Does testosterone or coloration affect growth rates of adult males of the lizard *Psammodromus algirus*?

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Abstract: Elevation of circulating testosterone levels during the breeding season stimulates the development of breeding coloration and favors traits that maximize reproductive success, but it may have several costs. Retardation of growth by testosterone has been proposed as one of these costs to males of long-lived species. We examine the results of a field experiment on the short-term (within the breeding season) and long-term (1 year) effects of heightening levels of testosterone on the growth of males of the lacertid lizard *Psammodromus algirus*. Testosterone did not affect the mass gain of males during the reproductive period or between years. The between-years increase in snout–vent length in testosterone-supplemented males tended to be less than in control individuals. Within-season individual mass gain was mainly determined by body mass at emergence: the lower the body mass the higher the mass gain. Our results do not support the idea that the production of ornamental traits limits growth, as the negative correlation between degree of head coloration and mass gain disappeared when body mass was controlled for.

Résumé : L'augmentation des concentrations de testostérone en circulation au cours de la saison de reproduction peut stimuler l'apparition de colorations nuptiales et favoriser des caractères qui maximisent le succès de la reproduction, mais peut également comporter des coûts élevés. A titre d'exemple, une croissance retardée par la testostérone peut être une conséquence coûteuse chez les mâles d'espèces à longévité élevée. Nous examinons ici les résultats d'une étude sur le terrain sur les effets à court terme (au cours de la saison de reproduction) et à long terme (1 an) de l'augmentation de la testostérone sur la croissance des lézards mâles *Psammodromus algirus*. La testostérone n'a pas affecté les gains de masse chez les mâles durant la saison de reproduction ou d'année en année. L'augmentation de la longueur museau-évent d'une année à l'autre chez les mâles qui ont pris de la testostérone avait tendance à être plus faible que celle enregistrée chez les témoins. Au cours d'une saison, les gains de masse sont généralement déterminés par mesure de la masse à l'émergence : plus la masse est élevée, plus le gain de masse est important. Nos résultats n'appuient pas l'hypothèse selon laquelle la production d'une ornementation limite la croissance, puisque la corrélation négative entre l'intensité de la coloration de la tête et le gain de masse disparaît lorsqu'on tient compte de la masse du corps.

[Traduit par la Rédaction]

Introduction

One of the most important components of the reproductive effort of breeding male lizards in temperate areas is the production of high levels of testosterone shortly before the reproductive season begins (Moore and Lindzey 1992). It has been shown experimentally that the elevation of circulating testosterone levels in adult male lizards activates the development of ornamental characters (Cooper et al. 1987) and courtship behavior (Ferguson 1966; Cooper et al. 1987) and improves sprint speed, burst stamina (Klukowski et al. 1998), and exercise endurance (John-Alder 1994), and increases activity, home-range size (De Nardo and Sinervo 1994), and aggressive behavior (Moore and Marler 1987). The action of the hormone through these characters may increase reproductive success, but evidence for this is largely lacking.

In many other lizards, however, male-typical color patterns are not seasonally activated by circulating androgens but are present year-round. Experimental evidence suggests that in the adults of these lizard species, androgens do not contribute much to the expression of ornamental nuptial traits (Moore et al. 1998). Sex differences in these traits in these species are probably due to the early organizational action, rather than seasonal activation, of androgens and this has been confirmed in manipulative studies on hatchlings in one species (Hews and Moore 1995).

Raising the testosterone level also has costs, however. Higher circulating testosterone levels may increase energy expenditure, owing to an increase in territorial defense (Marler et al. 1995), and may reduce immunocompetence (Saad et al. 1990, 1992) and increase susceptibility to ectoparasitic infestation (Salvador et al. 1996). Thus, testosterone-implanted lizards may experience greater mortality (Marler and Moore 1988; Salvador et al. 1996). Another cost associated with testosterone may be its adverse effect on growth. Reptiles continue to growth after sexual maturity until an asymptotic size is reached, after which growth is negligible (Andrews 1982).

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Testosterone limits the growth of both immature and mature males. Experimental work with the snake *Thamnophis sirtalis* has shown that body size may be reduced by administering testosterone to adult and young males (Crews et al. 1985); also, growth of the head may be inhibited in juveniles by this hormone (Shine and Crews 1988). In addition, administration of testosterone to juvenile lizards retarded body growth (Hews et al. 1994; Abell 1998).

