

Postautotomy tail activity in the Balearic lizard, *Podarcis lilfordi*

Panayiotis Pafilis · Valentín Pérez-Mellado ·
Efstratios Valakos

Received: 18 April 2007 / Revised: 24 September 2007 / Accepted: 16 October 2007 / Published online: 10 November 2007
© Springer-Verlag 2007

Abstract Caudal autotomy is an effective antipredator strategy widespread among lizards. The shed tail thrashes vigorously for long periods to distract the predator and facilitate the lizard's escape. This movement is maintained by energy supplied by the anaerobic conversion of glycogen into lactate. It has been suggested that lactate accumulation serves as an index for the vigor of tail thrashing. We made three predictions: (1) tail loss frequency should be higher under heavier predation regime, (2) the duration of postautotomy tail movement should be extended in populations under heavy predation pressure as an adaptation to the higher risk and the increased need for defense, and (3) as result, lactate in these tail tissues should be concentrated at higher levels. To eliminate the impact of phylogeny and environmental factors on the interpretation of our result, we focused exclusively on one species, the Balearic lizard (*Podarcis lilfordi*). We studied three populations under different predation pressure but sharing the same climatic conditions. We found no differences among the studied populations either in postautotomy duration of

tail movement or in levels of final lactate accumulation while autotomy frequency was higher where predation pressure was more intense. Tail loss effectiveness is directly influenced by the level of predation, while secondary features of the trait appear to remain independent from the impact of environment.

Keywords Predator–prey interactions ·
Evolutionary physiology · Lacertids · Inlands

Introduction

Tail autotomy is an effective defensive mechanism widely used by lizards in many families (Arnold 1984; Bellairs and Bryant 1985). Tail shedding may provide an immediate survival benefit, but the ensuing consequences have a direct impact on lizard's overall fitness (Arnold 1988). Caudal autotomy is related to reduced locomotory ability, lowered social status, higher mortality rates, and loss of energy storage with subsequent reduction in reproductive effort and growth (Fox and Rostker 1982; Vitt and Cooper 1986; Martin and Salvador 1992; Martin and Avery 1998; Fox and McCoy 2000; Chapple and Swain 2002).

Shed tails may thrash vigorously for extended periods to distract a predator from an escaping lizard (Clark 1971; Vitt et al. 1977). Postautotomy movement is fueled by glycogen, which is oxidized anaerobically into lactate, providing the required energy to the muscles (Dial and Fitzpatrick 1981, 1983; Gleeson 1996). Physiological pathways involved in tail muscle energetics have been reported as sharing a common pattern among species of the same family (Pafilis et al. 2005). However, interspecies divergences in phylogeny may introduce biases that can easily obscure a real pattern or even create a spurious one (Huey 1987; Pianka 2001).

P. Pafilis
Department of the Environment, University of the Aegean,
Lofos Xenia,
81100 Mytilene, Lesvos, Greece
e-mail: pafilis@aegean.gr

V. Pérez-Mellado
Department of Animal Biology, University of Salamanca,
Salamanca 37071, Spain
e-mail: valentin@usal.es

E. Valakos (✉)
Department of Biology, Section of Animal
and Human Physiology, University of Athens,
Panepistimiopolis, Ilissia,
157-84 Athens, Greece
e-mail: evalakos@biol.uoa.gr

Furthermore, differences in ecological factors, such as climate, may alter the interpretation of ecophysiological studies' findings, as the effect of the environment on physiological processes is deeply important (Goldstein and Pinshow 2006; Naya and Bozinovic 2006).

In this paper, we focus on populations of a single species, the Balearic lizard *Podarcis lilfordi*, to eliminate the confounding effects of phylogeny and environmental variation. The three populations studied share the same climatic conditions but are experiencing different predation pressure and are showing diverse postautotomy tail activity in regard to vigor and duration of movement (Cooper et al. 2004). Duration and vigor of tail thrashing are known to increase a lizard's escape time (Congdom et al. 1974; Dial and Fitzpatrick 1983). We predicted that first, where predation pressure is more intense, the frequency of voluntary autotomy should be higher; second, where autotomy is more frequent, the duration of movement should be more prolonged; third, where the movement is more intense, anaerobic metabolism should be activated more intensely, and because lactate concentration might be interpreted as an index for the vigor of postautotomy movement (Meyer et al. 2002), this should be accumulated at higher levels.

