Ethology Ecology & Evolution 18: 275-288, 2006

Experimental and molecular evidence that body size and ventral colour interact to influence male reproductive success in a lizard

J. STAPLEY [1] and J.S. KEOGH

School of Botany and Zoology, The Australian National University, Canberra, ACT 0200, Australia

Received 31 October 2005, accepted 25 August 2006

This study aimed to first investigate the relationship between male ventral colour, body size and ectoparasite load, and then test, using an enclosure experiment, how body size and ventral colour influence male reproductive success in a non-territorial lizard. Individuals of the sexually dimorphic Australian skink, *Pseudemoia entrecasteauxii*, were sampled in the field and male body size and ventral coloration were recorded. The frequency of orange and white ventral colour appeared bimodal; males either had orange or white ventral colour. While larger males were more likely to have orange ventral colour, there was considerable variation and small males with orange ventral colour and large males with white ventral colour were common. The number of mites each male had was positively correlated with snout vent length, weight and head depth corrected for body size and was greater for males with orange ventral colour. Mating trials in outdoor enclosures were used to investigate the separate and combined effects of male ventral colour, body size and male behavior on reproductive success. Behavioural observations revealed that males with orange ventral colour were dominant over males with white ventral colour and, contrary to expectations, male body size was not related to dominance. A total of 32 neonates were genotyped along with their mothers and potential fathers using three polymorphic DNA loci. Large males with orange ventral colour fathered the most neonates. These findings suggest that although males with orange ventral colour were dominant, both body size and ventral colour influenced male reproductive success.

KEY WORDS: sexual selection, status-signalling badge, microsatellites, skink, *Pseudemoia entrecasteauxii*.

---

[1] Corresponding author: Jessica Stapley, School of Botany and Zoology, Australian National University, Canberra, ACT 0200, Australia (E-mail: Jessica.stapley@anu.edu.au).

INTRODUCTION

Sexual selection is one of the most powerful selective agents acting on separate sex organisms (Shuster & Wade 2003). The study of male traits on reproductive success has provided insight into many areas of evolutionary ecology including mating system evolution, speciation, inheritance and quantitative genetics (Andersson 1994, Arnold & Duvall 1994, Kokko & Monaghan 2001, Shuster & Wade 2003). Male traits can influence male reproductive success through male-male competition and/or female mate choice (Andersson 1994, Kokko et al. 2003). Large body size, more elaborate armaments, or bright colours that correlate with fighting ability are under strong intra-sexual selection in a variety of taxa (Hoelzer 1990, Andersson 1994, Emlen 1996, Olsson & Madsen 1998, Forslund 2000, Hagelin 2002). Similarly, female choice favours a variety of male traits that may or may not be indicators of male quality (Kokko et al. 2003). In several species males have multiple secondary sexual traits and different traits can influence male reproductive success by influencing one or both of these processes of sexual selection (Marchetti 1998, Andersson et al. 2002). Here we consider how two male traits influence male reproductive success in a non-territorial lizard using a manipulative mating study. Although lizards represent a highly morphologically diverse group that have been used as model systems with which to address questions central to evolutionary theory for many decades (Cox et al. 2003, Pianka & Vitt 2003), evidence of sexual selection acting on male traits are rare, and no studies have considered how multiple male traits may influence reproductive success.

Male body size is the most common trait to influence reproductive success in lizards, and several studies have demonstrated that large males generally sire more neonates (Abell 1997, Gullberg et al. 1997, Lebas 2001, Calsbeek & Sinervo 2002, Morrison et al. 2002). Other factors such as alternative mating tactics had little direct effect on reproductive success (Zamudio & Sinervo 2000, Morrison et al. 2002, Stapley & Keogh 2005) and no effect of colour badges has been identified (Abell 1997, Lebas 2001). Evidence from field studies such as these are crucial and provide evidence of traits influencing reproductive success, however often multiple male traits can be highly correlated and it is difficult to tease apart the effects of each trait on male reproductive success. Therefore, manipulative enclosure studies, where particular combinations of males with different phenotypes can be used to tease apart the individual and combined effects of different male secondary sexual traits, offer the best opportunity for addressing this issue.

The Australian mountain log skink, *P. entrecasteauxii* is a sexually dimorphic lizard that would be useful for identifying the influence of sexual selection on male morphology and behaviour. During the breeding season some males develop orange coloration that covers their ventral surface, and forms a thin lateral line from behind the forelimb extending down their body (Pengilley 1972). The species occurs at high densities, and adult home ranges overlap extensively suggesting that the species is not territorial (Pengilley 1972). This allows for a clear test of the influence of male phenotypic traits on male reproductive success while controlling for the potentially confounding affects of territory quality or male defence of resources. In addition microsatellite markers useful for paternity testing have been developed for this species (Stapley et al. 2003). The aim of this study was to describe the relationship between male traits and body size in *P. entrecasteauxii* and investigate how they influence male reproductive success determined using genetic markers.

