

Female adaptation to developmental plasticity in male calling behavior

Oliver M. Beckers · Johannes Schul

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Abstract The katydid *Neoconocephalus triops* exhibits in North America substantial developmental plasticity of male mating calls. The AM rate of the summer calls is significantly faster than that of the winter calls at the same temperature. In the tropics, where *N. triops* originated, males express only the fast summer-call phenotype. We tested two alternative hypotheses: (1) call plasticity in the population from North America evolved in response to selection by female preference after *N. triops* colonized North America, or (2) call plasticity evolved before *N. triops* expanded into North America and its expression in the novel environment led to adaptive change of female preferences. First, we tested whether call plasticity was present in tropical populations of *N. triops*. Tropical males expressed the winter-call phenotype when reared under winter conditions, indicating that call plasticity did not evolve in response to temperate climates. Second, we compared female preferences among temperate and tropical populations. We found that the temperature dependence of preferred AM rate was significantly steeper in temperate *N. triops* than in a tropical population of *N. triops*. Third, we compared temperature dependence of female preference of the *N. triops* populations to three *Neoconocephalus* species

without call plasticity. Only temperate *N. triops* had significantly steeper temperature dependence than the other species. This steeper temperature dependence matched female preference to the fast summer call at high temperatures and to the slow winter call at low temperatures in temperate populations. These results support the hypothesis that female preference changed in *N. triops* in North America to compensate for the plasticity of male calls.

Keywords Phenotypic plasticity · *Neoconocephalus triops* · Communication · Mating system

Introduction

Developmental (or phenotypic) plasticity, the ability of a given genotype to produce different phenotypes in response to different environmental conditions, is a widespread phenomenon in nature (Stearns 1989; Schlichting and Pigliucci 1998; West-Eberhard 2003). The ability of organisms to express different phenotypes can be adaptive, especially in heterogeneous or novel environments (Yeh and Price 2004; Trussel and Smith 2000; Laurila et al. 2002). However, developmental plasticity may also be non-adaptive (Roy et al. 2006; Kenyon and Hunter 2007; Whitman and Agrawal 2009), resulting in decreased fitness of the environmentally induced phenotype in the new environment compared to the ancestral phenotype (Van Tienderen 1991; Ghalambor et al. 2007). If plasticity is detrimental, selection is expected to reduce this plasticity, which could possibly lead to the fixation of only one phenotype as a result of genetic accommodation (West-Eberhard 2003, 2005). Non-adaptive developmental plasticity may persist if it is genetically linked to other (adaptive) traits and thus cannot evolve independently

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O. M. Beckers (✉) · J. Schul
Department of Biological Sciences, University of Missouri,
204 Tucker Hall,
Columbia, MO 65211, USA
e-mail: beckersom@unlserve.unl.edu

Present Address:

O. M. Beckers
School of Biological Sciences, University of Nebraska,
55 Manter Hall,
Lincoln, NE 68588, USA

(Lande 1982; Loeschke 1987). In this case, it is conceivable that correlated traits may adapt to compensate for the environmentally induced variation.

Communication signals produced in the context of reproduction contain information about the (typically male) signaler, such as species identity or the genetic quality of the signaler, which the (typically female) receiver can use for mate choice. Plasticity of mating signals has been reported in many communication systems (e.g. Hill et al. 2002; Wagner and Hoback 1999; Hoikkala and Isoherranen 1997), however this plasticity is usually restricted to signal parameters that indicate the signaler's quality but not species identity. Signal parameters that females use to recognize the species identity of males are usually 'static call parameters' (Gerhardt 1991) with low variability both among the signals of one male and among different males. Accordingly, such call parameters are only rarely subject to developmental plasticity (e.g. in crickets Walker 2000; Grace and Shaw 2004; Olvido and Mousseau 1995). At least in one case, male calls and female preferences changed in parallel (i.e. exhibit developmental plasticity), ensuring a match of calls and preferences in variable environments (Grace and Shaw 2004).

A notable exception is the katydid *Neoconocephalus triops*, which is the only *Neoconocephalus* species that exhibits substantial developmental plasticity in the AM rate of the male calls (Whitesell and Walker 1978; Greenfield 1990). AM rate of male calls is the single most important parameter for female *N. triops* to recognize conspecific calls (Beckers and Schul 2008). *N. triops* has the life history typical of tropical katydids (direct egg development, Whitesell 1974), and it is widespread throughout the Neotropics (Greenfield 1990). Its range extends into temperate North America (Whitesell 1974; Walker and Greenfield 1983; Greenfield 1990), where *N. triops* has two distinct, seasonal generations. Males of these two generations exhibit such substantial developmental plasticity of calls that the two generations were initially described as different species (Whitesell 1974; Whitesell and Walker 1978). The principal difference is that the amplitude-modulation (AM) rate of summer calls is more than 20% faster than that of winter calls at the same ambient temperature (compared to 5–10% in the cricket systems mentioned above). The different call phenotypes are induced by the photoperiod experienced during juvenile development (Whitesell 1974; Whitesell and Walker 1978). Tropical populations of *N. triops* never experience the conditions inducing the winter-call phenotype and thus produce only the summer call.

Female *N. triops* from temperate populations have a stabilizing preference for AM rate (Beckers and Schul 2008). Female preference does not exhibit developmental plasticity that parallels that of male calls, i.e. preference does

not differ between summer and winter females (Beckers and Schul 2008). However, female preference for AM rate changes significantly more with ambient temperature than does the AM rate of male calls (for a discussion of the temperature dependence of call production and call preferences see Beckers and Schul 2008; review in Gerhardt and Huber 2002). This results in an overlap of the preference with the fast summer call only at high (=summer) temperatures and the slow winter call only at low (=winter) temperatures. In contrast, summer calls at low temperatures and winter calls at high temperatures are unattractive to females (Beckers and Schul 2008). Thus, despite the absence of developmental plasticity in females, their preferences match the call phenotypes produced by males during either season. Note that we refer to the change of AM rate of male calls and female preferences with ambient temperature as the 'temperature dependence' of either trait.

Beckers and Schul (2008) hypothesized that the developmental plasticity of *N. triops* calls evolved in the temperate population in response to the steep temperature dependence of female call preferences. In the tropics, communication likely takes place in a narrow temperature range so that a mismatch of male and female temperature dependence would not interfere with communication. In temperate climates, however, communication would take place in a wider temperature range (summer and winter), selecting for an adaptation of male calls (Beckers and Schul 2008). This hypothesis makes two predictions: (1) developmental plasticity of male calls should be limited to temperate *N. triops* populations, and (2) temperature dependence of female preferences should not differ between temperate and tropical *N. triops* populations.

