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SCIENCE DIRECT

Hormones and Behavior 48 (2005) 321 – 328

Hormones and Behavior

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Regulation of body weight and thermogenesis in seasonally acclimatized Brandt's voles (*Microtus brandti*)

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Received 8 October 2004; revised 25 February 2005; accepted 11 April 2005
Available online 1 June 2005

Abstract

Seasonal changes in an animal's morphology, physiology, and behavior are considered to be an adaptive strategy for survival and reproductive success. In the present study, we examined body weight and several behavioral, physiological, hormonal, and biochemical markers in seasonally acclimatized Brandt's voles (*Microtus brandti*) to test our hypothesis that Brandt's voles can decrease energy intake associated with decrease in body weight, body fat content, serum leptin level, and increasing thermogenesis in winter conditions. We found that the body weight of Brandt's voles was lowest in winter (December to February) and highest in spring and early summer (May to June). This seasonal variation in body weight was associated with changes in other markers examined. For example, the winter decrease in body weight was accompanied by increased energy intake and enhanced nonshivering thermogenesis (NST) as well as by decreased body fat mass and reduced levels of circulating leptin. Further, circulating levels of leptin were positively correlated with body weight and body fat mass, and negatively correlated with energy intake and uncoupling protein 1 contents. Together, these data do not support our hypothesis and suggest that leptin may be involved in this process and serve as a starvation signal in Brandt's voles.

Keywords: Energy intake; Basal metabolic rate (BMR); Nonshivering thermogenesis (NST); Leptin; Body fat; Uncoupling protein 1 (UCP1)

Introduction

Small mammalian species, such as rodents, usually show seasonal changes in their morphology, physiology, and behavior to cope with altered environmental conditions (Bartness et al., 2002; Concannon et al., 2001; Heldmaier et al., 1982; Klingenspor et al., 1996). For example, in response to decreased ambient temperature and a shortened photoperiod in winter, many rodent species reduce their overall body weights, decrease body fat, enhance thermogenic capacity, and cease reproductive behaviors (Bartness et al., 2002; Concannon et al., 2001; Demas et al., 2002; Drazen et al., 2000; Iverson and Turner, 1974; Klingenspor et al., 1996; Rafael et al., 1985; Voltura and Wunder, 1998). It has been demonstrated that, in winter, rodents may increase their nonshivering thermogenesis (NST) through uncoupling respiration in brown adipose tissues (BAT) (Jefimow et al., 2004; Li et al., 2001; Merritt, 1995; Merritt and Zegers, 1991; Steinlechner et al., 1983; Wang and Wang, 1996; Wang et al., 2003), and this process is usually associated with increases in BAT weights, mitochondria protein concentrations, and uncoupling protein 1 (UCP1) mRNA expression (Li et al., 2001; Nieminen and Hyvarinen, 2000; Praun et al., 2001; Rafael et al., 1985). UCP1, a 32-kDa molecule which is specifically located in the inner mitochondrial membrane, is responsible for adaptive thermogenesis in BAT during cold or winter. It can dissipate the proton motive force across the inner mitochondrial membrane as heat (Nicholls and Locke, 1984).

Leptin, a hormone derived primarily from the adipose tissue, plays an important role in controlling food intake, energy expenditure, and sexual behavior (Ahima and Flier, 2000; Dijk, 2001; Flier, 1998; Fox et al., 2000; Oates et al.,

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0018-506X/$ - see front matter © 2005 Elsevier Inc. All rights reserved.
doi:10.1016/j.yhbeh.2005.04.004