METHODS

Field sampling

Individuals were sampled from Namadgi National Park in the Australian Capital Territory in southeastern Australia during December-January 2001-2002. Lizards were caught by "mealworming" whereby a mealworm is attached to a fishing pole with string and placed in front of the lizard. When the lizard bites the mealworm the pole is gently lifted, with the lizard still attached, and placed over the bucket into which the lizard drops. Lizards were then transferred into individual cloth bags for transport back to the laboratory. Measurements were taken of male snout vent length (SVL) to the nearest mm using a ruler, head length, width and depth to the nearest 0.01 mm using digital callipers, and weight to the nearest 0.01 g using a digital balance. All measures of head size and weight were standardized for body size by using the residuals from a linear regression of each measure on SVL. As ectoparasites can be indicative of a reduction in immuncompetence (Salvador et al. 1996, Veiga et al. 1997), the number of mites (Trombiculidae: *Eutrombicula* sp.) within the evaginations of the skin, known as "mite pockets", were counted under a dissecting microscope to investigate if ectoparasite load co-varied with other male traits. Ventral and lateral colour score was recorded for all adult males (> 40 mm, Pengilley 1972). Only ventral colour was used for further analyses because ventral and lateral colour score were highly correlated ($F_{1,249} = 105.6$, $P < 0.001$, $r = 0.29$). This area of colour also is much larger and, because males inflate their gular region during agonistic encounters (similar to behaviours described in *Lampropholis guichenoti*; Torr & Shine 1996), it suggests that this area of colour is likely to be a stronger signal than the thin lateral line. Colour was identified by visual comparison to a Natural Colour Scheme (NCS) colour chart at normal incidence (full, morning sunlight). A total of 34 different ventral colours were identified. The colours varied from mostly white to strong orange. The frequency of each colour approximated a bimodal distribution with a colour score of 0-1 in the white-cream range and a colour score of 23-24 in the orange range (Fig. 1). Therefore most males could be characterized into those with orange ventral colour (orange males) and those with white ventral colour (white males). This method of colour assessment is biased toward the human visual system and we know from studies of *Anolis* lizards that the lizard visual system varies considerably to that of humans (Loew et al. 2002). Lizards have four colour photoreceptors and can detect light at lower (ultraviolet) wavelengths (Loew et al. 2002). We did consider the possibility that aspects of male colour may be reflected in the ultraviolet range. Spectral measurements of a sub-sample of males failed to identify any reflectance within the UV range (320-400 nm) and the orange colour of the male's ventral surface had a peak reflectance at 550-650 nm (J. Zeil & J. Stapley unpubl. data). We used males from the extreme ends of the colour continuum in our mating system experiments (below) to ensure that these two colour morphs were likely to be perceived as different, regardless of the visual system.

Mating study

The mating system study took place in large (2 m diameter) naturalistic outdoor enclosures that were enclosed within a bird-exclusion fence. Mating takes place in autumn, sperm is stored over winter and females ovulate, fertilize the eggs and gestate through spring and summer, giving birth to 3-6 offspring the following autumn (Pengilley 1972). Mating occurs within four weeks of giving birth, so we placed lizards into the enclosures in December 2001 and made behavioural observations of mating behaviour during January-March 2002 once females began to give birth to neonates that resulted from matings that occurred the previous January-March in the wild. The neonates remained in the enclosures to minimize disturbance and closely replicate field conditions but were removed from enclosures in April and released back into the field. Lizards were housed at two densities to explore the possible influence of density on male reproductive behaviours and male reproductive success. "Low" density enclosures contained 9 adult lizards (5 females and 4 males) and "high" density enclosures contained 18 adult lizards (10 females and 8 males). The low-density enclosure is similar to the natural density of these lizards (J. Stapley unpubl. data). High and low density treatments were replicated 12 times, resulting in 24 enclosures and 324 lizards. We used large initial sample sizes because little is known about annual survivorship or age, so we were uncertain about the number of animals that would survive for the length of the experiment. Lizards remained in these enclosures until January the following year, at which time they were caught and females were relocated to the laboratory to give birth and males were released into the filed at their point of capture. Lizards were provided with ad libitum water and food in the form of crickets and mealworms.

The males in both the low- and high-density enclosures were arranged into matched pairs to test the individual and combined effects of male body size and ventral colour on reproductive success. The low-density enclosure contained a set of 4 males and the high-density enclosure contained two sets of 4 males (i.e. 8 males total). A male set was formed using two small size matched males (one with orange and one with white ventral colour) and two large size matched males (one with orange and one with white ventral colour). Therefore, there were four male phenotypic categories: (1) small, white (SW) (mean SVL = 47.14 ± 0.70); (2) small, orange (SO) (mean SVL = 47.30 ± 0.71); (3) large, white (LW) (mean SVL = 51.82 ± 0.68); (4) large, orange (LO) (mean SVL = 51.85 ± 0.63). Mean male condition (residuals from linear regression of weight on SVL) did not differ between these phenotypic categories ($F_{1,141} = 2.74$, $P = 0.10$).