An alternative hypothesis is that the developmental plasticity evolved in a different context (e.g. as pleiotropic effect) before *N. triops* spread into temperate regions, and the steep temperature dependence of female preferences evolved as a response to the newly expressed winter calls in temperate *N. triops*. Tropical *N. triops* would not have expressed developmental plasticity, as they do not experience the necessary environmental conditions (i.e. short photoperiod). After moving into temperate regions, the slow winter-call phenotype caused a mismatch with female preferences and thus caused the evolution of a steeper temperature dependence of female preference. This scenario predicts a different pattern than the first hypothesis: (1) developmental plasticity of male calls should be widespread among tropical and temperate populations of *N. triops*, and (2) the preference of the temperate population of *N. triops* should have a steeper temperature dependence than tropical *N. triops* and also than other *Neoconocephalus* species without call plasticity.

Here, we test the predictions made by these two hypotheses. We raised males from tropical populations

under winter conditions to test whether they possess developmental plasticity for AM rate. We then compared the temperature dependence of female preference of the temperate *N. triops* population to that of two tropical *N. triops* populations, as well as to that of three *Neoconocephalus* species without male call plasticity.

Materials and methods

Developmental plasticity of male calls

We compared developmental plasticity of male calls among three *N. triops* populations: north-central Florida, a temperate population that expresses call plasticity (Whitesell and Walker 1978), and Puerto Rico and Costa Rica, two tropical populations that only express the summer-call phenotype under natural conditions (Greenfield 1990). We collected adult male and female *N. triops* from populations in the vicinity of Gainesville (Florida), Liberia (Costa Rica), Luquillo, and Naguabo (Puerto Rico), and brought the animals to the laboratory at the University of Missouri.

N. triops has long wings and is an excellent flier. The species is highly mobile (Reinhold 1983), and we have observed calling males more than 100 km from breeding populations (unpublished). This suggests that *N. triops* in North America has no distinct population structure, and that there is gene flow among different locales. This view has been supported by a recent phylogenetic analysis of *Neoconocephalus* (Snyder et al. 2009).

We collected from each population 20–30 adult females and 15–25 adult males to start a mass culture in the lab. Animals were kept at a light/dark cycle of 15/9 h and high/low temperatures of 30/20°C. We provided apples and puppy chow (Purina, MO) as food and grass as oviposition medium. The grass was replaced and checked for eggs every 1–2 weeks for a period of 8–10 weeks. We raised nymphs from the collected eggs using the protocol described in detail by Beckers and Schul (2008).

First-instar male nymphs from each population were haphazardly assigned for rearing under ‘summer conditions’ or ‘winter conditions’. Under ‘winter conditions’, males were reared to adulthood at a light/dark cycle of 11/13 h and daily high/low temperatures of 23/17°C, and then transferred to diapause conditions (9.5/14.5 h light/dark cycle, at 18/12°C). Diapause was terminated after a minimum of 7 weeks by increasing the day length to 13–15 h and high/low temperatures to 26/18°C. Under ‘summer conditions’, animals were reared to adulthood with a light/dark cycle of 15/9 h and high/low temperatures of 30/20°C.

Developmental plasticity of male calls is determined by the photoperiod experienced during juvenile development

(Whitesell 1974; Whitesell and Walker 1978). The photoperiods and temperatures used in this study correspond approximately to the natural conditions that each generation experiences in Gainesville, FL. These conditions reliably induced summer and winter calls in males from the north-central Florida population (Whitesell and Walker 1978; Beckers and Schul 2008). We recorded male calls 1 to 3 weeks after adult molt (summer males) or termination of diapause (winter males) to confirm that these conditions induced the corresponding call phenotypes.

Call recordings We recorded males in a temperature-regulated anechoic chamber at an ambient temperature of $25 \pm 1^\circ\text{C}$. Males were placed in screen cages, and a ¼" electret microphone (RadioShack 33-3028, frequency response 30 to 18,000 Hz) was attached to each cage. Calls were recorded with a custom-made A/D converter system (16-bit resolution, 250-kHz sampling rate) and analyzed using custom-made software with a temporal resolution of 0.1 ms. For each recorded call, we calculated the mean double-pulse (=AM; see below) rate of at least 100 consecutive double pulses.

In order to investigate the developmental plasticity of AM rate in terms of the proportional change of AM rate between summer and winter calls among populations, we log-transformed the AM rate of the summer and winter calls. The log-transformed AM rates were statistically compared using a two-factor ANOVA with ‘population’ and ‘generation’ as factors. Post-hoc comparisons were conducted using Tukey-HSD tests (note that this statistical test automatically corrects the significance levels for multiple comparisons, Day and Quinn 1989). The variance among data sets was not homogenous; the variance was smaller for two generation-population combinations compared to the others (Likelihood ratio test $\chi_1^2=23.93$, $p<0.0001$). However, when tested with separate variances, the results of the ANOVA did not change, i.e. the effects of ‘generation’ and ‘population’ remained significant (Likelihood Ratio test: population, $\chi_2^2=190.61$ $p<0.0001$; generation, $\chi_1^2=141.74$, $p<0.0001$), and the interaction remained not significant ($\chi_2^2=2.85$, $p=0.24$). Thus, we present the statistics of the two-factor ANOVA in the results.

Temperature dependence of male calls

We measured the temperature dependence of male calls for the three populations of *N. triops*, as well as from *N. affinis*. Male calls were recorded as described above at ambient temperatures that ranged between 22 and 28°C. Temperature at the male’s position was measured within 1 min after the recording was made. Since AM rate of male calls of many Orthopterans changes linearly with temperature (e.g.

Whitesell and Walker 1978; Walker et al. 1973; Walker 1975; Pires and Hoy 1992), we calculated for each population or species a linear regression line based on the AM rates of the recorded calls to determine its temperature dependence. Regression coefficients were normalized to the AM rate at 25°C for each population or species to determine the relative change of AM rate per degree Celsius [%/°C]. Previously reported call data for *N. bivocatus*, *N. robustus* (Büttner 2002) and *N. triops* (north-central Florida population; Whitesell and Walker 1978) was used for comparison with our call data (Table 1). If female preference for AM rate of *N. triops* from Florida evolved to compensate for the expression of call plasticity, we expect the temperature dependence of calls and preference to be mismatched in this population, whereas in tropical populations and other species (that do not express or have call plasticity) the temperature dependences should be similar for the communication system to function over a wide range of temperatures.

Temperature dependence of female preferences

We estimated the steepness of the temperature dependence of female preference of summer females for three populations of *N. triops* (north-central Florida, Puerto Rico, Costa Rica). Female *N. triops* from the Costa Rica and Puerto Rico populations were collected and raised under summer conditions as described above. For the Florida population, we used data of female preference from Beckers and Schul (2008). Selectivity (i.e. lower cutoff rates; see below) did not differ between summer and winter females from this population at 20°C and 25°C (Beckers and Schul 2008). Therefore, we pooled the data for the lower cutoff rates of the two generations (winter = 9 females, summer = 11 females) for the comparison with the tropical populations of *N. triops* and additional species. We started testing of

females 2 to 3 weeks after adult molt (summer generation) or the end of diapause (winter generation of Florida population, see above). Females were tested for up to 6 weeks, during which we detected no changes in their selectivity. Females of all populations and species were tested in the same experimental set-up and experimental conditions (except ambient temperature, see below) between August 2004 and October 2007.

