

Expression of breeding coloration in European Green Lizards (*Lacerta viridis*): variation with morphology and tick infestation

R. Václav, P. Prokop, and V. Fekiač

Abstract: According to the hypothesis of parasite-mediated sexual selection, for a communication system to work reliably, parasites should reduce the showiness of sexual signals of their host. In this study, we examined whether the expression of breeding coloration in free-ranging adult European Green Lizards (*Lacerta viridis* (Laurenti, 1768)) is linked with infestation by their common ectoparasite *Ixodes ricinus* (L., 1758) (Acari: Ixodidae). We found that tick infestation was higher in males than in females. Males showing relatively heavier body for their tail length (predominantly males with regenerated tails) and relatively thinner tail base experienced higher infestation rates. In turn, relatively heavier females for their snout–vent length were less tick infested. Although some components of throat and chest coloration varied significantly with relative tail length, tail-base thickness, body mass, and head size, a measure of male throat and female chest color saturation seemed independent of lizard morphology. After correcting for the effects of morphology on skin coloration and tick load, the saturation of blue throat color in male lizards decreased with increasing level of tick infestation. In contrast, yellow chest color saturation increased with residual tick numbers in females. Considering presumably different signaling functions of male and female lizard coloration, our work suggests that tick infestation might represent a handicap for Green Lizards.

Résumé : Selon l'hypothèse de la sélection sexuelle sous l'influence des parasites, il faut, pour qu'un système de communication fonctionne de façon fiable, que les parasites réduisent le déploiement des signaux sexuels de leur hôte. Dans notre étude, nous vérifions si l'expression de la coloration de reproduction chez des lézards verts d'Europe (*Lacerta viridis* (Laurenti, 1768)) adultes et libres en nature est affectée par l'infestation par l'un de leurs ectoparasites communs, *Ixodes ricinus* (L., 1758) (Acari : Ixodidae). L'infestation par les tiques est plus forte chez les mâles que chez les femelles. Les mâles qui possèdent un corps relativement plus lourd en fonction de leur longueur de queue (en majorité des mâles avec des queues régénérées) et des bases de la queue relativement plus fines souffrent de taux d'infestation plus élevés. En revanche, les femelles plus lourdes en fonction de leur longueur museau–évent ont des infestation moindres de tiques. Bien que certaines composantes de la coloration de la gorge et de la poitrine varient significativement en fonction de la longueur relative de la queue, de l'épaisseur de la base de la queue, de la masse corporelle et de la taille de la tête, les mesures de la saturation de couleur de la gorge des mâles et de la poitrine des femelles semblent indépendantes de la morphologie des lézards. Après avoir tenu compte des effets de la morphologie sur la coloration de la peau et de la charge de tiques, nous observons que la saturation de coloration bleue sur la gorge des mâles décroît en fonction de l'augmentation du niveau d'infestation des tiques. À l'inverse, la saturation de la coloration jaune de la poitrine des femelles augmente en fonction du nombre résiduel de tiques. En présumant que les colorations des lézards mâles et femelles ont probablement des fonctions de signalisation différentes, notre étude indique que les infestations de tiques représentent vraisemblablement un désavantage chez les lézards verts.

[Traduit par la Rédaction]

Introduction

Ornaments were proposed to represent handicaps that may reveal a male's ability to allocate limiting resources to somatic growth, immune defense, and the maintenance of sexual characters (Zahavi 1975; Folstad and Karter 1992). As an extension to the "Red Queen" hypothesis, Hamilton and Zuk (1982) hypothesized that parasites could be one of na-

ture's agents in maintaining the honesty of host ornament expression. Hamilton and Zuk's (1982) hypothesis makes several assumptions: a negative correlation between an individual's fitness and its parasite load, heritable variation in parasite resistance, variation in expression of secondary sexual characters in response to parasite infestation, and the female preference for less parasitized males. There is increasing evidence for the adverse effects of parasitism on

Received 1 May 2007. Accepted 4 September 2007. Published on the NRC Research Press Web site at cjz.nrc.ca on 30 November 2007.

R. Václav.^{1,2} Estación Experimental de Zonas Áridas (Consejo Superior de Investigaciones Científicas), Departamento de Ecología Funcional y Evolutiva, General Segura 1, E-04001 Almería, Spain.

P. Prokop. Institute of Zoology, Slovak Academy of Sciences, Dúbravská cesta 9, SK-84506 Bratislava, Slovakia.

V. Fekiač. Faculty of Natural Sciences, P.J. Šafárik University, Moyzesova 16, SK-041 54 Košice, Slovakia.

¹Corresponding author (e-mail: Radovan.Vaclav@savba.sk).

²Present address: Institute of Zoology, Slovak Academy of Sciences, Dúbravská cesta 9, SK-84506 Bratislava, Slovakia.

host condition (e.g., Booth et al. 1993; Oppliger et al. 1996) and for the heritability of parasite resistance (e.g., von Schantz et al. 1996; see Penn and Potts 1998). However, despite intense scientific enquiry, there is only weak overall evidence that parasites mediate sexual signals of their hosts (see Møller et al. 1999).

For communication most animals use multiple signals (see Candolin 2003), many of which, moreover, consist of several components (Rowe 1999). It is not clear whether multicomponent signals evolved through separate processes or in combination (Andersson 1994; Brooks and Coullidge 1999). Nevertheless, recent studies suggest that different components of a sexual signal may provide information about different aspects of the signaler's phenotypic quality (Badyaev et al. 2001), different components of a signal may be transmitted to different receivers (Brooks and Coullidge 1999), or that receivers use different components of a signal in different situations (Calkins and Burley 2003). Therefore, current knowledge, particularly of color traits, implies that sexual characters should be investigated as multicomponent signals (Grether et al. 2004).

