the diurnal plasma leptin rhythm is under direct control of the biological clock (suprachiasmatic nucleus, SCN). The SCN plays a key role in the induction, synchronization, and entrainment to the light-dark cycle of circadian rhythm among behavioral, metabolic, and endocrine functions (Rusak and Zucker, 1992). On the other hand, Kalsbeek et al. (2001) demonstrated that the SCN controls the diurnal rhythm of plasma leptin levels directly, independent of other clock-controlled rhythms (feeding, insulin), in rats. Although rats consume most of their food during the night, the nocturnal rise in circulating leptin levels in rats may be primarily due to increased food consumption at night, and changes in the daily pattern of food intake must affect plasma leptin levels (Saladin et al., 1995). Food restriction in Mongolian gerbils (*Meriones unguiculatus*) also suppresses serum leptin levels in both male and female gerbils (Karakas et al., 2005). Our animal model is a nocturnal animal, and food intake is more uniformly distributed throughout the day and night (Rowland, 1985). Differences in the phases of leptin rhythms are not due to different preferences in feeding time (Karakas and Gündüz, 2006). Although we did not estimate body fat content, the stability in body weight in response to photoperiods reflected changes in body fat stores and thereby confirmed that circulating leptin does not monitor body fat mass in male Syrian hamsters. Because leptin was suppressed by melatonin regardless of photoperiod and food intake was determined in the present study, it is reasonable to suggest that treatment with melatonin may have modified feed intake or mechanisms controlling fat storage (or the redistribution of energy stores to adipose tissue). The nature of the temporal information that melatonin treatment transmitted to the adipose cells is not known, but it modulates leptin secretion and expression, suggesting that the adipose tissue is a target for circadian regulation of the SCN. In the present study, which employed subcutaneous melatonin implants in pineal-intact hamsters, the marked diurnal variation in serum melatonin titers was abolished as a consequence of elevated daytime levels, but rhythmicity in serum melatonin persisted (Figure 3). It must be stressed that melatonin has receptors in the SCN (Williams et al., 1989), and it is possible that, similar to the rhythm in melatonin release, SCN control of leptin release is mainly stimulatory; thus, removal of this stimulatory SCN control results in decreased leptin levels (Karakas and Gündüz, 2006). Choi and Dallman (1999) reported that in SCN-lesion rats, increased plasma leptin levels are accompanied by obesity. Melatonin receptors of the SCN are important for the seasonal changes in reproductive status and adiposity in Siberian hamsters. The relationship between melatonin and leptin release seems to be different in seasonally and annually breeding animals. Thus, the circadian control of leptin secretion appears to be important and is controlled in a manner similar to melatonin.

In combination with a functional inverse relationship between plasma leptin levels and melatonin activity, the current results led to the proposition that the rhythm in plasma leptin levels might be due to the inhibitory effect of circulating melatonin on fat cells or the SCN. Interactions between melatonin and the leptin are probably species-specific and closely associated with the seasonal cycles of body weight and reproduction. SCN output pathways in seasonal animals are unclear. It appears that melatonin works via an intermediary, at least for the seasonal changes in adiposity (Ng and Wong, 1986). Whether leptin plays an important role in mediating seasonal changes in body weights and reproductive responses or signals to the metabolic state in different photoperiods remains to be determined. Our results confirm that continuously elevated levels of circulating melatonin do not inhibit nocturnal synthesis or release of endogenous melatonin but suppress circulating leptin levels. The nature of the temporal information that the melatonin continuous treatment sends to the adipose tissue is not known;

however, as demonstrated here, it modulates leptin secretion and expression, suggesting that the adipose tissue is a target for circadian regulation of the SCN. We think that understanding the regulation of leptin synthesis and secretion in seasonally breeding animals is a promising way to investigate the physiological role of melatonin and may allow for comprehensive knowledge of the biological rhythms.

References

Ahima RS, Prabakaran D, Mantzoros C, Qu DQ, Lowell B, Maratos-Flier E, Flier J (1996). Role of leptin in the neuroendocrine response to fasting. Nature 382: 250–252.

Baltaci AK, Mogulkoc R (2007). Pinealectomy and melatonin administration in rats: their effects on plasma leptin levels and relationship with zinc. Acta Biol Hung 58: 335–43.

Baltaci AK, Mogulkoc R (2012). Leptin and zinc relation: in regulation of food intake and immunity. Indian J Endocrinol Metab 16: 611–616.

Bartness TJ, Wade GN (1984). Photoperiodic control of body weight and energy metabolism in Syrian hamsters (*Mesocricetus auratus)*: role of the pineal gland, melatonin, gonads, and diet. Endocrinology 114: 492–498.

Bronson FH, Heideman PD (1994). The Physiology of Reproduction. New York, NY, USA: Raven Press.

Chehab FF, Lim M, Ronghua L (1996). Correction of sterility in omozygous female obese mice by treatment with the human recombinant leptin. Nature Genet 12: 318–320.

Choi SJ, Dallman MF (1999). Hypothalamic obesity: multiple routes mediated by loss of function in medial cell groups. Endocrinology 140: 4081–4088.

Dark J, Zucker I, Wade GN (1983). Photoperiodic regulation of body mass, food intake, and reproduction in meadow voles. Am J Physiol-Reg I 245: R334–R338.

Davis FC (1989). Daily variation in maternal and fetal weight gain in mice and hamster. J Exp Zool 250: 273–282.

Gündüz B (2002). Daily rhythm in serum melatonin and leptin levels in the Syrian hamster (*Mesocricetus auratus*). Comp Biochem Phys A 132: 393–401.