In addition, we estimated the degree of the temperature dependence of female preferences for three other *Neoconocephalus* species that do not have call plasticity (*N. affinis*, *N. bivocatus*, and *N. robustus*) for comparison with *N. triops*. If female preference of *N. triops* from Florida evolved in response to male call plasticity, we expect the temperature dependence of preference from this population not only being steeper than that of tropical *N. triops* populations but also than those of other *Neoconocephalus* species. Furthermore, the temperature dependence of tropical *N. triops* and the other species should be similar since these populations or species do not express or have call plasticity, respectively.

The tested species were chosen to represent a wide range of male calls and female preference functions. AM rates of male calls ranged from (all values for 25°C) 200 pulses/s in *N. robustus* over 90 double pulses/s in *N. bivocatus* (Deily and Schul 2004) down to 13 double pulses/s in *N. affinis* (Greenfield 1990). As in *N. triops*, females of *N. bivocatus* and *N. affinis* use the AM rate to identify conspecific calls (Deily and Schul 2004; Beckers and Schul 2008; Bush et al. 2009). Female *N. robustus* use the duration of the silent interval between pulses for call recognition, which likely represents the ancestral state in this genus (Deily and Schul 2004). Based on their life history, *N. affinis* represents a tropical species (Naskrecki 2000; Greenfield and Walker 1983), and *N. bivocatus* and *N. robustus* represent temperate species (Greenfield 1990). For the phylogenetic

Table 1 Temperature dependence of AM rate of male calls of *Neoconocephalus* species. The relative change of AM rate with temperature (%/°C) was calculated by normalizing the regression coefficient of each

population or species to the AM rate at 25°C. Formulas of the regression lines for the temperature dependence of AM rates and their literature sources are given in the center and right columns, respectively

Species	Relative change of AM rate of male calls [%/°C]	Temperature dependence of AM rate of male calls	Source of call data
<i>N. triops</i> (Florida)	S: 1.87	$y = 2.06x + 58.70 (N = 13)$	This study
	S: 3.57	S : $y = 3.99x + 12.13 (N = 15)$	Whitesell and Walker (1978)
	W: 2.98	W : $y = 2.61x + 22.19 (N = 16)$	
<i>N. triops</i> (Puerto Rico)	3.61	$y = 3.75x + 10.19 (N = 13)$	This study
<i>N. triops</i> (Costa Rica)	1.88	$y = 2.39x + 67.45 (N = 16)$	This study
<i>N. robustus</i>	2.27	$y = 4.70x + 89.9$	Büttner (2002)
<i>N. bivocatus</i>	4.32	$y = 3.75x - 6.9$	Büttner (2002)
<i>N. affinis</i>	3.62	$y = 0.45x + 1.19$	This study

relationships among the four species studied here, see Snyder et al. (2009).

Adult *N. affinis* were collected in Puerto Rico in the vicinity of Naguabo and Luquillo. Offspring of these animals were raised under ‘summer conditions’ (see above). Females of *N. bivocatus* and *N. robustus* were collected as last-instar nymphs in Boone County, Missouri (USA). The insects were kept at a light/dark cycle of 14/10 h at 28/20°C until maturity. We started testing of females of all species 2 to 3 weeks after adult molt.

Phonotaxis tests We tested female phonotaxis using a walking compensator (Kramer–Kugel; Weber et al. 1981) in a temperature-regulated anechoic chamber at ambient temperatures of 20 and 25°C (for *N. triops* populations), or 22, 25, and 29°C (for *N. affinis*, *N. bivocatus*, and *N. robustus*). For each tested temperature, females were kept for a minimum of 3 days before the start of the experiments and during the time the experiments were conducted in incubators at about +3/−3°C (high/low, respectively) of the tested temperature. Behavioral data was collected after the female was acclimatized to the ambient temperature of the anechoic test chamber for at least 5 min.

Females were placed on top of a sphere, free to walk but kept in place by compensatory sphere rotations, while acoustic signals were presented from loudspeakers located in the animal’s horizontal plane. The direction and speed of walking by a female were read from the control circuitry. All trials were performed in the dark, except for an infrared light that was used to monitor the insects’ position (for details see Schul 1998; Weber et al. 1981).

Experimental protocol Each test and control stimulus was presented for 60–90 s sequentially from each of two loudspeaker positions. The first stimulus (=sequential broadcast of the same stimulus from each loudspeaker position) of each test series was the control stimulus, followed by two to three test stimuli, another control stimulus, two to three test stimuli, and so on, until all stimuli of a series were tested. A period of 60 s of silence elapsed between stimuli. The phonotactic responses of the females to the control stimuli were used as a reference to estimate the response strength to each test stimulus (see ‘phonotaxis score’ below). The experimental series lasted between 30 and 60 min and consisted of up to six test stimuli (plus up to four controls). We haphazardly varied the sequence of stimuli within a series among individual females (for a detailed description of the experimental protocol see Schul 1998; Bush et al. 2002).

Stimulation We generated synthetic signals using a custom-developed DA-converter/amplifier system with 250-kHz

sampling rate and 16-bit resolution. Stimuli were delivered using one of two loudspeakers (Motorola KSN1218C) mounted at a distance of 150 cm in the horizontal plane of the animal and separated by an angle of 105°. Signal amplitude was set to 80±1 dB peak SPL (re. 20 µPa) using a Brüel and Kjaer sound level meter (B and K 2231) and a ¼" condenser microphone (G.R.A.S 40BF), which was positioned 1 cm above the sphere (Beckers and Schul 2008).

We used sinusoids of approximately the dominant frequency of each species call as carrier signals (*N. robustus*, 7 kHz; *N. bivocatus*, 10 kHz; *N. triops*, 11 kHz; and *N. affinis*, 12.5 kHz; dominant frequencies taken from Schul and Patterson 2003; Beckers and Schul 2008; Bush et al. 2009) to which we subsequently applied amplitude modulations. For *N. affinis*, stimuli also contained the second harmonic (25 kHz) at a relative amplitude of −6 dB. Calls of *N. triops*, *N. bivocatus*, and *N. affinis* consist of double pulses (Walker et al. 1973; Whitesell and Walker 1978; Walker and Greenfield 1983). Females of these three species respond equally strong to stimuli with double pulses merged to long pulses (Deily and Schul 2004; Beckers and Schul 2008; Bush et al. 2009). In our experiments, we used stimuli for these species that consisted of pulses equivalent to a ‘merged double pulse’ of the male call. We refer to the double-pulse rate of male calls and the equivalent rate of the long pulses in our stimuli as AM rate. Male *N. robustus* produce calls with a single-pulse structure (Walker et al. 1973). Here, stimuli mimicked this pattern, and the AM rate refers to the single-pulse rate of both male calls and stimuli used. For each species, we adjusted pulse duration and interval duration to generate stimuli that varied in AM rate for the experiments conducted at different ambient temperatures.

