

# Dietary protein constraint on age at maturity: an experimental test with wild deer mice

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## Summary

1. Five populations of *Peromyscus maniculatus borealis* (Mearns) were live-trapped in the Kananaskis Valley, Alberta, throughout the breeding seasons of 1996 and 1997, to test the hypothesis that growth and maturation of young-of-the-year (YY) females are limited by dietary protein.

2. In 1996, two populations received a high protein (30%) food supplement, one received a high energy, but low protein (14%) food supplement and two others were unsupplemented. In 1997, three populations received the protein (30%) supplement and two served as controls.

3. In 1996, control populations had high nestling growth rates and many (43%) YY females bred. In 1997, nestling growth rates were lower in control populations and no YY females bred.

4. Supplementation of high protein food resulted in consistently high nestling growth rates in both 1996 and 1997, and a significantly higher proportion of YY females bred than controls in both years.

5. Supplementation of high energy, but low protein food had no effect on YY growth or maturation.

6. Neither protein nor energy supplementation had any effects on the number of litters conceived per season, litter success, litter size or sex ratio of litters born to over-wintered females.

7. We conclude that growth and maturation of YY female *Peromyscus maniculatus* in this area are limited by the availability of dietary protein.

*Key-words:* age at first reproduction, food quality, food supplementation, maturation, *Peromyscus maniculatus borealis*.

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## Introduction

Since Lack (1954) first suggested that most vertebrate populations are limited by food resources, many studies have examined the degree to which food quantity affects life-history parameters (Boutin 1990). Several life-history traits of mammals, such as age at first reproduction, length of the breeding season, breeding intensity and litter size have been shown to be limited by food availability (e.g. Craig & Bunn 1989; Boutin 1990; Sullivan 1991; Dobson 1995; Duquette & Millar 1995; Risch, Dobson & Murie 1995; Hubbs & Boonstra 1997; but see Becker, Boutin & Larsen 1998). It has been suggested that many populations may be

limited by dietary protein rather than energy (White 1993), although few studies (Cole & Batzli 1978; Dobson & Kjelgaard 1985; Bomford & Redhead 1987; Bomford 1987b) have examined the effects of food quality on life-history parameters in wild populations.

Female *Peromyscus maniculatus* (deer mice) in the Kananaskis Valley, Alberta, rarely breed during the summer of their birth, despite having a breeding season that is usually long enough to permit them to do so. Limited data indicate that the few females who breed as young-of-the-year (YY) do not incur reproductive costs of increased over-winter mortality or decreased future reproductive success (Teferi & Millar 1993). As a result, it appears that these populations may be subject to some social or ecological constraint preventing maturation as YY.

Several experimental studies have been conducted over the past 10 years to determine why so few YY

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females breed, but there has been no clear explanation (Lusk & Millar 1989; Teferi & Millar 1993; MacDonald 1997; Woolfenden & Millar 1997). Breeding YY females have consistently exhibited high growth rates as nestlings, suggesting that individual quality may be important in determining which females mature as YY (Lusk & Millar 1989; Teferi & Millar 1993; A. G. McAdam & J. S. Millar, unpublished). Quality of YY in these populations appears to vary both annually and among litters. There are 'poor' years in which nestling growth is low and YY breeding does not occur, and 'good' years in which nestling growth is high and YY breeding is more frequent (A. G. McAdam & J. S. Millar, unpublished). Within 'good' years, some adult females are able to raise litters with high growth rates, while others are not, but nestling growth does not appear to vary among offspring within breeding litters (A. G. McAdam & J. S. Millar, unpublished). Supplementation of food (sunflower seeds, oats and rodent chow) resulted in modest, but consistent increases in the number of breeding YY, but failed to increase nestling growth rates and many YY still did not breed (Teferi & Millar 1993). The limited response to this supplement may have occurred because it failed to mimic the natural diet of mice in the area (Woolfenden & Millar 1997).

*Peromyscus maniculatus* are omnivores, consuming vegetation, seeds, berries, fungi and animal matter in the form of arthropods (Jameson 1952). The high protein contents of some of these foods suggest that protein may influence diet choice (Vickery *et al.* 1994). Deer mice show a high preference for many insects (Bellocq & Smith 1994) and incorporate large amounts of insects into their diet during spring (Jameson 1952; Martell & Macaulay 1981), when protein requirements for reproduction and early growth are high (Robbins 1983). Insects not only provide high absolute levels of dietary protein, but animal protein may also be more easily digested than plant proteins which are often associated with digestion inhibitors which can reduce the amount of assimilable protein (Robbins 1983). The abundance of flying insects is low in the Kananaskis Valley (Barclay 1990), suggesting that the abundance of insects available to non-volant small mammals may also be limited. As a result, breeding by mice may be constrained by the availability of animal protein.