2000; Rayner and Trayhurn, 2001; Scarpace and Metheny, 1998; Schneider et al., 2000; Zhang et al., 1994). Exogenous leptin treatments decrease adiposity by decreasing food intake and increasing energy utilization (Abelenda et al., 2003; Polleymounter et al., 1995; Scarpace et al., 1997). In several field rodent species, annual cycles of food intake and adiposity have been found to be associated with fluctuation of serum leptin and of leptin gene expression in the white adipose tissue, implicating leptin in the regulation of seasonal changes in animal's body weight and energy balance (Concannon et al., 2001; Klingenspor et al., 1996; Rousseau et al., 2003). For example, Djungarian hamsters decreased their body weight, body fat mass, and food intake, and these decreases are associated with a decrease in leptin gene expression in white adipose tissue under short day and/or winter conditions (Bartness et al., 2002; Ebling et al., 1998; Klingenspor et al., 2000; Steinlechner et al., 1983). Klingenspor et al. (2000) further demonstrated that leptin sensitivity increased in short day and/or winter conditions even through leptin gene expression in WAT decreased, which might cause a satiety effect. The role of leptin in mammals in the field might be different from that in laboratory animals. Unlike mammals in the field, some laboratory rodents such as rats and mice increased their food intake in association with decreases in body weight, body fat, and serum leptin levels under cold acclimation (Abelenda et al., 2003; Bing et al., 1998). Thus, leptin might be a starvation signal to control food intake (Flier, 1998). Furthermore, leptin administration increased animal's body temperature, basal metabolic rates (BMR), NST, and UCP1 mRNA expression in BAT, indicating a potential involvement of leptin in thermogenesis (Scarpace et al., 1997; Polleymounter et al., 1995). However, contradictory results have also been reported. For example, in cold-acclimatized rats, leptin administration reduced BAT thermogenesis (Abelenda et al., 2003) and low serum leptin levels were accompanied by an increase in UCP1 gene expression (Bing et al., 1998).

Brandt's voles (*Microtus brandti*) live mainly in grasslands of Inner Mongolia of China, the Republic of Mongolia, and the region of Beigaer Lake in Russia (Zhang and Wang, 1998). In these habitats, climate shows remarkable seasonal variation with winter lasting for 5 months. Therefore, this rodent species could serve as an ideal model to study winter survival and adaptive strategy. It has been reported that Brandt's voles showed relatively stable BMR and seasonal changes in NST (Wang et al., 2003), and increased NST was associated with an increase in UCP1 mRNA expression (Li et al., 2001). The role of leptin in seasonal changes of body weight, energy balance, and thermogenesis in the Brandt's voles is still unknown. We hypothesized that Brandt's voles, like Djungarian hamsters, can decrease energy intake in association with decreases in serum leptin levels, body weight, body fat, and increases in thermogenesis in winter condition. In the present study, we examined several behavioral, morphological, physiological, hormonal, and biochemical measurements in seasonally acclimatized Brandt's voles. We particularly focused on leptin to study not only its seasonal changes but also its correlation with changes in other measurements indicative of an animal's thermogenic capacity to investigate the role of leptin in the regulation of seasonal changes of energy balance and thermogenesis.

Materials and methods

Subjects

Subjects were offspring of adult Brandt's voles that were captured in Inner Mongolia in May 1999 and then transported to the Institute of Zoology, Chinese Academy of Sciences in Beijing, China. Subjects were housed in groups (3–5) in plastic cages (30 × 15 × 20 cm) that contained sawdust bedding. All animals were maintained under 16L: 8D photoperiod with lights on at 0400 h, and room temperature was kept around 23 ± 1°C. All animal procedures were approved by the Institutional Animal Care and Use Committee of the Institute of Zoology, Chinese Academy of Sciences. Subjects were fed ad libitum with standard rabbit food and water. At about 70–90 days of age, all voles were moved from the animal house to an outdoor enclosure (natural condition) at the same time (July, 2001) and housed individually. After 1-month stabilization, subjects were randomly assigned into one of six experimental groups, and experiments were performed in August, October, and December of 2001, and in February, May, and June of 2002, in other words, after 1, 3, 5, 7, 10, and 11 months acclimatization in natural environmental conditions, respectively. First, subjects' energy intake, basal metabolic rate (BMR), and nonshivering thermogenesis (NST) were measured. They were then sacrificed via decapitation, and blood and tissue samples were taken for measurement of physiological parameters (see below). An additional group of subjects was also housed in an outdoor enclosure (natural condition) individually in July of 2001, and their body weights were measured at 15-day intervals throughout a year.

