



Display behaviour and dewlap colour as predictors of contest success in brown anoles

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Many animals display visual signals in male contests for access to females and territories. These visual signals can be multimodal and stimulate different aspects of a signal receiver's visual system. Over two summers, we tested whether aspects of behaviour and dewlap colour might function as signals that predict contest success when males compete for access to either mates or territories in male brown anole lizards. We found that behaviour (PC1, a correlated composite of head-bob, push-up, and dewlap extension frequency) and an aspect of dewlap colour (PC3, the relative amounts of ultraviolet, yellow, orange, and red of the dewlap margin) were retained in the minimum adequate model predicting contest success across years and social contexts. Winners showed significant differences in behaviour (winners displayed more) and dewlap margin PC3 (winners had lower PC3 scores) compared to contest losers. These findings suggest that display behaviour and dewlap colour might serve as signals indicating a male's ability to win contests for access to females and territories. © 2014 The Linnean Society of London, *Biological Journal of the Linnean Society*, 2014, **111**, 646–655.

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INTRODUCTION

Male competition and male contests are important aspects of sexual selection (Darwin, 1871). Displays of conspicuous traits during such contests can directly and indirectly influence male reproductive success because winners establish territories and obtain access to food as well as mates (Andersson, 1994).

Male traits that are displayed in contests can be multimodal and stimulate different aspects of a signal receiver's visual system (Partan & Marler, 2006). For example, male traits can be postural, and the postural signal is perceived by a signal receiver because motion-sensitive nerve cells are temporally stimulated by movement (Fleishman, 1986; Fleishman, Marshall & Hertz, 1995). This mode of signal communication is common in birds (Searcy & Nowicki, 2005) and lizards (Carpenter, 1967, 1978; Jenssen,

1977; Fleishman, 1992; Stapley & Whiting, 2005; Sacchi *et al.*, 2009). Postural displays are known to enhance contest and/or reproductive success in many bird and lizard species.

Displayed traits can also be colourful and function by stimulating a signal receiver's photoreceptor cones (Fleishman *et al.*, 1998; Fleishman & Persons, 2001). These colourful traits can be shown to others in a variety of social contexts, including agonistic encounters, during which the traits function as an armament (Berglund, Bisazza & Pilastro, 1996). Armament traits in lizards are frequently combined with postural displays in ways that influence contest success (Hover, 1985; Thompson & Moore, 1991a, b; Sinervo & Liveley, 1996; Stapley & Whiting, 2005; Lappin *et al.*, 2006; Sacchi *et al.*, 2009).

Colourful traits, including those used as armaments, are often red, a colour that contrasts maximally with a green background (Grether, Kolluru & Nersissian, 2004). Red colours are often generated by carotenoid pigments in fishes (Kodric-Brown, 1985;

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Houde, 1987; Candolin, 1999), birds (Hill & McGraw, 2006), and lizards (Macedonia *et al.*, 2000; Steffen & McGraw, 2007; Fitze *et al.*, 2009; Cote *et al.*, 2010). These pigments have become common in the literature because of their roles in free-radical scavenging and animal coloration (Vershinin, 1999), as well as their function as honest signals (Lozano, 1994; Hill, 2002). Ultraviolet (UV)-coloured signals are common among fishes (Loosey *et al.*, 1999), and birds (Eaton & Lanyon, 2003) and serve a prominent role as armaments in male agonistic interactions among male lizards (Molina-Borja, Padron-Fumero & Alfonso-Martin, 1998; LeBas & Marshall, 2000; Stapley & Whiting, 2005; Lappin *et al.*, 2006; Whiting *et al.*, 2006; Bajer *et al.*, 2011).

Lizards frequently employ a polygynous and territorial mating system in which male–male contests are important for mate and territory acquisition (Cooper & Greenberg, 1992; Pianka & Vitt, 2003). The brown anole (*Norops sagrei*) is a dactyloid (*sensu* Nicholson *et al.*, 2012) that exemplifies this polygynous, territorial mating system. Adult males vigorously defend a space and/or access to several females (Evans, 1938a, b; Tokarz, 1998, 2002; Calsbeek & Marnocha, 2006) and males mate with more than one female (Schoener & Schoener, 1980; Tokarz, 1998). Direct physical contests are common among males, although males can signal to rivals using two non-exclusive means: (1) a stereotypical display of head-bob and push-up behaviours (Scott, 1984; McMann, 2000; Partan *et al.*, 2011; Simon, 2011) and (2) presentation of sexually-dimorphic dewlap colour (Steffen & McGraw, 2009). Brown anole dewlaps are coloured: orange and yellow in the centre and yellow and UV along the margin. Carotenoids create yellow colours of the central and marginal regions of the dewlap, and interact with pterins to create shades of orange and red in the dewlap's centre (Steffen & McGraw, 2009).

At least two studies (Nicholson *et al.*, 2007; Vanhooydonck *et al.*, 2009) have suggested that dactyloid dewlap colour diversity is partially influenced by sexual selection. Several studies have attempted to investigate female preference for dewlap colour in single species (Crews, 1975; Sigmund, 1983; MacDonald & Echternacht, 1991), although no research has examined how the combined influence of dewlap colour and display behaviour interact during intrasexual selection.

The present study aimed to use UV-visible spectrometry, in conjunction with a detailed ethological analysis, to assess the relative importance of brown anole dewlap colour versus postural display behaviours for predicting contest success in two intrasexual contexts: (1) when males compete for access to a solitary perch and (2) when males compete for access to mates. Given that postural displays are so evident

in brown anole social interactions, we predict that males who display more will win contests compared to males who display less, and this will be true in the presence and absence of females. Given that brown anole dewlap colour is red, orange, and yellow, as well as UV, we predict that males possessing dewlaps with higher spectral scores in these colours will win contests in both contest types.