The honesty of sexual signaling, particularly in avian systems, is most often studied in the framework of female mate choice (e.g., Hamilton and Zuk 1982). However, sexual signals that evolved through male–male or female–female competition may even more strongly indicate the overall quality of their bearer because they are constantly tested in combat (Berglund et al. 1996). Lizards represent a group where coloration or other visual cues are typically associated with status signals (Cooper and Greenberg 1992), with dominant individuals achieving higher fitness (see Olsson and Madsen 1998). In contrast to most avian systems, color signals in lizards are used during aggressive displays not only by males but also by females (see Cooper and Greenberg 1992; Hager 2001). Thus, reptiles provide a suitable system to investigate the role of parasites in the expression of breeding coloration.

The aim of this study was to examine whether different components of color signals of two body patches reflect different phenotypic traits and the level of tick parasitism of European Green Lizards (*Lacerta viridis* (Laurenti, 1768)). In this species, the throat and chest achieve vibrating blue and yellow colors during the mating period, with the two patches being displayed during inter- and intra-sexual social interactions via stereotypic head bobbing and “push-ups”. Although coloration, particularly that of the throat, is more conspicuous in male Green Lizards, we examined the coloration of the throat and chest in both sexes. This is because mature female Green Lizards also obtain faint bluish and vivid yellow colors during the breeding season, and they are territorial and display their throat and chest during aggressive encounters with conspecifics.

Materials and methods

Study system and area

The European Green Lizard is a 35–40 cm (total length) long lizard, inhabiting warm and dry areas of central Europe and the Balkans. Green Lizards become sexually mature by the age of 3 years, with their breeding season beginning after hibernation in April and lasting until June when fe-

males lay about 7–20 eggs. Male Green Lizards do not care for offspring, so females only obtain sperm from males. The species belongs to the largest European lacertids and shows a relatively long lifespan (>9 years).

Hard ticks of the genus *Ixodes* Latreille, 1795 (Acari: Ixodidae) are common ectoparasites of reptiles worldwide (Barnard and Durden 2000). Owing to their high density, reptilian species may locally function as the main host species for subadult *Ixodes* ticks (e.g., Casher et al. 2002). The activity of nymphal and larval *Ixodes* ticks usually exhibits two peaks per year, with the first peak occurring in spring months. The peak of *Ixodes* tick parasitism on Green Lizards coincides with the mating phase of the host species, with virtually all Green Lizards being infested with ticks in some areas (R. Václav, unpublished data).

During the spring of 2004, we sampled Green Lizards breeding in two sites within the Slovak Karst National Park, Slovakia. Each of the replicate sites was ~10 ha large, with a distance of 30 km between them. Lizards of both study populations bred in a more or less open habitat on south-facing rocky slopes (~200 to 400 m above sea level) dominated by broad-leaved scrub vegetation (*Quercus petraea* (Mattuschka) Liebl., *Carpinus betulus* L., *Cornus mas* L., *Acer campestre* L., *Crataegus monogyna* Jacq., *Prunus mahaleb* L., *Rosa canina* L.). While the lizards of one of the sites bred relatively homogeneously distributed throughout an open scrubland, the lizards from the other population were concentrated on the rocky outcrops of grassland–woodland ecotones.

Data collection

Sixty-five adult Green Lizards were captured by slip-noosing during two different days, 20 May and 1 June. The captures were always conducted on both sites on each date. Each of the 65 captured lizards was visually sexed according to throat coloration, head shape, the shape of the ventral side of tail base (male tail base has two bulges), and bite marks on the head and belly (males bite the belly of females during copulation; head bite marks usually come from territorial disputes between males). Right after capture, we counted the number of ticks that the lizard carried and measured its snout–vent length (SVL), snout–tail length (STL; from the tip of the snout to the rear of the tail), body mass, head width and height (to estimate head size, a sexually selected trait in some reptiles; Olsson and Madsen 1998), and, only in males, tail-base circumference (TB), i.e., the size of a body part containing the hemipenes (an estimate of reproductive state based on reproductive tissue swelling; see Holmes and Wade 2004). We stored lizards in separate containers and later took colorimetric measurements on their throat and chest. All lizards were released at the point of capture the following morning after their capture. Although tail autotomy was ubiquitous in our population, especially among males, all lizards sampled had their tail either intact or regenerated. Tail length (STL – SVL) was included in the analyses to consider the effect of tail autotomy on body coloration and tick load (see below).

Color measurement and analysis

The skin reflectance of all captured lizards (SVL ± 1 SE: males, 102.9 ± 1.33, $n = 45$; females, 101.36 ± 1.27, $n = 20$)

was measured under standardized conditions with a portable spectrophotometer (USB2000; Ocean Optics, Dunedin, Florida) sensitive to wavelengths between 350 and 700 nm, which uses a deuterium-halogen lamp as a light source (Top Sensor Systems DH-2000, Eerbeek, the Netherlands) and is calibrated to a white and black standard before measuring every individual lizard. We collected 3–4 reflectance measurements on the throat and chest of each lizard (see Figs. 1A, 1B). In total, 451 reflectances were recorded. The reflectance measurements are expressed relative to a Spectralon® white standard. The spectral data for each reading were grouped into 10 nm bins (e.g., Cuthill et al. 1999), finally obtaining 35 data points for every spectral reading. To examine variation in skin reflectances, we used principal component analysis (PCA; see Endler 1990; Cuthill et al. 1999). To examine differences in reflectances between male and female Green Lizards, a single PCA was run for each body patch (i.e., throat and chest) by including reflectances from lizards of both sexes (see Endler 1990). Each PCA extracted three principal components (PCs) with eigenvalues >1, which together explained >94% and >96% of the variations in the spectral reflectances of the chest and throat, respectively. There was a high repeatability in color variables in terms of PC1–PC3 factor scores between measurements of the same body patch within individual lizards (interclass correlation coefficient (ICC) >0.89). Therefore, we averaged the reflectances taken on the same patch for each individual.