Energy intake

Energy intake for each experimental group was measured for 3 days as described previously (Liu et al., 2002; Song and Wang, 2001). During each test, subjects were housed individually in stainless steel mesh metabolic cages (0.24 × 0.24 × 0.24 m), where food was provided in excess of the animals' needs and water was provided ad libitum. The uneaten food and feces were collected after the 3-day test, separated manually, and oven-dried at 70°C for at least 72 h. The caloric contents of food and feces were determined by Parr1281 oxygen bomb calorimeter (Parr Instrument USA; Liu et al., 2002; Song and Wang, 2001).

Metabolic trials

The basal metabolic rate (BMR) was measured by using an established closed-circuit respirometer at 29°C (Song and Wang, 2001; Wang et al., 2000). Briefly, the metabolic chamber volume is 3.6 l and the temperature inside the chamber was maintained by a water bath (±0.5°C). KOH and silica gel were used to absorb carbon dioxide and water in the metabolic chamber. Subjects were fasted for 3 h prior to being put into the metabolic chamber. After a 40-min adaptation, oxygen consumption was recorded for 60 min at 5-min intervals. The two stable consecutive lowest readings were used to calculate BMR and corrected to standard temperature and pressure (STP). Subjects' body temperature was measured before and after each test. All metabolic measurements were performed between 1000 and 1700 h.

NST was induced by scapular subcutaneous injections of 0.2 mg/ml norepinephrine bitartrate (NE) (Shanghai Harvest Pharmaceutical Co. LTD) at 25 ± 1°C. The dosage of NE was calculated based on the equation described by Helamaier (1971): NE dosage (mg/kg) = 6.6 $M_b^{-0.458}$ (g). The two consecutive highest recordings of oxygen consumption were taken to calculate the maximum NST (Song and Wang, 2003).

Measurement of UCP1 and serum leptin

The interscapular BAT was removed immediately after subjects were sacrificed, and each pad was then trimmed of connective tissue and weighed. Mitochondrial protein was prepared as described in Wiesinger et al. (1989). Mitochondrial protein concentrations were determined by the Folin phenol method (Lowry et al., 1951) with bovine serum album as the standards. Total BAT protein (20 μg per lane) was separated in a discontinuous SDS–polyacylamide gel (12.5% running gel and 3% stacking gel) and blotted to a nitrocellulose membrane (Hybond-C, Amersham). To check for the efficiency of protein transfer, gels and nitrocellulose membranes were stained after transferring with Coommassie brilliant blue and Ponceau red, respectively. UCP1 was detected using a polyclonal rabbit anti-hamster UCP1 (1:5000) as a primary antibody and goat anti-rabbit (1:5000) as the secondly antibody (Klingenspor et al., 1996). We used enhanced chemoluminescence (ECL, Amersham) for detection and unspecific binding sites were saturated with 5% non-fat dry milk in PBS. UCP1 concentration was expressed as relative units (RU), as determined from area readings by using Scion Image.

All subjects were sacrificed between 0900 and 1100 by puncture of the posterior vena cava. Blood was centrifuged at 4000 rpm for 30 min, and serum was sampled and stored at −75°C. Serum leptin levels were measured by radioimmunoassay (RIA) using the Linco ^{125}I Multi-species Kit (St. Louis, MO) and leptin values were determined in a single RIA. The lowest level of leptin that can be detected by this assay was 1.0 ng/ml when using a 100μl sample size (Instructions for Multi-species leptin RIA Kit). Inter- and intra-assay variability for leptin RIA was <3.6% and 8.7%, respectively.

Carcass composition analysis

The entire gastrointestinal tract was removed and the eviscerated carcass (not including BAT) was dried to constant weight at 60°C for determination of dry body weight. Total body fat was extracted from the dried carcass by ether extraction in a Soxhlet apparatus.

Data analysis

Data were analyzed using SPSS software (SPSS, 1988). Distributions of all variables were tested for normality using the Kolmogorov–Smirnov test. Non-normally distributed data were transformed to natural logarithms. To standardize body weight influences, BMR, NST, and energy intake were scaled to the 0.67 power of body mass ($BW^{0.67}$) as proposed for rodents (Heusner, 1984; Pei et al., 2001). Data were analyzed by one-way analysis of variance (ANOVA) and significant group differences were further evaluated by LSD post hoc test. To detect possible associations of serum leptin with body weight, body fat mass, energy intake, or UCP1, we used Pearson correlation analyses. All values were expressed as mean ± SEM, and $P < 0.05$ was considered to be statistically significant.

