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Effects of nutritional factors on juvenile development in male European ground squirrels (*Spermophilus citellus*)

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Abstract

In European ground squirrels (*Spermophilus citellus*) as in other obligate hibernators, physiological processes underlie strict time constraints. Especially for the juveniles growth and prehibernatory fattening are temporally very limited. The aim of this study was to determine the effects of specific nutritional factors on development in juvenile males. Male ground squirrels were investigated for a 7-week period starting at natal emergence. Individuals were assigned to 1 of 3 experimental groups and were provided with a diet either high in proteins or polyunsaturated fatty acids (PUFAs). The third group was used as a control and was fed with a standard rat diet. Food quality affected growth in that males of the high-protein group had significantly larger head lengths in week 4, 5, and 6 after natal emergence than those of the other 2 groups. However, body mass did not differ significantly between the groups. All juvenile males showed an increase in plasma testosterone levels from week 1–2 to week 3–4 post-emergence, thereafter testosterone secretion remained stable until week 5–6. The daily testosterone increase after the first 2 weeks was significantly higher in the PUFA group compared to the other 2 groups. The results underline the importance of diet composition for growth rates and the timing of gonadal activation during early development in this species.

Key words: *Spermophilus citellus*, nutrition, juvenile development, testosterone

Introduction

In hibernating animals, reproduction is strongly limited by energetic and temporal constraints (Heaney 1984). Therefore, reproduction often competes with energy allocation to developmental processes like structural growth or prehibernatory fattening. European ground squirrels (*Spermophilus citellus*) are obligate hibernators, and their mating period is limited to 3 weeks in spring. Females produce only 1 litter per year. The juveniles are born in early May after a 4-week gestation period and emerge from the natal burrow at an age of about 32 days (Millesi et al. 1999a, b). Females continue lactation after juvenile emergence for up to 3 weeks depending on maternal condition and litter size (Huber et al. 1999, 2001). Juvenile European ground squirrels enter hibernation in late September (Millesi et al. 1999b), and hence time for growth and fattening is

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limited. Under these energetic and temporal constraints, nutrition in the first weeks of life may be crucial for further development.

In ground squirrels, specific nutritional components have been shown to be critical for developmental processes and hibernation patterns (Frank and Storey 1995; Pulawa and Florant 2000). Numerous studies have demonstrated that polyunsaturated fatty acids (PUFAs) affect hibernation, as PUFAs can enhance the duration and depth of mammalian torpor (Bruns et al. 2000; Florant 1998; Florant et al. 1993; Frank and Storey 1995; Geiser and Kenagy 1987; Geiser et al. 1992; Harlow and Frank 2001; Hill and Florant 2000). PUFA content of plants can vary with species, season, and among different parts of the plant (Florant et al. 1990; Quinn 1988). Free-ranging ground squirrels try to maintain the optimal depot-fat composition for hibernation through active diet selection (Frank 1994). European ground squirrels hibernate in individual burrows, and during hibernation, they rely completely on the stored body fat (Millesi et al. 1999b). Our study focused on the effects of 2 nutritional factors: Proteins, which are known to be essential for growth and development (Armstrong et al. 2001; Matsuo et al. 1995; Swanson 1989), and PUFAs that determine fat content and composition.

In many ground squirrel species including *S. citellus*, natal dispersal starts about 30–40 days after natal emergence (Hoffmann et al. 2004; Holekamp 1984). The study concentrated on the time period between natal emergence and the potential onset of dispersal. This period can be viewed as crucial for developmental processes. Structural growth in the first active season has been found to be terminated in juveniles at an age of approximately 100 days (Duschek unpubl. data), i.e. about 10 weeks after natal emergence. Juvenile European ground squirrels start to depart from their birth sites at 9 weeks of age, and males tend to move with higher travel speed than females (Hoffmann et al. 2004). The departure of the juveniles from their natal burrow involves an increased risk of predation (Hoffmann et al. 2004). Furthermore, dispersed juveniles have to find and adapt hibernacula in the new area.

Free-living male European ground squirrels have a maximum life span of 4 years (Hoffmann et al. 2003), and the lifetime reproductive success can be increased by precociousness. Age at sexual maturity varies with population density (Hoffmann et al. 2003; Millesi et al. 2004). At high population densities with high intrasexual competition, most yearling males were sexually immature, while in low-density years with high female availability, all yearling males were reproductively active. As sexually active males emerge from hibernation with developed testes (Millesi et al. 1998), the decision on sexual maturity is made before or during hibernation. Therefore, nutrition in juveniles may be important for the timing of puberty. Proteins may enhance growth and improve body condition, resulting in increased competitive ability in agonistic interactions (Schwagmeyer and Brown 1983). Optimal body-fat composition enables optimal timing of hibernation patterns, which in turn may stimulate gonadal development.

The aim of this study was to determine the effects of specific nutritional factors on the juvenile development in male European ground squirrels during a 7-week period starting at natal emergence.

Material and methods

Animals, study area, and capture techniques

26 juvenile male European ground squirrels (*Spermophilus citellus*), living in 3 outdoor enclosures in the north of Vienna (Austria), were investigated from their natal emergence (late May/early June) until week 7 thereafter (July). The enclosures were located within the distribution range of the species, and the ground squirrels lived in natural, self-made burrows.

All animals were permanently marked with transponder chips injected subcutaneously in the neck region (RFID-tag, Datamars Comp.). For distant recognition the fur was painted with commercial hair dye in individual patterns. Males were captured in weekly intervals with Tomahawk live traps. At each capture animals were weighed ($\pm$0.5 g, Sartorius laboratory scale), and head length was measured using a caliper ($\pm$0.1 mm, condylo-basal length), if the animal kept calm enough to get reliable data.