Statistical analyses

A head-size index was calculated based on PCA with head width and head height (PCA extracted a single factor explaining 98% of variance, eigenvalue = 1.96). A multiple linear regression with a backward stepwise method was used to determine the predictors of tick infestation rate and lizard coloration. Tick loads were $\log(x + 1)$ -transformed to achieve normality of distribution. Instead of using residuals of body mass after controlling for the effect of body size, we use raw values and multiple regressions (Freckleton 2002). Consequently, in the regression models, we consider the covariation between body mass and SVL as an indication of a body-condition effect, while the covariation between body mass and tail length is considered to arise because of the effect of tail autotomy on body-mass variation. All the variables and relationships were checked for normality and linearity. Also, we tested the assumption that there is no interaction between categorical and continuous predictors with homogeneity-of-slopes GLM. The repeatabilities for reflectance measurements refer to intraclass correlation coefficients. The values presented in the Results are means \pm 1 SE. All tests were calculated with STATISTICA® version 7 (StatSoft Inc. 2004).

Results

We observed the first engorged ticks on Green Lizards in the last week of March. Thereafter, tick infestation increased and remained high throughout May and June. All the ticks collected from Green Lizards were subadult ticks, i.e., larvae and (or) nymphs, with around 90% of cases represented by *I. ricinus* and 10% of the genus *Dermacentor* Koch, 1844.

Ixodes trianguliceps Birula, 1895 and the genus *Haemaphysalis* Koch, 1844 also have been found on lizards. The tick-engorgement sites were around the shoulders, behind and inside the ear, and in the forelimb axillae. *Ixodes trianguliceps* ticks were found on the toes only.

Individual determinants of tick infestation

While controlling for the confounding effects of capture site and a measure of body size (SVL), we found that tick loads were higher on males (raw data: 20.64 ± 2.20 ticks per male, range = 2–64 ticks per male, $n = 45$ males) than females (2.70 ± 0.66 ticks per female, range = 0–8 per female, $n = 20$ females) (GLM — main effect: sex, $F_{[1,61]} = 223.43$, $P < 0.001$; random effect: capture site, $F_{[1,61]} = 5.92$, $P = 0.018$; covariate: SVL, $\beta = 0.05$, $F_{[1,61]} = 0.50$, $P = 0.50$).

Tick burden was greater in those males that were relatively heavier for their tail length and tail-base thickness (Table 1). In turn, relatively heavier females for their SVL carried less ticks than relatively lighter females (Table 1). After removing from analysis three gravid females in an advanced stage of gestation, we obtained a model with the same results (body mass, $\beta = -1.77$, $t_{[14]} = -2.68$, $P = 0.018$; SVL, $\beta = 1.60$, $t_{[14]} = 2.42$, $P = 0.030$).

Individual determinants of coloration

The first principal component (PC1) from a PCA including male and female skin reflectances described achromatic variation (i.e., brightness) for throat and chest colorations; factor loadings at wavelengths across the whole spectrum showed high absolute values (Figs. 2A, 2B). Because all PC1 loadings were negative, a patch's brightness was negatively related to PC1 scores. The second principal components (PC2) showed positive loadings for short wavelengths and negative loadings for long wavelengths (Figs. 2A, 2B). Therefore, greater throat PC2 scores imply more saturated bluish throat colors in males and females or less saturated yellowish throat colors in females, respectively (Figs. 1A, 2A). In turn, greater chest PC2 scores indicate less saturated yellow colors in both males and females (Figs. 1B, 2B). PC3 loadings showed high and positive values for the wavelengths between 350 and 390 nm (Figs. 2A, 2B). Therefore, throat and chest PC3 scores were positively related to UV reflectance.

Green Lizards exhibited significant sexual dimorphism in terms of throat PC1–PC2 scores and chest PC1–PC3 scores (MANCOVA, throat PC1–PC3 as dependent variables, sex and capture site as independent predictors, and SVL as a covariate; whole model: sex, Wilks' $\lambda = 0.16$, $P < 0.001$; capture site, Wilks' $\lambda = 0.95$, $P = 0.371$; SVL, Wilks' $\lambda = 0.84$, $P = 0.015$; univariate results for sex: throat PC1, $F_{[1,61]} = 114.67$, $P < 0.001$ (throat darker in males); throat PC2, $F_{[1,61]} = 14.45$, $P < 0.001$ (more saturated blue in males); throat PC3, $F_{[1,61]} < 0.01$, $P = 0.968$; MANCOVA, chest PC1–PC3; whole model: sex, Wilks' $\lambda = 0.83$, $df = 3$, 59 , $P < 0.001$; capture site, Wilks' $\lambda = 0.90$, $P = 0.095$; SVL, Wilks' $\lambda = 0.84$, $P = 0.016$; univariate results for sex: chest PC1, $F_{[1,61]} = 6.23$, $P = 0.015$ (chest darker in males); chest PC2, $F_{[1,61]} = 9.62$, $P = 0.003$ (more saturated yellow in females); chest PC3, $F_{[1,61]} = 12.51$, $P < 0.001$ (UV reflectance higher in males)).