Results

Body weight

The Brandt's voles showed marked seasonal variations in their body weight ($F_{(19,140)} = 25.07$, $P < 0.001$, Fig. 1). Body weights dropped significantly and showed a 25.8% decrease from August to October, stayed at this level until March, increased rapidly, and then reached a peak in May. The voles' body weights in June increased 44.0% compared to March and 28.1% compared to August of the previous year.

BMR and NST

The BMR and NST data are summarized in Table 1. No group differences were found for BMR ($F_{(5,35)} = 1.83$, $P = 0.13$). However, a marked group difference was found in NST ($F_{(5,35)} = 9.66$, $P < 0.001$). The post hoc test indicated the highest NST in December and February, which were significantly higher than that in August, May, and June. In June, voles had the lowest NST compared to other time points examined.

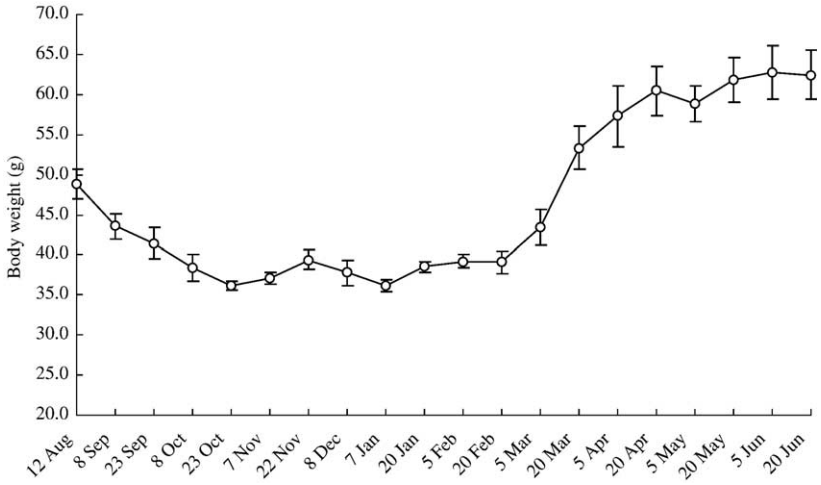

Fig. 1. Seasonal changes of body weight in Brandt's voles. Body weights of Brandt's voles began to decrease in August and reached the lowest level in October, then stayed at stable levels until March, and increased again and reached the highest levels in May. Values are expressed as mean ± SEM. $P < 0.05$ was considered to be statistically significant ($n = 8$ for seasonal acclimatization).

Energy intake, body composition, and UCP1 contents

Brandt's voles showed seasonal variations in their energy intake ($F_{(5,35)} = 11.27$, $P < 0.001$, Table 1). The highest energy intakes were found in December and February compared to any other time points. In addition, energy intakes in October and May were significantly higher compared to those in August.

Body composition, in particular body fat mass, also displayed seasonal variation ($F_{(5,35)} = 9.41$, $P < 0.001$, Table 1). Body fat mass was significantly higher in May and June than in October, December, and February. In addition, body fat mass in August was higher than in December and February. Similarly, lean dry body weight also showed seasonal changes ($F_{(5,35)} = 10.80$, $P < 0.001$, Table 1). The voles had the lowest lean dry body weights in October, December, and February, and the highest lean dry body weights in May and June. Finally, no group differences were found in UCP1 contents of BAT (Table 1).

Serum leptin levels

Serum leptin levels also showed marked fluctuations during seasonal acclimatization ($F_{(5,35)} = 13.24$, $P < 0.001$, Fig. 2). Serum leptin levels were lowest in August, increased significantly in October, remained at this level until February, and then increased again and reached a peak in May and June. Correlation analysis indicated that this seasonal change in serum leptin levels was positively correlated with changes in overall body weight ($r = 0.57$, $P < 0.001$; Fig. 3A), body fat mass ($r = 0.42$, $P < 0.01$; Fig. 3B), and negatively correlated with energy intake ($r = 0.34$, $P < 0.05$; Fig. 3C) and UCP1 contents ($r = 0.36$, $P < 0.05$; Fig. 3D).