In the stimuli used for tests of *N. affinis*, *N. bivocatus*, and *N. robustus*, the pulse pattern was continuously repeated. In the stimuli for *N. triops* the pulses were grouped into verses, interrupted by silent pauses of 50-ms duration. Verse duration was 950 ms for the Florida (Beckers and Schul 2008) and Puerto Rico populations, and 650 ms for the Costa Rica population (unpublished data).

The control stimuli mimicked the temporal pattern of the conspecific call for each species. The AM rate of the control stimulus was adjusted to match that of male calls of the species or population of the female being tested at the ambient temperature of each experiment. In previous experiments, such control stimuli were highly attractive to females of each species, eliciting responses comparable to those during stimulation with conspecific calls (Deily and Schul 2004; Beckers and Schul 2008; Bush et al. 2009).

Data analysis We quantified female response to stimuli that varied in AM rate by calculating a phonotaxis score (Schul 1998; Deily and Schul 2004; Bush et al. 2009). The phonotaxis score included three measures indicating the relative strength of phonotaxis: (1) the walking speed relative to the walking speed during the control stimulus, describing the elicited locomotion activity; (2) the vector length, describing the accuracy of orientation; and (3) the angular orientation relative to that during the control stimulus. The phonotaxis score can range from approximately +1 to -1, representing perfect positive or negative phonotaxis, respectively. Scores close to 0 indicate random or no responses (for details see Schul 1998). For each female, we plotted phonotaxis scores as a function of AM rate for each temperature. These response functions were smoothed by calculating a gliding average through three neighboring values and then normalized to the maximum value for each female (Beckers and Schul 2008).

Because of the different call recognition mechanisms, the shape of female response functions (=preference) differed between species (Fig. 2). Preference functions of species using AM rate for call recognition peaked at certain AM rates and decreased towards both lower and higher AM rates, i.e. had band-pass characteristics (Fig. 2a, all species but *N. robustus*). *N. robustus* uses the duration of the silent interval between pulses for call recognition (Deily and Schul 2004), and the preference function decreased only towards lower AM rates (=longer intervals) but not towards higher AM rates (=shorter intervals), i.e. had high-pass characteristics (Fig. 2b). In species with band-pass characteristics, we fitted parabolic functions ($y = c - a*(x - b)^2$) to the preference function of each female; the parts of the preference function with responses close to zero were excluded from the curve fitting. For *N. robustus*, sigmoidal functions ($y = 1/(1 + e^{-(x+a)*b})$) were fitted to individual response functions. Best-fit functions were determined by minimizing the sum of squared errors. These two types of functions matched well the distribution of our data, respectively (see Fig. 2). Female preference of all species tested had the characteristics of rate filters (i.e. band-pass or high-pass filters). To quantify the tuning of the rate filters, we used the technical definition of the lower cutoff rate (or frequency), which defines it as the rate at which the response falls below 70% towards lower AM rates (Speaks 1999). We determined the lower cutoff rate from the best-fit curve of each female, and used its change with temperature to estimate the temperature dependence of female preference. This approach allowed us to compare the ambient-temperature dependence of female preference between species independent of shape of their preference functions.

Statistics We obtained preference data for each *N. triops* female only at one temperature (20°C or 25°C), i.e. the temperature data for this species consisted of independent data points. Since we were interested in the proportional change of female preference for AM rate at the two test temperatures, we log-transformed the lower cutoff rates of each female. The transformed cutoff rates were statistically compared using a two-factor ANOVA with the factors ‘ambient temperature’ and ‘population’. Post-hoc comparisons were conducted using Tukey-HSD tests. The variance of the data among populations was not homogeneous among treatment combinations (population/temperature); the variance was smaller for two treatment combinations compared to the others (Likelihood ratio test: $\chi_1^2=25.26$, $p<0.0001$). However, when tested with separate variances, the results of the ANOVA did not change, i.e. the effects of ‘temperature’ and ‘population’ remained significant (Likelihood Ratio test: ‘temperature’, $\chi_1^2=116.30$, $p<0.0001$; ‘population’, $\chi_2^2=98.12$, $p<0.0001$), and the interaction between the two effects remained significant ($\chi_2^2=14.04$, $p<0.001$), supporting the results of the ANOVA.

For *N. affinis*, *N. bivocatus*, and *N. robustus*, we determined preference functions at two to three temperatures (22, 25, and 29°C) for each female in a repeated-measures design. We determined the temperature dependence of female preference by normalizing the lower cutoff rate of each female to the mean cutoff rate of each species at 25°C and calculating a linear regression line for each female based on this normalized data. We then calculated for each species the mean value of these individual regression coefficients as a measure of the temperature dependence of female preference [%/°C]. This procedure reduced the data collected at multiple temperatures for each female to only one data point (= regression coefficient) for each female and thus permitted statistical comparison of the data sets among species.

Because the behavioral data of the three *N. triops* populations was collected differently (see above), we had to transform the *N. triops* data to generate comparable data sets to the three other species. We normalized the lower cutoff rate of each female to the mean cutoff rate of the population at 25°C. We then calculated for each *N. triops* female tested at 20°C the difference between its normalized lower cutoff rate at this temperature and the mean normalized rate at 25°C (=1). From this difference, we calculated the relative change of the lower cutoff rate per degree Celsius [%/°C]. Since the data did not meet the assumptions of parametric statistical tests, we compared population/species means with a Kruskal–Wallis test and post-hoc Mann–Whitney test. Significance levels were corrected for multiple comparisons (sequential Bonferroni correction; Sokal and Rohlf 1997).

Statistics were calculated using JMP (SAS institute, Inc., version 7.0.1) and statistical procedures described in Zar (1999). All data are given as mean \pm SEM.