Materials and methods

Study organism and site

P. lilfordi is a small-bodied (approx. 7.5 g), diurnal lizard. Its distribution is restricted to coastal islets around Mallorca and Menorca (Balearic Islands, Spain). Our investigation was conducted in the islets of Colom (51 ha), Aire (34.3 ha), and Sargantana (2.3 ha) close to Menorca coast. The three islets harbor different predators and experienced different levels of human activities (Pérez-Mellado 1989; Pérez-Mellado et al. 2003; Table 1). Although tail-break frequency in natural populations has often been misinterpreted (Schoener 1979), it still remains a useful indication for predation pressure (Turner et al. 1982). To assess the frequencies of regenerated tails, we made a survey of 663 lizards from the three populations (Table 1).

Table 1 Presence/absence (+/–) of potential predators and proportion of regenerated tails in the three islets under study (number of examined individuals is presented in parenthesis)

	Aire	Sargantana	Colom
Breeding kestrels	–	+	+
Breeding seagulls	+	–	+
Snakes	–	–	+
Ship rats	–	–	+
Long time human settlement	–	+	+
Percentage of regenerated tails	48.57% (560)	59.65% (57)	73.91% (46)

Predation simulation

To simulate predatory attacks, we adopted the technique described by Pérez-Mellado et al. (1997). Lizards (52 individuals, 30 males and 22 females) were placed on a cork substrate (offering traction), and their tail was grasped with a digital caliper at a distance of 20 mm from the cloaca. The grasp force was standardized by taking into account the diameter of the tail and then reducing the opening calipers to one half of that. Each trial lasted no more than 15 s, and if the tail was shed, the time to cessation of movement was measured (samples termed “exhausted tail”). In 15 individuals (five from each population), the tail was removed rapidly with the manual use of forceps, to obtain an estimate of baseline metabolite concentrations at time zero (termed “resting tails”). Immediately after shedding, the tails were placed in dry ice to avoid changes in physiological metabolites concentrations.

Tissue metabolites determination

Lactate Muscle tissue (150 mg) was homogenized (1:3 w/v) with ice-cold perchloric acid, and the homogenate was centrifuged at 4°C and 5,000×g for 10 min. The pellet was used in protein analysis. The supernatant was neutralized (with 0.5 M Tris/0.5 M KOH) and centrifuged again. The pellet was discarded, and the supernatant was used for lactate determination using the method of Hohorst (1965). Measurements were read at 340 nm using a spectrophotometer (Novaspec II, Pharmacia Biotech).

Glycogen The indirect method by Seifter et al. (1950) against a glucose standard was followed. Tailpieces, after the tissue had been minced, were boiled for 20 min in 30% KOH. The absorbance was read at 620 nm.

Results

Proportion of regenerated tails was significantly different in the three natural populations studied (*G* test: $G=13.12$, $P<0.01$). Pairwise comparisons indicate that only the population

