



Re-feeding evokes reproductive overcompensation of food-restricted Brandt's voles

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ABSTRACT

In natural conditions, animals have to cope with fluctuations of food resources. Animals having experienced prolonged decrease in feeding opportunities may increase their reproductive success when meeting abundant food. Though food restriction is well known to reduce reproductive success of animals, it is not clear whether re-feeding can restore or even overcompensate the reproductive success. In this study, we investigated the differences in reproductive parameters between food-restricted and re-fed (FR–RF) group and control group of Brandt's vole (*Lasiopodomys brandtii*). For 4 weeks, FR–RF voles were provided with 70% of their normal daily food intake and then they were fed ad libitum for the next 4 weeks. Voles of control group were fed ad libitum for 8 weeks. Females (FR–RF or control) were mated to non-littermate males of the same group (FR–RF or control), and we found that the mean litter size and survival rate of F1 pups of FR–RF group were significantly higher than those of control group. We also provided a field example showing that the litter size of Brandt's voles tended to be higher if they experienced two consecutive dry and wet months than that of voles didn't have this experience. Our results suggest that re-feeding may have evoked an overcompensatory mechanism of food-restricted voles in reproductive success. This may be an adaptive strategy for Brandt's voles (with oscillating populations) to cope with the fluctuating food resources in natural conditions by adjusting their reproductive success.

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1. Introduction

Due to spatial and temporal variations in food availability, rodents often face food shortage in a certain stage of their life cycles [1,2]. Food availability has been shown to be an important factor in causing variations in population numbers of animals [3–5]. Pulsed resources from precipitation often cause rapid growth of plants, and then the growth of herbivores, or even carnivores at higher trophic levels in many terrestrial and aquatic ecosystems [3,6]. Abundant precipitation, which often increases food supply to rodents, has been shown to promote population growth of small rodents, e.g. in Australia [7,8], South America [9,10], Africa [11] and Asia [12]. Animals having experienced prolonged periods without sufficient food may increase their reproductive success (defined as high reproductive output and offspring survival) when they encounter abundant food resources [13,14].

Food restriction can result in the body weight loss of rats [15]; suppress the function of testis, decrease the concentration of serum testosterone and influence the breeding of rodents [16,17]; for example, Wistar rats, diet with 23% of protein in restricted quantities, presented a significant decrease in testis (20%) and epididymis (14%) weights, in serum (78%) and testicular (68%) testosterone concentrations as well as in copulatory efficiency (26%). But it enhances the endogenous diurnal peak of plasma total corticosterone in young rats [18]. The effects of food restriction can also influence the sex ratio, body growth, reproduction and survival of their F1 or F2 offspring through maternal effects [19–21]. Maternal effect is the effect on offspring originated from parents not through genetic changes [22]. Mothers may pass on traits or characteristics to their offspring (thereby maternal effects), and that sometimes these traits are adaptive and beneficial, but sometimes mothers face experiences (e.g., reduction in food availability) that may be detrimental to their offspring [23–26]. Past or current food restriction often results in reduced reproductive success [27], but longer life span [28]. The potential benefit is that an increased chance of survival under food restriction may permit reproductive value when the famine ends [29,30].

Re-feeding can restore body weight and physiological conditions of food-restricted animals [31]. Re-feeding can reverse the

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suppression of copulatory behaviors of food-restricted female musk shrews (*Suncus murinus*) [32], or increase the litter size of red-backed vole (*Clethrionomys gapperi*) experienced malnutrition [33]. However, it is not clear whether re-feeding could evoke overcompensation in reproductive success in animals after food restriction (Reproductive Overcompensation Hypothesis). We define overcompensation as that the reproductive success (reproduction and/or offspring survival) of food-restricted and refed animals is significantly higher than that of control animals with normal feeding (supplied with abundant food and water). Such an overcompensatory mechanism in reproductive success may be important for animals to match food resources in natural conditions.

Brandt's vole (*Lasiopodomys brandtii*), is widely distributed in the grasslands of Inner Mongolia (China), Mongolia, and the Beigaer region of Russia [34]. It is one of several species in the grasslands of China that require management, to reduce competition with livestock, reverse degradation of the grassland, or to prevent outbreaks of zoonotic diseases such as bubonic plague [35,36]. Brandt's vole usually breeds from April to August [37]. Grass vegetation is an important factor acting as both food and shelter, which affects the population density [34,38]. The population density fluctuates greatly from year to year, and it varies from 1000 to 3000 per hm^2 in Mongolia [34,36]. In the semi-arid grasslands of Inner Mongolia, the growth of grass is largely dependent on precipitation in spring and summer [34]. High rainfall was found to benefit sharp increase in population density of Mongolia gerbils (*Meriones unguiculatus*) [12], and Brandt's voles [39] in this region.

In Inner Mongolia, precipitation in breeding seasons changes greatly from month to month. Previous field observations indicate that reproduction of Brandt's vole is significantly influenced by monthly precipitation in the breeding seasons (Table 2), suggesting that pulsed food abundance induced by abundant precipitation may evoke high reproductive success and then result in rapid population growth of Brandt's vole in the semi-arid grasslands.

In this study, we wanted to explore: (1) the effect of re-feeding on reproductive output and offspring survival in a laboratory colony of Brandt's vole having experienced food restriction, by manipulating the food availability of adult males and females; (2) the changes of hormone (testosterone and corticosterone) levels of male voles between experimental voles after re-feeding and Control voles; (3) whether there is a long-term fitness correlation between maternal generation and the progeny of Brandt's vole after the food restriction and re-feeding experience; (4) the relationship between monthly variation of precipitation and reproduction of Brandt's voles in field conditions to verify our time scale of food restriction and re-feeding experiment.

2. Materials and methods

2.1. Animals and housing conditions

We used 30 pairs of Brandt's voles captured in the grasslands of Inner Mongolia to build up the initial colony of our laboratory. The adult voles used in this study were the third generation from this colony.

Before experiments, voles were housed individually in plastic cages (25 cm \times 14 cm \times 14 cm) with sawdust as bedding and a wire mesh cover on the top of the cage. All voles used in this study were 120–150 days of age, sexually naïve, and were normally fed in sibling groups after weaning. All experimental female voles had similar reproductive status without experiences of pregnancy, lactating or both. The room temperature was kept at $23 \pm 1^\circ\text{C}$, and maintained with a 14 h: 10 h light–dark cycle. Commercial rabbit pellets (Beijing KeAo Feed Co., Beijing, China) and water were provided ad libitum.