Deficiencies in dietary protein have been shown to slow growth and delay maturation in laboratory rodents (Glass, Harrison & Swerdloff 1976; Pau & Milner 1984; Bronson 1985), and many species of small mammals alter their foraging habitat or type of forage to increase levels of dietary protein (see Cameron & Eshelman 1996; Bomford 1987a). In addition, natural increases in dietary protein have been associated with increased breeding intensity (Bomford 1987a) and increased litter sizes (Hoffmann 1958). Supplementation of the diet of small mammals with high protein food has been found to alter habitat selection

(Berger 1991; Eshelman & Cameron 1996), increase breeding intensity (Cole & Batzli 1978; Bomford & Redhead 1987), increase litter size (Cole & Batzli 1978; Dobson & Kjelgaard 1985) and decrease the age at first reproduction (Dobson & Kjelgaard 1985).

The objective of this study was to test the hypothesis that nestling growth and breeding by female YY deer mice in the Kananaskis Valley are constrained by a lack of dietary protein. A series of food supplementation experiments was conducted during two successive breeding seasons, to examine the effects of dietary protein supplementation, while controlling for the addition of energy. We predicted that if nestling growth and breeding by YY females are limited by dietary protein, nestling mice would have higher growth rates, and a higher proportion of them would breed on protein supplemented areas than on energy-supplemented and control areas. However, if YY growth and breeding is limited by energy, growth rates and levels of breeding would be similar in mice on both protein and energy-supplemented areas, but higher than controls. If neither energy nor protein is limiting, mice on control areas would have similar nestling growth rates and rates of maturation, as those on energy and protein supplemented areas.

## Methods

Five populations of *P. maniculatus borealis* were monitored through live-trapping in the Kananaskis Valley, Alberta (51°N; 115°W), during the breeding seasons of 1996 and 1997. Trapping grids ranging in size from 1.2 to 2.4 ha were established in rocky creek beds and open aspen habitat which represent prime deer mouse habitat (Millar, Innes & Loewen 1985). Each grid consisted of an array of trapping stations  $\approx$  20 m apart. One Longworth live-trap, baited with sunflower seeds and oats, and supplied with cotton bedding, was set at each trapping station. Each grid was trapped two nights per week from May to September, in both years. Traps were locked open between trapping nights.

Individual mice were ear tagged at first capture, and sex, weight, reproductive status and location were recorded at the initial and at each subsequent capture. Age was determined by pelage characteristics (grey = young-of-the-year, brown = over-wintered), weight was measured to the nearest 0.5 g using a Pesola spring balance, and reproductive status was assessed as testes scrotal (large and descended) or non-scrotal for males, and as non-perforate, perforate, pregnant (swollen abdomen), lactating (large and prominent nipples), or pregnant and lactating for females. All over-wintered females were coated with fluorescent powder at each capture and powder trails were followed to identify nest sites (Lemen & Freeman 1985).

Parturition was evident from changes in female weight and reproductive condition. Parturition dates were estimated as the median date between a female's

last capture as pregnant and first subsequent capture as lactating. Additional traps were set near a female's nest and at the trapping station where she was most frequently captured, beginning 21 days after parturition, in order to capture her offspring upon first emergence from the natal nest. Emerged offspring were assigned to dams based on the date and location of first capture, and by the presence of fluorescent powder in their pelage (Millar, Derrickson & Sharpe 1992). Any juveniles that could not be assigned to a resident dam were considered to be immigrants.

Animals were designated as residents if they were present on a grid for at least 14 days (Lusk & Millar 1989) and were captured at least three times in a 2-week period. Trapability was estimated for each trapping grid as the number of captures (minus the first and last, and summed for all individuals), divided by the number of potential captures (minus the first and last, and summed for all individuals; Lusk & Millar 1989).