Fig. 1. Spectral reflectance of the (A) throat and (B) chest patches of male ($n = 45$; ○) and female ($n = 20$; ●) European Green Lizards (*Lacerta viridis*). The curves are median reflectances and error bars denote 95% CI.

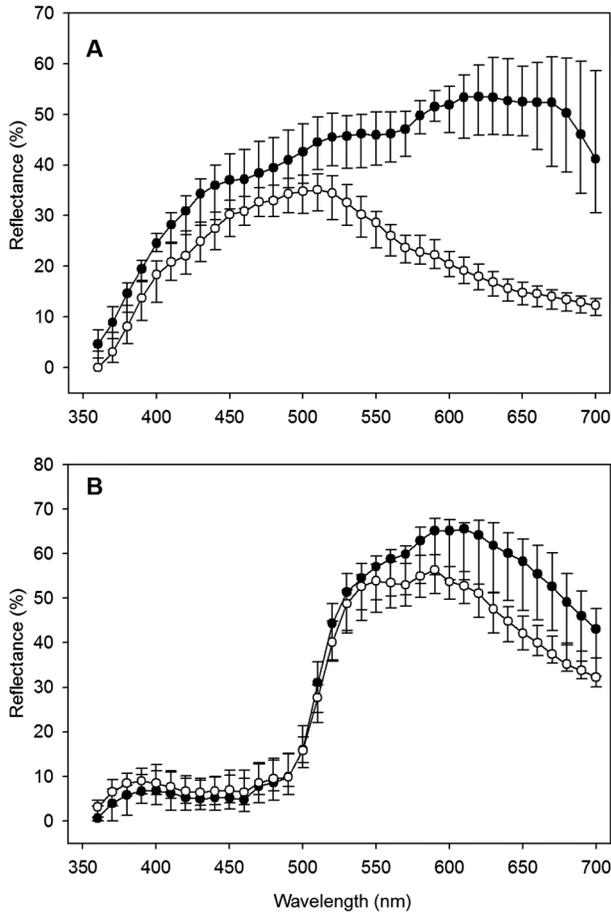


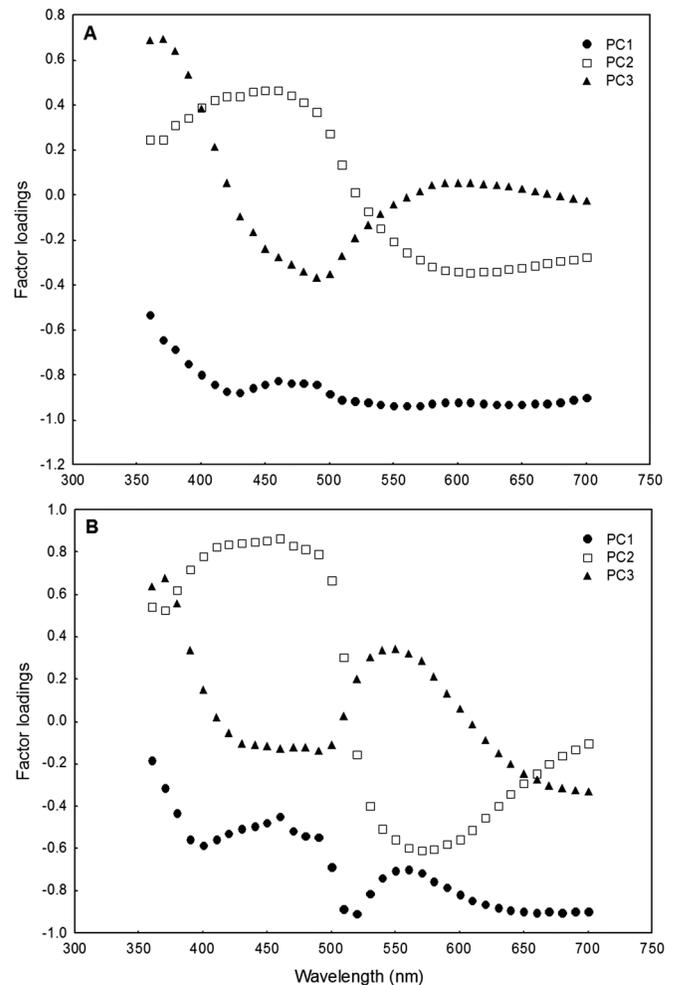
Table 1. Backward stepwise regression on the morphological predictors of tick (*Ixodes ricinus*) parasitism of European Green Lizards (*Lacerta viridis*).

Source of variation	Slope	SE	t	P
Males				
Body mass	1.08	0.33	3.26	0.002
Tail base	-0.90	0.28	-3.25	0.002
Tail length	-0.51	0.18	-2.75	0.009
Trap site			2.80	0.008
Females				
Body mass	-2.58	0.78	-3.28	0.004
SVL	2.24	0.78	2.86	0.011

Note: SVL, snout-vent length.

Because of sexual color dimorphism, for further analyses on color predictors we used separate PCA to compute factor scores for each sex. With this procedure, however, the relationships between factor loadings and wavelengths were reversed for male and female throat PC3 and male chest PC2 compared with the corresponding PCs shown in Fig. 2. Consequently, the interpretation regarding these principal com-

Fig. 2. Relationships between factor loadings and wavelengths for the reflectances of (A) throat and (B) chest patches in European Green Lizards.



ponents is also reversed. The morphological predictors of male and female colorations are summarized in Tables 2 and 3.