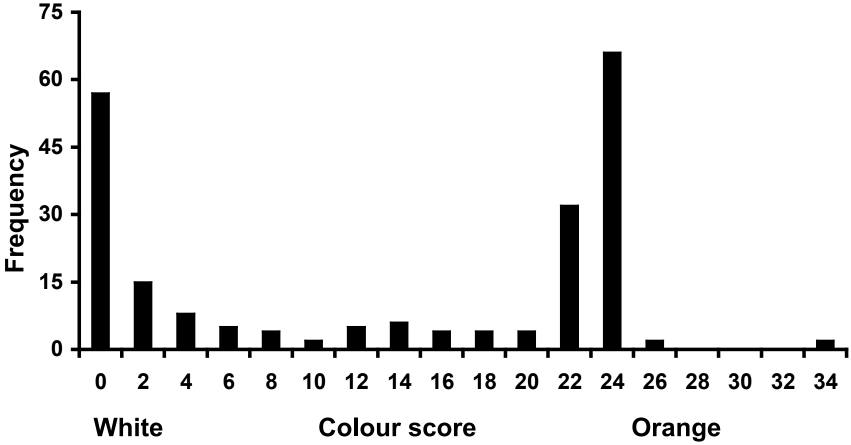

Fig. 1. — Frequency histogram of male ventral colors for each colour score identified from the NCS colour chart.

Behavioural observations

We carried out detailed 20-min observations on each enclosure in a rotation, such that each enclosure was observed 3 times during the breeding season, resulting in a total of 72 observation sessions. During the 20-min observations we recorded all lizards that were active and all interactions between lizards. An overall activity ratio was calculated for each male category. This was calculated by dividing the number of times a male from each phenotypic category was active by the total possible activity score for that phenotypic category. The total possible activity score for each male phenotypic category was the number of males in that category (24 for low density and 48 for high density) times the number of 20-min observations for each enclosure (3). We scored an interaction when lizards were within 2 cm of each other. Interactions between males were classified as aggressive or non-aggressive. The interaction was scored as aggressive when lizard A approached lizard B and lizard B retreated rapidly, or if either lizard displayed any aggressive behaviours such as a slow walk approach with head arched and gular region expanded, biting, wrestling or chasing (the sequence of aggression is very similar to behaviours described in *Lampropholis guichenoti*; TORR & SHINE 1994). Following an initial aggressive encounter, biting and wrestling were rarely observed and most encounters lasted less then 2 sec. For this reason we did not record the level or duration of an aggressive interaction but simply scored an interaction as either "non-aggressive" (lizards next to each other with no obvious interaction) or "aggressive" (any one of the aggressive displays noted above with one lizard running away). We calculated an overall aggressive behaviour score (ABS) for each male category in each enclosure. This was calculated as the total number of "aggressive" interactions a male made minus the total number of "aggressive" interactions that the male received (CARPENTER 1995).

Interactions between males and females were recorded and the initiator was noted. When a lizard approached another of the opposite sex their behaviour was scored as an "association" if no tail twitch was observed, a "courtship" if the male performed a tail twitch as he approached, which is a species typical courtship behaviour, and a "bite" if the male attempted a mating grasp one the female (TORR & SHINE 1994). We did not record female behaviour in this study because there were few aggressive behaviours between females, or between females and males and because the female's response to male courtship was almost invariably the rejection behaviour (one acceptance i.e. copulation was observed). From observations in captivity on 40 females paired with males in which mating was observed, females reject males with a tail wave, when a female accepted a male's courtship approach there was no tail wave and the male would proceed to hold her in a mating grasp for up to 45 min before mating (J. STAPLEY unpubl. data). Female acceptance/rejection behaviour was obvious and acceptance always results in a mating. As only one mating was observed during this study there was virtually no variation in female behaviour and it could not be analyzed. It is possible that females perform subtle behaviours towards males that entice them to court more however, it was not possible to quantify these subtle responses in these semi-natural conditions.

Paternity analysis

Females were caught in January 2003 and housed individually in plastic containers (420 mmL × 320 mmW × 220 mmH) in a temperature-controlled room (18-20 °C) with a natural light cycle. Paper was provided as bedding and cardboard rolls provided refuge. A temperature gradient was established by placing heating tape under one half of the container to allow animals to thermoregulate freely. Animals were provided ad libitum water and food. Females were checked daily until parturition. On their day of birth, neonates were weighed, sexed by everting the hemipenes and a small tissue sample (tail tip) was taken for DNA analysis. Mothers and neonates were released at the mother's point of capture 1-2 weeks after parturition. All mothers, neonates and males from the female's enclosure were genotyped for three microsatellite loci (Pe31, Pe134, Pe242) (STAPLEY et al. 2003). Paternity was assigned using the CERVUS program (MARSHALL et al. 1998). A conservative approach was taken to paternity assignment, sires were only assigned paternity if there were no genetic mismatches and if only a single father was assigned to each neonate. The likelihood estimates were not used as these can lead to erroneous conclusions (Jones & Ardren 2003).

Data analysis

The relationship between male ventral colour (presence/absence) and other male traits was analyzed with a logistic regression. The relationship between the number of mites and other male traits was analyzed with a generalized linear regression with a Poisson model because the data was count data (Quinn & Keough 2002). Male traits and male ABS in high- and low-density enclosures was analyzed with an ANOVA. Comparisons of the activity index, the total number of associations, total courtship interactions and number of neonate was compared between each of the four male phenotypic categories and across the two densities using a 2-Way Contingency table (Quinn & Keough 2002). Data was analyzed with Genstat Release 5.0 and R 2.0.1 (Ihaka & Gentleman 1996). Post hoc power calculations were calculated for non-significant results using G*Power. Power was estimated for a small effect size of 0.15 (15% variation) (Cohen 1988). This was chosen as it provides a conservative estimate of power and is comparable to the average effect size of evolutionary and ecological studies as calculated from 44 meta-analyses (Møller & Jennions 2002).