Nestling growth indices were estimated for each mouse born on the trapping grids, as the weight at first capture (g) divided by the estimated age at first capture (days; Teferi & Millar 1993). Six of 348 juveniles had very large ( $> 0.7 \text{ g day}^{-1}$ ) or very small ( $< 0.2 \text{ g day}^{-1}$ ) growth indices. These mice were considered to have been improperly matched with dams and were therefore reclassified as immigrants. Young-of-the-year females were assumed to have bred if they were identified as pregnant or lactating. Young-of-the-year females who could have bred but did not, were defined as those females who emerged prior to the last conception of the season and remained on the trapping grid until the final parturition of the season (A. G. McAdam & J. S. Millar, unpublished).

Two of the five trapping grids were supplemented with a high protein cat food (Co-op Cat food #320C, Interprovincial Cooperative Ltd, Saskatoon, Saskatchewan;  $3.7 \text{ kcal g}^{-1}$ , 30% crude protein) in both 1996 and 1997. Two grids were part of a long-term study examining deer mouse population dynamics and remained unsupplemented. The fifth grid was supplemented with a 3:1 mixture by volume of whole oats and sunflower seeds in 1996, and high protein cat food in 1997. The seed mixture supplemented in 1996 was a high energy, but low protein supplement ( $\approx 4 \text{ kcal g}^{-1}$ ; 14% protein; Scherz & Senser 1994). Supplementation treatments could not be assigned to grids randomly because of the requirements of the concurrent long-term study.

Food was supplemented in large (1.5- or 3-L) metal cans covered by a sheet-metal lid with a small hole, large enough for a deer mouse to enter, but small enough to exclude larger rodents. Feeding stations, consisting of a single can, were spaced regularly throughout the supplemented trapping grids at a density of  $\approx 20$  stations  $\text{ha}^{-1}$ . Supplementation of food began on 25 May 1996, and 26 April 1997, and continued until trapping ceased at the end of the breeding

season. No supplementation occurred over winter. Throughout the period of supplementation, each feeding station was replenished with  $\approx 150 \text{ g}$  of food at least once per week, but some were regularly depleted. In several cases, bears (*Ursus americanus*, *Ursus horribilis*) repeatedly disturbed feeding stations. In these cases, feeding stations were abandoned and  $\approx 3 \text{ kg}$  of food was broadcast twice-weekly throughout the disturbed grid. A total of 110 and 190 kg of cat food were supplemented on the protein supplemented grids in 1996 and 1997, respectively. The amount of sunflower seeds and oats added to the energy-supplemented grid in 1996 was estimated at 45 kg.

Because it was not possible to supplement individuals in this study, individual trapping grids were used as the unit of replication. In some cases, however, data are presented as means of individuals or litters. Comparisons of nestling growth rates were made using averages for each grid by year combination to avoid potential problems associated with pseudo-replication (Hurlbert 1984).

Simple transformations did not remove the heterogeneity of variance in the number of litters conceived by over-wintered (OW) females during the breeding season, so a non-parametric test (Kruskal-Wallis) was used in this comparison (Zar 1996). The effects of year and food supplementation on the proportion of YY females breeding were analysed using a three-dimensional contingency table (Zar 1996). Statistical tests were performed with the STATISTICA software package (StatSoft Inc. 1994), using two-tailed probabilities and a Type I error rate of 0.05. Values are presented as means  $\pm 1 \text{ SE}$ .

## Results

There were no differences among trapping grids within a given treatment for any of the variables examined (proportion of YY breeding, number of litters conceived, litter success, litter size, sex ratio, summer survival and over-winter survival), so data were pooled for each treatment. Trapability was consistently high among all treatments. Approximately 75% of mice in each population were captured on any given trap night (Table 1). The presence of faeces containing fluorescent powder and the depletion of supplemented food within the feeding stations indicated that resident female deer mice were consuming the supplemented food.

Spring population densities appeared to be higher in 1997 ( $12.1 \text{ individuals ha}^{-1}$ ) than in 1996 ( $7.5 \text{ individuals ha}^{-1}$ ; Table 1). Breeding started later in 1996 than 1997, but also ended later, resulting in similar lengths of the breeding season in both years. Protein supplemented and control populations had similar spring densities in both years. Breeding began earlier on protein supplemented grids than control grids in 1996, but not in 1997. In both years, however, first conceptions occurred prior to the start of sup-

**Table 1.** Spring density, trapability, dates of first and last parturitions, and length of breeding seasons for *Peromyscus maniculatus* populations supplemented with high energy food, high protein food and unsupplemented populations in the Kananaskis Valley, AB