Tick infestation and host coloration

After correcting for morphological determinants of tick load and lizard coloration, we found that tick load significantly accounted only for male throat and female chest color saturations ($P > 0.20$ for other color variables for the two patches). In particular, after controlling tick load for body mass, tail-base circumference, tail length, and capture-site and throat color saturation for capture-site differences (see Tables 1, 2), the saturation of male blue throat color decreased with increasing tick numbers (linear regression: $\beta = -0.37$, $SE = 0.14$, $t_{[43]} = -2.62$, $P = 0.012$; Fig. 3A). Regarding females, the saturation of yellow chest color increased with residual tick load ($\beta = -0.63$, $SE = 0.18$, $t_{[18]} = -3.44$, $P = 0.003$; Fig. 3B).

Discussion

We found that relatively heavier males for their tail length and tail-base thickness and relatively lighter females for

Table 2. Backward stepwise regression on the morphological predictors of throat and chest colorations in male European Green Lizards.

Source of variation	Slope	SE	<i>t</i>	<i>P</i>	Relationship
Throat					
PC1					
Body mass	-1.59	0.53	-3.01	0.004	Relatively lighter for SVL, TL, and TB predicts darker throats
Snout-vent length (SVL)	1.21	0.41	2.97	0.005	
Tail length (TL)	0.55	0.19	2.98	0.005	
Tail base (TB)	0.65	0.26	2.51	0.016	
PC2					
Trap site			2.76	0.008	Site difference
PC3					
Body mass	-1.00	0.41	-2.43	0.019	Relatively heavier for SVL predicts higher UV reflectance
SVL	0.77	0.41	1.88	0.068	
Chest					
PC1					
TB	0.26	0.14	1.84	0.073	Thicker TB predicts darker chests
TL	0.35	0.14	2.41	0.020	Greater TL predicts darker chests
PC2					
Trap site			3.79	0.001	Site differences
Body mass	-1.15	0.37	-3.11	0.003	Relatively heavier for SVL predicts less saturated yellow in the chests
SVL	1.07	0.39	2.77	0.008	
PC3					
Trap site			3.47	0.001	Site differences
Body mass	0.91	0.36	2.54	0.015	Relatively heavier for SVL predicts higher UV reflectance
SVL	-0.91	0.37	-2.42	0.020	

Table 3. Backward stepwise regression on the morphological predictors of throat and chest colorations in female European Green Lizards.

Source of variation	Slope	SE	<i>t</i>	<i>P</i>	Relationship
Throat					
PC1					
Body mass	0.49	0.22	2.28	0.036	Relatively heavier for TL predicts darker throats
Tail length (TL)	-0.22	0.22	-0.99	0.334	
PC2					
Head size index	0.76	0.17	4.47	0.001	Larger head predicts more saturated blue in the throats
PC3					
	—	—	—	—	
Chest					
PC1					
Body mass	0.74	0.17	4.46	0.001	Relatively heavier for TL predicts darker chests
TL	-0.37	0.17	-2.23	0.039	
PC2					
	—	—	—	—	
PC3					
Head-size index	1.11	0.50	2.24	0.039	Relatively larger head predicts higher UV reflectance
Snout-vent length	-0.91	0.50	-1.84	0.084	

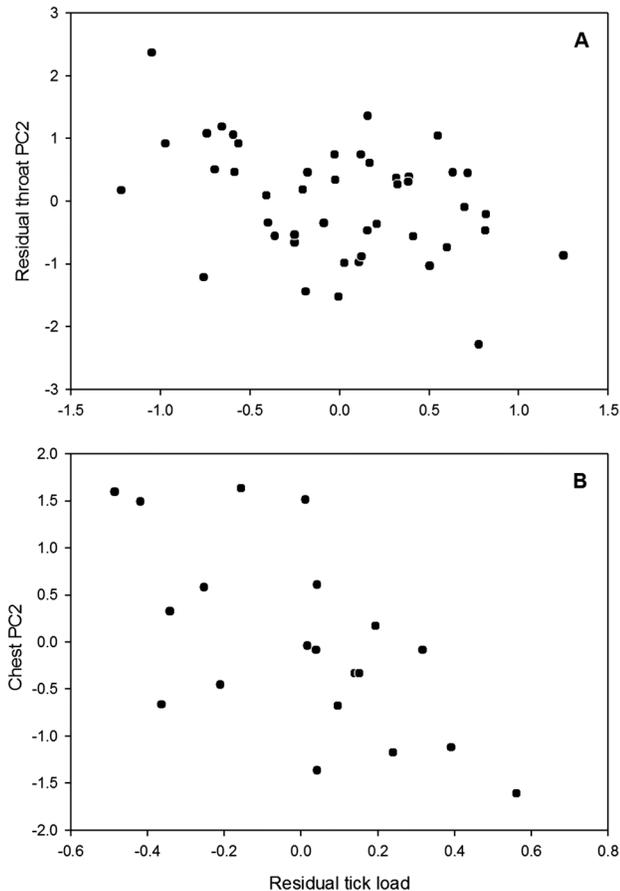
their SVL experienced higher tick infestation levels. While some components of lizard coloration were related to lizard morphology (cf. Salvador and Veiga 2000), a measure of saturation of male throat and female chest color and female throat UV reflectance seemed more flexible with respect to an animal's morphology. After controlling for the morphological determinants of skin coloration and tick infestation intensity, a measure of saturation of blue throat color in males and yellow chest color in females varied significantly with tick infestation intensity.