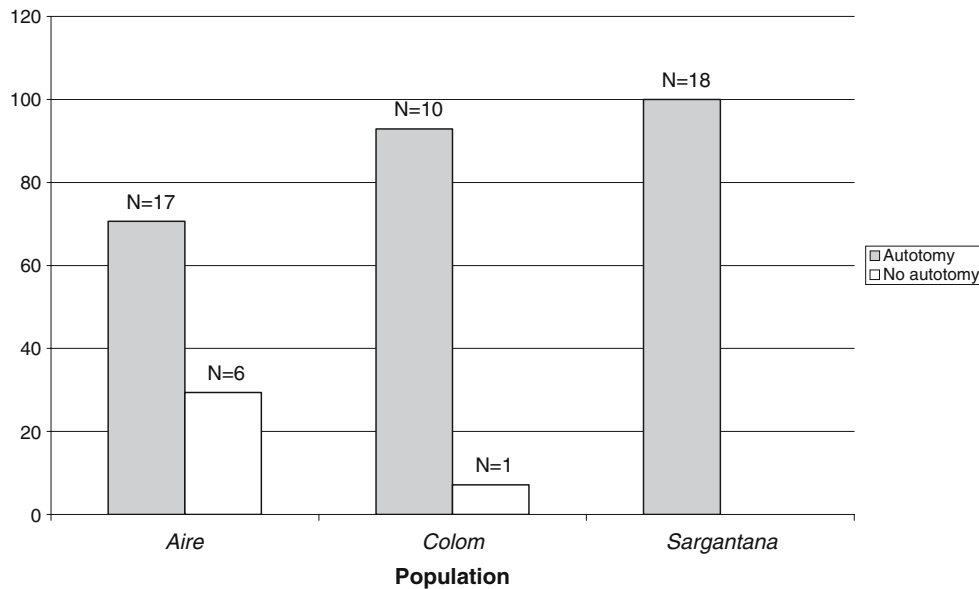


Fig. 1 Percentage of autotomized tails in the three populations under study. *N* on the bar stands for the examined individuals

of highest (Colom) and lowest (Aire) predation pressure really showed a different proportion of regenerated tails ($G=11.36$, $P<0.01$) in the expected direction. No differences were detected between Aire and Sargantana ($G=2.55$, $P>0.05$) or Sargantana and Colom ($G=2.34$, $P>0.05$).

A clear difference in autotomy performance after predation simulation emerged from the comparison of the three populations ($G=218.013$, $P<0.05$, $df=2$; Fig. 1). When the Aire population was excluded from the analysis, this difference disappeared ($G=0.742$, $P>0.05$, $df=1$).

Mean duration of tail movement was around 3 min in all cases (Table 2). No significant differences were detected among populations examined (two-way analysis of variance: $F_{2, 26}=0.719$, $P=0.49$) or between the two sexes ($F_{1, 26}=0.168$, $P=0.68$); the interaction terms were not significant either ($F_{2, 26}=0.278$, $P=0.78$).

Significant differences were detected in lactate accumulation between the exhausted and the resting tails

(Table 2; $F_{1, 33}=56.56$, $P<0.00001$). However, we did not find any difference between the examined populations ($F_{2, 33}=0.15$, $P=0.85$) or between sexes ($F_{1, 33}=0.021$, $P=0.88$). The interaction between the three factors was nonsignificant (*population–sex*: $F_{2, 33}=0.175$, $P=0.40$; *population–movement duration*: $F_{2, 33}=0.23$, $P=0.79$; *sex–movement duration*: $F_{1, 33}=0.04$, $P=0.84$; *between the three factors*: $F_{2, 33}=0.18$, $P=0.82$).

Glycogen concentrations in all populations examined differed between exhausted and resting tails and were higher in the former (Table 2; $F_{1, 21}=1279.1$, $P<0.00001$). In contrast, we found no significant differences either between the examined populations ($F_{2, 21}=0.46$, $P=0.64$) or between the sexes ($F_{1, 21}=0.05$, $P=0.83$; interactions: *population–sex*: $F_{2, 21}=0.022$, $P=0.98$; *population–movement duration*: $F_{2, 21}=0.48$, $P=0.63$; *sex–duration status*: $F_{1, 21}=0.01$, $P=0.91$; *between the three factors*: $F_{2, 21}=1.242$, $P=0.33$).