MATERIAL AND METHODS

LIZARD COLLECTION AND CARE

Brown anoles were collected during 23–26 April in 2005 and 20–25 April in 2006 from Lake Eaton, Mill Dam Lake, and Silver Springs, Marion County, Florida, USA (29°15'43.48"N, 81°52'12.07"W). Each of these sites is no greater than 5 km from the other. All lizards were collected by hand or noose. In both years, all lizards were transported to a laboratory at Auburn University where we recorded sex, weight (nearest 0.01 g), and snout-to-vent length (nearest 0.5 mm). Once measured, lizards were assigned a unique toe clip (no more than two toes removed; antiseptic applied to each clipped toe). Lizards were then placed in pre-contest housing made of 37.9-L terraria (50.8 × 20.4 × 30.5 cm) partitioned into four equal-sized compartments (25.4 × 10.2 × 30.5 cm), each containing a wood dowel for a perch, a water dish, and a small plant. Full spectrum fluorescent bulbs (Vitalite T8, 32 W) were suspended 30.5 cm above each terrarium top and all terraria received additional natural light via a large window.

Lizards were sprayed with water daily, fed crickets three times each week (three per feeding) and meal worms *ad libitum*. All food items were dusted with Repta-vite vitamin powder (Zoo Med Laboratories) before being offered to a lizard. The laboratory was maintained at 32.2 °C and 60% relative humidity. No lizard was used in trials more than once.

SIZE-MATCHED MALES AND DISPLAY HOUSING

Size determines the outcomes of contests among male brown anoles (Tokarz, 1985). Nevertheless, in social interactions in which mature adult males compete for access to a territory or a female, a large proportion of fights are expected to occur between males of similar size because small males avoid fights with larger males after initial encounters (J. E. Steffen, pers. observ). To test the effects of dewlap colour and display behaviour, irrespective of body size, we size-matched (nearest 0.5 mm) pairs of males and placed each pair into contest housing 1 day before a contest. Contest housing consisted of 37.9-L terraria that were partitioned with particle boards into three compartments. The largest compartment (25.4 × 20.4 × 30.5 cm)

represented half of the terrarium divided along its width. The other half was further divided into two small compartments ($25.4 \times 10.2 \times 30.5$ cm) along the long axis of the aquarium. Each compartment contained a water dish (filled daily) and housed one of the two males. The outside walls of the terraria were lined with green construction paper to facilitate visual detection of each male's red and yellow dewlap colours (Endler, 1992). The partitions prevented lizards from seeing any other compartment until the partitions were removed. Lighting was similar to lighting described for pre-contest housing.

SPECTROMETRY

We measured the colour of the dewlap in two distinct regions: the centre (appears red to the unaided human eye) and the margin (appears yellow or white to the unaided human eye). Spectral measurements were taken with an Ocean Optics S2000 UV-visible spectrometer (OOI Base32 software) 1 day before initiation of the experiment (always starting at 10.00 h CST) on lizards that showed no signs of imminent shedding. All reflectance data were generated relative to a white reflectance standard and were taken in an unlit room with tightly drawn blinds covering the window. We placed a small black rubber stopper on the tip of the reflectance probe, creating a 2-mm gap between the probe tip and the dewlap, ensuring a constant distance between probe and dewlap. To measure dewlaps with the spectrometer, we placed each lizard ventral side up on a flat black table and immobilized the animal with two pieces of athletic tape: one placed across its belly and the other across its mandible. The dewlap was maximally extended by grasping it with a small clamp and adjusting the height of the clamp via its attachment to a horizontal metal arm on a ring stand. We placed the spectrometer probe at a 90° angle, flush with the exposed skin of the dewlap. We measured spectral reflectance along the centre and margin of the dewlap, taking four, non-overlapping spectral measurements per dewlap region and averaging them for each lizard.

Spectral measurements were gathered as percent reflectance at 1-nm wavelength increments from 300–700 nm (representing the lower range of photon absorption by UV-sensitive cones; Fleishman, Loew & Leal, 1993). Spectral measurements were smoothed using CLR, version 1.0 (Montgomerie copyright 2008). Each smoothed file was standardized (mean reflectance subtracted as described by Cuthill *et al.*, 1999) and then reduced to the means of 20-nm bandwidths. Principal components analysis (PCA) was performed on these standardized spectral files and the resulting PCs described the spectral shape (for details, see below).

CONTESTS AND BEHAVIOURAL ANALYSIS

Males interacted with each other in two different social contexts. First, males engaged in agonistic behaviours to compete for access to a solitary perch (MM contests). Fourteen pairs of males were used in such contests in 2005 and 32 pairs of males were used in 2006. Second, males engaged in agonistic interactions to compete for access to, and copulation with, an adult female (MMF contests). Twenty-one pairs of males were used in such contests in 2005 and 16 pairs were used in 2006. MM contests were performed in terraria in which partitions were lifted to expose a single perch present in the largest compartment. MMF contests were performed in terraria in which a perch existed in each of the three separate compartments and partitions were lifted to expose the two males to each other, as well as to a mature female in the large compartment. All females were assumed to be sexually receptive because behavioural trials were conducted from 1 May to 15 July, when this species is known to be reproductively active (Lee *et al.*, 1989).

At the beginning of each trial, we turned on an 8-mm video camera and removed partitions separating the three compartments. We immediately left the room and allowed the camera to record all behaviours. At a later date, we used ETHOLOG, version 2.2 (Ottoni, 2000) to record, count, and time all behavioural events of all participating lizards. We defined each individual dewlap extension, head-bob, push-up, bite, and copulation as a separate behavioural event. We quantified the behavioural events of each trial from the ETHOLOG files to: (1) enumerate head-bob (HB; head and neck moved dorsoventrally by extension and flexion of forelimbs), push-up (PU; extension and flexion of all four limbs), and dewlap extensions (DEW; extension of throat fan); (2) calculate display rates for each of these behaviours; and (3) measure time spent on the perch for each male (MM trials), or count copulations with a female for each male (in MMF trials). For each behavioural trial, we noted the elapsed time from the start of a trial to the first appearance of a behaviour (latency to display). We defined a particular male to be dominant in MM trials when its occupancy of the solitary perch went uncontested for 10 min. For MMF trials, the first male to intromit a hemipenis was defined to be dominant. We calculated display rates for each type of male behaviour (HB, PU, DEW) by enumerating each behaviour and dividing the number by the total time (starting at time of first behavioural event and ending when dominance of one male was achieved). After each trial, lizards were euthanized with MS222, fixed in formalin, and deposited in the Auburn University Natural History Museum Collections. All housing and protocols for care of lizards were approved by the

Institutional Animal Care and Use Committee (protocol numbers 2005-0961 and 2006-0961).