The allocation of energetic resources to the production of testosterone and the elaboration of nuptial coloration may divert resources necessary for growth. The cost of reduced growth may be low in short-lived species but significant in longer lived species in which larger size and associated social dominance may increase reproductive success in subsequent breeding seasons. There may be a trade-off between growth and coloration that varies with age: older males invest a higher proportion of resources in coloration than in growth but the opposite occurs in younger males (Olsson 1994; Olsson and Silverin 1997). However, a seasonal increase in testosterone levels might have a greater effect on this trade-off in species in which the effects of androgens in activating sexual ornaments are significant than in species in which sex differences arise primarily from the early organizational actions of androgens.

The lacertid lizard *Psammodromus algirus* is an appropriate model in which to examine the relationships between testosterone, breeding coloration, and growth because growth continues after sexual maturity and male breeding coloration varies with size. In previous work (Salvador et al. 1996, 1997), we examined the effect of testosterone supplementation on male breeding coloration. We examine here the results of a field experiment on the effects of heightening testosterone levels on male growth within and between 2 consecutive years. We predicted that males treated with exogenous testosterone would grow less, both during the reproductive season and between years. We also examine the relationships between mass growth and area of nuptial coloration on the head during the breeding season. According to the trade-off hypothesis, we would expect an inverse relationship between mass gain and the area size of nuptial coloration, irrespective of body mass.

Methods

Our study was conducted at a site in a deciduous oak (*Quercus pyrenaica*) forest near Navacerrada, Madrid Province, Spain, during 4 consecutive years (1994–1997). We established a 2-ha grid with markers every 10 m and captured adult males by noose shortly after lizards emerged from hibernation (February–March). Individuals were weighed with a Pesola spring balance to the nearest 0.1 g (range = 7–16 g) and marked by toe-clipping for individual recognition; snout–vent length (SVL) was measured to the nearest millimetre (range = 70–86 mm).

Captured males were alternately assigned to a control group (C) or a testosterone-implanted group (T). Both C and T males received a subcutaneous implant of Silastic tubing (outer diameter 1.95 mm, inner diameter 1.47 mm; Dow Corning), the end of which was plugged with wooden caps and sealed with silicon adhesive. Males were cold-anaesthetized and implanted through a small dorsal incision that was closed with a suture. C males received an empty implant, while the implant of T males contained 3 mm (for males smaller than 80 mm SVL) or 5 mm (for males larger than 80 mm SVL) of packed crystalline testosterone propionate (Sigma Chemical Company) (Salvador et al. 1996, 1997). Males were released at the capture site during the same day. All males recaptured at the end of the reproductive season still had the implant, but most males recaptured the following year had apparently lost the implant before emergence. Males that lost their tail during the breeding season were excluded from the analyses.

C (n = 57) and T (n = 40) males recaptured at the end of the breeding season (May) were weighed and the pattern of color spots on the throat was drawn using a camera lucida or photographed. We classified the orange throat coloration of males as one of five types, from minimum to maximum coloration: 0, no coloration; 1, spots on the fourth pair of submaxillary scales; 2, spots on the third and fourth pairs of submaxillary scales; 3, spots on all submaxillary scales; and 4, extensive, nonfragmented coloration (Veiga et al. 1997) (Fig. 1). When coloration was included as a factor in ANCOVAs, we considered only two groups: (1) individuals with no or reduced coloration (types 0–2) and (2) individuals with well-developed coloration (types 3 and 4). The number of days that elapsed between initial capture and final recapture within the breeding season (42.7 ± 0.9 days; mean ± SE) did not vary significantly between coloration types (F = 0.30, P = 0.87) or experimental treatments (F = 0.90, P = 0.34).

We did not measure plasma testosterone levels during the experiment, so we cannot exclude the possibility that we recorded the effects of pharmacological doses. However, the relation of testosterone level with body mass was similar to that used in several studies in which no pharmacological effects were detected (Marler and Moore 1988). We observed no abnormality in the behavior of the experimental males.

During the reproductive season, we used the change in mass rather than the change in SVL as an index of growth. Because the period between initial capture and final recapture during the reproductive season was relatively short (25–65 days), increments in SVL were small and often within measurement error. In contrast, mass could be measured more accurately than SVL, therefore changes in mass better reflected actual growth. However, growth in SVL from one season to the next was substantial, at least for individuals with SVL of 80 mm or less (Fig. 2). We computed mass change in grams per day between the day of initial capture and the day of final recapture. We termed the two mass values initial mass and final mass, respectively.