RESULTS

Field sampling

A total of 245 mature males were sampled from the field in 2001-2002. Males with orange ventral colour were slightly larger than males with white ventral colour (mean SVL ± standard error (SE) orange = 49.79 ± 0.34 and white = 47.79 ± 0.44, $F_{1,245} = 6.54$, $P = 0.013$). Condition and head size did not differ between white and orange males (condition: $F_{1,245} = 0.01$, $P = 0.88$; head width: $F_{1,245} = 0.93$, $P = 0.33$; head depth: $F_{1,245} = 1.00$, $P = 0.31$, head length: $F_{1,245} = 2.74$, $P = 0.09$: Power = 1.0). The number of mites was positively correlated with male body size, male condition, head depth and colour (Fig. 2. SVL: $F_{1,245} = 475.92$, $P < 0.001$; condition: $F_{1,245} = 117.31$, $P < 0.001$; head depth: $F_{1,245} = 29.40$, $P < 0.001$) and greater for orange males (mean number of mites ± SE orange = 11.62 ± 0.65 and white = 10.29 ± 0.64, $F_{1,245} = 8.88$, $P = 0.003$). One observation had large residuals (i.e. one lizard had 99 mites), but removing this observation from the analysis did not alter the main findings and so it has been included in the final analysis.

Mating study

Males from each of the four categories were equally likely to be active during the 20-min observations in the high- and low-density enclosures ($\chi^2_1 = 0.03$, df = 1, $P = 0.99$, Power = 0.32) and this is also true when the data was pooled across the two densities ($\chi^2_1 = 0.04$, df = 1, $P = 0.82$, Power = 0.42). The ABS did not differ between high- and low-density enclosures but differed between males of each phenotypic category (density: $F_{1,44} = 0.77$, $P = 0.384$; phenotype: $F_{3,44} = 4.27$, $P = 0.04$. Fig. 3). When males were grouped according to body size, mean ABS did not differ between large and small males ($F_{1,44} = 1.04$, $P = 0.313$, Power = 0.71), but was greater in males that had orange ventral color ($F_{1,44} = 6.66$, $P = 0.013$). The most aggressive interactions were initiated by large males with orange ventral colour (LO) and directed towards large males with white venters (LW) (Table 1). In the high-density enclosures there was the potential for interactions between males of the same size and badge status, however these were rare (Table 1). This suggests that animals of similar phenotype did not engage in more aggressive interactions.

The most numerous interactions observed between males and females were associations and courtship interactions. Attempted mating grasps (bites) by males were observed 9 times and were not formally analyzed. Males from each of the four phenotypic categories were observed associating with, and courting females (Table 2) at similar frequencies in the low-density (association: $\chi^2_3 = 3.84$, $P = 0.21$, Power = 0.79; courtship: $\chi^2_3 = 6.73$, $P = 0.08$, Power = 0.47) and high-density (association: $\chi^2_3 = 3.83$, $P = 0.22$, Power = 0.73; courtship: $\chi^2_3 = 5.58$, $P = 0.13$, Power = 0.44) enclosures.

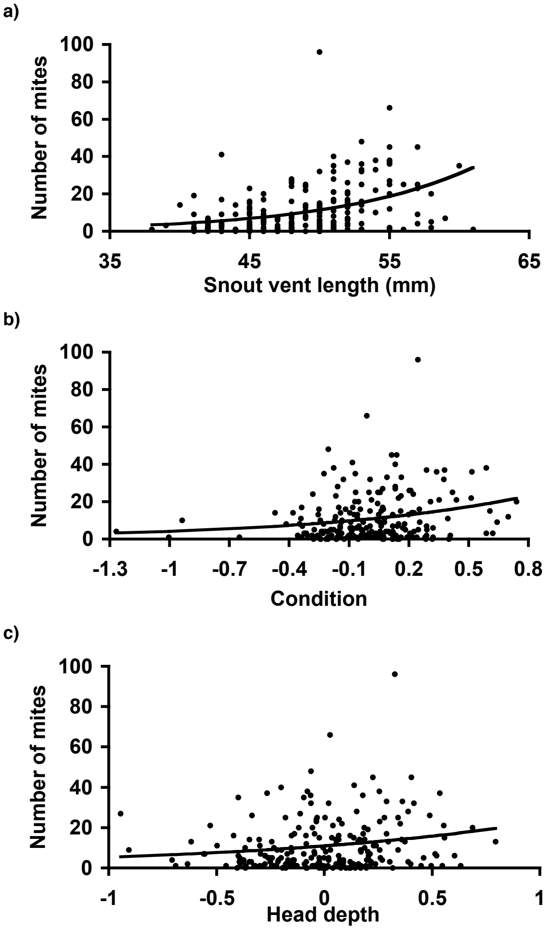

Fig. 2. — Observed (circles) and fitted (line) relationship between the number of mites and a) snout vent length (SVL), b) condition (the residuals from a regression of weight on SVL), c) residuals from a regression of head depth on SVL.