Discussion

Seasonal changes in body weight especially in small mammals are considered to be adaptive strategies essential for survival and reproductive success (Concannon et al., 2001; Drazen et al., 2000; Iverson and Turner, 1974; Merritt, 1995; Merritt and Zegers, 1991; Millar, 1987). In the present study, we found that the body weight of Brandt's voles was lowest in winter (December to February) and highest in spring and early summer (May to June). This seasonal variation in body weight was associated with changes in several behavioral, physiological, hormonal, and biochemical markers, all indicative of altered thermogenic

Table 1
Carcass composition, energy budgets, and thermogenic parameters in seasonal acclimatized Brandt's voles (mean ± SEM)

	15 August	17 October	11 December	24 February	8 May	26 June	*P*
Sample size	8	6	6	7	7	7	
Dry carcass mass (g)	19.4 ± 1.8^{a}	14.3 ± 1.6^{b}	9.8 ± 1.0^{b}	13.8 ± 0.7^{b}	25.4 ± 2.9^{c}	22.9 ± 1.5^{ac}	0.01
Fat mass (g)	10.8 ± 1.4^{ac}	7.5 ± 1.3^{ab}	3.8 ± 1.0^{b}	5.4 ± 0.4^{b}	13.4 ± 1.7^{c}	12.1 ± 1.4^{c}	0.01
Fat free mass (g)	8.6 ± 0.5^{a}	6.8 ± 0.5^{ac}	6.0 ± 0.8^{c}	8.4 ± 0.4^{a}	12.0 ± 1.2^{b}	10.8 ± 0.7^{b}	0.01
Energy intake (kJ $g^{-0.67}$ day^{-1})	17.5 ± 2.3^{a}	28.6 ± 3.2^{b}	43.6 ± 7.1^{c}	45.7 ± 2.9^{c}	28.9 ± 1.7^{b}	20.1 ± 3.1^{ab}	0.01
Basal metabolic rate (ml $O_2/g^{0.67}$·h)	9.9 ± 0.6	9.9 ± 1.0	9.4 ± 1.1	11.9 ± 0.9	9.7 ± 0.6	8.8 ± 0.4	ns
Nonshivering thermogenesis (ml $O_2/g^{0.67}$·h)	19.2 ± 1.1^{ad}	21.0 ± 0.7^{ab}	24.8 ± 0.9^{b}	23.6 ± 1.2^{b}	17.4 ± 2.0^{cd}	14.7 ± 0.6^{c}	0.01
UCP1 (relative unit, RU)	1.30 ± 0.12	2.35 ± 1.28	1.85 ± 1.06	1.76 ± 0.34	1.57 ± 0.28	1.35 ± 0.24	ns

Means with the same superscripts within rows are not significantly different ($P > 0.05$).

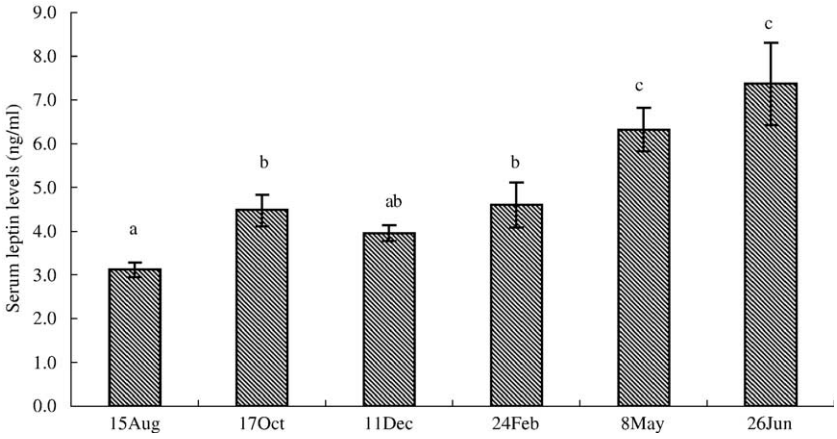

Fig. 2. Seasonal changes of serum leptin concentration in Brandt's voles. Serum leptin levels also showed remarkable fluctuations during seasonal acclimatization and were lowest in August, increased significantly in October, remained at this level until February, and then increased again and reached a peak in May and June. Values are expressed as mean ± SEM. $P < 0.05$ was considered to be statistically significant.