Results

Developmental plasticity of male calls in *N. triops*

The AM rates of summer and winter generation of the three populations of *N. triops* are shown in Fig. 1. Summer males produced calls with a significantly higher AM rate than winter males in all three populations, (two-factor ANOVA: ‘generation’, $F_{1, 118}=547.55$, $p<0.0001$; all post-hoc Tukey-HSD tests: $p<0.05$). We could not detect a significant difference among populations in their response to summer and winter rearing conditions (‘generation \times population’, $F_{2, 118}=1.99$, $p=0.1408$). Thus, all three populations of *N. triops* expressed significant developmental plasticity of the AM rate, and the amount of plasticity (i. e. the difference of AM rate between generations) did not differ among them. The AM rate of the winter calls

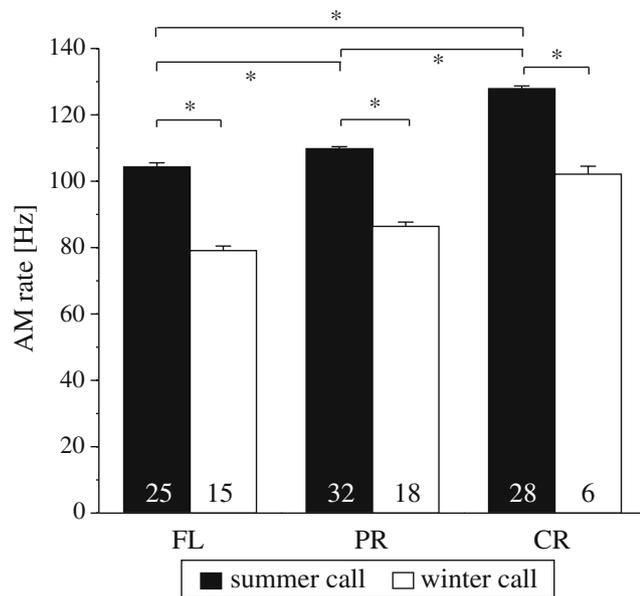


Fig. 1 AM (= double pulse) rates (mean \pm SEM) of males raised under summer (black bars) and winter conditions (open bars) from Florida (FL), Puerto Rico (PR), and Costa Rica (CR). The AM rates of summer calls differed significantly among the three populations. In all three populations, winter calls were significantly slower than the corresponding summer calls. The change of AM rate between generations did not differ significantly among populations. Asterisks indicate significant differences. Note that the statistical comparisons were conducted using log-transformed data. Calls were recorded at an ambient temperature of $25\pm 1^\circ\text{C}$

decreased by 20.16% (Costa Rica) to 24.13% (Florida) relative to the summer calls of the corresponding population. We also found a significant ‘population’ effect on the AM rate of each call phenotype ($F_{2, 118}=135.70$, $p<0.0001$, all post-hoc Tukey-HSD tests: $p<0.05$).

Temperature dependence of male calls

The temperature dependence of AM rate of male calls varied for different measurements and populations of *N. triops* between 1.88 and $3.60\%/^\circ\text{C}$ (Table 1). Differences between studies were likely due to relatively small sample sizes ($N=13\text{--}16$). The temperature dependence of *N. triops* calls was in the same range as those of other *Neconocephalus* species (2.27 to $3.61\%/^\circ\text{C}$, Table 1).

Temperature dependence of female preferences

Female preferences were strongly temperature-dependent (Fig. 2). We first compared the temperature dependence of female preference among the three populations of *N. triops* (Fig. 3). In all populations, preference functions shifted with increasing ambient temperature significantly towards higher AM rates (two-factor ANOVA: ‘ambient temperature’, $F_{1, 64}=412.24$, $p<0.0001$). We found a significant interaction between ‘population’ and ‘ambient temperature’ ($F_{2, 64}=7.15$, $p<0.0016$), indicating that the temperature dependence of female preference differed among the three populations. Post-hoc tests revealed a significant difference in ambient-temperature dependence between the Florida and Costa Rica populations (Tukey-HSD test: $q=4.94$, $p<0.005$) but not between the Puerto Rico population and either the Florida ($q=2.56$, $0.2>p>0.1$) or the Costa Rica population ($q=2.38$, $0.5>p>0.2$). We also found a significant effect of ‘population’ on the preferred AM rate of females (two-factor ANOVA: ‘population’, $F_{2, 64}=76.22$, $p<0.0001$), corresponding to the population differences in the AM rate of male calls (see above).

We next compared the temperature dependence of the three *N. triops* populations to that of three other *Neconocephalus* species (see methods for treatment of the *N. triops* data). For the Florida population, the relationship between temperature and preference for AM rate was significantly steeper (relative change of preferred AM rate: $5.82\%/^\circ\text{C}$) than that of the Costa Rica population and the three other species ($3.10\text{--}4.10\%/^\circ\text{C}$; Kruskal–Wallis test: $H_5=34.45$, $p=0.0001$, all post-hoc Mann–Whitney tests significant: $p\leq 0.0043$ at corrected $\alpha\leq 0.0064$; Fig. 4). We could not detect a difference between either of the tro-

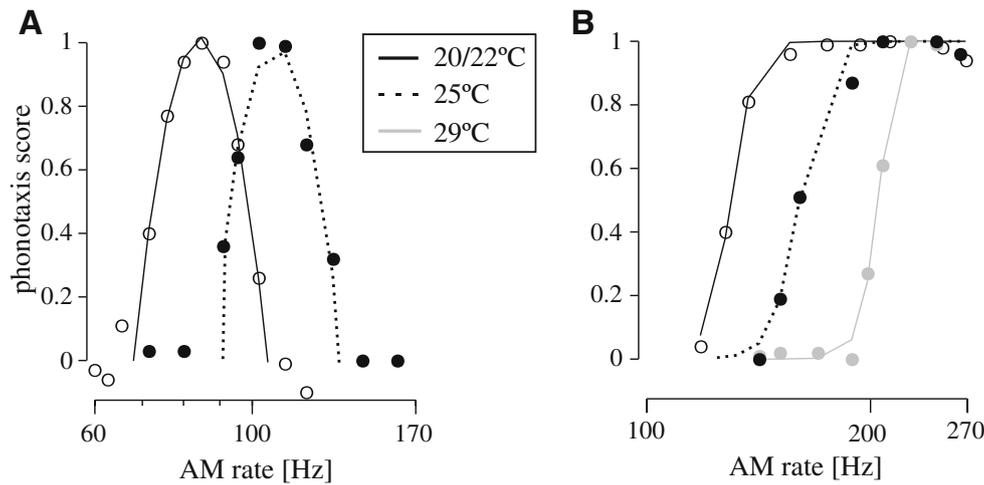


Fig. 2 Representative examples of female preference data and best-fit functions of species using AM rate (a) or interval duration (b) for call recognition at different ambient temperatures. We fitted either a parabolic function ($y = c - a^*(x - b)^2$) to the preference function of populations/species that use AM rate for call recognition (a), or a

sigmoidal function ($y = 1/(1 + e^{(-x+a)^*b})$) for species that use the interval duration for call recognition (b). The lower cutoff rate was determined as the AM rate at which the fitted function fell below 70% towards lower AM rates. Note the shift of the preferred AM rate towards higher rates with increasing ambient temperatures

pical *N. triops* populations and the remaining species (all post-hoc Mann–Whitney tests not significant: $p \geq 0.0142$ at corrected $\alpha \leq 0.0073$), except for the comparison between *N. triops* from Puerto Rico and *N. affinis* (post-hoc Mann–Whitney test: $p = 0.0029$ at corrected $\alpha = 0.0057$).