2.2. Food restriction and re-feeding treatment

We measured the daily food intake of voles based on free food consumption. Voles were supplied with quantitative food (more than their daily consumption) everyday, and we reclaimed the left food next day at the same time to measure the daily food consumption of each animal. This was done for seven successive days.

We did three replicates (the 1st, 2nd and 3rd experiments) to examine the effect of food-restricted and refed experience on reproductive success. Food-restricted voles were provided with 70% food of normal food intake. Our preliminary study indicated that this food level for 4 weeks caused significant decline of body weight and reproduction, but little decline in survival of adult voles as compared with controls. Further, in the breeding seasons, the reproduction of voles is significantly related to the monthly (about 4 weeks) variations of food as represented by precipitation (Table 2).

2.3. FR–RF effect on maternal voles and F1 offspring

In the 1st experiment, voles were weighed and randomly assigned to two groups: food-restricted and refed group (FR–RF), and control group. There was no significant difference in initial body weight between the two groups. Voles of the FR–RF group (20 males and 21 females) were provided with 70% of normal daily food intake in the first 4 weeks, and then they were fed ad libitum in the following 4 weeks. Voles of control group (18 males and 21 females) were fed ad libitum throughout the 8 weeks. When the 4-week re-feeding period was finished, males and females of the same group (FR–RF or control) were paired in cages (30 cm \times 15 cm \times 20 cm) for seven days. Males were removed from the cages when females showed the appearance of copulatory plugs, and then females were housed individually and fed ad libitum. We used pregnancy rate, mean litter size of female voles; sex ratio, body weight at birth and weaning, and survival rate at weaning of litters to measure the reproductive success.

The 2nd and 3rd experiments (as replicates of the 1st one) were conducted under the same conditions of temperature, humidity and photoperiod, but in different months of the year. There was no significant difference in initial body weight between the FR–RF group (48 males and 59 females) and the control group (40 males and 40 females) in the second experiment, and between the FR–RF group (25 males and 25 females) and the control group (25 males and 25 females) in the third experiment.

2.4. Serum testosterone and corticosterone levels

Male voles of both groups from experiment 3 were sacrificed after being mated with females. We randomly selected 40 males to examine changes in the testosterone and corticosterone concentrations. Whole blood sample was collected in 2 mL eppendorf tube, allowed to clot at room temperature and centrifuged at 4000 rpm for 20 min for serum harvest. Testosterone and corticosterone concentrations were determined by rat ELISA (enzyme-linked immunosorbent assay) kit of testosterone (Cat. No. 3B930, RapidBio Lab. Calabasas, CA, USA) and kit of corticosterone (Cat. No. HR083). Detailed procedures were done following the manufacturer's instructions of the kit when using a 125 μL sample. Intra- and inter-assay coefficients of variation were less than 5% and 10%, respectively.

2.5. Effect on F1 and F2 offspring:maternal effect

We designed the 4th experiment to examine the maternal effect or cross-generation effect of Brandt's voles caused by the nutrition changes. In the 4th experiment the reproductive success of F1 generation and the survival of F2 offspring were examined. Male and female F1 offsprings of the same group (FR–RF experimental or control but from different families) from the third experiment were

paired for mating for seven successive days when they were 120 days old. We also used pregnancy rate, mean litter size of female voles; sex ratio, body weight at birth and weaning, and survival rate at weaning of litters to measure the reproductive success. All experimental procedures on housing and use of laboratory animals were performed in accordance with guidelines of the Institute of Zoology, Chinese Academy of Sciences.

2.6. Monthly variation of litter size and precipitation

To understand variation of litter size and precipitation in Inner Mongolia, we analyzed historical data with detailed information on litter size in the breeding seasons of Brandt's vole. This data was obtained by a research team led by Dazhao Shi (Unpublished data) in Xinbahuzuo Qi (48°13'N, 118°16'E, 644 m in altitude), Inner Mongolia from 1983 to 1985. The mean monthly precipitation (M) and its standard deviation (SD) during 1980 to 2009 are shown in Table 1. The monthly precipitation can be classified into five categories by referring to SD (it is spaced by 20% SD). We define the normal precipitation range (equal to normal food, F^{00}) as follow: $M - 0.2 \times SD$ (low value) and $M + 0.2 \times SD$ (high value). Precipitation of above or below this range is taken as wet (equal to abundant food, F^{++}) or dry (equal to food shortage, F^{--}) conditions. Female voles were captured with kill-traps. The number of embryos of pregnant females was recorded and used for calculating mean litter size of pregnant female voles. The monthly variation of litter size and precipitation in the Xinbahuzuo Qi, Inner Mongolia from 1983 to 1985 are shown in Table 2. The food availability conditions are defined as F^{00} , F^{--} or F^{++} regimes based on the monthly precipitation by referring to the normal precipitation range in Table 1. Two consecutive months with F^{--} and F^{++} regimes can be taken as the treatment of FR–RF group to examine the response of litter size of voles to abundant precipitation after drought in field condition. The data were collected during the main breeding seasons of the Brandt's vole (from April to July).

2.7. Statistical analyses

Data were analyzed by using SPSS 13.0 statistical Package (SPSS Inc, Chicago, IL, USA) and R software (version 2.11.0). Differences of maternal body weight were analyzed by using *t*-test (when data distribution was normal), or Mann–Whitney *U* test (when the normality prerequisite was violated). Differences in pregnancy rate (% pregnant females), mean litter size, sex ratio (% males), body weight at birth and at weaning of F1 pups and survival rate (% survived pups to weaning) between FR–RF and control groups (including 1st, 2nd and 3rd experiments) were analyzed by using R software (version 2.11.0) with group as fixed factors, and time of experiment as covariate, using Generalized Linear Mixed Model. Differences between FR–RF and control group of the 4th experiment were analyzed by using SPSS 13.0. Results were presented as the mean \pm S.E.M, and $p < 0.05$ was considered to be statistically significant.

Table 1

Monthly precipitation (mm) in the breeding seasons of Brandt's vole from April to August in the Xinbahuzuo Qi, Inner Mongolia from 1980 to 2009.