STATISTICAL ANALYSIS

PCA

We used PCA to transform a large number of correlated behavioural and colorimetric variables into a few orthogonal variables (Cuthill *et al.*, 1999). We pooled spectral files of all males from both social contexts and both years, and we performed PCA. This generated colorimetric PC coefficients that were used to aid in interpretation of the PCs. We interpreted the PC coefficients by graphing the PC coefficients for each PC against wavelength and determining how positive aspects of a curve vary with negative aspects of that curve (Cuthill *et al.*, 1999). Colorimetric PC scores were also generated for each male and served as the transformed data that were used in the subsequent analysis of colour and contest success. Each spectral PC was interpreted using the wavebands: UV = 300–399 nm, blue = 400–474 nm, green = 475–549 nm, yellow = 550–599 nm, orange = 600–649 nm, and red = 650–700 nm.

We also used PCA on behavioural data because HB, PU, and DEW were highly correlated. We pooled all HB, PU, and DEW frequencies for each male in contests from each year and social context into a single file and performed PCA. We used varimax rotation on dewlap colorimetric and behavioural data to maximize variance of PC scores, which maximizes high correlations between correlated variables and minimizes low correlations. This facilitates interpretation of a factor by making the variables that correlate with it unambiguous (Tabachnik & Fidell, 2001).

INTERPRETATION OF PCs

Spectral variation of the dewlap centre was reduced to three PCs that explained 94.0% of the total spectral variation. PC1 (63.1% of variance) represented amounts of yellow, orange, red, and UV wavelengths relative to blue and green wavelengths (Fig. 1). PC2 (24.8% of variance) represented amounts of UV, blue, orange, and red wavelengths relative to green and yellow wavelengths. PC3 (6.0% of variance) represented blue, green, and yellow wavelengths relative to UV, orange, and red wavelengths.

Spectral variation of the dewlap margin was reduced to three PCs that explained 96.2% of the total spectral variation. PC1 (72.7% of variance) represented amounts of green, yellow, orange, and red wavelengths relative to UV and blue wavelengths (Fig. 1). PC2 (17.0% of variance) represented amounts of upper UV (380–400 nm), blue, and green relative to lower UV (300–379 nm), yellow, orange, and red

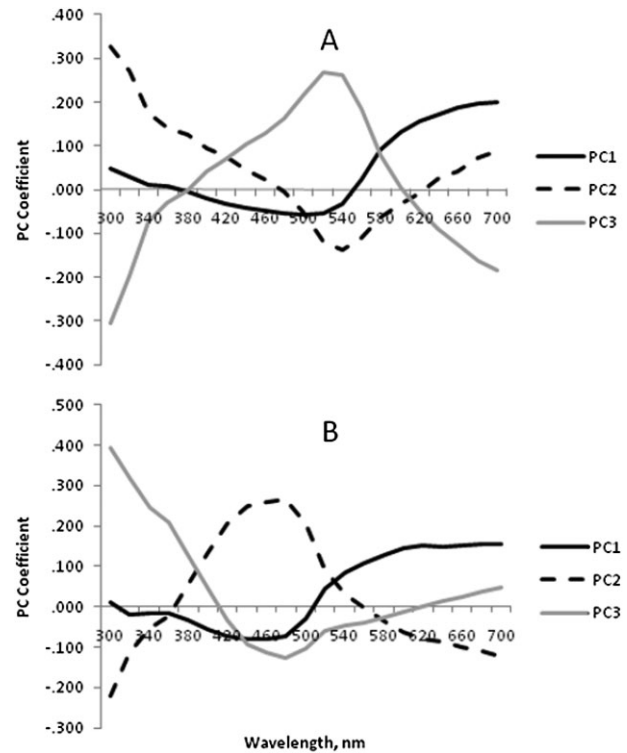


Figure 1. Principal component (PC) analysis of brown anole dewlap spectral variability in the centre and margin regions from males engaged in contests for access to perches (male–male; MM) and females (male–male–female; MMF). These illustrations show the influence of each wavelength on the identity of each PC by graphing PC coefficients against wavelength. A, PCs 1–3 of the dewlap centre. B, PCs 1–3 of the dewlap margin. For details, see text.

wavelengths. PC3 (6.5% of variance) represented UV (and, to a lesser extent, orange and red wavelengths) relative to blue, green, and yellow wavelengths.

All of the variation in HB, PU, and DEW frequencies among winners and losers across years and social contexts was described in a single PC that explained 51.6% of the variance in behaviour. Behavioural PC1 represents a correlated composite variable of HB, PU, and DEW frequency because PC1 had a PC coefficient of 0.507 for HB rate, 0.446 for PU rate, and 0.435 for DEW rate.

BINARY LOGISTIC MULTIPLE REGRESSION

We used backwards stepwise logistic multiple regression to determine: (1) the minimum adequate model (i.e. the simplest model that best describes the data; Crawley, 1993; Hardy & Field, 1998) and (2) the behavioural and colorimetric factors that predict contest success between males competing for access to a perch or a female. A minimum adequate model was

achieved by retaining all factors that significantly changed the log-likelihood of the full model and by removing all factors that did not (Tabachnik & Fidell, 2001). We performed maximum likelihood ratio, backwards, stepwise, logistic regression using the binary logistic regression procedure in SPSS (IBM Corp). We entered the behavioural and colorimetric PCs as continuously-distributed independent variables, year and social context as categorical covariates, and contests success (0 = contest loss, 1 = contest win) as the dependent variable. We investigated whether colour or behaviour varied significantly by year (2005 and 2006) or social context (MM and MMF) by including all possible main effects and interaction terms in an initial logistic regression model. This was carried out in SPSS by assigning main effect variables as block 1, and potentially interactive variables (e.g. year, or social context) as successive blocks thereafter. In our analysis, block 2 included the potential interactions of main effect variables with year, and block 3 included the potential interactions of main effect variables with social context. The end result was a binary logistic regression table retaining main effect and interactive variables that served as the minimum adequate model to predict contest success in brown anoles in two different social contexts (MM and MMF). When we arrived at the initial minimum adequate model, we re-ran the binary logistic regression analysis with all nonsignificant main effect and interactive terms removed (i.e. we made a final, full model) to obtain the most accurate statistical values for a final minimum adequate model predicting contest success. P to enter was 0.05 and P to remove was 0.10.