capacity. For example, in comparison to spring/summer, the winter decrease in body weight was accompanied by increased food intake and NST as well as by decreased body fat mass and serum leptin contents. Serum leptin was positively correlated with body weight and body fat mass, and negatively correlated with energy intake and UCP1 contents. Together, these data indicate that altered thermogenesis underlies seasonal changes in body weight of Brandt's voles and that leptin may be involved in this process.

Seasonal changes of body weight and thermogenic capacity

Our finding of winter-associated decreases in body weight of Brandt's voles is consistent with the previous finding in the same species (Wang et al., 2003) as well as in other species of rodents (Concannon et al., 2001; Iverson and Turner, 1974; Merritt, 1995; Merritt and Zegers, 1991). To cope with winter stress and energy requirements, animals may reduce their body weight (Merritt, 1995; Merritt and Zegers, 1991; Steinlechner et al., 1983) but increase their

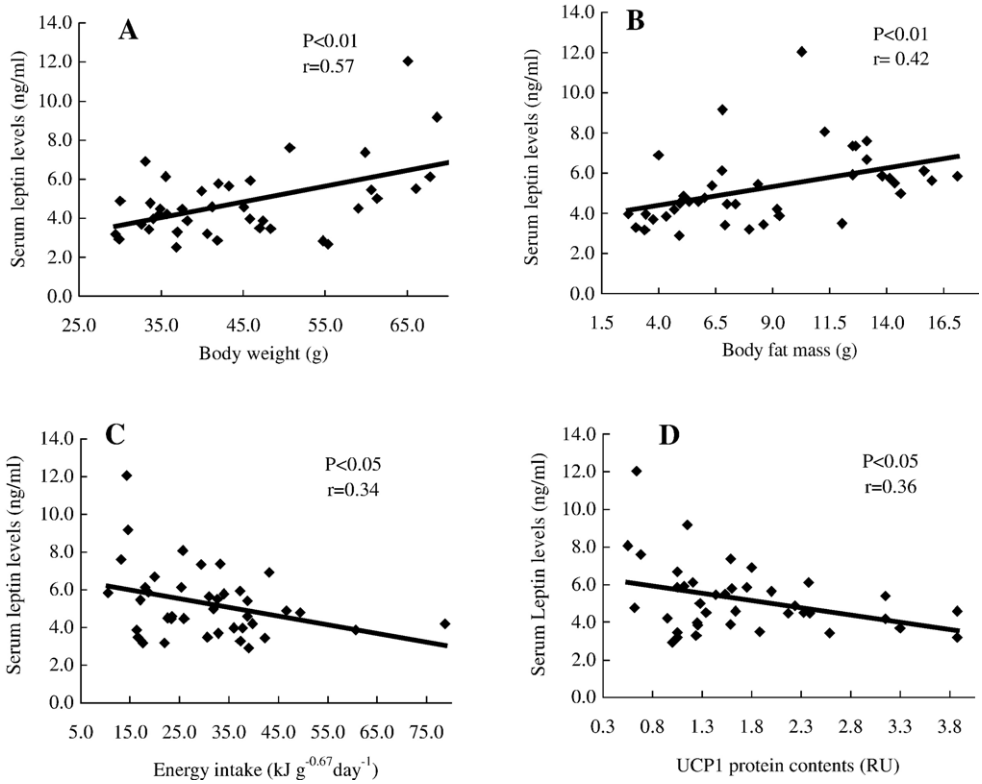

Fig. 3. Correlation of serum leptin levels with body weight (A), body fat mass (B), energy intake (C), and UCP1 contents (D) in seasonal acclimatized Brandt's voles. Serum leptin level was positively correlated with changes in overall body weight ($r = 0.567$, $P < 0.001$), body fat mass ($r = 0.423$, $P < 0.01$), and negatively correlated with energy intake ($r = 0.340$, $P < 0.05$,) and UCP1 contents ($r = 0.360$, $P < 0.05$).