Month	N	Minimum	Maximum	Mean	Std. Deviation	Normal range (Mean $\pm 0.2 \times SD$)	
						Low	High
April	30	0.8	28.4	11.0	7.0	9.6	12.4
May	30	0	55.1	17.7	13.5	15.0	20.4
June	30	3.8	137.7	44.2	28.3	38.6	49.9
July	30	27.8	209.9	76.4	39.2	68.6	84.2
August	30	21.2	187.6	66.2	41.9	57.8	74.6

Table 2

The litter size and precipitation in the Xinbahuzuo Qi, Inner Mongolia (Data provided by Dazhao Shi). F^{--} , F^{++} and F^{00} are defined based on below, above or within the normal precipitation range as shown in Table 1. For each year, two consecutive food abundance indices (as indicated by F^{--} , F^{++} and F^{00}) represent the food treatment on voles for two months which may affect the litter size of the next month. This is because changes of food abundance (as indicated by monthly precipitation) significantly affect changes of litter size of the next month (see Fig. 5).

Year	Month	N	Litter size	Std. Deviation	Precipitation (mm)	Food abundance
1983	4	34	7.00	1.23	9.2	F^{--}
	5	88	6.97	1.33	2.2	F^{--}
	6	16	6.06	1.18	55.3	F^{++}
	7	45	6.89	1.09	64.9	F^{++}
1984	4	36	6.64	1.40	9.2	F^{--}
	5	34	6.59	1.10	24.6	F^{++}
	6	21	6.90	1.51	48.1	F^{00}
	7	35	6.51	0.98	111.9	F^{++}
1985	4	30	6.70	1.24	14.4	F^{++}
	5	31	7.03	1.08	5.2	F^{--}
	6	24	6.58	1.25	75.8	F^{++}
	7	21	6.81	1.33	59.9	F^{++}

3. Results

3.1. FR–RF effect on maternal voles and F1 offspring

The initial body weight of maternal voles (day 0) showed no significant difference between FR–RF and control groups in the three experiments. During the food restriction period (the first 4 weeks), body weight showed significant decline in FR–RF group for all three experiments. During the re-feeding period (the last 4 weeks), body weight of FR–RF group increased rapidly; the differences in body weight between the two groups became non-significant by the end of re-feeding ($p > 0.05$; Fig. 1a–c).

There were no significant differences in pregnancy rate (% pregnant females) (FR–RF: 62.12%, $n = 66$; Control: 52.70%, $n = 74$; $z = 1.449$, $p = 0.147$); body weight at birth of F1 pups (FR–RF: 2.70 ± 0.04 g, $n = 40$; Control: 2.74 ± 0.04 g, $n = 38$; $z = -0.837$, $p = 0.405$); sex ratio (% males) of F1 pups (FR–RF: 44.83%, $n = 38$; Control: 40.00%, $n = 33$; $z = 1.117$, $p = 0.264$) between FR–RF and control group.

However, mean litter size (average number of pups per maternal females) in FR–RF group was higher than that of control group (FR–RF: 7.07 ± 0.48 , $n = 41$; Control: 5.64 ± 0.38 , $n = 39$; $z = 2.53$, $p = 0.011$, Fig. 2). Body weight at weaning of F1 pups (21 days after birth) of FR–RF group was significantly lower than that of control group (FR–RF: 14.82 ± 0.55 g, $n = 35$; Control: 17.48 ± 0.61 g, $n = 29$; $z = -2.59$, $p = 0.009$, Fig. 3). Survival rate of F1 pups at weaning of FR–RF group was significantly higher than that of control group (FR–RF: 80.69%, $n = 41$; Control: 66.82%, $n = 39$; $z = 3.051$, $p = 0.002$, Fig. 4).

3.2. Effect on serum testosterone and corticosterone of male voles

The concentrations of testosterone (FR–RF: 4.8581 ± 0.9110 ng/mL, $n = 21$; Control: 3.5076 ± 0.7678 ng/mL, $n = 19$; $z = -0.826$, $p = 0.409$), and corticosterone (FR–RF: 29.1415 ± 5.5052 nmol/L, $n = 21$; Control: 21.4386 ± 4.5138 nmol/L, $n = 19$; $z = -0.637$, $p = 0.524$) of male voles were non-significant between FR–RF and control groups.

3.3. Effect on F1 and F2 offspring: maternal effect

For data from the 4th experiment, we found that mean litter size of FR–RF group tended to be higher than that of control group (FR–RF: 7.82 ± 0.31 , $n = 22$; Control: 6.95 ± 0.43 , $n = 21$; $z = -1.654$, $p = 0.098$). However, we did not find significant differences between FR–RF and control groups in pregnancy rate of F1 females