Year	Treatment	(n) <sup>a</sup>	Spring density (individuals ha <sup>-1</sup> ) <sup>b</sup>	Trapability <sup>c</sup>	Date of first parturition	Date of last parturition	Length of breeding season (days)
1996	Control	2	6.57	0.69	163.5	229.0	65.5
	Energy	1	15.79	0.82	— <sup>d</sup>	191	— <sup>d</sup>
	Protein	2	8.50	0.80	147.5	228.0	80.5
1997	Control	2	13.46	0.74	132.5	215.5	83.0
	Protein	3	10.69	0.72	138.0	216.7	78.7

Note: Data are presented as means of all populations within each treatment.

<sup>a</sup>Number of monitored populations.

<sup>b</sup>Number of over-wintered individuals known to be present in population during the first 2 weeks of trapping.

<sup>c</sup>Number of captures (minus first and last) summed for all individuals in the population divided by the number of potential captures (minus first and last) summed for all individuals in the population.

<sup>d</sup>Young-of-the-year were present at the start of trapping in the energy-supplemented population.

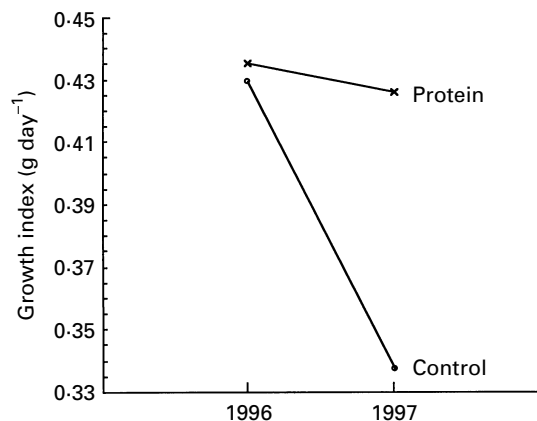
plementation. The spring density on the energy-supplemented grid was higher than the other grids in 1996 and breeding appeared to end very early.

There was a significant interaction between the growth indices of nestlings born on protein supplemented and control areas, and year ( $F_{1,5} = 10.522$ ,  $P = 0.023$ ; Fig. 1). On control grids, nestling growth was very high in 1996 ( $0.431 \pm 0.010$  g day<sup>-1</sup>), but low in 1997 ( $0.338 \pm 0.021$  g day<sup>-1</sup>), while growth remained consistently high on protein supplemented areas in both 1996 ( $0.433 \pm 0.014$  g day<sup>-1</sup>) and 1997 ( $0.425 \pm 0.009$  g day<sup>-1</sup>). Nestling growth on the energy-supplemented grid ( $0.39$  g day<sup>-1</sup>) was lower than on control and protein supplemented grids in 1996.

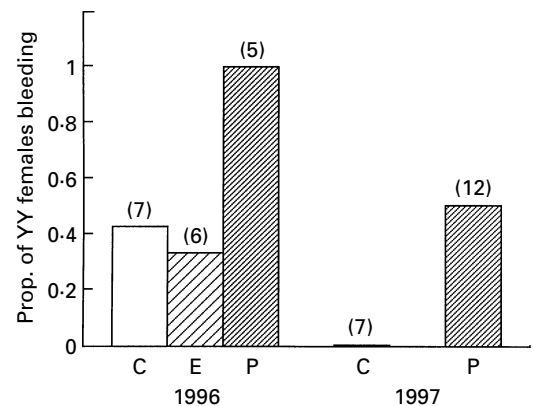
A total of 113 YY females were recruited into the monitored populations over both years. Sixty-five of these 113 (58%) were born early enough in the season

to have bred, of which 37 (33%) remained in the trapable population long enough to have been detected as having bred. The frequency of breeding among these 37 YY females was dependent on both the year and the supplement received ( $\chi^2_3 = 11.345$ ,  $P < 0.01$ ; Fig. 2). Breeding by YY females was more common in 1996 than 1997 ( $\chi^2_3 = 8.776$ ,  $P < 0.05$ ), and more common on protein supplemented grids than on control grids ( $\chi^2_3 = 10.384$ ,  $P < 0.025$ ). The frequency of breeding by YY females on the energy-supplemented grid did not differ from that of the control grids in 1996 (Fisher's exact test,  $n = 13$ ,  $P = 0.59$ ).