We detected considerable inter- and intra-sexual variation in tick loads on Green Lizards. A male-biased parasite infestation

rate is common in many vertebrate hosts and is attributed to the effect of male reproductive physiology on behavior and immune system (see Zuk and McKean 1996). The sex-biased tick encounter rate could explain why male lizards carry more ticks than females because male lizards have larger home ranges than females (Perry and Garland 2002).

Tick loads were higher in relatively heavier males for their tail length and tail-base thickness. As relatively heavier males with shorter tails inevitably include males with regenerated tails, it appears that males that experienced tail autotomy at some point in their life and (or) showed regressed

Fig. 3. Relationship between residual tick load intensity and (A) blue throat PC2 scores in male and (B) yellow chest PC2 scores in female Green Lizards. Tick load intensity was corrected for lizard morphology. Male throat PC2 was corrected for trap-site differences; for blue and yellow colors, greater positive PC2 scores mean more and less saturated colors, respectively.



tail base for their body mass suffered higher tick loads. Lower quality male Green Lizards increase their reproductive success by visiting more female territories (Elbing 1999; *Psammodromus algirus* (L., 1758), Salvador et al. 1996a; *Sceloporus virgatus* Smith, 1938, Abell 2000), thereby increasing their exposure to questing parasites. Therefore, it is possible that more intensely tick-infested male lizards recruit from lower quality males that travel longer distances to obtain copulation with a female. A different microhabitat use between lizards with intact and regenerated tails also could account for variation in tick load rates among male lizards. Yet, in *Lacerta monticola* Boulenger, 1905, Martín and Salvador (1992) did not detect a significant change in habitat use during the reproductive season in relation to tail autotomy. Finally, a trade-off between immune defense and tail regeneration could be responsible for higher parasite loads in males that underwent tail autotomy. The only work examining the effect of autotomy on parasite infestation rates, however, did not reveal a different number of blood parasites between tailless and intact Common Lizards (Oppliger and Clobert 1997).

Relatively heavier females for their SVL were less in-

festated by ticks than relatively lighter females. This is in accordance with a positive effect of nutrition on immune system (Chandra 1997) and a suggested positive impact of nutrition on the level of parasitism (Folstad and Karter 1992). Alternatively, even after excluding females in a very advanced stage of gravidity from analysis, it is still possible that the level of parasitism could be related to egg development rather than body condition of females. For example, gravid female lizards reduce locomotory activity and remain close to their refuge (Cooper et al. 1990). Thus, relatively heavier females might have been less parasitized because of their decreased spatial activity and tick encounter rates.

Males with greater tick burden exhibited a less saturated blue color in their throat patch. Because we controlled for the confounding effects of lizard morphology and trap site on coloration and tick loads, it is unlikely that this relationship could be attributed to variation in male phenotypic quality or regional differences in the progress of the mating season. Thus, this result is consistent with Hamilton and Zuk's (1982) hypothesis in that parasites negatively impact the expression of male sexual characters within species.

The breeding coloration of dewlaps of male lizards is frequently elicited by the increment of androgen levels (see Cooper and Greenberg 1992). Salvador et al. (1996b) found in *P. algirus* a link between coloration and parasitism owing to a dual effect of testosterone on ornament expression and immune system; testosterone-implanted males increased their ornament size, as well as their tick load (Salvador et al. 1996b; but see Oppliger et al. 2004). The work by Salvador et al. (1996b), however, did not show whether elevated tick infestation was associated with showier coloration and higher testosterone titers also under natural conditions. This information is essential because testosterone supplementation did not affect male coloration and tick loads in all males (Salvador et al. 1996b, 1997). A recent review suggests that the response of the immune system to changing testosterone levels may be more important than testosterone itself for the elaboration of sexual characters (see Roberts et al. 2004). Indeed, Olsson et al. (2005) found variation in tick load in Sand Lizards (*Lacerta agilis* L., 1758) to be related to genetically based tick resistance and to vary with corticosterone rather than testosterone plasma levels. As ticks are highly abundant parasites of the study species, genetically based parasite resistance might account for low tick loads in male Green Lizards showing a more vivid blue throat color.

To our knowledge, this is the first work that examines the link between parasite load and female host coloration. Tick load significantly contributed to explaining variation in female coloration; females with greater tick burden showed more saturated yellow chest colors. Development of vivid yellow, orange, or red female coloration in lizards is elicited in response to changing hormone levels, particularly progesterone (Medica et al. 1973), and is often closely correlated with aggressive rejection display behavior (see Cooper and Greenberg 1992). Several hypotheses have been proposed to explain female color change, including sex recognition, aggression avoidance, courtship rejection, courtship stimulation, or conditional signaling (see Cooper and Greenberg 1992). Cooper (1986) suggested that the vivid coloration of females serves to reduce agonistic or sexual encounters, thus

decreasing the risk of predation. Given that tick parasitism entails energetic costs (e.g., Dunlap and Mathies 1993), we propose that female Green Lizards with greater tick burdens may produce more vivid chest colors so as to avoid aggression or courtship and thus reduce investment in energy-consuming activities.