ONE-WAY ANALYSIS OF VARIANCE (ANOVA)

We performed one-way ANOVAs within social context and year to determine whether behaviour or colour of winners as a group differed from losers as a group. Alpha for critical significance was $P < 0.05$ in all of these statistics. We performed all statistics using SPSS, version 21 (IBM Corp.).

RESULTS

All dewlap colorimetric and behavioural variables were put into an initial full model that predicted contest success for MM and MMF interactions in 2005 and 2006 (Table 1). The minimum adequate model contained a dewlap colorimetric score (PC3 of the dewlap margin) and the behavioural PC score. Dewlap margin PC3 (relative amounts of UV, yellow, orange, and red) was a significant predictor of contest success ($B = -0.453 \pm 0.183$ SE, $P = 0.013$). Behaviour PC1 (relative rates of HB, PU, and DEW) was a

significant predictor of contest success ($B = 0.674 \pm 0.199$ SE, $P = 0.001$).

The significant predictors from the initial minimum adequate model were entered into a final, full model to determine which predictors are retained as a final minimum adequate model predicting contest success. This final model, with the simplest ability to predict contest success among pairs of males, was identical to Block 1 of the initial minimum adequate model (Table 2). The final model predicted contest success with an overall mean of 63.4% accuracy (lose accuracy = 68.4%; win accuracy = 58.5%).

In one-way ANOVA comparisons between winners and losers, males that won contests had higher mean behavioural PC1 scores (0.231 ± 0.123) than males that lost contests (-0.242 ± 0.088 ; $F_{162,1} = 9.651$, $P = 0.002$) (Fig. 2). Males that won contests had marginally significant lower mean dewlap margin PC 3 scores (-0.176 ± 0.098) than males that lost contests (0.110 ± 0.120 ; $F_{160,1} = 3.441$, $P = 0.065$) (Fig. 2).

DISCUSSION

We found that variation in anole behaviour (head-bobs, dewlap extensions, and push-ups), as well as dewlap colour (UV and, to a lesser extent, yellow–orange–red brightness and chroma), predicted success in male contests for females and perches. These variables predicted contest success in both years and social contexts.

Presumably, these outcomes indicate that these traits are candidates for sexual selection; if colours and behaviours are sexually selected, then we expect expression of these traits to differ between winners and losers. In the present study, males that displayed at higher rates won contests compared to males that displayed at lower rates, a finding consistent with the results of the study by Simon (2011), who found that higher head-bob rates were displayed by winning male brown anoles. Contest-based push-up behaviour is common among lizards (Carpenter, 1986) and males that perform more push-ups gain greater access to mates in many species investigated (Hover, 1985; Zucker, 1994a, b; LeBas & Marshall, 2000).

We found that UV and, to a lesser extent, orange and red wavelengths in dewlap colour predicted success in contests for females and perches. In brown anoles, reds, oranges, and yellows are generated by two pigment classes, carotenoids, and pterins (Bagnara & Hadley, 1973). Coloration derived from carotenoids is often used by animals as a signal associated with sexual selection (Andersson, 1994; Hill, 2007; Chen *et al.*, 2013). In common wall lizards, carotenoid-based ventral red coloration appears to be polymorphic and, among individuals that exhibit a red colour, there is a negative association between

Table 1. Results of initial backwards stepwise logistic multiple regression predicting contest success among male brown anoles competing for access to perches and females

Block	Step	Variable type	Variables in equation	B	SE	d.f.	Significance	Exp(B)	-2 LL
0	Step 0		Constant	0.037	0.158	1	0.813	1.038	223.137
	Step 1	Dewlap colour	PC1 centre	0.021	0.202	1	0.918	1.021	203.775
		Dewlap colour	PC2 centre	0.017	0.226	1	0.938	1.018	
		Dewlap colour	PC3 centre	-0.168	0.165	1	0.308	0.845	
		Dewlap colour	PC 1 margin	0.117	0.203	1	0.564	1.124	
		Dewlap colour	PC2 margin	0.067	0.169	1	0.692	1.069	
		Dewlap colour	PC3 margin	-0.503	0.262	1	0.054	0.605	
		Behaviour	PC1 behaviour	0.748	0.230	1	0.001	2.113	
			Constant	0.060	0.170	1	0.724	1.062	
		Dewlap colour	PC3 margin	-0.453	0.183	1	0.013	0.636	
2	Step 6	Behaviour	PC1 behaviour	0.674	0.199	1	0.001	1.961	205.8
			Constant	0.048	0.168	1	0.773	1.050	
	Step 1	Dewlap colour	PC3 margin	0.177	0.713	1	0.804	1.193	202.315
		Behaviour	PC1 behaviour	0.319	0.716	1	0.656	1.376	
		Dewlap colour	PC1 centre × YR	0.041	0.115	1	0.721	1.042	
		Dewlap colour	PC2 centre × YR	-0.017	0.150	1	0.910	0.983	
		Dewlap colour	PC3 centre × YR	-0.114	0.094	1	0.225	0.892	
		Dewlap colour	PC1 margin × YR	0.057	0.125	1	0.648	1.059	
		Dewlap colour	PC2 margin × YR	-0.014	0.105	1	0.894	0.986	
		Dewlap colour	PC3 margin × YR	-0.493	0.529	1	0.351	0.611	
3	Step 8	Behaviour	PC1 behaviour × YR	0.357	0.539	1	0.507	1.429	205.8
			Constant	-0.036	0.255	1	0.889	0.965	
	Step 1	Dewlap colour	PC3 margin	-0.453	0.183	1	0.013	0.636	202.906
		Behaviour	PC1 behaviour	0.674	0.199	1	0.001	1.961	
			Constant	0.048	0.168	1	0.773	1.050	
		Dewlap colour	PC3 margin	0.177	0.713	1	0.804	1.193	
		Behaviour	PC1 behaviour	0.319	0.716	1	0.656	1.376	
		Dewlap colour	PC1 centre × SOC	0.041	0.115	1	0.721	1.042	
		Dewlap colour	PC2 centre × SOC	-0.017	0.150	1	0.910	0.983	
		Dewlap colour	PC3 centre × SOC	-0.114	0.094	1	0.225	0.892	
7	Step 7	Dewlap colour	PC1 margin × SOC	0.057	0.125	1	0.648	1.059	205.8
		Dewlap colour	PC2 margin × SOC	-0.014	0.105	1	0.894	0.986	
		Dewlap colour	PC3 margin × SOC	-0.493	0.529	1	0.351	0.611	
		Behaviour	PC1 behaviour × SOC	0.357	0.539	1	0.507	1.429	
			Constant	-0.036	0.255	1	0.889	0.965	
		Dewlap colour	PC3 margin	-0.453	0.183	1	0.013	0.636	
		Behaviour	PC1 behaviour	0.674	0.199	1	0.001	1.961	
			Constant	0.048	0.168	1	0.773	1.050	