Table 1.

Frequency of non-aggressive (NA) and aggressive (A) interactions within high- and low-density enclosures, between males of each of the four phenotypic categories; small with white ventral colour (SW), small with orange ventral colour (SO), large with white ventral colour (LW) and large with orange ventral colour (LO).

Initiator	Receiver	High		Low		Total	
		NA	A	NA	A	NA	A
SW	SW	0	1	—	—	1	0
SO	SO	0	2	—	—	0	2
LW	LW	0	0	—	—	0	0
LO	LO	0	2	—	—	0	2
SW	SO	0	1	1	0	1	1
SW	LW	4	5	2	1	6	6
SW	LO	0	1	3	2	3	3
SO	SW	1	2	0	1	1	3
SO	LW	1	3	0	3	1	6
SO	LO	1	1	0	0	1	1
LW	SW	0	5	1	2	0	7
LW	SO	4	4	0	4	4	4
LW	LO	2	2	3	1	5	3
LO	SW	0	9	0	3	0	12
LO	SO	0	5	2	2	2	7
LO	LW	0	6	1	10	1	16

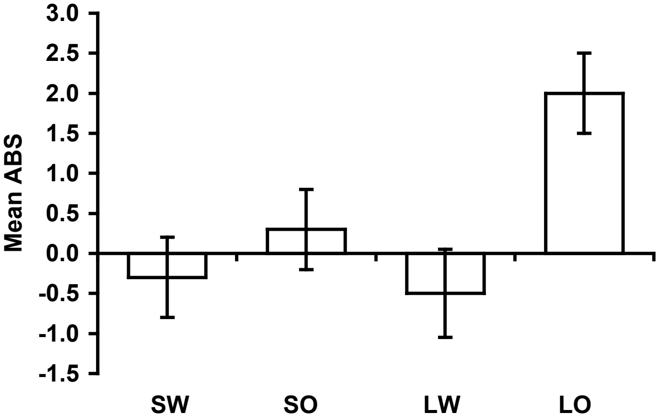

Fig. 3. — Mean aggressive behavior score (± standard errors of difference (SED)) of males from each of the four phenotypic categories. The data has been pooled for high and low-density enclosures.

Table 2.

Number of female-male associations and number of courtships observed for each male phenotypic category (SW – small with white venter, SO – small with white venter, LW – large with orange venter, LO – large with orange venter) in high- and low-density enclosures.

	Associations		Courtship	
	High	Low	High	Low
SW	12	10	6	2
SO	20	13	7	8
LW	12	13	3	5
LO	10	21	10	9

A total of 33% (107) of the lizards survived over winter until December (summer) 2003 when the females were relocated to the laboratory to give birth. Annual survival estimates for other lizards vary from 5% to 80% (Andrews & Nichols 1990, Díaz 1993) but no data is available for wild populations of our species. Survival was random with respect to sex (χ^2_1 = 0.00, df = 1, P = 0.990, Power = 0.64) and male phenotypic category (χ^2_3 = 0.06, df = 3, P = 0.804, Power = 0.53). Although the power of the paternity analysis was reduced the lizards used in the paternity analysis are likely to provide a random subset of the animals used in the experiment. Most deaths occurred during winter and as such it was unlikely to effect the behavioural observations during autumn in the previous year.

A total of 32 neonates (15 female, 17 male) were born in 2003 from 14 mothers and paternity was assigned to all. Three clutches had two sires, which resulted in 27% multiple paternity, excluding 3 clutches that had a clutch size of 1. Because of the low sample sizes the paternity data was pooled across density treatments, as such density was not analyzed. The number of neonates sired by each male phenotype differed (χ^2_3 = 21.75, $P < 0.001$. Fig. 4). Based on ventral colour alone, the number of neonates sired by males with orange ventral colour was greater then for males with white (χ^2_1 = 12.5, $P < 0.001$). Based on body size alone, larger males sired more neonates (χ^2_1 = 6.12, P = 0.013). There was no evidence of size-assortative mating ($F_{1,16}$ = 0.23, P = 0.63, Power = 0.30). No measured female or male traits influenced neonate weight (clutch size: $F_{1,11}$ = 2.71, P = 0.12; mother SVL: $F_{1,11}$ = 4.12, P = 0.06; neonate sex: $F_{1,11}$ = 0.07, P = 0.78; father SVL: $F_{1,11}$ = 0.17, P = 0.68; father colour: $F_{1,11}$ = 0.66, P = 0.43, Power = 0.48) or neonate sex (clutch size: $F_{1,11}$ = 2.71, P = 0.12; neonate weight: $F_{1,11}$ = 0.01, P = 0.92; mother SVL: $F_{1,11}$ = 0.20, P = 0.66; father SVL: $F_{1,11}$ = 0.34, P = 0.54; father color: $F_{1,11}$ = 0.00, P = 0.92, Power = 0.48).