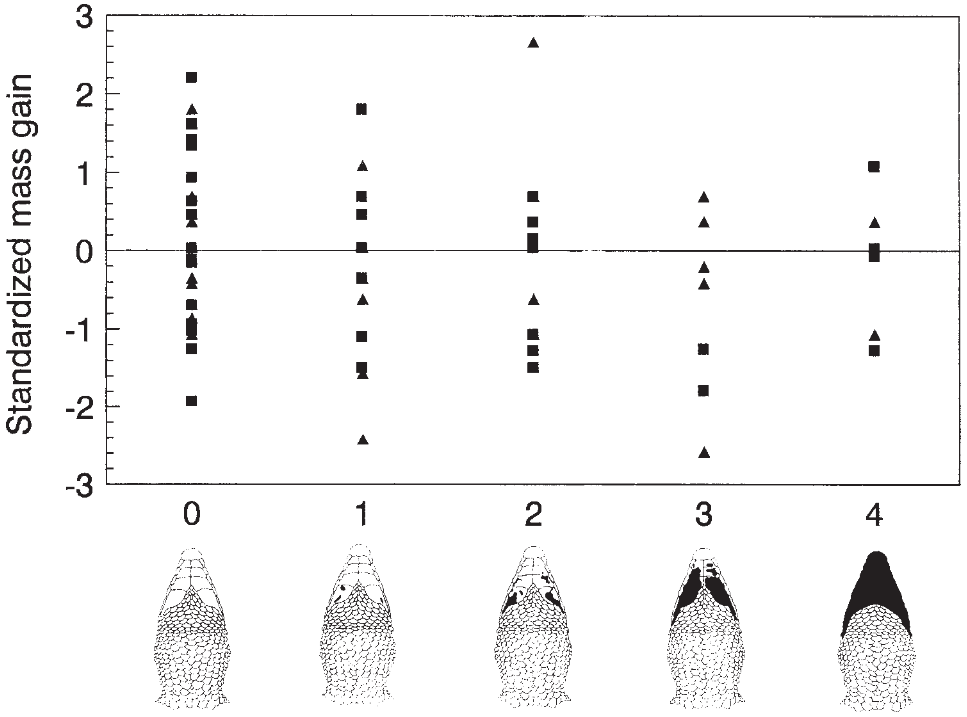
We also examined the delayed effects of testosterone on growth. For males captured in 2 consecutive years, we examined the differences in SVL and mass (measured at emergence) between the year of treatment and the following year (n = 10 for C males; n = 5 for T males).

As morphological and growth variables showed significant interannual variation, we standardized the values for each year independently in order to have 0 mean and 1 standard deviation when they were pooled in statistical analyses. Standardized coloration values were logarithmically transformed for use in correlation analyses.

Results

Table 1 presents SVLs, masses, and mass growth rates for C males and T males during the reproductive period. The three variables varied significantly between years (ANOVA, P < 0.001 in all cases). Figure 1 shows mass gain during the reproductive period for the different coloration types considered. Table 2 shows SVL and mass in relation to coloration type. Significant positive correlations were found between coloration type and SVL (r = 0.46, P < 0.001) and coloration type and mass (r = 0.41, P < 0.001).

Fig. 1. Relationship between standardized mass gain during the reproductive season and coloration type in control (■) and experimental (▲) male lizards (*Psammodromus algirus*).



Interannual growth

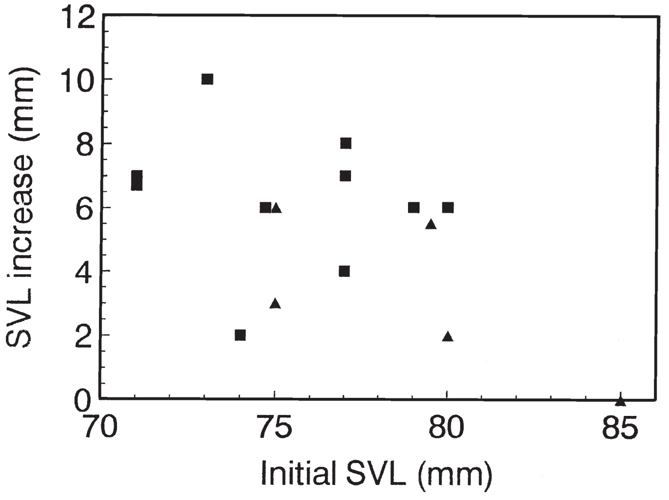
Mass gain between years did not reveal significant effects of treatment (analysis of covariance (ANCOVA), $F_{[1,12]}$ = 0.49, P = 0.49, power = 0.11; covariate initial mass, $F_{[1,12]}$ = 2.49, P = 0.14, power = 0.31). SVL growth between years was not affected by treatment, but individuals treated with testosterone tended to grow less (3.3 ± 1.11 mm; mean ± SE) than control individuals (6.3 ± 0.68 mm) ($F_{[1,12]}$ = 2.47, P = 0.14, power = 0.30; covariate initial SVL, $F_{[1,12]}$ = 2.25, P = 0.16, power = 0.28) (Fig. 2).