Dewlap colour variables, as well behaviour variables, were entered as independent variables; year and social context were entered as categorical covariates; and contest success was the discrete dependent variable. Block 0 = the constant only model; Block 1, Step 1 = the fully predictive model (i.e. the 'full model') containing all possible main effect variables; Block 1, Step 6 = the minimum adequate model containing significant main effect predictors; Block 2, Step 1 and Step 8 = the full and minimum adequate models, respectively, containing all significant main effect predictors and possible interactions with year; Block 3, Step 1 and Step 7 = the full and minimum adequate models, respectively, containing all significant main effect predictors and possible interactions with year and social context. Significance = statistical significance where $P < 0.05$. *Variables that significantly change the log-likelihood (LL) ratio at $P < 0.05$. *Italicized significance values are marginally significant, $P < 0.10$. For details, see text. YR, year; SOC, social context.

Table 2. Results of final backwards stepwise logistic multiple regression predicting contest success among male brown anoles competing for access to perches and females

Block	Step	Variable type	Variables in equation	B	SE	d.f.	Significance	Exp(B)	-2 LL
0	0		Constant	0.037	0.158	1	0.813	1.038	223.137
1	1	Dewlap colour	PC3 margin	-0.453	0.183	1	0.013*	0.636	205.8
		Behaviour	PC1 behaviour	0.674	0.199	1	0.001*	1.961	
			Constant	0.048	0.168	1	0.773	1.050	

The significant dewlap colour variable (PC3 of dewlap margin) and the behaviour variable (PC1, both from the initial backwards stepwise logistic multiple regression) were entered as IVs, and contest success was the discrete DV. Significance = statistical significance where $P < 0.05$. *Variables that significantly change the log-likelihood (LL) ratio at $P < 0.05$. See text for details.

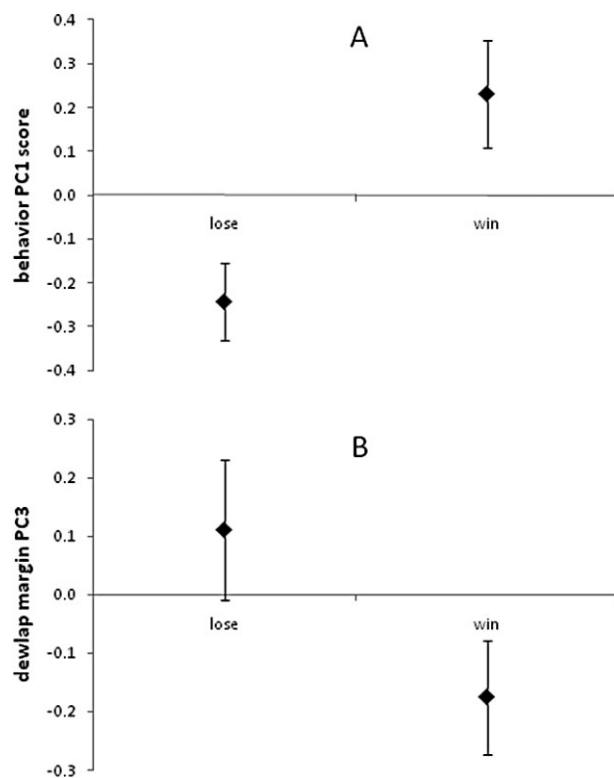


Figure 2. One-way analysis of variance comparisons of principal components (PCs) retained by the minimum-adequate logistic regression model predicting contest success between losers and winners in male–male (MM) and male–male–female (MMF) brown anole contests. A, winners have a higher behavioural PC1 score than losers ($F_{162,1} = 9.651$, $P = 0.002$). B, winners have a lower dewlap margin PC3 scores than losers ($F_{160,1} = 3.441$, $P = 0.065$). Error bars indicate the SEM. For details, see text.

the percentage of red coloration and sprint speed (Zajitschek *et al.*, 2012). Ventral colour polymorphism does not influence fighting ability and contest outcome in this species (Sacchi *et al.*, 2009). However, the Australian frillneck lizard has a large and exten-

sible red, orange, and yellow coloured frill that is carotenoid dependent and is a reliable predictor of competitive ability in contests (Hamilton, Whiting & Pryke, 2013).