Table 2 Duration of movement and final concentrations of lactate and glycogen in resting and exhausted tails for the three populations examined

Populations	Time (min), mean±SD; <i>N</i> ; (range)	Lactate (mg/g tissue)		Glycogen (mg/g tissue)	
		Resting tails, mean±SD; <i>N</i> ; (range)	Exhausted tails, mean±SD; <i>N</i> ; (range)	Resting tails, mean±SD; <i>N</i> ; (range)	Exhausted tails, mean±SD; <i>N</i> ; (range)
Aire	3.46±1.33; 13; (1.50–6.20)	0.85±0.03; 5; (0.80–0.87)	1.85±0.39; 13; (1.24–2.59)	7.45±0.36; 4; (6.95–7.74)	2.51±0.36; 9; (2.04–3.25)*
Colom	3.55±1.73; 6; (1.20–6.32)	0.84±0.07; 5; (0.025–0.034)	1.85±0.48; 6; (1.12–2.42)	7.56±0.22; 4; (7.32–7.85)	2.55±0.49; 4; (2.04–3.12)*
Sargantana	2.87±1.21; 13; (1.45–5.19)	0.85±0.06; 5; (0.79–0.91)	1.68±0.37; 13; (1.02–2.20)	7.71±0.27; 5; (7.26–7.98)	2.48±0.34; 7; (2.02–3.01)*

*Denotes significance at $P<0.05$

Discussion

The differences in predation pressure are reflected in the fraction of autotomized tails (Fig. 1). As previous studies indicated, the duration of tail thrashing is especially important for snake predators (Dial and Fitzpatrick 1983), and we would expect longer thrashing periods in Colom, while the predator-free population from Aire should show the briefest duration of movement. However, duration of movement did not differ among the three populations (Table 2). We believe that the reason for this lack of diversity is the recent invasion of predators. The Balearic Islands were free from saurophagous terrestrial and avian predators starting with the Messinian Salinity Crisis (5.3 million years ago) until the arrival of humans in the Holocene Period (6,000–8,000 years ago; Pérez-Mellado et al. 1997). Because predation pressure was low, the effectiveness of autotomy was reduced (Pérez-Mellado et al. 1997). Hence, it seems that the isolation time has been too short for a selective adaptation regarding postautotomic tail activity. This point is further indicated by the longer duration of tail thrashing of lacertids from eastern Mediterranean islands (Pafilis et al. 2005) where predation pressure has been always heavy, as fossil records indicate (Kotsakis 1981; Caloi et al. 1988). Postautotomy movement had to be more prolonged in eastern species so as to offset the severe predation strain. This also applies to continental Spanish species, which show longer durations as a result of the higher predation to which they are subject (Pérez-Mellado et al. 1997). Alternatively, it is possible that duration of thrashing is a conservative trait that does not easily change in response to predation pressure. The limited predation in Aire may be responsible for the lowest observed autotomy percentages among the three islets. The higher autotomy frequency was observed in Colom, and we believe that the discrepancy between field (Table 1) and experimental data (Fig. 1) can be explained by the limited sample size in the case of predation simulation). Yet, the value for Aire (74%) is still high as compared to other lacertids (Pérez-Mellado et al. 1997; Pafilis et al. 2005). We believe that the reason must be attributed to the extremely dense population of the islet, estimated at around 4,100 individuals/ha (as opposed to 1,600 for Colom and 690 for Sargantana, Pérez-Mellado 1998). This density, in combination with the small size of the islet and the low food availability (Brown and Pérez-Mellado 1994), increases competitive interaction between the lizards, even to the point of attacking the tails of their rivals (Salvador 1986). Cannibalism is quite common in *P. lilfordi* (Arnold 1988; Pérez-Mellado 1998), with this effect further exacerbated in the extreme conditions of Aire. Although tail shedding because of intraspecific competition seems to be uncommon among lacertids (Arnold

1988), there is evidence from dense insular populations of other *Podarcis* species supporting the opposite theory (Chondropoulos et al. 1993; Adamopoulou et al. 1999).

According to our predictions, as a consequence of the higher autotomy rate and the more intense movement, individuals from Colom and Sargantana should accumulate higher lactate levels than those from Aire. Nevertheless, we failed to detect any difference among populations just as recorded in other lacertids species (Pafilis et al. 2005). Anaerobic metabolism is a common energy production mechanism among reptiles (Pough and Andrews 1985). It seems that lactate production through anaerobiosis is a thoroughly conservative pathway that has been fixed during evolution, and it is not influenced by external factors. Glycogen converts into lactate during tail movement and thus final levels change inversely between exhausted and resting tails. Average lactate and glycogen concentrations were found at levels similar to those published elsewhere (Meyer et al. 2002; Pafilis et al. 2005). These results further support the conservative nature of the feature.