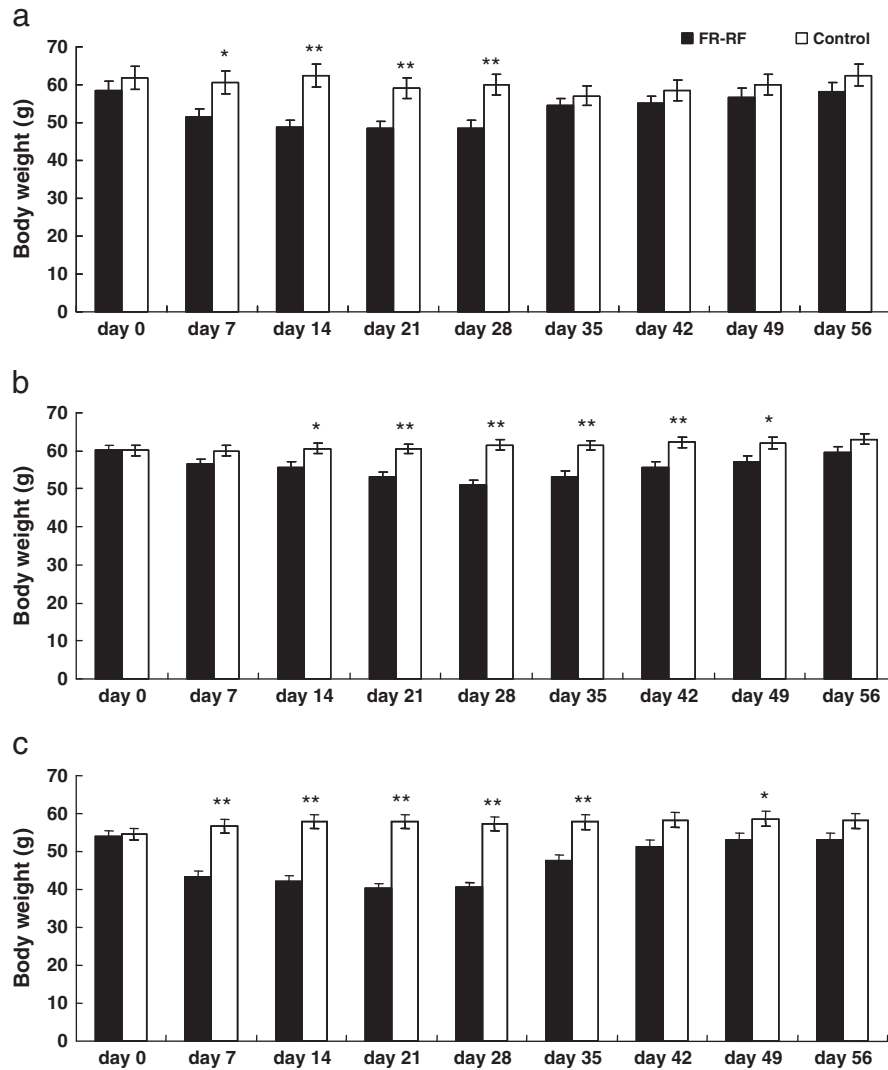


Fig. 1. Differences in body weight (mean \pm S.E.M, g) of maternal voles between FR–RF and control groups during food restriction and re-feeding period. FR–RF: food-restricted and refed group; Control: control group. The letters from a to c represent the results of the first, second and third experiments, respectively. *, $p < 0.05$; **, $p < 0.01$.

($\chi^2 = 0.432$, $p = 0.511$); sex ratio (% males, $\chi^2 = 0.101$, $p = 0.751$) and survival rate at weaning of F2 pups ($\chi^2 = 2.555$, $p = 0.110$).

The differences of body weight at birth of F2 pups (FR–RF: 2.62 ± 0.03 g, $n = 22$; Control: 2.67 ± 0.03 g, $n = 20$; $t = -1.256$, $p = 0.216$), and body weight at weaning of F2 pups (FR–RF: 15.62 ± 0.39 g,

$n = 22$; Control: 14.88 ± 0.64 g, $n = 20$; $t = 1.029$, $p = 0.310$) were all non-significant between the two groups. Body weight at birth of F2 pups (maternal body weight as covariate) was non-significant ($p = 0.396$) between the two groups, and body weight at weaning (body weight at birth as covariate) was also non-significant ($p = 0.059$).

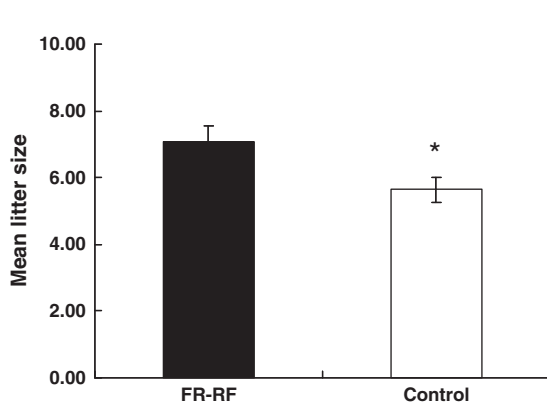


Fig. 2. Difference in mean litter size (mean \pm S.E.M) of pregnant females. FR–RF: food-restricted and refed group; Control: control group. *, $p < 0.05$.

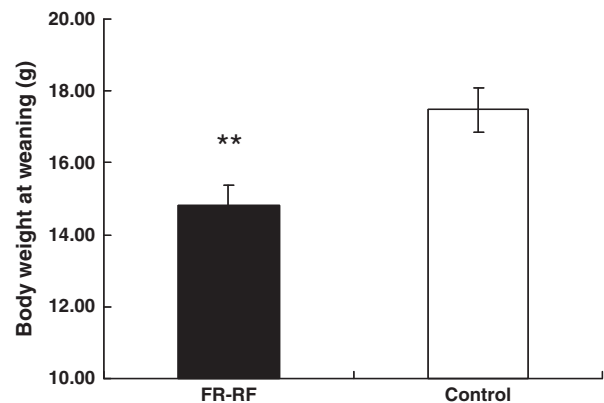


Fig. 3. Difference in body weight at weaning (21 days after birth, mean \pm S.E.M, g) of F1 pups: FR–RF compared to controls; **, $p < 0.01$.

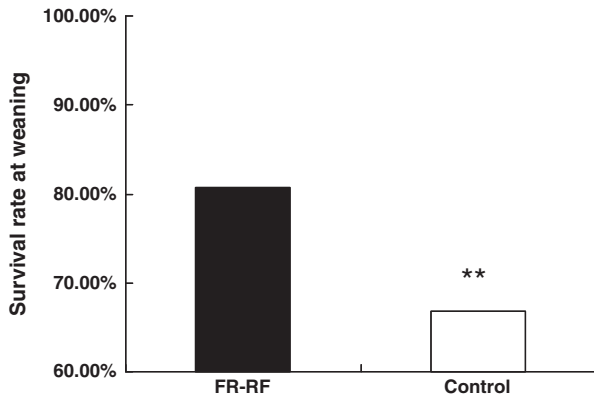


Fig. 4. Difference in survival rate at weaning (21 days old) of F1 pups between FR–RF and control groups. FR–RF: food-restricted and refed group; Control: control group. **, $p < 0.01$.

3.4. Response of litter size to precipitation in field condition

We found that monthly litter size (L_t) of voles corresponded very well with variation of precipitation (P_t) with one month time-lag (Fig. 5a); the change of litter size ($dL_t = L_{t+1} - L_t$) was positively correlated to the change of precipitation of previous month ($dP_{t-1} = P_t - P_{t-1}$)

(P_{t-1}) in the Xinbahuzuo Qi, Inner Mongolia ($r = 0.943$, $p = 0.005$; Fig. 5b). The correlation between change of litter size and the average precipitation of previous two months (representing the food level) was non-significant ($r = -0.086$, $p = 0.872$). These results suggest that in the breeding seasons, litter size is highly dependent on changes, not the levels of food abundance (as represented by precipitation). Voles experienced increase of monthly precipitation showed increase of litter size than no increase or decrease of monthly precipitation.

Table 2 shows effect of food condition of two consecutive months (reflected by combination of F^{00} , F^{--} or F^{++}) on mean litter size of next month in the Xinbahuzuo Qi. We found the following relationships: mean litter size in June = 6.90 in 1984 (experiencing $F^{--}F^{++}$ in April and May, equal to FR–RF treatment) > 6.58 in 1985 ($F^{++}F^{--}$) > 6.06 in 1983 ($F^{--}F^{--}$); mean litter size in July = 6.89 in 1983 (experiencing $F^{--}F^{++}$ in May and June, equal to FR–RF treatment) > 6.81 in 1985 ($F^{--}F^{++}$) > 6.51 in 1984 ($F^{++}F^{00}$). These results suggest that the litter size of voles tended to be higher when they meet abundant food after being experienced food shortage.