UV integument colour has been shown to be a widespread signal in the behaviour of many fishes (Losey *et al.*, 1999) and birds (Eaton & Lanyon, 2003). UV throat colour, in particular, has been shown to be an important intraspecific signal in the behaviour of several territorial lizards. For example, high UV reflectance determined the outcome of male–male fights in the ornate crevice dragon (Bajer *et al.*, 2011). Throat UV was a signal of fighting ability in male *Augrabies* flat lizards and contests were more likely to escalate when one contestant had reduced UV (Stapley & Whiting, 2005; Whiting *et al.*, 2006).

Surprisingly, we found that males with lower amounts of UV along with orange and red wavelengths along the dewlap margin won more contests than males with higher amounts of these colours. Both carotenoids and pterins reside in this portion of the male brown anole dewlap but pterins are relatively low in abundance (Steffen & McGraw, 2009). Therefore, the yellow and UV reflectance spectrum is presumably a by-product of differential photon absorption by xanthophylls (predominantly), iridophores, and melanins (Grether *et al.*, 2004). How xanthophyll concentration variation generates the height and shape of the UV peak is not well understood in brown anoles, although simulations of variation in xanthophyll abundance in great tit feathers suggest a mechanism because increased xanthophyll abundance in feathers reduced the overall reflectance of UV peaks (Andersson & Prager, 2006). Another possibility is that, as anoles age, dewlaps become less colourful and the contest success associated with reduced dewlap margin UV is a result of age-related differences in scale wear or pigment concentrations.

Similar to the findings reported for other iguanians (Hover, 1985; Thompson & Moore, 1991a, b; Zucker, 1994a, b), our results show that behavioural and

colour traits have a combined signalling function in male–male combat that gives winners access to territories and, therefore, mates. In the contests for mating opportunities reported in the present study, males achieved access to a female by first winning a fight with a rival male.

Female preference for male traits (including dewlap colour and dewlap function) has been investigated in the green anole and appears to play a limited role (Sigmund, 1983; MacDonald & Echternacht, 1991). In laboratory studies of brown anoles, female movement never led to mate decisions or copulations, and females did not show preferences for males that varied in behaviour or dewlap colour (J. E. Steffen, pers. observ.). Tokarz (1998) suggested that male–male competition is more important than female mate choice in determining female mating partners in brown anoles because females almost entirely mated with males who possessed a territory where the female resided, rather than mating with males outside of this territory.

Moreover, ecologically relevant traits, such as dewlap size and bite force, are not preferred by female green anoles (Lailvaux & Irschick, 2006a). Indeed, the majority of performance traits that have been shown to be relevant to sexual selection (e.g. metabolic rate, energy reserves, endurance, sprint speed) in a variety of animal taxa show positive associations with male–male combat but no association with female choice (Lailvaux & Irschick, 2006b).

Our study suggests that behavioural traits are a stronger signal of contest success than dewlap colour, at least for animals that are in close proximity, because the behaviour PC had a higher logistic regression coefficient than colour PCs. Behavioural traits such as head-bobs and push-ups are probably more effective long distance cues because the visual system of anoles is specialized for detecting high velocity and accelerated movements (Fleishman, 1992). These behavioural displays are performed at a variety of distances from a receiver and are detectable under a variety of light environments. Indeed, Fleishman (1992) suggested that dewlap colour might only be an adequate cue to the conspecific of its identity and territorial status at very close range. Interestingly, our findings suggest that the margin of the dewlap is the region with a possible signal function. A colourful and moving margin of the extending dewlap is highly conspicuous because of the contrast created by the spectra of the leading edge displayed against the background vegetation (Fleishman, 1992). Moreover, motion-sensitive neurones respond when an object or edge moves through an anole's visual field. Motion-sensitive cells have been studied in many animal species and, in almost every case, they respond to edges created by contrast in brightness

(Fleishman, 1992). It would be interesting to determine whether neurones and visual photoreceptors respond to variation in UV brightness in brown anoles, and whether males appear to attend to this spectral signal in the wild. It would also be interesting to determine whether dewlap colour pattern variation (as described by Endler, 2012) predicts contest success in anoles.

In conclusion, several studies in birds and lizards have found that armament colour and display behaviour are signals that predict contest success and access to mates, and are, therefore, sexually selected traits. Our findings add the brown anole, a model squamate, to this list.

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REFERENCES

- Andersson M. 1994.** *Sexual selection*. Princeton, NJ: Princeton University Press.
- Andersson S, Prager M. 2006.** Quantifying colors. In: Hill GE, McGraw KJ, eds. *Bird coloration*, Vol. 1. *Mechanisms and measurements*. Cambridge, MA: Harvard University Press, 41–89.
- Bagnara JT, Hadley ME. 1973.** *Chromatophores and colour change, the comparative physiology of pigmentation*. Englewood Cliffs, NJ: Prentice Hall.
- Bajer K, Molnár O, Török J, Herczeg G. 2011.** Ultraviolet nuptial colour determines fight success in male European green lizards (*Lacerta viridis*). *Biology Letters* **7**: 866–868.
- Berglund A, Bisazza A, Pilastro A. 1996.** Armaments and ornaments: an evolutionary explanation of traits of dual utility. *Biological Journal of the Linnean Society* **58**: 385–399.
- Calsbeek R, Marnocha E. 2006.** Context dependent territorial defense: the importance of habitat structure in *Anolis sagrei*. *Ethology* **122**: 537–543.
- Candolin U. 1999.** Male–male competition facilitates female choice in Sticklebacks. *Proceedings of the Royal Society of London Series B, Biological Sciences* **266**: 785–789.