We conclude that, when the effect of individual phenotypic quality is controlled for, higher levels of tick infestation are associated with less vivid throat coloration in males, but more vibrating chest coloration in females. While female and male breeding colorations are likely to occur through different mechanisms and signal different information, we propose that the link between tick infestation and breeding coloration reflects the cost of tick infestation and shapes investment in energy-consuming activities. Because tick infestation rates appear to be higher in lower quality males, it remains to be tested whether lower parasite loads and, correspondingly, more vibrant male throat colors are due to higher tick resistance and (or) lower locomotory activity.

Acknowledgements

We thank F. Valera, M. Labuda, and two anonymous reviewers for their invaluable comments on the manuscript. We are indebted to H. Hoi for the loan of a spectrophotometer. We are also indebted to V. Taragelová and J. Křištofík for the determination and quantification of collected tick specimens. P.P. and R.V. were supported by the VEGA grant No. 2/7080/27 of the Slovak Academy of Sciences. R.V. was also supported by grant SB2003-0333 of the Ministry of Education and Science of Spain. The study was conducted under approval from the Ministry of Environment of the Slovak republic (license No. 1430/467/04-5.1). This article is dedicated to the memory of Milan Labuda.

References

- Abell, A.J. 2000. Costs of reproduction in male lizards, *Sceloporus virgatus*. *Oikos*, **88**: 630–640. doi:10.1034/j.1600-0706.2000.880320.x.
- Andersson, M. 1994. Sexual selection. Princeton University Press, Princeton, N.J.
- Badyaev, A.V., Hill, G.E., Dunn, P.O., and Glen, J.C. 2001. Plumage color as a composite trait: developmental and functional integration of sexual ornamentation. *Am. Nat.* **158**: 221–235. doi:10.1086/321325.
- Barnard, S.M., and Durden, L.A. 2000. A veterinary guide to the parasites of reptiles. Vol. 2. Arthropods (excluding mites). Krieger, Malabar, Fla.
- Berglund, A., Bisazza, A., and Pilastro, A. 1996. Armaments and ornaments: an evolutionary explanation of traits of dual utility. *Biol. J. Linn. Soc.* **58**: 385–399. doi:10.1006/bjil.1996.0043.
- Booth, D.T., Clayton, D.H., and Block, B.A. 1993. Experimental demonstration of the energetic cost of parasitism in free-ranging hosts. *Proc. R. Soc. Lond. B Biol. Sci.* **253**: 125–129.
- Brooks, R., and Coullidge, V. 1999. Multiple sexual ornaments coevolve with multiple mating preferences. *Am. Nat.* **154**: 37–45. doi:10.1086/303219.
- Calkins, J.D., and Burley, N.T. 2003. Mate choice for multiple ornaments in the Californian quail, *Callipepla californica*. *Anim. Behav.* **65**: 69–81. doi:10.1006/anbe.2002.2041.
- Candolin, U. 2003. The use of multiple cues in mate choice. *Biol. Rev. (Camb.)*, **78**: 575–595.
- Casher, L.E., Lane, R.S., Barrett, R.H., and Eisen, L. 2002. Relative importance of lizards and mammals as hosts for ixodid ticks in northern California. *Exp. Appl. Acarol.* **26**: 127–143. doi:10.1023/A:1020911306291. PMID:12475082.
- Chandra, R.K. 1997. Nutrition and the immune system: an introduction. *Am. J. Clin. Nutr.* **66**: 460S–463S. PMID:9250133.
- Cooper, W.E., Jr. 1986. Chromatic components of female secondary sexual coloration: influence on social behavior of male keeled earless lizards (*Holbrookia propinqua*). *Copeia*, 1986: 980–986. doi:10.2307/1445295.
- Cooper, W.E., Jr., and Greenberg, N. 1992. Reptilian coloration and behaviour. In *Hormones, brain, and behaviour. Biology of the Reptilia*. Vol. 18, Physiology E. Edited by C. Gans and D. Crews. University of Chicago Press, Chicago. pp. 299–400.
- Cooper, W.E., Jr., Vitt, L.J., Hedges, R., and Huey, R.B. 1990. Locomotor impairment and defense in gravid lizards (*Eumeces laticeps*): behavioral shift in activity may offset costs of reproduction in an active forager. *Behav. Ecol. Sociobiol.* **27**: 153–157.
- Cuthill, I.C., Bennett, A.T.D., Partridge, J.C., and Maier, E.J. 1999. Plumage reflectance and the objective assessment of avian sexual dichromatism. *Am. Nat.* **160**: 183–200.
- Dunlap, K.D., and Mathies, T. 1993. Effects of nymphal ticks and their interaction with malaria on the physiology of male fence lizards. *Copeia*, 1993: 1043–1048.
- Elbing, K. 1999. Variabilität von Sozialverhalten und Fortpflanzungssystem — Freilandbeobachtungen an Smaragdeidechsen (*Lacerta viridis*). *Z. Feldherpetol.* **6**: 135–146.
- Endler, J.A. 1990. On the measurement and classification of colour in studies of animal colour patterns. *Biol. J. Linn. Soc.* **41**: 315–352.
- Folstad, I., and Karter, A.J. 1992. Parasites, bright males, and the immunocompetence handicap. *Am. Nat.* **139**: 603–622. doi:10.1086/285346.
- Freckleton, R.P. 2002. On the misuse of residuals in ecology: regression of residuals vs. multiple regression. *J. Anim. Ecol.* **71**: 542–545. doi:10.1046/j.1365-2656.2002.00618.x.
- Grether, G.F., Kolluru, G.R., and Nersissian, K. 2004. Individual colour patches as multicomponent signals. *Biol. Rev. (Camb.)*, **79**: 583–610.
- Hager, S.B. 2001. The role of nuptial coloration in female *Holbrookia maculata*: evidence for a dual signaling system. *J. Herpetol.* **35**: 624–632. doi:10.2307/1565901.
- Hamilton, W.D., and Zuk, M. 1982. Heritable true fitness and bright birds: a role for parasites? *Science (Washington, D.C.)*, **218**: 384–387. doi:10.1126/science.7123238. PMID:7123238.
- Holmes, M.M., and Wade, J. 2004. Seasonal plasticity in the copulatory neuromuscular system of green anole lizards: a role for testosterone in muscle but not motoneuron morphology. *J. Neurobiol.* **60**: 1–11. doi:10.1002/neu.10334. PMID:15188267.
- Martín, J., and Salvador, A. 1992. Tail loss consequences on habitat use by the Iberian rock lizard, *Lacerta monticola*. *Oikos*, **65**: 328–333. doi:10.2307/3545026.
- Medica, P.A., Turner, F.B., and Smith, D.D. 1973. Hormonal induction of color change in female leopard lizards, *Crotophytus wislizenii*. *Copeia*, 1973: 658–661. doi:10.2307/1443064.
- Møller, A.P., Christe, P., and Lux, E. 1999. Parasitism, host immune function, and sexual selection: a meta-analysis of parasite-mediated sexual selection. *Q. Rev. Biol.* **74**: 3–20. doi:10.1086/392949. PMID:10081812.
- Olsson, M., and Madsen, T. 1998. Sexual selection and sperm competition in reptiles. In *Sperm competition and sexual selection in reptiles*. Edited by T.R. Birkhead and A.P. Møller. Academic Press, London. pp. 503–564.
- Olsson, M., Madsen, T., Wapstra, E., Silverin, B., Ujvari, B., and