Discussion

Our experiments tested two hypotheses about the co-evolution of male-call plasticity and female preferences in *N. triops*. We found that tropical populations exhibited developmental plasticity of male calls to a similar degree as

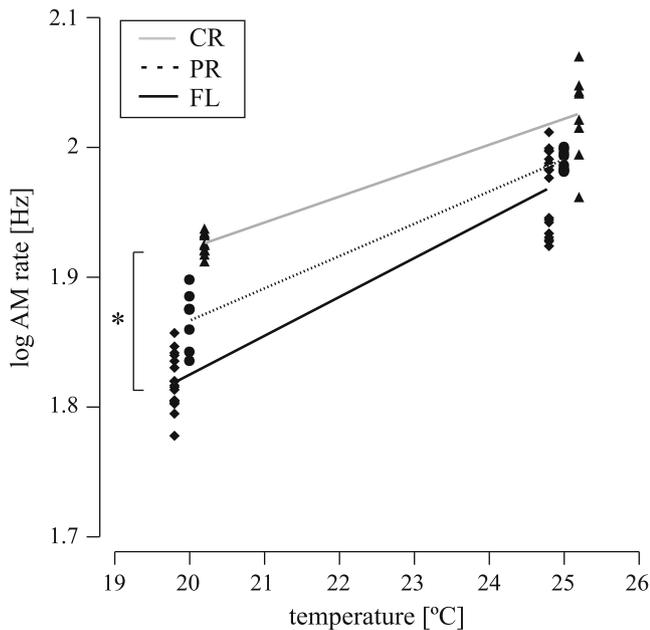


Fig. 3 Individual lower cutoff rates (log-transformed) of female preference for AM rate of *N. triops* from Florida, Puerto Rico, and Costa Rica tested at 20°C and 25°C ambient temperature. For each population, the mean cutoff rates between the two temperatures are connected by lines for visual presentation only. Cutoff rates of the Florida population changed significantly more with temperature than those of the Costa Rica population (indicated by an asterisk)

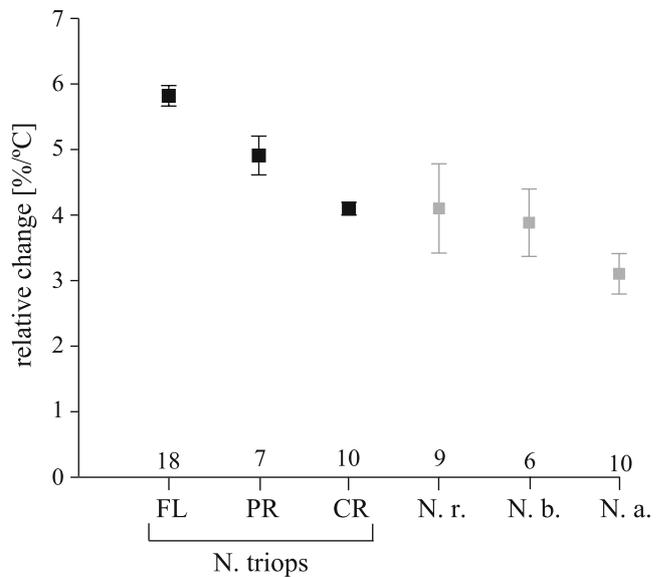


Fig. 4 Average relative change (\pm SEM) of female preference for AM rate per degree Celsius of *N. triops* from Florida (FL), Puerto Rico (PR), Costa Rica (CR), *N. robustus* (*N. r.*), *N. bivocatus* (*N. b.*), and *N. affinis* (*N. a.*). Temperature dependence of female preference of Florida *N. triops* was significantly steeper than those of the Costa Rica population and all other species of *Neoconocephalus* tested. *N* for each population/species is given at the bottom of the figure

the temperate population when reared under summer and winter conditions. In contrast, temperate *N. triops* differed significantly in the temperature dependence of female preference from the Costa Rica population, as well as from other *Neoconocephalus* species that do not express call plasticity. These results support the hypothesis that male call plasticity drove the evolution of female temperature-dependent call preferences in temperate *N. triops*, and thus rejects the hypothesis that the temperature dependence of female preferences drove the evolution of male call plasticity

Origin of male call plasticity

The genus *Neoconocephalus* (and its likely old-world sibling *Ruspolia*) is considered to be a tropical group with some species having secondarily moved into temperate regions (Greenfield 1990). The range of *N. triops* encompasses most of tropical South and Central America and the Caribbean (Walker and Greenfield 1983; Greenfield 1990). At its northern edge, the range extends into the southern part of North America (Whitesell 1974); whether *N. triops* also extends into temperate regions in South America is not known. Accordingly, *N. triops* has a life history typical for tropical katydids: eggs develop immediately after oviposition (Whitesell 1974). In tropical habitats, adult *N. triops* are present throughout the year (Greenfield 1990), while in temperate habitats, *N. triops* overwinter as adults (Whitesell 1974). In contrast, other temperate *Neoconocephalus* species overwinter in the egg stage and have only one reproductive generation per year (Greenfield 1990). The life history data (Whitesell 1974) thus indicate a tropical origin of *N. triops*.

Tropical *N. triops* in the field do not produce the winter call with the slow AM rate (Greenfield 1990; Walker and Greenfield 1983) because they do not experience photoperiods short enough to induce this call phenotype. However, when reared under these conditions in our experiments, tropical males displayed a similar developmental plasticity of AM rate as males from temperate populations (Fig. 1). Thus, the developmental plasticity of the AM rate of male calls is a trait shared among temperate and tropical *N. triops* populations. This suggests that call plasticity is an ancestral trait of *N. triops* that originated in the tropics, rather than having evolved as response to the temperate environment. The developmental plasticity of the calls is most likely not adaptive, as one phenotype (i.e. winter call) is never expressed in the environment where the developmental plasticity likely evolved (tropics).

It is possible that developmental plasticity of male calls evolved in temperate regions (where males actually express the different call phenotypes) and spread through gene flow into tropical populations. This, however, appears unlikely.

The significant differences in AM rate of male calls (Fig. 1) and the differences in temperature dependence of female preference (Fig. 3) among the three populations suggest that there is only a low level of gene flow among the *N. triops* populations studied.

Temperature dependence of male calls and female preferences

The temporal parameters of insect calls are in most cases strongly temperature-dependent (e.g. Walker 1957, 1975; Martin et al. 2000; Fonseca and Revez 2002). Female preferences have usually a similar ambient-temperature dependence (e.g. von Helversen and von Helversen 1981; Doherty 1985; Pires and Hoy 1992) to maintain the functionality of the communication system across the relevant range of temperatures.