DISCUSSION

The population sampling revealed that the frequency of males in the population with orange ventral colour was bimodal, thus males were categorized according to their ventral colour and males belonging to these two categories were used in the mating trials. Males with orange ventral colour were on average larger than their white counterparts but this difference was small (1.5 mm) in relation to population variation in adult male body size (38-61 mm), and both large and small individuals with orange ventral colour were present in the population. Male mite load was positively related to his body size, condition, head depth and was slightly larger in males with orange venters. The enclosure experiment revealed that large, males with orange ventral colour were more aggressive and had a higher reproductive success compared to males in the other three phenotypic categories. Although the findings of the paternity tests are limited by low survival, it does provide empirical evidence to demonstrate that the combined effects of male traits, i.e. ventral colour and body size influenced reproductive success in this non-territorial lizard. Male reproductive success was not related to male-female associations or male courtship rates as measured relatively crudely in this experiment and this is consistent with the findings of other field studies that use molecular techniques to measure paternity (Hughes 1998, Double & Cockburn 2000, Lewis et al. 2000, Stapley & Keogh 2005).

Males with orange ventral colour were more aggressive than males with white, but large males were not more aggressive than small males. Male aggression rarely escalated to actual fights; instead subordinate males were submissive towards dominant males and retreated on their approach. These observations are consistent with a dominance hierarchy (Boyd & Silk 1983). The recognition of hierarchies and subsequent reduced aggression helps to reduce the potential costs of aggressive encounters and appears to be widespread among vertebrates (Bradbury & Vehrencamp 1998), including lizards (Whiting 1999, López & Martín 2001). Theory predicts that contests between males of a similar size or coloration should be longer or more escalated (Maynard Smith & Parker 1976). However, there were very few interactions observed between males of the same phenotypic category in this study. A similar result was found in tree lizards, where contests never escalated between males of the same colour morph (Thompson & Stewart 1994). Lizards were placed into enclosures in mid-December and observations did not begin until January, so it is possible that contests between males of a similar phenotypic category occurred before observations were made. As dominance is correlated with male ventral colour, the orange ventral colour

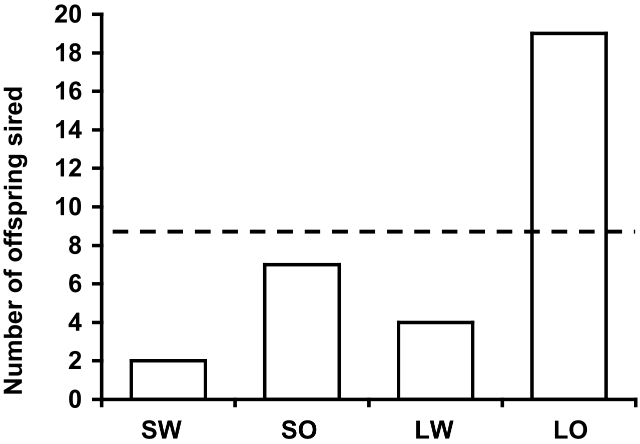

Fig. 4. — Total number of neonates sired by males from each of the four phenotypic categories. The data has been pooled for high- and low-density enclosures. The dashed line indicates the predicted reproductive success if it was random with respect to male phenotype.

of *P. entrecasteauxii* may provide a reliable cue to aggression and dominance status. Increased plasma testosterone has been found to influence aggression and development of colour badges in lizards (SALVADOR et al. 1996, VEIGA et al. 1997) suggesting a possible physiological link between aggression and colour. Increased plasma testosterone of males with orange venters is further supported by the finding that these males had a greater ectoparasite load, which may be indicative of a reduction in immunocompetence (SALVADOR et al. 1996, VEIGA et al. 1997). Mite load was also positively correlated with male size, condition and head depth.

We found no differences in dominance hierarchy or male-female interactions between high- and low-density enclosures. Population density has been identified as one of the major ecological variables influencing the structure of mating systems (for review see BIRKHEAD & MØLLER 1998) and female and male mating behaviours (CROWLEY et al. 1991, CADE & CADE 1992, JIROTKUL 1999). In none of these previous studies did a dominance hierarchy characterize the social system and it is possible that dominance hierarchies are relatively robust to changes in density. The fact that no differences in male reproductive success were detected between the two densities may simply be the result of low power. However, several field studies also have failed to identify density effects on male reproductive success or levels of multiple paternity (DUNN et al. 1994, WEATHERHEAD & BOAG 1997, GRIFFITH et al. 1999, JONES et al. 2001).

Male ventral colour affected male dominance, but the combined effects of body size and ventral colour influenced male reproductive success. This would suggest that there is a male body size advantage to paternity that is not related to male contest or dominance. This advantage may come from pre- or post-zygotic paternity bias. If females preferred larger males then they could bias paternity by either selecting to mate with larger males more often or at peak fertility, or by selecting larger male sperm (KOKKO et al. 2003). Studies testing female mating bias are needed to confirm this hypothesis. The results of this study suggest that male traits may interact in somewhat unpredictable ways to influence male reproductive success.

ACKNOWLEDGEMENTS

We would like to thank Chris Hayes for valuable assistance in the genetics lab. Thanks also go to all the members of the Keogh Lab and Stuart Dennis who helped in the construction of the outdoor enclosures. Funding was provided by grants from the ASIH Gaige Fund, the Ecological Society of Australia, Australian Geographic Society and the Australian Federation of University Women to Jessica Stapley and by the Australian Research Council to J. Scott Keogh. The research described in this paper was approved by the Animal Experimentation and Ethics Committee of the Australian National University (Protocol number: F.BTZ.17.00).