energy intake and thermogenesis (Bing et al., 1998; Jefimow et al., 2004; Heldmaier et al., 1982; Li et al., 2001; Merritt, 1995; Merritt and Zegers, 1991; Song and Wang, 2001; Wang et al., 2003). This notion is supported by our data from the present study. For Brandt's voles, the decrease in body weight can increase the ratio of surface area to volume and therefore cause more heat loss than large animals, which might increase energy usage to maintain body temperature. Brandt's voles in winter (December–February) showed a decrease in their overall body weight and an increase in energy intake compared to those in spring/summer (May–June). Further, the voles in winter showed an elevated level of NST compared to the voles in summer, indicating a winter-associated enhancement in thermogenic capacity (Wang et al., 2003). Such seasonal changes in thermogenesis have also been documented in several other rodent species including Djungarian hamsters (Heldmaier et al., 1982; Rafael et al., 1985), golden hamsters (Jefimow et al., 2004), and Mongolian gerbils (Li et al., 2001; Wang et al., 2003).

Brandt's voles also displayed a significant decrease in body fat mass in winter compared to spring/summer. Seasonal regulation of body fat mass has been found to be species-specific. For example, ground squirrels increase their body fat mass before hibernating for winter survival (Ormseth et al., 1996). On the other hand, Djungarian hamster and common shrew (Nieminen and Hyvarinen, 2000; Rafael et al., 1985; Rousseau et al., 2003) display winter-associated decreases in body fat mass, possibly due to increased catabolism (Bartness et al., 2002; Bing et al., 1998; Voltura and Wunder, 1998). Therefore, species-specific patterns of seasonal changes in body fat may serve as specific strategies in dealing with seasonal changes in climate and food availability (Mercer, 1998). Interestingly, Brandt's voles in winter also displayed a significant decrease in fat free body weight, indicating that these animals have involved a physiological adaptation to maintain a low body fat content in winter to face the periods of insufficient energy resources (Flier, 1998).

Finally, UCP1 contents or mRNA expression may serve as an indicator of NST capacity. It has been demonstrated that cold- or short-photoperiod-induced elevation in NST was accompanied by increased UCP1 expression in several rodent species (Abelenda et al., 2003; Demas et al., 2002; Praun et al., 2001). In Brandt's voles, cold was found to induce concurrent increases in both NST and UCP1 (Li et al., 2001; Yang et al., 2003). In the present study, however, no seasonal differences were found in UCP1 contents in the BAT of the Brandt's voles. The inconsistency between these results and results from the earlier studies could not be easily explained. One possibility is that changes of multiple environmental factors (such as temperature, photoperiod, and food quality, etc.) during acclimatization in the present study exerted more complicated effects on the UCP1 contents compared to manipulations of a single factor. Nevertheless, this speculation needs to be tested in further studies.

Seasonal changes of serum leptin levels

Leptin, a hormone that is predominantly produced in adipose tissues, plays important roles in the regulation of body weight and energy balance. In the present study, the serum leptin level displayed seasonal fluctuations; it was significantly lower in fall/winter than in spring/summer. Similar seasonal changes in serum leptin levels and in leptin gene expression have also been found in other animals. For example, common shrew showed a decrease in BAT secretion of leptin during later autumn and winter (Nieminen and Hyvarinen, 2000). In Djungarian hamsters, leptin gene expression in adipose tissues was significantly reduced in winter (Klingenspor et al., 1996). It has been suggested that decreased leptin secretion and/or leptin gene expression were due to the inhibitory effects of cold temperature that increases the adrenergic tone (Hardie et al., 1996; Trayhurn et al., 1995) and sympathetic nerve activity (Li et al., 1997).