Hoffman RA (1983). Seasonal growth and development and the influence of the eyes and pineal gland on body weight of golden hamsters (*M. auratus*). Growth Factors 47: 109–121.

Hoffman RA, Davidson K, Steinberg K (1982). Influence of photoperiod and temperature on weight gain, food consumption, fat pads, and thyroxine in male golden hamsters. Growth Factors 46: 150–162.

Horton TH, Ray SL, Rollag MD, Yellon SM, Stetson MH (1992). Maternal transfer of photoperiodic information in Siberian hamsters. 5. Effects of melatonin implants are dependent on photoperiod. Biol Reprod 47: 291–296.

Kalsbeek A, Fliers E, Romijn JA, LaFleur SE, Wortel J, Bakker O, Endert E, Buijs RM (2001). The suprachiasmatic nucleus generates the diurnal changes in plasma leptin levels. Endocrinology 142: 2677–2685.

Karakas A, Camsari C, Serin E, Gündüz B (2005). Effects of photoperiod and food availability on growth, leptin, sexual maturation and maintenance in the Mongolian gerbils (*Meriones unguiculatus*). Zool Sci 22: 665–670.

Karakas A, Gündüz B (2006). Suprachiasmatic nuclei may regulate the rhythm of leptin hormone release in Syrian hamsters (*Mesocricetus auratus*). Chronobiol Int 23: 225–236.

Klingenspor M, Dickopp A, Heldmaier G, Klaus S (1996). Short photoperiod reduces leptin gene expression in white and brown adipose tissue of Djungarian hamsters. FEBS Lett 399: 290–294.

Klingenspor M, Niggemann H, Heldmaier G (2000). Modulation of leptin sensitivity by short photoperiod acclimation in the Djungarian hamster, *Phodopus sungorus*. Am J Physiol-Reg I B 170: 37–43.

Matthew JC, Donald LJ, Roberst AS (1999). Leptin's actions on the reproductive axis: perspectives and mechanisms. Biol Reprod 60: 216–222.

Nieminen P, Mustonen AM, Asikainen J, Hyvarinen H (2002). Seasonal weight regulation of the raccoon dog (*Nyctereutes procyonoides*): interactions between melatonin, leptin, ghrelin and growth hormone. J Biol Rhythm 17: 75–83.

Ng TB, Wong CM (1986). Effects of pineal indoles and arginine vasotocin on lipolysis and lipogenesis in isolated adipocytes. J Pineal Res 3: 55–66.

Rasmussen DD, Boldt BM, Wilkinson CW, Yellon SM, Matsumoto AM (1999). Daily melatonin administration at middle age suppresses rat visceral fat, plasma leptin and plasma insulin to youthful levels. Endocrinology 110: 1009–1012.

Reiter RJ (1980). The pineal gland and its hormones in the control of reproduction in mammals. Endocr Rev 1: 109–131.

Reiter RJ (1991). Melatonin: that ubiquitously acting pineal hormone. News Physiol Sci 6: 223–227.

Rowland NE (1985). Ingestive behavior of Syrian hamsters—advantages of the comparative approach. Brain Res 15: 417–423.

Rusak B, Zucker I (1992). Neural regulation of circadian rhythms. Physiol Rev 59: 449–526.

Saladin R, Devos P, Guerremillo M, Leturque A, Girard J, Staels B, Auwerx J (1995). Transient increase in obese gene-expression after food-intake or insulin administration. Nature 377: 527–529.

Steinlechner S, Niklowitz P (1992). Impact of photoperiod and melatonin on reproduction in small mammals. Anim Reprod Sci 30: 1–28.

Stetson MH, Rollag MD, Watson-Whitmyre M, Tate-Ostroff B (1983). The effects of daily injections and constant release melatonin on the endogenous pineal melatonin rhythm in golden hamsters. Exp Biol M 74: 119–122.

Turek FW (1979). Effect of melatonin photic-independent and photic-dependent testicular growth in juvenile and adult male golden hamsters. Biol Reprod 20: 1119–1122.

Wade GN, Schneider JE, Li HY (1996). Control of fertility by metabolic cues. Am J Physiol-Reg I 270: E1–E19.

Wang J, Liu R, Hawkins M, Barzilai N, Rossetti L (1998). A nutrient-sensing pathway regulates leptin gene expression in muscle and fat. Nature 393: 684–688.

Watson-Whitmyre M, Stetson MH (1985). A mathematical method for estimating paired testes weight from in situ testicular measurements in three species of hamster. Anat Rec 213: 473–476.

Williams LM, Morgan PJ, Hastings MH, Lawson W, Davidson G, Howell HE (1989). Melatonin receptor sites in the Syrian hamster brain and pituitary: localization and characterization using [^{125}I]iodomelatonin. J Neuroendocrinol 1: 315–320.

Wolden-Hansen T, Mitton DR, McCants RL (2000). Daily melatonin administration to middle-aged male rats suppresses body weight, intraabdominal adiposity, and plasma leptin and insulin independent of food intake and total body fat. Endocrinology 141: 487–497.

Zalatan F, Krause JA, Blask DE (2001). Inhibition of isoproterenol-induced lipolysis in rat inguinal adipocytes *in vitro* by physiological melatonin via receptor-mediated mechanism. Endocrinology 142: 3783–3790.

Ziylan YZ, Baltaci AK, Mogulkoc R (2009). Leptin transport in the central nervous system. Cell Biochem Funct 27: 63–70.