4. Discussion

Phenotypic plasticity has become an important topic in evolutionary ecology [40]. Through phenotypic plasticity, organisms are able to adapt to novel or changing environments [41,42]. If food supply

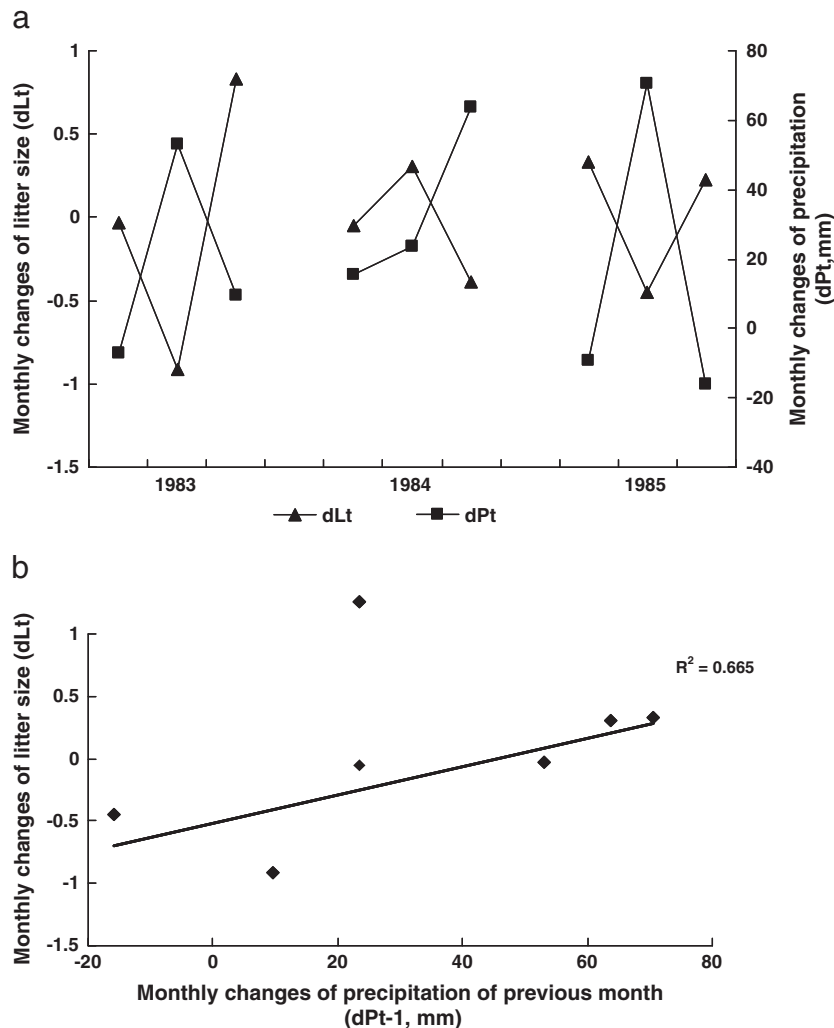


Fig. 5. Relationship between litter size (L_t) and precipitation (P_t) in the Xinbahuzuo Qi from 1983 to 1985. a. monthly changes of litter size (dL_t) and precipitation (dP_t). b. the relationship between changes in litter size (dL_t) and changes in precipitation (dP_{t-1}) of the previous month. $dL_t = L_{t+1} - L_t$, $dP_{t-1} = P_t - P_{t-1}$.

fluctuates greatly among seasons or years, animals may have evolved the phenotypic plasticity in reproduction and survival to match the fluctuations of food resources, and evolved the reproductive and survival strategies to adjust their reproductive output based on food availability [13,14].

Food restriction often results in reduced reproductive success of animals. From studies in experimental species, it decreases the rate of conception and milk yield, increases the likelihood of premature delivery and the birth of small offspring [27,43–45]. Camargo and Urbina reported that a 35% reduction in food given to *matrinxã* (*Brycon amazonicus*) during 6 months prior to spawning reduced the number of spawned females and the egg amount [46]. Food restriction may result in retarded fertile ability, lowered sperm count and motility, and increased abnormal sperms of male mice [47]. Spermatogenesis and testosterone are generally viewed as being resistant to reduced food intake in adult mammals. On the contrary, re-feeding can reverse the suppression of reproduction resulted from food restriction [31–33].

The trade-off between litter size and quality of offspring is central to many hypotheses and models of optimal litter size [48,49]. Quality of offspring potentially affects parental fitness through the probability of successful recruitment and reproduction of their offspring [50,51]. At present, there were no consistent results on the relationship between litter size and juvenile survival. In birds, studies have shown that juvenile survival and clutch size can be unrelated [52,53], negatively related [52], or positively related [54,55]. It was reported that increased litter size led to reduced survival of offspring on day 21 in *Peromyscus polionotus* [56]. König et al. reported that house mice (*Mus musculus*) had an 11% death rate at weaning when the litter size was 12, while no juvenile died when the litter size was 6 [57]. In the study of reproductive costs for free-ranging bank vole (*Clethrionomys glareolus*) in eight 0.25 ha enclosures by manipulating litter size, it was found that the number of offspring at weaning did not differ significantly between the treatment group (enlarged litters) and Control group (control litters) [58]. Similarly, in a study on wild muskrats (*Ondatra zibethicus*) in a 600 ha cattail (*Typha* sp.) marsh, it was found that survival of young at weaning in successful litters was not significantly correlated with litter size [59]. In our study, we found that food restriction and re-feeding increased the litter size of maternal voles, and also increased the survival of F1 offspring, showing a positive association between survival of juvenile and litter size. This overcompensatory property is likely a phenotypic plasticity used by Brandt's voles for adaptation to the fluctuation of food resources in natural conditions. In arid Inner Mongolia, food abundance changes greatly due to large variation of monthly precipitation (Table 2), the reproductive overcompensatory capacity may help population abundance of Brandt's voles to match fluctuation of food resources. The difference between our experiments and previous studies may be caused by experimental procedures or species-specific reasons. To achieve such overcompensation effect in the Brandt's vole, firstly, the breeding of rodents should vary greatly with variation of climate or food (Table 2); and secondly, the experimental duration of food restriction and re-feeding should match with field conditions.