Growth within the breeding season

A two-factor ANCOVA for mass gain with initial mass as covariate indicated that (*i*) testosterone treatment did not significantly affect mass growth rates during the breeding season (C males, 0.03 ± 0.12 g/d (mean ± SE); T males, –0.05 ± 0.18 g/d; $F_{[1,92]}$ = 0.06, P = 0.8, power = 0.04) and (*ii*) coloration type was not associated with mass growth ($F_{[1,92]}$ = 0.08, P = 0.78, power = 0.05).

The correlation between initial mass and mass gain when coloration type and treatment were factored out was significant (ANCOVA, $F_{[1,86]}$ = 7.75, P = 0.007, power = 0.78) (Fig. 3), indicating that initial mass is a major predictor of mass gain.

Fig. 2. Relationship between increase in snout–vent length (SVL) between 2 consecutive years and initial SVL in control (■) and experimental (▲) male lizards.



Discussion

We predicted that male *P. algirus* with testosterone implants would have lower growth rates than controls. This prediction was not supported by our observations. It would be expected that males with the highest testosterone levels would have negative energetic budgets, because testosterone can stimulate aggressiveness, patrolling, and activity in several lizard species (*Sceloporus jarrovi*, Moore and Marler 1987; *Uta stansburiana*, DeNardo and Sinervo 1994). However, testosterone did not affect mass gain in male *P. algirus*, at least during the reproductive period, although the low statistical power of the test makes this conclusion tentative. In several other lizard species, sexually mature males grow little, if at all, during the reproduction period (Tinkle 1967; Dunham 1981). As body size can be crucial for maximizing reproductive success (e.g., Gullberg et al. 1997), males could overcome the constraints imposed by reproduction by delaying growth until the annual postreproductive period. This has

Table 1. Initial snout–vent lengths (SVL) and masses of male lizards (*Psammodromus algirus*) measured at emergence from hibernation and growth rates of both control and experimental males in different years.

		SVL (mm)		Mass (g)		Growth rate (g/d)	
Year	*n*	Mean ± SE	Range	Mean ± SE	Range	Mean ± SE	Range
1994	22	80.5±0.99	73–85	12.6±0.50	9.0–16.0	–0.005±0.003	–0.030 to 0.02
1995	19	75.3±0.88	70–85	9.1±0.35	7.0–12.0	0.020±0.004	–0.020 to 0.05
1996	27	80.0±0.62	75–86	11.1±0.30	7.8–14.7	0.009±0.003	–0.030 to 0.05
1997	29	75.5±0.70	70–85	9.5±0.30	7.1–13.5	0.020±0.002	–0.003 to 0.05

Table 2. Snout–vent lengths (SVL) and masses of male lizards measured at emergence, according to coloration types (see Fig. 1).

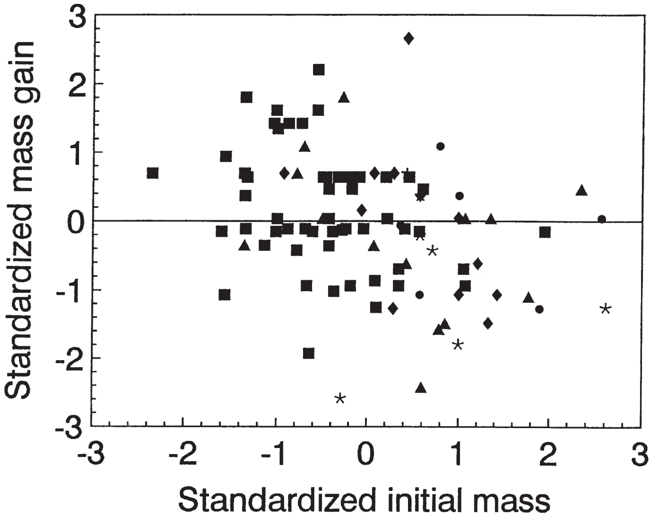
		SVL (mm)		Mass (g)	
Coloration type	*n*	Mean ± SE	Range	Mean ± SE	Range
0	56	75.0±0.40	70–83	9.2±0.2	7.0–12.4
1	15	81.1±0.80	75–86	11.5±0.3	9.5–14.5
2	12	81.7±0.80	77–85	12.5±0.5	9.8–16.0
3	7	82.9±1.10	78–85	13.0±0.6	10.7–15.0
4	7	83.6±0.43	82–85	14.1±0.4	12.0–15.0