- Witzell, H. 2005. MHC, health, color, and reproductive success in sand lizards. *Behav. Ecol. Sociobiol.* **58**: 289–294. doi:10.1007/s00265-005-0921-y.
- Oppliger, A., and Clobert, J. 1997. Reduced tail regeneration in the Common Lizard, *Lacerta vivipara*, parasitized by blood parasites. *Funct. Ecol.* **11**: 652–655.
- Oppliger, A., Célrier, M.L., and Clobert, J. 1996. Physiological and behaviour changes in common lizards parasitized by haemogregarines. *Parasitology*, **113**: 433–438.
- Oppliger, A., Giorgi, M.S., Conelli, A., Nembrini, M., and John-Alder, H.B. 2004. Effect of testosterone on immunocompetence, parasite load, and metabolism in the common wall lizard (*Podarcis muralis*). *Can. J. Zool.* **82**: 1713–1719. doi:10.1139/z04-152.
- Penn, D., and Potts, W.K. 1998. Chemical signals and parasite-mediated sexual selection. *Trends Ecol. Evol.* **13**: 391–396. doi:10.1016/S0169-5347(98)01473-6.
- Perry, G., and Garland, T.G. 2002. Lizard home ranges revisited: effects of sex, body size, diet, habitat, and phylogeny. *Ecology*, **83**: 1870–1885.
- Roberts, M.L., Buchanan, K.L., and Evans, M.R. 2004. Testing the immunocompetence handicap hypothesis: a review of the evidence. *Anim. Behav.* **68**: 227–239. doi:10.1016/j.anbehav.2004.05.001.
- Rowe, C. 1999. Receiver psychology and the evolution of multi-component signals. *Anim. Behav.* **58**: 921–931. doi:10.1006/anbe.1999.1242. PMID:10564594.
- Salvador, A., and Veiga, J.P. 2000. Does testosterone or coloration affect growth rates of adult males of the lizard *Psammmodromus algirus*? *Can. J. Zool.* **78**: 1463–1467. doi:10.1139/cjz-78-8-1463.
- Salvador, A., Martín, J., López, P., and Veiga, J.P. 1996a. Long-term effect of tail loss on home-range size and access to females in male lizards (*Psammmodromus algirus*). *Copeia*, 1996: 208–209. doi:10.2307/1446960.
- Salvador, A., Veiga, J.P., Martín, J., López, P., Abelenda, M., and Puerta, M. 1996b. The cost of producing a sexual signal: testosterone increases the susceptibility of male lizards to ectoparasitic infestation. *Behav. Ecol.* **7**: 145–150. doi:10.1093/beheco/7.2.145.
- Salvador, A., Veiga, J.P., Martín, J., and López, P. 1997. Testosterone supplementation in subordinate small male lizards: consequences for aggressiveness, colour development, and parasite load. *Behav. Ecol.* **8**: 135–139. doi:10.1093/beheco/8.2.135.
- StatSoft Inc. 2004. STATISTICA® (data analysis software system). Version 7 [computer program]. StatSoft Inc., Tulsa, Okla.
- von Schantz, T., Wittzell, H., Göransson, G., Grahn, M., and Persson, K. 1996. MHC genotype and male ornamentation: genetic evidence for the Hamilton-Zuk model. *Proc. R. Soc. Lond. B Biol. Sci.* **263**: 265–271.
- Zahavi, A. 1975. Mate selection — a selection for a handicap. *J. Theor. Biol.* **67**: 603–605.
- Zuk, M., and McKean, K.A. 1996. Sex differences in parasite infections: patterns and processes. *Int. J. Parasitol.* **26**: 1009–1023. doi:10.1016/S0020-7519(96)00086-0. PMID:8982783.