Litter size is often negatively correlated to the quality of offspring. The body mass of offspring is often lower in enlarged litters or higher in reduced litters [58,60]. The birth weight of golden-mantled ground squirrels (*Spermophilus saturatus*) was not found to be related to litter size, but the average body weight of juveniles in the smallest litters was significantly greater than that in the largest litters by day 35 [61]. In this study, we found no significant difference in birth weight of F1 pups between FR–RF and control groups. However, body weight at weaning of FR–RF group was significantly lower than that of control group, showing a negative relation between the number of offspring and weaning body weight. This was likely caused by limitation of milk of maternal female voles to larger litters in FR–RF

groups. It is notable that although the body weight at weaning in FR–RF was smaller than that of control group, the survival rate of FR–RF group was higher. This was likely caused by the overcompensation mechanism on F1 offspring induced by maternal voles which experienced FR–RF treatment.

Food restriction can suppress the function of testis and the concentration of testosterone of male rodents [16,17]. However, this suppression could be rapidly reversed by re-feeding [62,63]. Chronic food restriction potentiates the diurnal elevation of plasma corticosterone in rats and mice [64–66]. The major response to stress such as fasting of Mongolian gerbils (*M. unguiculatus*) and food deprivation of water vole (*Arvicola terrestris*) is the increase of corticosterone [67,68]. In this study, we did not find significant differences of testosterone and corticosterone concentrations in male voles between FR–RF and control groups, suggesting that re-feeding can reverse the effect of food restriction on testis and hypothalamo-pituitary-adrenal axis (HPA) in Brandt's vole.

Food restriction experience may have a long-term effect on reproduction of mammals, or even have a cross-generation effect. For example, Bondrup-Nielsen and Foley reported that food restriction on female infants of meadow voles (*Microtus pennsylvanicus*) showed a higher reproductive success of their subsequent offspring, measured as pregnancy rate and litter size [33]. Huck et al. also reported that F2 generation from food-restricted female golden hamsters (*Mesocricetus auratus*) had a smaller litter size and a lower sex ratio from birth to day 25 [20]. Our data showed that F1 and F2 offsprings of Brandt's vole had a similar, but non-significant, increase in reproduction and/or survival. Admittedly, the fitness of an offspring can be affected by the food availability of a mother [69]. The effect of environment on mothers can be transmitted to offspring via cytoplasmic factors (e.g. hormones and mRNAs) directly (via maternal programming) or indirectly (via offspring sensitivity to maternally transmitted factors) [23]. In our study, such cross-generation effect was obvious in F1 offspring of voles, but not significant in F2 generation though it show similar trend. It is likely that the cross-generation effect of re-feeding after starvation also declines with increasing generation as previous found in other species (e.g. in the Greater long-tailed hamster [21]).

Sex ratio could be skewed in relation to a variety of environmental or parental conditions in mammals. Physiological condition of individual females during their periods of maternal investment might influence sex ratio of progeny [70]. Females in poor condition might be expected to produce an excess of female offspring if male juveniles are energetically more expensive to raise [71]. Female golden hamsters (*M. auratus*) food-restricted on days 1–50 postpartum, did produce their own female-biased litters [20]. But food-restricted female rat-like hamsters (*Cricetulus triton*) produced significantly more male than female F1 and F2 offspring [21]. In this study, we found that the sex ratio of F1 and F2 voles in FR–RF group was non-significant with that of control group. It was concluded that re-feeding could restore the physiological condition of parental voles after food restriction.

There may be a strong link between energy balance and reproductive success in physiological mechanisms [72]. Primary sensory stimuli may arise from mechanical distention of the lumen, gut contractions and chemical changes within the lumen of the gut [73]. The metabolic stimuli may be produced by changes in the oxidation of metabolic fuels due to changes of food intake, which then influences reproduction [74,75]. Reproductive processes and behaviors are sensitive to the levels of oxidizable metabolic fuels; deficits in such fuels create metabolic sensory stimuli that inhibit reproduction, and vice versa [72]. Previous studies suggest that metabolic fuels may act on the gonadotropin-releasing hormone (GnRH) system in a steroid-independent manner in small rodents [76,77]. In our study, treatment of food restriction may cause deficits in oxidizable metabolic fuels which inhibit reproduction performance of voles. Re-feeding

of food-restricted voles may create a surplus of the oxidizable metabolic fuels which thus cause overcompensation in reproduction of FR–RF females. We speculate that for animals whose oscillating populations are driven by fluctuating food resources, their reproduction systems may be more sensitive to changes of oxidizable metabolic fuels, not the level of these fuels. This may help to explain why re-feeding animals (showing rapid increase of fuel level) show higher reproductive success than that of control animals (showing constant high fuel level). It is notable that in field condition, we found changes of litter size of the voles were closely related to food changes (as represented by precipitation). This observation suggests that the reproduction of Brandt's voles is indeed sensitive to food changes, not only to food level. Thus the above speculation is worth to be testified.

In summary, our results indicate that treatment of food restriction and re-feeding can cause overcompensation in parental reproduction and offspring survival as compared to normal feeding voles ad libitum. This overcompensation capacity may be a significant adaptive strategy for Brandt's vole to cope with the fluctuating food resources in field conditions. But due to the limited observations of reproductive overcompensation in field, we need to look for more evidence in Brandt's vole or other species whose populations show strong oscillations. It is also necessary to reveal the reproductive differences between males and females, as well as the underlying molecular mechanism behind this overcompensatory phenomenon in Brandt's vole.