been demonstrated in the desert lizards *Sceloporus merriami* and *Urosaurus ornatus*, which exhibited reduced foraging success in late spring, while in summer, after reproduction concluded, they increased their foraging efficiency coincidentally with higher prey abundance (Dunham 1981). These results may not apply to *P. algirus*, however, because the temperate regions in which it lives are ecologically very different from desert regions.

In any case, the notion that testosterone may affect short- or long-term growth cannot be completely rejected on the basis of these results, because the null hypothesis was accepted with rather low statistical power. It is possible that no effect of testosterone on mass loss was detected because it was compensated for by muscle hypertrophy, a common effect of this hormone (Mooradian et al. 1987; John-Alder 1994). A trade-off between testosterone production and growth would imply that, once a critical size is attained, an individual should decide between investing in reproduction by increasing its circulating hormone levels, thereby restricting its growth and the possibility of future reproduction, and delaying reproductive effort. Previous evidence showed that in contrast to the effect observed in larger males, giving exogenous testosterone to small adult males did not affect several reproductive variables (Salvador et al. 1996, 1997), suggesting that in individuals which have not yet attained a size favoring reproductive success, a better strategy would be to invest in growth rather than in reproduction.

The variation of mass gain in *P. algirus* depended mainly on the individual's size, or, in other words, on the growth already attained, as occurs in many other lizards (Andrews 1982). Contrary to what has been suggested recently (Olsson and Silverin 1997), our results do not support the idea that the development of ornamental traits limits growth. The correlation between the size of the head-coloration area and mass gain disappeared when the individual's size was controlled for statistically. Previous results showed that in *P. algirus* and other lizards, developing larger patches of color required an increment in circulating testosterone levels (Cooper et al. 1987; Salvador et al. 1996). However, only larger males were able to increase coloration by means of this mechanism, suggesting that coloration is not useful for small males (Salvador et al. 1997). The development of advertising coloration by small subordinate individuals may render them more easily detectable by dominant individuals, thus increasing their risk of being attacked and (or) suffering injuries (Rohwer 1975; Rohwer and Ewald 1981; Cooper et al. 1987).

Fig. 3. Relationship between standardized mass gain by male lizards during the reproductive season and standardized initial mass. Coloration types are as follows: ■, type 0; ▲, type 1; ◆, type 2; *, type 3; ●, type 4.



The development of nuptial coloration could be a terminal investment for old individuals with reduced survival expectancies. Of seven males captured at the end of the reproductive season in 2 consecutive years, six did not develop coloration during the first year and only one exhibited type 1 coloration (Fig. 1). No male with well-developed coloration (types 3 and 4) was recaptured the following year. Also, in a nearby population, Díaz (1993) reported higher mortality among the most-colored males. However, the wide SVL range for each coloration class considered in this study suggests that other factors, such as individual variation in the ability to produce colors and (or) in mass-gain rates, are also involved in the variability of coloration recorded. Alternatively, males might develop coloration only after reaching a certain body size and condition that enable them to reproduce successfully. Thus, the apparent elevated mortality of individuals with fully developed colors would result from a high level of reproductive effort rather than as a direct consequence of advanced age.

In conclusion, the better predictor of growth in *P. algirus* is the size of the individual, although the growth rate seems to be determined to a considerable extent by factors not considered in this study. Testosterone had no detectable effect on within-season changes in mass or between-season growth in SVL, but the results are not completely conclusive. The development of color traits, however, does not seem to constrain growth. On the contrary, coloration seemed to develop only after individuals had reached a critical size that presumably confers on the individual considerable opportunities to reproduce.

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