Taken together, our results suggest that duration of postautotomy movement is not subject to phylogenetic or ecological impact, at least for the case of *P. lilfordi*. Physiological metabolites related to anaerobiosis change evenly among populations or species. The duration of tail thrashing is predation dependent according to previous studies (Pérez-Mellado et al. 1997; Pafilis et al. 2005), as tail loss performance, but it seems that this change takes place at a slower rate. Comparative studies using species from different families would further clarify the actual nature of the effect.

Acknowledgements We would like to express our deep gratitude for the linguistic revision to Johannes Foufopoulos. All experiments comply with Spanish and Greek legislation for the Protection of Wildlife and Environment. All animals were released after the experimental procedure, and none was killed.

References

- Adamopoulou C, Pafilis P, Valakos E (1999) Diet composition of *Podarcis milensis*, *Podarcis gaigeae* and *Podarcis erhardii* (*Sauria: Lacertidae*) during summer. *Bonn Zool Beitr* 48: 275–282
- Arnold EN (1984) Evolutionary aspects of tail shedding in lizards and their relatives. *J Nat Hist* 18:127–169
- Arnold EN (1988) Caudal autotomy as a defense. In: Gans C, Huey RB (eds) *Biology of the reptilia 16, ecology B: defense and life history*. Alan R Liss, New York, pp 235–273
- Bellairs DA, Bryant SV (1985) Autotomy and regeneration in reptiles. In: Gans BC, Billet F (eds) *Biology of the reptilia 15, development*. Wiley, New York, pp 301–410
- Brown RP, Pérez-Mellado V (1994) Ecological energetics and food acquisition in dense Menorcan islet populations of the lizard *Podarcis lilfordi*. *Funct Ecol* 8:427–434