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References

- Boonstra R. Population cycles in microtines: the senescence hypothesis. *Evol Ecol* 1994;8:196–219.
- Xiong WH, Liang H, Yu XH, Cong L, Zhang ZB. Food limitation and low-density populations of sympatric hamster species in North China. *Contrib Zool* 2009;78(2):65–75.
- Brown JH, Ernest SKM. Rain and rodents: complex dynamics of desert consumers. *Bioscience* 2002;52:979–87.
- Eeneest SKM, Brown JH, Parmenter RR. Rodents, plants, and precipitation: spatial and temporal dynamics of consumers and resources. *Oikos* 2000;88:470–82.
- Huitu O, Jokinen I, Korpimäki E, Koskela E, Mappes T. Phase dependence in winter physiological condition of cyclic voles. *Oikos* 2007;116:565–77.
- Ostfeld RS, Keesing F. Pulsed resources and community dynamics of consumers in terrestrial ecosystems. *Trends Ecol Evol* 2000;15:232–7.
- Pech RP, Hood GM, Singleton GR, Salmon E, Forrester RI, Brown PR. Models for predicting plagues of house mice (*Mus domesticus*) in Australia. In: Singleton GR, Hinds LA, Leirs H, Zhang Z, editors. Ecologically-based management of rodent pests. Canberra, Australia: ACIAR; 1999. p. 81–112.
- Singleton GR. Population dynamics of an outbreak of house mice (*Mus domesticus*) in the mallee wheatlands of Australia—hypothesis of plague formation. *J Zool* 1989;219:495–515.
- Armesto JJ, Vidiella PE, Gutierrez JR. Plant communities of the fog-free coastal desert of Chile: plant strategies in a fluctuating environment. *Rev Chil Hist Nat* 1993;66:271–82.
- Fuentes ER, Campusano C. Pest outbreaks and rainfall in the semi-arid region of Chile. *J Arid Environ* 1985;8:67–72.
- Leirs H. Populations of African rodents: models and the real world. In: Singleton GR, Hinds LA, Leirs H, Zhang Z, editors. Ecologically-based management of rodent pests. Canberra, Australia: ACIAR; 1999. p. 388–408.
- Li ZL, Zhang WR. Analysis on the relation between population of *Meriones unguiculatus* and factors of meteorological phenomena. *Acta Theriol* 1993;13(2):131 [in Chinese].
- Shanley DP, Kirkwood TBL. Calorie restriction and aging: a life-history analysis. *Evolution* 2000;54(3):740–50.
- Huck UW, Labov JB, Lisk RD. Food-restriction first generation juvenile female hamsters (*Mesocricetus auratus*) affects sex ratio and growth of third generation offspring. *Biol Reprod* 1987;37:612–7.
- Even PC, Nicolaïdis S. Adaptive changes in energy expenditure during mild and severe feed restriction in the rat. *Br J Nutr* 1993;70:421–31.
- Blank JL, Desjardins C. Differential effects of food restriction on pituitary–testicular function in mice. *Am J Physiol* 1985;248(2):R181–9.
- de Souza Santos AM, Ferraz MR, Teixeira CV, Sampaio FJB, da Fonte Ramos C. Effects of undernutrition on serum and testicular testosterone levels and sexual function in adult rats. *Horm Metab Res* 2004;36(1):27–33.
- Han ES, Evans TR, Shu JH, Lee S, Nelson JF. Food restriction enhances endogenous and corticotropin-induced plasma elevations of free but not total corticosterone throughout life in rats. *J Gerontol Biol Sci* 2001;56(9):B391–7.
- Ferkin MH, Sorokin ES, Johnston RE, Lee CJ. Attractiveness of scents varies with protein content of the diet in meadow vole. *Anim Behav* 1997;53:133–41.
- Huck UW, Labov JB, Lisk RD. Food restricting young hamsters (*Mesocricetus auratus*) affects sex ratio and growth of subsequent offspring. *Biol Reprod* 1986;35:592–8.
- Liang H, Zhang ZB. Food restriction affects reproduction and survival of F1 and F2 offspring of rat-like hamster (*Cricetulus triton*). *Physiol Behav* 2006;87:607–13.
- Boonstra R, Hochachka WM. Maternal effects and additive genetic inheritance in the collared lemming *Dicrostonyx groenlandicus*. *Evol Ecol* 1997;11:169–82.
- Mousseau TA, Fox CW. The adaptive significance of maternal effects. *Trends Ecol Evol* 1998;13(10):403–7.
- Agrawal AA. Transgenerational consequences of plant responses to herbivory: an adaptive maternal effect? *Am Nat* 2001;157(5):555–69.
- Kofman O. The role of prenatal stress in the etiology of developmental behavioural disorders. *Neurosci Biobehav Rev* 2002;26:457–70.
- Groothuis TGG, Müller W, von Engelhardt N, Carere C, Eising CM. Maternal hormones as a tool to adjust offspring phenotype in avian species. *Neurosci Biobehav Rev* 2005;29:329–52.
- McGuire MK, Littleton AW, Schulze KJ, Rasmussen KM. Pre- and postweaning food restriction interact to determine reproductive success and milk volume in rats. *J Nutr* 1995;125:2400–6.
- Kirkwood TBL, Shanley DP. Food restriction, evolution and ageing. *Mech Ageing Dev* 2005;126:1011–6.
- Harrison DE, Archer JR. Natural selection for extended longevity from food restriction. *Growth Dev Aging* 1989;53(1–2):3.
- Phelan JP, Austad SN. Natural selection, dietary restriction, and extended longevity. *Growth Dev Aging* 1989;53:4–6.
- Grand TC, Millar JS. The effects of intermittent dietary restriction on weight gain and body fat in white-footed mice, *Peromyscus leucopus*. *Physiol Behav* 1990;48:221–4.
- Jennifer LT, Rissman EF. Brief refeeding restores reproductive readiness in food-restricted female Musk Shrews (*Suncus murinus*). *Horm Behav* 2000;38:21–8.
- Bondrup-Nielsen S, Foley PM. Long-term effects of malnutrition on reproduction: a laboratory study with meadow voles, *Microtus pennsylvanicus*, and red-backed voles, *Clethrionomys gapperi*. *Can J Zool* 1994;72(2):232–8.
- Zhang ZB, Wang ZW. Ecology and management of rodent pests in agriculture. Beijing: Ocean Press; 1998. p. 209. [in Chinese].
- Shi D, Wan XR, Davis SA, Pech RP, Zhang ZB. Simulation of lethal control and fertility control in a demographic model for Brandt's vole *Microtus brandti*. *J Appl Ecol* 2002;39:337–48.
- Zhong WQ, Wang MJ, Wan XR. Ecological management of Brandt's vole (*Microtus brandti*) in Inner Mongolia, China. In: Singleton GR, Hinds LA, Leirs H, Zhang Z, editors. Ecologically-based management of rodent pests. Canberra, Australia: ACIAR; 1999. p. 199–214.
- Wan XR, Wang MJ, Wang GH, Liu W, Zhong WQ. The reproductive parameters in the marked populations of Brandt's vole. *Acta Theriol* 2002;22:116–22 [in Chinese].
- Zhong WQ, Zhou QQ, Wang GH, Sun CL, Zhou PY, Liu WZ, et al. The design for the ecological management of Brandt's vole pest and its application. *Acta Theriol* 1991;11:204–12 [in Chinese].
- Zhang ZB, Pech R, Davis S, Shi DZ, Wan XR, Zhong WQ. Extrinsic and intrinsic factors determine the eruptive dynamics of Brandt's voles *Microtus brandti* in Inner Mongolia, China. *Oikos* 2003;100:299–310.
- Schlichting CD, Pigliucci M. Phenotypic evolution: a reaction norm perspective. Sunderland, MA: Sinauer Associates; 1998.
- Agrawal AA. Phenotypic plasticity in the interactions and evolution of species. *Science* 2001;294:321–6.
- Vezina F, Williams TD. Plasticity in body composition in breeding birds: what drives the metabolic costs of egg production? *Physiol Biochem Zool* 2003;76:716–30.
- Galler JR, Zartarian G. Reproductive performance in rats with different histories of malnutrition. *Br J Nutr* 1981;45:251–5.
- Young CM, Rasmussen KM. Effects of varying degrees of chronic dietary restriction in rat dams on reproductive and lactational performance and body composition in dams and their pups. *Am J Clin Nutr* 1985;41:979–87.
- Alexander MH, Lazan KS, Rasmussen KM. Effect of chronic protein-energy malnutrition on fecundability, fecundity and fertility in rats. *J Nutr* 1988;118:883–7.
- Camargo ACS, Urbinat EC. Influence of food restriction on the reproduction and larval performance of matrinxã, *Brycon amazonicus* (Spix and Agassiz, 1829). *Braz J Biol* 2008;68:869–73.
- Wu AG, Wan FD, Sun XF, Liu YG. Effects of dietary restriction on growth, neurobehavior, and reproduction in developing Kunming mice. *Toxicol Sci* 2002;70:238–44.