REFERENCES

ABELL A.J. 1997. Estimating paternity with spatial behaviour and DNA fingerprinting in the striped plateau lizard, *Sceloperus virgatus* (Phrynsomatidae). *Behavioral Ecology and Sociobiology* 41: 217-226.

ANDERSSON M. 1994. Sexual selection. *Princeton. Princeton University Press*.

ANDERSSON S., PRYKE S.R., ÖRNBORG J., LAWES M.J. & ANDERSSON M. 2002. Mulitple recievers, multiple ornaments, and a trade-off between agonistic and epigamic signalling in a widowbird. *The American Naturalist* 160: 683-691.

Andrews R.M. & Nichols J.D. 1990. Temporal and spatial variation in survival rates of the tropical lizard *Anolis limifrons*. *Oikos* 57: 215-221.

Arnold S.J. & Duvall D. 1994. Animal mating systems — a synthesis based on selection theory. *The American Naturalist* 143: 317-348.

Birkhead T.R. & Møller A.P. 1998. Sperm competition and sexual selection. *Cambridge: Academic Press*.

Boyd R. & Silk J.B. 1983. A method for assessing cardinal dominance ranks. *Animal Behaviour* 31: 45-58.

Bradbury J.W. & Vehrencamp S.L. 1998. Principles of animal communication. *Sunderland: Sinauer Associated Inc*.

Cade W.H. & Cade E.S. 1992. Male mating success, calling and searching behaviour at high and low densities in the field cricket, *Gryllus integer*. *Animal Behaviour* 43: 49-56.

Calsbeek R. & Sinervo B. 2002. Uncoupling direct and indirect components of female choice in the wild. *Proceedings of the National Academy of Sciences of the United States of America* 99: 14897-14902.

Carpenter G.C. 1995. Modeling dominance: the influence of size, colouration, and experience on dominance in tree lizards (*Urosaurus ornatus). Herpetolgical Monographs* 9: 88-101.

Cohen J. 1988. Statistical power analysis for the behavioural sciences, 2nd ed. *New Jersey: Lawerance Erlbaum*.

Cox R.M., Skelly D.K. & John-Alder H.B. 2003. A comparative test of adaptive hypothesis for sexual size dimorphism in lizards. *Evolution* 57: 1653-1669.

Crowley P.H., Travers S.E., Linton M.C., Cohn S.L., Sih A. & Sargent R.C. 1991. Mate density, predation risk, and the seasonal sequence of mate choice: a dymanic game. *The American Naturalist* 137: 567-596.

Díaz J.A. 1993. Breeding coloration, mating opportunities, activity, and survival in the lacertid lizard *Psammodromus algirus*. *Canadian Journal of Zoology* 71: 1104-1110.

Double M. & Cockburn A. 2000. Pre-dawn infidelity: females control extra-pair mating in superb fairy-wrens. *Proceedings of the Royal Society of London (B, Biological Sciences)* 267: 465-470.

Dunn P.O., Whittingham L.A., Litfeld J.T., Robertson R.J. & Boag P.T. 1994. Effects of breeding density, synchrony and experience on extra-pair paternity in tree swallows. *Behavioral Ecology* 5: 124-129.

Emlen D.J. 1996. Artificial selection on horn lenght-body size allometry in the horned beetle, *Onthophagus acuminatus* (Coleoptera: Scarabaeidae). *Evolution* 50: 1219-1230.

Forslund P. 2000. Male-male competition and large size advantage in the European earwigs, *Forficula auricularia. Animal Behaviour* 59: 753-762.

Griffith S.C., Stewart I.R.K., Dawson D.A., Owens I.P.F. & Burke T. 1999. Contrasting levels of extra-pair paternity in mainland and island populations of the house sparrows (*Passer domesticus*): is there an 'island effect'. *Biological Journal of the Linnean Society* 68: 303-316.

Gullberg A., Olsson M. & Tegelstrom H. 1997. Male mating success, reproductive success and multiple paternity in a natural population of sand lizards — behavioural and molecular genetics data. *Molecular Ecology* 6: 105-112.

Hagelin J.C. 2002. The kinds of triats involved in male-male competition: a comparision of plumage behavior, and body size in quail. *Behavioral Ecology* 13: 32-41.

Hoelzer G.A. 1990. Male-male competition and female choice in the Cortez damselfish, *Stegastes rectifraenum*. *Animal Behaviour* 40: 339-349.

Hughes C. 1998. Integrating molecular techniques with field methods in studies of social behaviour: A revolution results. *Ecology* 79: 383-399.

Ihaka R. & Gentleman R. 1996. R: A language for data analysis and graphics. *Journal of Computational and Graphical Statistics* 5: 299-314.

Jirotkul M. 1999. Population density influences male-male competition in guppies. *Animal Behaviour* 58: 1169-1175.

Jones A.G. & Ardren A.R. 2003. Methods for parentage analysis in natural populations (Invited Review). *Molecular Ecology* 12.