There was no effect of supplementation on the number of litters conceived by OW females during the



**Fig. 1.** Significant two-way interaction ( $F_{1,5} = 9.12$ ,  $P = 0.029$ ) of nestling growth indices (g day<sup>-1</sup>) for young-of-the-year *Peromyscus maniculatus* in protein supplemented (x) and control populations (o) during the breeding seasons of 1996 and 1997.



**Fig. 2.** Proportion of young-of-the-year (YY) female *Peromyscus maniculatus* breeding in protein (P) and energy (E) supplemented populations, and in control (C) populations during the breeding seasons of 1996 and 1997. A higher proportion of YY females bred in protein supplemented populations than controls ( $\chi^2_3 = 10.384$ ,  $P < 0.025$ ) and a higher proportion bred in 1996 than 1997 ( $\chi^2_3 = 8.776$ ,  $P < 0.05$ ). The number of potential breeders are shown in parentheses.

**Table 2.** Number of litters, litter success, litter size and sex ratio of offspring for resident, over-wintered (OW) *Peromyscus maniculatus* in populations supplemented with high energy food, high protein food and unsupplemented populations in the Kananaskis Valley, AB

Treatment	Number of litters <sup>a</sup>	Litter success <sup>b</sup>	Litter size <sup>c</sup>	Sex ratio <sup>d</sup>
Control	1.68 ± 0.16 (37 <sup>e</sup> )	0.55 (62 <sup>f</sup> )	3.50 ± 0.31 (34 <sup>f</sup> )	0.41 (34 <sup>f</sup> )
Energy	1.23 ± 0.12 (13)	0.44 (16)	4.14 ± 0.63 (7)	0.41 (7)
Protein	1.63 ± 0.13 (48)	0.65 (76)	3.80 ± 0.24 (49)	0.48 (49)
All	1.59 ± 0.09 (98)	0.58 (154)	3.70 ± 0.18 (90)	0.45 (90)

<sup>a</sup>Number of litters attempted per females (Kruskal–Wallis:  $H_2 = 2.37$ ,  $P = 0.31$ ).

<sup>b</sup>Number of litters in which at least one offspring was weaned divided by the total number of attempted litters (Chi-square test:  $\chi^2_2 = 2.89$ ,  $P > 0.1$ ).

<sup>c</sup>Number of weaned offspring per successful litter (ANOVA:  $F_{2,87} = 0.616$ ,  $P = 0.54$ ).

<sup>d</sup>Proportion of females in litters (Chi-square test:  $\chi^2_2 = 1.47$ ,  $P > 0.25$ ).

Data are presented as means ± 1 SE. Sample sizes are in parentheses.

<sup>e</sup>Number of females.

<sup>f</sup>Number of litters.

breeding season (Kruskal–Wallis,  $H_2 = 2.37$ ,  $P = 0.31$ ), the proportion of those litters, which were successful to weaning ( $\chi^2_2 = 2.89$ ,  $P > 0.10$ ), the number of offspring weaned per successful litter ( $F_{2,87} = 0.616$ ,  $P = 0.54$ ), nor their sex ratio ( $\chi^2_2 = 1.47$ ,  $P > 0.25$ ; Table 2). On average, OW females had 1.6 litters per season, of which 58% were successful. Each successful female weaned an average 3.7 offspring per litter, 45% of which were female.

Supplementation of food also did not have a significant effect on the survival of either YY ( $\chi^2_2 = 1.30$ ,  $P > 0.50$ ) or OW mice ( $\chi^2_2 = 5.05$ ,  $P > 0.05$ ) during the breeding season, although female mice supplemented with protein tended to survive better than control mice (Table 3). Very few (1/13) OW females on the energy-supplemented grid survived the summer. Survival of YY females over winter differed among treatments. *Post hoc* pairwise comparisons suggested that survival of YY over-winter was lower on the energy-supplemented grid than controls (Fisher's exact test, energy  $n = 28$ ,  $P = 0.022$ ; protein  $n = 28$ ,

$P = 0.103$ ). Overall, 70.8% of all weaned females survived to the end of their first summer, but only 18.9% survived to the following breeding season. Only three mice survived their second winter to breed as 2-year-olds, one of which survived to the end of her third summer.