- Carpenter CC. 1967.** Aggression and social structure in iguanid lizards. In: Milstead WW, ed. *Lizard ecology: a symposium*. Columbia, MO: University of Missouri Press, 87–105.
- Carpenter CC. 1978.** Ritualistic social behavior in lizards. In: Greenberg N, MacLean PD, eds. *Behavior and neurology of lizards*. Bethesda, MD: National Institute of Mental Health, 253–267.
- Carpenter CC. 1986.** An inventory of the display-action-patterns in lizards. *Smithsonian Herpetological Literature* **68**: 1–18.
- Chen I, Symonds MRE, Melville J, Stuart-Fox D. 2013.** Factors shaping the evolution of colour patterns in Australian agamid lizards (Agamidae): a comparative study. *Biological Journal of the Linnean Society* **109**: 101–112.
- Cooper WE Jr, Greenberg N. 1992.** *Reptilian coloration and behavior*. Chicago, IL: University of Chicago Press.
- Cote J, Meylan S, Clobert J, Voituron Y. 2010.** Carotenoid-based coloration, oxidative stress and corticosterone in common lizards. *Journal of Experimental Biology* **213**: 2116–2124.
- Crawley MJ. 1993.** *GLIM for ecologists*. Boston, MA: Blackwell Scientific Publications.
- Crews D. 1975.** Effects of different components of male courtship behaviour on environmentally induced ovarian recrudescence and mating preferences in the lizard, *Anolis carolinensis*. *Animal Behaviour* **23**: 349–356.
- Cuthill IC, Bennett ATD, Partridge JC, Maier EJ. 1999.** Plumage reflectance and the objective measurement of avian sexual dichromatism. *American Naturalist* **153**: 183–200.
- Darwin C. 1871.** *The descent of man, and selection in relation to sex*. London: Murray.
- Eaton MD, Lanyon SM. 2003.** The ubiquity of avian ultraviolet plumage reflectance. *Proceedings of the Royal Society of London Series B, Biological Sciences* **270**: 1721–1726.
- Endler JA. 1992.** Signals, signal conditions, and the direction of evolution. *American Naturalist* **139**: 1–27.
- Endler JA. 2012.** A framework for analyzing colour pattern geometry: adjacent colours. *Biological Journal of the Linnean Society* **107**: 233–253.
- Evans LT. 1938a.** Cuban field studies on territoriality in the lizard *Anolis sagrei*. *Journal of Comparative Psychology* **25**: 97–125.
- Evans LT. 1938b.** Courtship and sexual selection of *Anolis*. *Journal of Comparative Psychology* **26**: 475–498.
- Fitze PS, Cote J, San-Jose LM, Meylan S, Isaksson C, Andersson S, Rossi JM, Clobert J. 2009.** Carotenoid-based colours reflect the stress response in the common lizard. *PLoS ONE* **4**: e5111.
- Fleishman LJ. 1986.** Motion detection in the presence and absence of background motion in an *Anolis* lizard. *Journal of Comparative Physiology A* **159**: 711–720.
- Fleishman LJ. 1992.** The influence of the sensory system and the environment on motion patterns in the visual displays of anoline lizards and other vertebrates. *American Naturalist* **139S**: 36–61.
- Fleishman LJ, Loew ER, Leal M. 1993.** Ultraviolet vision in lizards. *Nature* **365**: 397.
- Fleishman LJ, Marshall CJ, Hertz PE. 1995.** Comparative study of temporal response properties of the visual system of three species of anoline lizards. *Copeia* **1995**: 422–431.
- Fleishman LJ, McClintock WJ, D'eath RB, Brainard DH, Endler JA. 1998.** Color perception and the use of video playback experiments in animal behaviour. *Animal Behaviour* **56**: 1035–1040.
- Fleishman LJ, Persons M. 2001.** The influence of stimulus and background colour on signal visibility in the lizard *Anolis cristatellus*. *Journal of Experimental Biology* **204**: 1559–1575.
- Grether GF, Kolluru GR, Nersissian K. 2004.** Individual colour patches as multicomponent signals. *Biological Review* **79**: 583–610.
- Hamilton DG, Whiting MJ, Pryke SR. 2013.** Fiery frills: carotenoid-based coloration predicts contest success in frillneck lizards. *Behavioral Ecology* **24**: 1138–1149.
- Hardy ICW, Field SA. 1998.** The logistic analysis of animal contests. *Animal Behaviour* **56**: 787–792.
- Hill GE. 2002.** *Red bird in a brown bag: the function and evolution of colorful plumage in the house finch*. New York, NY: Oxford University Press, Inc.
- Hill GE. 2007.** Melanins and carotenoids as feather colorants and signals. In: Jamieson BGM, ed. *Reproductive biology and phylogeny of birds*, Vol. 6B. *Sexual selection, behavior, conservation, embryology, and genetics*. Enfield, NH: Science Publishers, Inc., 41–73.
- Hill GE, McGraw KJ. 2006.** *Bird coloration*, Vol. 1. *Mechanisms and measurements*. Cambridge, MA: Harvard University Press.
- Houde AE. 1987.** Mate choice based upon naturally occurring color-pattern variation in a guppy population. *Evolution* **41**: 1–10.
- Hover EL. 1985.** Differences in aggressive behavior between two throat color morphs in a lizard, *Urosaurus ornatus*. *Copeia* **1985**: 933–994.
- Jenssen TA. 1977.** Evolution of Anoline lizard display behavior. *American Zoologist* **17**: 203–215.
- Kodric-Brown A. 1985.** Female preference and sexual selection for male coloration in the guppy (*Poecilia reticulata*). *Behavioral Ecology and Sociobiology* **17**: 199–205.
- Lailvaux SP, Irschick DJ. 2006a.** No evidence for female association with high-performance males in the green anole lizard, *Anolis carolinensis*. *Ethology* **112**: 707–715.
- Lailvaux SP, Irschick DJ. 2006b.** A functional perspective on sexual selection: insights and future prospects. *Animal Behaviour* **72**: 263–273.
- Lappin AK, Brandt Y, Husak JF, Macedonia JM, Kemp DJ. 2006.** Gaping displays reveal and amplify a mechanistically based index of weapon performance. *American Naturalist* **168**: 100–113.
- LeBas NR, Marshall NJ. 2000.** The role of colour in signaling and male choice in the agamid lizard *Ctenophorus ornatus*. *Proceedings of the Royal Society of London Series B, Biological Sciences* **267**: 445–452.