- [48] Lack D. The significance of clutch-size. *Ibis* 1947;89:302–52.
- [49] Sikes RS. Tradeoffs between quality of offspring and litter size: differences do not persist into adulthood. *J Mammal* 1998;79:1143–51.
- [50] Clutton-Brock TH, Harvey PH, Rudder B. Sexual dimorphism, socioeconomic sex ratio and body weight in primates. *Nature* 1977;269:797–800.
- [51] Myers P, Master LL. Reproduction by *Peromyscus maniculatus*: size and compromise. *J Mammal* 1983;64:1–18.
- [52] Klomp H. The determination of clutch-size in birds: a review. *Ardea* 1970;58:1–124.
- [53] Nur N. The consequences of brood size for breeding blue tits. II. Nestling weight, offspring survival and optimal brood size. *J Anim Ecol* 1984;53:497–517.
- [54] Högstedt G. Evolution of clutch size in birds: adaptive variation in relation to territory quality. *Science* 1980;210:1148–50.
- [55] Perrins CM, Moss D. Reproductive rates in the great tit. *J Anim Ecol* 1975;44:695–706.
- [56] Kaufman DW, Kaufman GA. Reproduction by *Peromyscus polionotus*: number, size, and survival of offspring. *J Mammal* 1987;68(2):275–80.
- [57] König B, Riester J, Markl H. Maternal care in house mice (*Mus musculus*). II. The energy cost of lactation as a function of litter size. *J Zool* 1988;216:195–210.
- [58] Mappes T, Koskela E, Ylönen H. Reproductive costs and litter size in the bank vole. *Proc R Soc Lond B* 1995;261:19–24.
- [59] Boutin S, Moses RA, Caley MJ. The relationship between juvenile survival and litter size in wild muskrats (*Ondatra zibethicus*). *J Anim Ecol* 1988;57:455–62.
- [60] Sikes RS. Unit pricing: economics and the evolution of litter size. *Evol Ecol* 1998;12:179–90.
- [61] Kenagy GJ, Stevenson RD, Masman D. Energy requirements for lactation and postnatal growth in captive golden-mantled ground squirrels. *Physiol Zool* 1989;62(2):470–87.
- [62] Cameron JL, Nosbisch C. Suppression of pulsatile luteinizing hormone and testosterone secretion during short term food restriction in the adult male rhesus monkey (*Macaca mulatta*). *Endocrinology* 1991;128(3):1532–40.
- [63] Young KA, Zirkin BR, Nelson RJ. Testicular regression in response to food restriction and short photoperiod in white-footed mice (*Peromyscus leucopus*) is mediated by apoptosis. *Biol Reprod* 2000;62:347–54.
- [64] Stewart J, Meaney MJ, Aitken D, Jensen L, Kalant N. The effects of acute and life-long food restriction on basal and stress-induced serum corticosterone levels in young and aged rats. *Endocrinology* 1988;123(4):1934–41.
- [65] Sabatino F, Masoro EJ, McMahan CA, Kuhn RW. Assessment of the role of the glucocorticoid system in aging processes and in the action of food restriction. *J Gerontol Biol Sci* 1991;46:B171–9.
- [66] Klebanov S, Diais S, Stavinoha WB, Suh Y, Nelson JF. Hyperadrenocorticism, attenuated inflammation, and the life-prolonging action of food restriction in mice. *J Gerontol Biol Sci* 1995;50A(2):B78–82.
- [67] Xu DL, Wang DH. Fasting suppresses T cell-mediated immunity in female Mongolian gerbils (*Meriones unguiculatus*). *Comp Biochem Physiol A Mol Integr Physiol* 2010;155:25–33.
- [68] Bazhan NM, Makarova EN, Yakovleva TV. Deprivation of food during pregnancy and reproduction in the water vole (*Arvicola terrestris*). *J Mammal* 1996;77(4):1078–84.
- [69] Liang H, Zhang JJ, Zhang ZB. Food restriction in pregnant rat-like hamsters (*Cricetulus triton*) affects endocrine, immune function and odor attractiveness of male offspring. *Physiol Behav* 2004;82:453–8.
- [70] Trivers RL, Willard DE. Natural selection of parental ability to vary the sex ratio of offspring. *Science* 1973;179:90–2.
- [71] Labov JB, Huck UW, Vaswani P, Lisk RD. Sex ratio manipulation and decreased growth of male offspring of undernourished golden hamsters (*Mesocricetus auratus*). *Behav Ecol Sociobiol* 1986;18:241–9.
- [72] Schneider JE. Energy balance and reproduction. *Physiol Behav* 2004;81:289–317.
- [73] Schwartz GJ. The role of gastrointestinal vagal afferents in the control of food intake: current prospects. *Nutrition* 2000;16(10):866–73.
- [74] Friedman MI. An energy sensor for control of energy intake. *Proc Nutr Soc* 1997;56:41–50.
- [75] Friedman MI. Fuel partitioning and food intake. *Am J Clin Nutr* 1998;67:513S–8S.
- [76] Bronson FH. Food-restricted, pre-pubertal, female rats: rapid recovery of luteinizing hormone pulsing with excess food, full recovery of pubertal development with gonadotropin-releasing hormone. *Endocrinology* 1986;118:2483–7.
- [77] Bronson FH. Effect of food manipulation on the GnRH-LH-estradiol axis of young female rats. *Am J Physiol* 1988;254:R616–21.