- Caloi L, Kotsakis T, Palombo MR (1988) La fauna a vertebrati terrestri del Pleistocene delle isole del Mediterraneo. *Bull Ecol* 19:131–151
- Chapple DG, Swain R (2002) Effect of caudal autotomy on locomotor performance in a viviparous skink, *Niveoscincus metallicus*. *Funct Ecol* 16:817–825
- Chondropoulos BP, Maragou P, Valakos ED (1993) Food consumption of *Podarcis taurica ionica* (Lehrs, 1902) in the Ionian islands (Greece). In: Valakos ED, Boehme W, Pérez -Mellado V, Maragou P (eds) *Lacertids of the Mediterranean Region: a biological approach*. Hellenic Zoological Society, Athens, pp 173–182
- Clark DR (1971) The strategy of tail autotomy in the ground skink *Lygosoma laterale*. *J Exp Zool* 176:295–302
- Congdom JD, Vitt LJ, King WW (1974) Geckos: adaptive significance and energetics of tail autotomy. *Science* 184:1379–1380
- Cooper WE, Pérez-Mellado V, Vitt LJ (2004) Ease and effectiveness of costly autotomy vary with predation intensity among lizard populations. *J Zool* 262:243–255
- Dial BE, Fitzpatrick LC (1981) The energetic costs of tail autotomy to reproduction in the lizard *Coleonyx brevis* (Sauria: Gekkonidae). *Oecologia* 51:310–317
- Dial BE, Fitzpatrick LC (1983) Lizard tail autotomy: function and energetics of postautotomy tail movement in *Scinella lateralis*. *Science* 219:391–393
- Fox SF, McCoy JK (2000) The effect of tail loss on survival, growth, reproduction, and sex ratio of offspring in the lizard *Uta stansburiana* in the field. *Oecologia* 122:327–334
- Fox SF, Rostker MA (1982) Social cost of tail loss in *Uta stansburiana*. *Science* 218:692–693
- Gleeson TT (1996) Post-exercise lactate metabolism: a comparative review of sites, pathways, and regulation. *Annu Rev Physiol* 58: 565–581
- Goldstein DL, Pinshow B (2006) Taking physiology to the field: using physiological approaches to answer questions about animals in their environment. *Physiol Biochem Zool* 79:237–241
- Hohorst HJ (1965) L- (+) lactate determination with lactate dehydrogenase and DNP. In: Bergmeyer HU (ed) *Methods of enzymatic analysis*. Academic, New York, pp 266–270
- Huey RB (1987) Phylogeny, history and the comparative method. In: Feder ME, Bennett AF, Burggren WW, Huey RB (eds) *New directions in ecological physiology*. Cambridge University Press, Cambridge, pp 76–101
- Kotsakis T (1981) Le lucertole (Lacertidae, Squamata) del Pliocene, Pleistocene e Olocene delle Baleari. *Boll Soc Hist Nat Balears* 25: 135–150
- Martin J, Avery RA (1998) Effects of tail loss on the movement patterns of the lizard, *Psammotromus algirus*. *Funct Ecol* 12: 794–802
- Martin J, Salvador A (1992) Tail loss consequences on habitat use by the Iberian rock-lizard, *Lacerta monticola*. *Oikos* 65:328–333
- Meyer V, Preest MR, Lochetto SM (2002) Physiology of original and regenerated lizard tails. *Herpetologica* 58:75–86
- Naya DE, Bozinovic F (2006) The role of ecological interactions on the physiological flexibility of lizards. *Funct Ecol* 20:601–608
- Pafilis P, Valakos ED, Foufopoulos J (2005) Comparative postautotomy tail activity in six Mediterranean lacertid species. *Physiol Biochem Zool* 78:828–838
- Pérez-Mellado V (1989) Estudio ecologico de la lagartija balear *Podarcis lilfordi* (Gunther, 1874) en Menorca. *Rev Menorca* 53:455–511
- Pérez-Mellado V (1998) *Podarcis lilfordi* (Günther, 1874). In: Ramos MA et al (ed) *Fauna Ibérica 10, Reptiles*. Museo Nacional de Ciencias Naturales, Madrid, pp 272–282
- Pérez-Mellado V, Corti C, LoCascio P (1997) Tail autotomy and extinction in Mediterranean lizards. A preliminary study of continental and insular populations. *J Zool* 243:553–541
- Pérez-Mellado V, Perera A, Cortázar G (2003) La Lagartija balear, *Podarcis lilfordi* (Günther, 1884) de l' Illa d' en Colom, Parc Natural de s' Albufera des Grau (Menorca). Situación actual y estado de conservación. *Bul Cien Esp Prot Bal* 1:23–34
- Pianka E (2001) The role of phylogenetics in evolutionary ecology. In: Lymberakis P, Valakos E, Pafilis P, Mylonas M (eds) *Herpetologia Candiana*. S.E.H, Iraklion, pp 1–20
- Pough FH, Andrews RM (1985) Use of anaerobic metabolism by free-ranging lizards. *Physiol Zool* 58:205–213
- Salvador A (1986) *Podarcis lilfordi* (Gunther, 1874)—Balearen—Eidechse. In: Boehme W (ed) *Handbuch der Reptilien und Amphibien Europas 2, Echsen 3*. Aula, Wiesbaden, pp 83–110
- Schoener TW (1979) Inferring the properties of predation and other injury-producing agents from injury frequencies. *Ecology* 60: 1110–1115
- Seifter U, Dayton S, Novic B, Muntwyler E (1950) The estimation of glycogen with anthrone reagent. *Arch Biochem* 24:191–200
- Turner FB, Medica PA, Jennrich RI, Maza BG (1982) Frequencies of broken tails among *Uta stansburiana* in southern Nevada and a test of the predation hypothesis. *Copeia* 1982:835–840
- Vitt LJ, Cooper WE (1986) Tail loss, tail color, and predator escape in *Eumeces* (Lacertilia: Scincidae): age-specific differences in costs and benefits. *Can J Zool* 64:583–592
- Vitt LJ, Congdom JD, Dickson NA (1977) Adaptive strategies and energetics of tail autotomy in lizards. *Ecology* 58:326–337