All females who bred as YY (13) conceived only one litter during their first breeding season. Almost half (6/13) were successful at raising offspring to weaning. Successful litters by YY breeders had  $2.67 \pm 0.67$  offspring, of which 44% were female. These characteristics of YY reproduction differed from reproduction by OW females only in the number of litters conceived during the breeding season. Only two of nine females who bred as YY survived until the following spring, but this was not different from the over-winter survival of non-breeding YY females (Fisher's exact test,  $n = 45$ ,  $P = 0.73$ ). These two females conceived a total of seven litters as OW mice of which four were successful. These four successful litters had an average of  $3.5 \pm 0.5$  offspring.

**Table 3.** Survival to the end of the summer, over-winter and reproduction the following spring, of young-of-the-year (YY) and over-wintered (OW) female *Peromyscus maniculatus* in populations supplemented with high energy food, high protein food and unsupplemented(control) populations in the Kananaskis Valley, AB

Treatment	YY summer	YY winter	OW spring	OW summer	OW winter
Control	0.64 <sup>a</sup> (33)	0.55 (11)*	0.64 (39)	0.37 (38)	0.50 (6)
Energy	0.71 (24)	0.12 (17)*	— <sup>b</sup>	0.08 (13)	1.00 (1)
Protein	0.75 (56)	0.24 (17)*	0.78 (45)	0.41 (49)	0.17 (6)
All	0.71 (113)	0.27 (45)	0.71 (84)	0.35 (100)	0.39 (13)

Note: sample sizes are in parentheses.

\*Survival of YY over winter was different among treatments ( $\chi^2_2 = 6.389$ ,  $P < 0.05$ ).

<sup>a</sup>Survival values are presented as the proportion of females alive at the current life stage divided by the number of females alive at the previous life stage.

<sup>b</sup>Eight of 14 OW females were lactating at first capture.

## Discussion

Supplemented populations appeared to be similar to the control areas in density, trapability, and initiation and cessation of breeding. Breeding began in supplemented populations prior to the addition of food, so we were unable to examine the possible effects of food addition on the initiation of breeding, but the end of breeding did not appear to be extended in supplemented populations. Early initiation and late cessation of breeding have been reported in *P. maniculatus* in response to food addition in southern British Columbia (Fordham 1971; Taitt 1981), but not in another northern population of *P. maniculatus* (Gilbert & Krebs 1981).

The two breeding seasons examined in this study had many characteristics of 'good' and 'poor' years for YY breeding. The breeding season ended in control populations 2 weeks later in 1996 than 1997. In 1996, nestling growth rates were higher than any of the past 13 years studied in this area (A. G. McAdam & J. S. Millar, unpublished). Both a later cessation of breeding and high nestling growth rates have been previously associated with an increased frequency of breeding by YY females in these populations (Lusk & Millar 1989; Teferi & Millar 1993; A. G. McAdam & J. S. Millar, unpublished). In 1996, YY breeding in control populations was more frequent than in all years since 1985 (A. G. McAdam & J. S. Millar, unpublished). In 1997, however, nestling growth rates in control populations decreased to levels typical of poor years (A. G. McAdam & J. S. Millar, unpublished) and no YY breeding occurred, despite the breeding season being longer than 1996.

Supplementation of high protein food resulted in high nestling growth rates in both 1996 and 1997, and increased the frequency of YY breeding compared to control populations. Such decreases in age at first reproduction have been noted in several species of small mammals with supplemented food (see Boutin 1990), including Columbian ground squirrels supplemented with high protein food (Dobson & Murie 1987).

We did not expect that the protein supplementation would enable all females to breed in the summer of their birth. Many females in these populations are born too late in the season to breed as YY (A. G. McAdam & J. S. Millar, unpublished). In addition, there was likely variation among the remaining females in the quality of food received from their mothers due to natural variation in the foraging and food processing ability of dams, and variation in the competitiveness of littermates for milk. As a result, some early-born females may still not mature as YY, even though their mothers have access to abundant high quality food. In a laboratory experiment, only 52% of deer mice raised on the same high protein diet (cat food) were found to be mature at 42 days of age (McAdam 1998).