Jones A.G., Walker D., Lindstrom K., Kvarnemo C. & Avise J.C. 2001. Surprising similarity of sneaking rates and genetic mating patterns in two populations of sand goby experiencing disparate sexual selection regimes. *Molecular Ecology* 10: 461-469.

Kokko H., Brooks R., Jennions M.D. & Morley J. 2003. The evolution of mate choice and mating biases. *Proceedings of the Royal Society of London (B, Biological Sciences)* 270: 653-664.

Kokko H. & Monaghan P. 2001. Predicting the direction of sexual selection. *Ecology Letters* 4: 159-165.

Lebas N.R. 2001. Microsatellite determination of male reproductive success in a natural population of the territorial ornate dragon lizard, *Ctenophorus ornatus*. *Molecular Ecology* 10: 193-203.

Lewis A.R., Tirado G. & Sepulveda J. 2000. Body size and paternity in a teiid lizard (*Ameiva exsul*). *Journal of Herpetology* 34: 110-120.

Loew E.R., Fleishman L.J., Foster R.G. & Provencio I. 2002. Visual pigments and oil droplets in diurnal lizards: a comparative study of Caribbean anoles. *Journal of Experimental Biology* 205: 927-938.

López P. & Martín J. 2001. Fighting rules and rival recognition reduce costs of aggression in male lizards, *Podarcis hispanica*. *Behavioral Ecology and Sociobiology* 49: 111-116.

Marchetti K. 1998. The evolution of multiple male traits in the yellow-browed leaf warbler. *Animal Behaviour* 55: 361-376.

Marshall T.C., Slate J., Kruuk L.E.B. & Pemberton J.M. 1998. Statistical confidence for likelihood-based paternity inference in natural populations. *Molecular Ecology* 7: 639-655.

Maynard Smith J. & Parker G.A. 1976. The logic of asymmetrical contests. *Animal Behaviour* 32: 564-578.

Møller A.P. & Jennions M.D. 2002. How much variation can be explained by ecologists and evolutionary biologists? *Oecologia* 132: 492-500.

Morrison S., Keogh J.S. & Scott I.A.W. 2002. Molecular determination of paternity in a natural population of the multiply mating polygynous lizard *Eulamprus heatwolei*. *Molecular Ecology* 11: 535-545.

Olsson M. & Madsen T. 1998. Sexual selection and sperm competition in reptiles, pp. 503-564. In: Birkhead T.R. & Møller. A.P., Edits. Sperm competition and sexual selection. *Cambridge: Academic Press*.

Pengilley R. 1972. Systematic relationships and ecology of some lizards from Southeastern Australia. *PhD Dissertation*, *Department of Zoology*, *Australian National University, Canberra*.

Pianka E.R. & Vitt L.J. 2003. Lizards: windows to the evolution of diversity. *Berkeley: University of California Press*.

Quinn G.P. & Keough M.J. 2002. Experimental design and data analysis for biologists. *Cambridge: Cambridge University Press*.

Salvador A., Veiga J.P., Martín J., López P., Abelenda M. & Puerta M. 1996. The cost of producing a sexual signal — testosterone increases the susceptibility of male lizards to ectoparasitic infestation. *Behavioral Ecology* 7: 145-150.

Shuster S.M. & Wade M.J. 2003. Mating systems and strategies. *New Jersey: Princeton University Press*.

Stapley J., Hayes C.M. & Keogh J.S. 2003. Population genetic differentiation and multiple paternity determined by novel microsatellite markers from the Mountain Log Skink (*Pseudemoia entrecasteauxii*). *Molecular Ecology Notes* 3: 291-293.

Stapley J. & Keogh J.S. 2005. Behavioural syndromes influence mating systems: floater pairs of a lizard have heavier offspring. *Behavioral Ecology* 16: 514-520.

Thompson M.B. & Stewart J.R. 1994. Egg and clutch size of the viviparous Australian skink, *Pseudemoia pagenstecheri* and the identity of species with type ii allantoplacentae. *Journal of Herpetology* 28: 519-521.

Torr G.A. & Shine R. 1994. An ethogram for the small lizard *Lampropholis guichenoti*. *Amphibia-Reptilia* 15: 21-34.

Torr G.A. & Shine R. 1996 Patterns of dominance in the small scincid lizard *Lampropholis guichenoti*. *Journal of Herpetology* 30: 230-237.

Veiga J.P., Salvador A., Martín J. & López P. 1997. Testosterone stress does not increase asymmetry of a hormonally mediated sexual ornament in a lizard. *Behavioral Ecology and Sociobiology* 41: 171-176.

Weatherhead P.J. & Boag P.T. 1997. Genetic estimates of annual and lifetime reproductive success in male red-winged blackbirds. *Ecology* 78: 884-896.

Whiting M.J. 1999. When to be neighbourly: differential agonistic responses in the lizard *Platysaurus broadleyi*. *Behavioral Ecology and Sociobiology* 46: 210-214.

Zamudio K.R. & Sinervo E. 2000. Polygyny, mate-guarding, and posthumous fertilization as alternative male mating strategies. *Proceedings of the National Academy of Sciences of the United States of America* 97: 14427-14432.