A positive correlation between serum leptin levels and body fat mass in Brandt's voles was observed in the present study. Similar results have also been found in collared lemmings and Djungarian hamsters (Klingenspor et al., 2000; Nagy et al., 1995). Interestingly, seasonal adaptation adjustments of Brandt's voles are different from those in Djungarian hamsters, even though both species showed low leptin levels in association with low body weight and body fat. Energy intake in Brandt's voles increased significantly in winter, and on the contrary, Djungarian hamsters presented a decrease in energy intake in winter, which implied that the role of leptin in the regulation of energy intake in Djungarian hamsters is different from Brandt's voles. Further experiment demonstrated that Djungarian hamsters that were exposed to short photoperiod had a higher sensitivity in response to exogenous leptin administration than the ones that were exposed to long photoperiod, and this increased sensitivity to leptin may play an important role for the animal's winter survival (Klingenspor et al., 2000). Furthermore, reduced leptin concentrations were permissive of torpor in Siberian hamsters, which implied that leptin was also important for torpor or winter adaptations (Freeman et al., 2004; Nelson, 2004). Brandt's vole is a non-hibernating small mammal, and this may be the reason for the different roles of leptin in the regulation of energy intake from other animals such as Djungarian hamsters. Indeed, decreased serum leptin in winter was found to be associated with increased energy intake in Brandt's voles in our study. Even though there was a negative relationship between leptin and energy intake, in the absence of an exogenous leptin injection experiment, we cannot be sure whether the decrease of serum leptin contributes to energy intake. To determine the role of leptin in the regulation of energy intake in winter, it is necessary to detect whether Brandt's voles could be sensitive to

exogenous leptin when their endogenous leptin decreased in winter. Flier (1998) hypothesized that leptin may act as a starvation signal to increase energy intake in laboratory rats and mice. Leptin acts on food intake and energy expenditure via interactions with receptors in the hypothalamus (Good, 2000; Pelleymounter et al., 1995; Shwartz et al., 2000). Our data indicated that Brandt's voles were more like laboratory animals, and not like Djungarian hamsters, in responding to the low serum leptin in winter by significantly increasing energy intake to compensate for the increased energy expenditure associated with enhanced thermogenesis.

Although leptin is involved in energy storage and expenditure, its interactions with UCP1 in BAT are still controversial. Leptin administration could increase UCP1 expression in rats by increasing sympathetic outflow to BAT (Scarpace and Metheny, 1998; Scarpace et al., 1997). However, low serum leptin levels were found to accompany increased UCP1 gene expression in rats during cold acclimation (Bing et al., 1998), and leptin administration to cold-acclimated rats reduced BAT thermogenesis (Abelenda et al., 2003). The divergent reports about leptin and UCP1 expression point out a complex regulation of energy balance under cold ambient temperature, indicating that leptin can regulate thermogenesis by monitoring energy demands and expenditures (Abelenda et al., 2003; Bing et al., 1998). It is a common phenomenon that UCP1 plays a key role in the molecular thermogenesis of BAT in small mammals under short photoperiod and/or cold winter conditions. In the present study, although there was a negative correlation between serum leptin levels and UCP1 contents, UCP1 contents of BAT did not show significant seasonal changes in Brandt's voles, perhaps because of the small sample sizes. It is possible that failure to detect seasonal changes in UCP1 contents prevented us from detecting a clear possible interaction between leptin and UCP1 in the present study. Alternatively, leptin may act through different pathways to regulate energy balance and thermogenesis in Brandt's voles. This scenario needs to be examined in further studies.

In summary, Brandt's voles increased energy intake and thermogenesis in association with decreases in body weight, body fat mass, and serum leptin levels in winter conditions, which do not support our hypothesis. Leptin may act as a starvation signal to permit the increase in energy intake for energy exhaust mainly as thermogenesis for winter adaptation.

Acknowledgments

We wish to thank Dr. M. Klingenspor at Department of Biology, Phillips-University Marburg, Germany, for supplying the hamster UCP1 antibody and for his helpful suggestions during this study. Thanks to Dr. Thomas J. Curtis and Dr. Zuoxin Wang at the Psychology Department of Florida State University for constructive suggestions and correcting the English expression. Two anonymous reviewers made numerous helpful suggestions. This study was supported by grants from the National Natural Science Foundation of China (No. 30170151 and 30430140) and the Chinese Academy of Sciences (No. KSCX2-SW-103) to DHW.

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