Supplementation with high energy, but low protein food did not affect either growth rates of nestlings or the frequency of breeding by YY relative to controls in 1996. In fact, nestling growth rates on the energy-supplemented grid were lower than on any other grid in that year. Feeding stations were regularly replenished and so it is unlikely that high population densities in the energy-supplemented population affected the amount of resources available to each mouse. The availability of naturally occurring food on the energy-supplemented area was not known relative to controls. Breeding by OW females also ended very early in the energy-supplemented population, but this was primarily due to their very poor summer survival. Because of poor survival, the length of time during which environmental conditions were suitable for breeding was likely under-estimated in that area. Although the energy supplementation was not repeated in 1997, the results from 1996 are similar to those of Teferi & Millar (1993) who found no increase in nestling growth and only modest increases in YY breeding in populations supplemented with sunflower seeds, oats and rodent chow over three seasons.

The growth and breeding responses of mice in this study suggest that YY females in these populations are subject to dietary constraints on both growth as nestlings and sexual maturation. In a good year when nestling growth was high and YY breeding was frequent, supplementation with high protein food had only a limited effect. However, in a poor year when nestling growth was low, protein supplementation dramatically increased nestling growth and facilitated YY breeding. The results of the energy supplementation in 1996 also suggest that protein, rather than energy, was the dietary component limiting growth and maturation. It appears that there was sufficient high protein food naturally available in 1996, but not in 1997, to support high rates of nestling growth and female maturation.

While we attempted to test the growth and breeding responses of YY females to dietary protein, some minerals also differed between the two supplements (sodium, calcium, iodine and selenium). Previous supplementation with sodium suggests that mice in these populations obtain sufficient salt in their natural diets (Woolfenden & Millar 1997). Very low levels of calcium have been found to slow growth in laboratory *Mus musculus* (Ornoy, Wolinsky & Guggenheim 1974; Wolinsky & Guggenheim 1974), but the effects of calcium availability has not been examined in our field populations of *P. maniculatus*.

None of the reproductive characteristics of OW mice were influenced by either energy or protein supplementation in this study. The number of litters conceived in each season, litter success, litter size and sex ratio were similar in all treatment populations, and similar to values previously reported for these populations of *P. maniculatus* (Millar & Innes 1983, 1985;

Millar *et al.* 1992; Havelka & Millar 1997; MacDonald 1997; Woolfenden & Millar 1997). Survival tended to be higher for both OW and YY females on protein supplemented grids during the breeding season when food was added, although these differences were not significant. However, during the non-breeding season when food was not added, populations previously supplemented with protein suffered higher winter mortality. Survival of YY in supplemented populations was less than half of that reported previously for this population (Millar 1994). Survival is rarely increased during food supplementation (Boutin 1990), and few studies have examined survival after the removal of supplemental food (but see Duquette 1992; Dobson 1995).

Young-of-the-year and OW females differed in the number of litters conceived per season, but not in litter success or litter size. Although sample sizes were small, over-winter survival and reproductive success (number of offspring raised to weaning) as yearlings did not appear to differ among those females who bred as YY and those who delayed first reproduction until 1 year of age. These results are consistent with Teferi & Millar (1993), who also found that females who bred as YY could potentially raise 50% more offspring in their lifetime than females breeding for the first time as yearlings.

Breeding by YY females can have a large effect on the number of offspring produced during the breeding season. If all 10 OW females in a population attempt at least one litter, a total of 9.7 YY females would be produced in first litters of the season (10 litters \* 58% success \* 3.70 offspring litter<sup>-1</sup> \* 45% female; Table 2). The remaining 5.7 YY females would be produced in subsequent litters for a total of 15.4 females produced per season (10 \* 1.59 litters season<sup>-1</sup> \* 58% success \* 3.70 offspring litter<sup>-1</sup> \* 45% female; Table 2). If, however, the 9.7 YY females expected to be weaned from the first litters of the season were able to breed as YY, an additional 5.2 YY females (9.7 \* 1 litter season<sup>-1</sup> \* 46% success \* 2.67 offspring litter<sup>-1</sup> \* 44% female) would be produced. As a result, breeding by YY females can result in a 34% increase (5.2/15.4) in the number of females produced during the breeding season.

Food abundance clearly has a large influence on population density and individual life-history characteristics (Boutin 1990), but aspects of diet other than energy content may also have important effects on population and individual processes. Dietary protein has the potential to limit many mammalian systems (White 1993), especially during reproduction and early growth when protein requirements are highest (Robbins 1983). In addition to absolute protein requirements, the source of dietary protein may also influence both the availability of specific amino acids and their digestibility. A better understanding of the relative importance of food quantity and food quality will provide new insight into the way in which ver-

tebrate populations are limited by the availability of food resources.

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