AM rates changed to a similar degree with temperature among the three populations of *N. triops* and the three other species reported here ($\sim 2.0\text{--}3.5\%/^{\circ}\text{C}$; Table 1). Temperature dependence of female preference appeared to be well-matched to that of male calls in *N. triops* from Costa Rica and the other three *Neoconocephalus* species tested here (Fig. 4), indicating compatibility between male calls and female preferences over a wide range of ambient temperatures.

In contrast, female preference of the north-central Florida population of *N. triops* changed substantially more with temperature ($5.82\%/^{\circ}\text{C}$; Fig. 4) than did the AM rate in male calls. Thus, male calls and female preference overlap only in a narrow temperature range for each of the two call phenotypes (Beckers and Schul 2008). However, because of the developmental plasticity of the AM rate, the slower winter calls are matched to female preferences at low temperatures, and the faster summer calls to female preferences at higher temperatures (Beckers and Schul 2008). Thus, developmental plasticity of AM rate, together with the steeper temperature dependence of female preferences, results in a communication system that is well-matched in the diverse environments experienced by the two reproductive generations of *N. triops* in Florida (Beckers and Schul 2008).

The evolution of plasticity

Based on our results and the above reasoning, the following scenario appears likely for the evolution of call plasticity and female preferences in *N. triops*. Ancestral tropical populations of *N. triops* were similar to today's Costa Rica population: males possessed the developmental plasticity for AM rates (but did not express the winter AM rate due to a lack of short photoperiod), and the ambient-temperature dependence of female preference was matched to the AM

rate of the summer call. When this ancestral population expanded its range into temperate zones (e.g. north-central Florida), males expressed in the winter the winter-call phenotype with a 20% slower AM rate than the summer-call phenotype. This resulted in a distinct mismatch between male winter calls and female preference (Fig. 5a), as female preference was unaffected by the environmental conditions during development (Beckers and Schul 2008). This mismatch would lead to male winter calls being unattractive to females and female responses to this call phenotype in turn being slow and ineffective. Responses of female katydids to unattractive calls are slower and less directed than to attractive calls (e.g. Schul 1998; Schul et al. 1998). Acoustic communication would still succeed to bring males and females together, but in an inefficient way. Over time, selection on female preferences for more rapid mating (see below) resulted in an increase in the steepness of temperature dependence and hence, eliminated this mismatch (Fig. 5b). As a result, matches between female preference and AM rate of male calls were achieved in both seasons.

The proposed evolutionary scenario requires strong selective pressure for effective female phonotaxis, as well as some constraint limiting changes of the developmental plasticity of male calls. Efficient phonotaxis should always be advantageous for females because it decreases the cost of phonotaxis, such as energetic costs, predation risk, and

the cost of lost fecundity due to delayed mating. Additionally, females may compete for the most attractive males if males mate only once per night. The environmental conditions experienced by the winter generation may exacerbate selection for efficient phonotaxis because colder temperatures limit the daily time for calling and mating and thus the time available each night for a female to mate, potentially resulting in increased competition among females. Also, predation risk in the winter may be increased because of the absence of other katydids during this time of the year. Efficient phonotaxis may thus be a strong selective advantage for winter females.

It is commonly suggested that male calls would be under strong selection exerted by female preferences (see below). In temperate *N. triops* this has apparently not happened. Plasticity of traits may evolve through random changes not driven by selection (i.e. drift) or by selection on correlated traits (Schlichting 1986). In *N. triops*, developmental plasticity of the AM rate may be genetically linked to other (adaptive) traits through pleiotropy. The magnitude and sign of correlations among traits can influence the outcome of selection on these traits (Schlichting 1986), possibly limiting the ability of one trait (plasticity of male calls) to evolve (Lande 1982; Loeschke 1987; Newman 1994). If the gene(s) causing developmental plasticity of the calls is also involved in other, adaptive processes, it might be impossible for the call properties to adapt to female

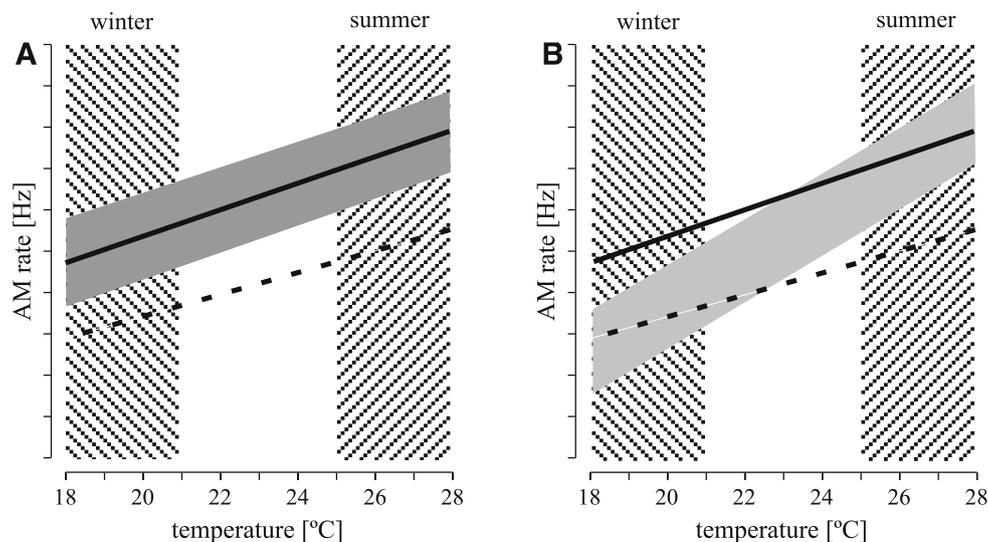


Fig. 5 Hypothetical model of the evolution of female preference in the population of *N. triops* from north-central Florida. The temperature dependence of the summer call is indicated by the *solid line*, that of the winter call by the *dashed line*. *Hatched vertical bars* indicate approximate temperature ranges after sunset for the summer and winter generations. **a** Hypothetical situation immediately after *N. triops* moved into temperate regions. Female preferences (*dark gray area*) are as in tropical populations: they match the AM rate of the

summer call over a broad temperature range, while the AM rate of the winter call is outside of the attractive range of female preferences. **b** Extant situation in temperate populations. *Light gray area* indicates temperature dependence of female preference after evolutionary change in response to expressed male-call plasticity. Female preference matches the summer AM rate at high temperatures and the winter AM rate at low temperatures (modified from Beckers and Schul 2008)

preferences. The change of AM rate between summer and winter generations was similar in the three *N. triops* populations (Fig. 1), suggesting that this trait has not diverged among these populations.

Developmental plasticity has been recognized as a potent source for phenotypic change (review in West-Eberhard 2003). The role of developmental plasticity in adaptive evolution has usually been described in the context of generating the necessary phenotypic variation for selection to act upon (West-Eberhard 2003). However, in *N. triops* adaptive evolution seemingly did not occur in the trait that exhibits developmental plasticity (male calls) but in a correlated trait (female preference) to compensate for the plasticity. Thus, the communication system of *N. triops* exemplifies a case in which developmental plasticity led to adaptive phenotypic change in an unusual way.