- Lee JC, Clayton D, Eisenstein S, Perez I. 1989. The reproductive cycle of *Anolis sagrei* in southern Florida. *Copeia* 1989: 930–937.
- Losey GS, Cronin TW, Goldsmith TH, Hyde D, Marshall NJ, McFarland WN. 1999. The UV visual world of fishes: a review. *Journal of Fish Biology* 54: 921–943.
- Lozano GA. 1994. Carotenoids, parasites and sexual selection. *Oikos* 70: 309–311.
- MacDonald D, Echtenacht AC. 1991. Red-throated and gray throated *Anolis carolinensis*: do females know the difference? *Anolis Newsletter* IV: 92–100.
- Macedonia JM, James S, Wittle LW, Clark DJ. 2000. Skin pigments and coloration in the Jamaican radiation of *Anolis* lizards. *Journal of Herpetology* 34: 99–109.
- McMann S. 2000. Effects of residence time on displays during territory establishment in a lizard. *Animal Behaviour* 59: 513–522.
- Molina-Borja M, Padron-Fumero M, Alfonso-Martin T. 1998. Morphological and behavioral traits affecting the intensity and outcome of male contests in *Gallotia galloti* (Family Lacertidae). *Ethology* 104: 314–322.
- Montgomerie R. 2008. *CLR*, Version 1.0. Kingston, ON: Queen's University.
- Nicholson KE, Crother BI, Guyer C, Savage JM. 2012. It is time for a new classification of anoles (Squamata: Dactyloidae). *Zootaxa* 3477: 1–108.
- Nicholson KE, Harmon LJ, Losos JB. 2007. Evolution of *Anolis* lizard dewlap diversity. *PLoS ONE* 2: e274.
- Ottoni EB. 2000. Etholog 2.2: a tool for the transcription and timing of behavior observation sessions. *Behavioral Research Methods, Instructional Companion* 32: 446–449.
- Partan SR, Marler P. 2006. Issues in the classification of multimodal communication signals. *American Naturalist* 166: 231–245.
- Partan SR, Otovic P, Price VL, Brown SE. 2011. Assessing display variability in wild brown anoles *Anolis sagrei* using a mechanical lizard model. *Current Zoology* 57: 140–152.
- Pianka ER, Vitt LJ. 2003. *Lizards: windows to the evolution of diversity*. Berkeley, CA: University of California Press.
- Sacchi R, Pupin F, Gentili A, Rubolini D, Scali S, Fasola M, Galeotti P. 2009. Male–male combats in a polymorphic lizard: residency and size, but not color, affect fighting rules and contest outcome. *Aggressive Behavior* 35: 274–283.
- Schoener TW, Schoener A. 1980. Densities, sex ratios, and population structure in four species of Bahamian *Anolis* lizards. *Journal of Animal Ecology* 49: 19–53.
- Scott MP. 1984. Agonistic and courtship displays of male *Anolis sagrei*. *Breviora* 479: 1–22.
- Searcy WA, Nowicki S. 2005. *The evolution of animal communication: reliability and deception in signalling systems*. Princeton, NJ: Princeton University Press.
- Sigmund WR. 1983. Female preference for *Anolis carolinensis* males as a function of dewlap color and background coloration. *Journal of Herpetology* 17: 137–143.
- Simon VB. 2011. Communication signal rates predict interaction outcome in the brown anole lizard, *Anolis sagrei*. *Copeia* 2011: 38–45.
- Sinervo B, Liveley CM. 1996. The rock-paper-scissors game and the evolution of alternative mate strategies. *Nature* 380: 240–243.
- Stapley J, Whiting MJ. 2005. Ultraviolet signals fighting ability in a lizard. *Biology Letters* 2: 169–172.
- Steffen JE, McGraw KJ. 2007. Contributions of pterin and carotenoid pigments to dewlap coloration in two anole species. *Comparative Biochemistry and Physiology. Part B, Biochemistry and Molecular Biology* 146: 42–46.
- Steffen JE, McGraw KJ. 2009. How dewlap color reflects its carotenoid and pterin content in male and female brown anoles (*Norops sagrei*). *Comparative Biochemistry and Physiology. Part B, Biochemistry and Molecular Biology* 154: 334–340.
- Tabachnik BG, Fidell LS. 2001. *Using multivariate statistics*. New York, NY: Allyn and Bacon.
- Thompson CW, Moore MC. 1991a. Syntopic occurrence of multiple dewlap color morphs in male tree lizards, *Urosaurus ornatus*. *Copeia* 1991: 493–503.
- Thompson CW, Moore MC. 1991b. Throat colour reliably signals status in male tree lizards, *Urosaurus ornatus*. *Animal Behaviour* 42: 745–753.
- Tokarz RR. 1985. Body size as a factor determining dominance in staged agonistic encounters between male brown anoles (*Anolis sagrei*). *Animal Behaviour* 33: 746–753.
- Tokarz RR. 1998. Mating pattern in the lizard *Anolis sagrei*: implications for mate choice and sperm competition. *Herpetologica* 54: 388–394.
- Tokarz RR. 2002. An experimental test of the importance of the dewlap in male mating success in the lizard *Anolis sagrei*. *Herpetologica* 58: 87–94.
- Vanhooydonck B, Herrell A, Meyers JJ, Irschick DJ. 2009. What determines dewlap diversity in *Anolis* lizards? An among-island comparison. *Journal of Evolutionary Biology* 22: 293–308.
- Vershinin A. 1999. Biological functions of carotenoids: diversity and evolution. *Biofactors* 10: 99–104.
- Whiting MJ, Stuart-Fox DM, O'Connor D, Firth D, Bennett NC, Blomberg SP. 2006. Ultraviolet signals ultra-aggression in a lizard. *Animal Behaviour* 72: 353–363.
- Zajitschek SRK, Zajitschek F, Miles DB, Clobert J. 2012. The effect of coloration and temperature on sprint performance in male and female wall lizards. *Biological Journal of the Linnean Society* 107: 573–582.
- Zucker N. 1994a. A dual status-signalling system: a matter of redundancy or differing roles? *Animal Behaviour* 47: 15–22.
- Zucker N. 1994b. Social influence on the use of a modifiable status signal. *Animal Behaviour* 48: 1317–1324.