Our results indicate that the developmental plasticity of male calls led to changes in female preference. This finding contradicts the conventional view that male signals should evolve in response to female preferences rather than vice versa (e.g. Andersson 1994; Endler and Basolo 1998; Rodriguez et al. 2006). We interpreted our initial findings (Beckers and Schul 2008) accordingly arguing that the developmental plasticity of male calls likely evolved in response to the temperature dependence of female preferences. However, the comparative study conducted here, which includes several populations of *N. triops*, as well as three closely related species, led to the opposite (and unexpected) conclusion that female preferences changed in response to male calls.

Our results indicate that female adaptations to male traits may be more pronounced than male adaptation to female traits, even in systems with classical sex roles. This highlights the need to consider alternative hypotheses, even if they appear unlikely, when designing studies of evolutionary processes.

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References

- Andersson M (1994) Sexual selection. Princeton University Press, NJ
- Beckers OM, Schul J (2008) Developmental plasticity of mating calls enables acoustic communication in diverse environments. *Proc R Soc Lond B* 275:1243–1248
- Bush SL, Gerhardt HC, Schul J (2002) Pattern recognition and call preferences in treefrogs (Anura: Hylidae): a quantitative analysis using a no-choice paradigm. *Anim Behav* 63:7–14
- Bush SL, Beckers OM, Schul J (2009) A complex mechanism of call recognition in the katydid *Neoconocephalus affinis* (Orthoptera: Tettigoniidae). *J Exp Biol* 212:648–655
- Büttner UK (2002) Charakterisierung der Gesänge von fünf in Missouri (USA) heimischen *Neoconocephalus*-Arten (Orthoptera, Tettigoniidae). Diploma thesis, University of Erlangen, Germany
- Day RW, Quinn GP (1989) Comparisons of treatments after an analysis of variance in ecology. *Ecol Monogr* 59(4):433–463
- Deily JA, Schul J (2004) Recognition of calls with exceptionally fast pulse rates: female phonotaxis in the genus *Neoconocephalus* (Orthoptera: Tettigoniidae). *J Exp Biol* 201:3523–3529
- Doherty JA (1985) Temperature coupling and “trade-off” phenomena in the acoustic communication system of the cricket, *Gryllus bimaculatus* De Geer (Gryllidae). *J Exp Biol* 144:17–35
- Endler JA, Basolo AL (1998) Sensory ecology, receiver biases and sexual selection. *Trends Ecol Evol* 13(10):415–420
- Fonseca PJ, Revez MA (2002) Temperature dependence of cicada song (Homoptera, Cicadoidea). *J Comp Physiol A* 187:971–976
- Gerhardt HC (1991) Female mate choice in treefrogs: static and dynamic acoustic criteria. *Anim Behav* 42:615–635
- Gerhardt HC, Huber F (2002) Acoustic communication in insects and anurans. University of Chicago Press, Chicago
- Ghalambor CK, McKay JK, Carroll SP, Reznik DN (2007) Adaptive versus non-adaptive phenotypic plasticity and the potential for contemporary adaptation in new environments. *Funct Ecol* 21:394–407
- Grace JL, Shaw KL (2004) Effects of developmental environment on signal preference coupling in a Hawaiian cricket. *Evolution* 58:1627–1633
- Greenfield MD (1990) Evolution of acoustic communication in the genus *Neoconocephalus*: discontinuous songs, synchrony, and heterospecific interactions. In: Bailey WJ, Rentz DCF (eds) *The Tettigoniidae: biology, systematics and evolution*. Springer, Heidelberg, pp 71–97
- von Helversen D, von Helversen O (1981) Korrespondenz zwischen Gesang und auslösendem Schema bei Feldheuschrecken. *Nova acta Leopold* 54(245):449–462
- Hill GE, Inouye CY, Montgomerie R (2002) Dietary carotenoids predict plumage coloration in wild house finches. *Proc R Soc Lond B* 269:1119–1124
- Hoikkala A, Isoherranen E (1997) Variation and repeatability of courtship song characters among wild-caught and laboratory-reared *Drosophila montana* and *D. littoralis* males (Diptera: Drosophilidae). *J Insect Behav* 10:193–202
- Kenyon SG, Hunter MS (2007) Manipulation of oviposition choice of the parasitoid wasp, *Encarsia pergandiella*, by the endosymbiotic bacterium *Cardinium*. *J Evol Biol* 20:707–716
- Lande R (1982) A quantitative genetic theory of life history evolution. *Ecology* 63:607–615
- Laurila A, Karttunen S, Merilä J (2002) Adaptive phenotypic plasticity and genetics of larval histories in two *Rana temporaria* populations. *Evolution* 56(3):617–627
- Loeschke V (1987) Genetic constraints on adaptive evolution. Springer, Berlin
- Martin SD, Gray DA, Cade WH (2000) Fine-scale temperature effects on cricket calling song. *Can J Zool* 78:706–712
- Naskrecki P (2000) Katydid of Costa Rica. Systematics and bioacoustics of the cone-head katydids (Orthoptera: Tettigoniidae: Conecephalinae sensu lato). Orthopterists Society, Philadelphia, Pennsylvania
- Newman RA (1994) Genetic variation for phenotypic plasticity in the larval life history of spadefoot toads (*Scaphiopus couchii*). *Evolution* 48(6):1773–1785

- Olvido AE, Mousseau TA (1995) Effect of rearing environment on calling song plasticity in the striped ground cricket. *Evolution* 49(6):1271–1277
- Pires A, Hoy RR (1992) Temperature coupling in cricket acoustic communication I. Field and Laboratory studies of temperature effects on calling song production and recognition in *Gryllus firmus*. *J Comp Physiol A*: 69–78
- Reinhold R (1983) Houston is at the five plagues and counting. *New York Times*: 11/13/1983.
- Rodriguez RL, Ramaswamy K, Cocroft RB (2006) Evidence that female preferences have shaped male signal evolution in a clade of specialized plant-feeding insects. *Proc R Soc Lond B* 273:2585–2593
- Roy HE, Steinkraus DC, Eilenberg J, Hajek AE, Pell JK (2006) Bizarre interactions and endgames: entomopathogenic fungi and their arthropod hosts. *Annu Rev Entomol* 51:331–357
- Schlichting CD (1986) The evolution of phenotypic plasticity in plants. *Ann Rev Ecol Syst* 17:667–693
- Schlichting CD, Pigliucci M (1998) Phenotypic evolution: a reaction norm perspective. Sinauer Associates, Sunderland
- Schul J (1998) Song recognition by temporal cues in a group of closely related bushcricket species (Genus *Tettigonia*). *J Comp Physiol A* 183:401–410
- Schul J, Patterson AC (2003) What determines the tuning of hearing organs and the frequency of calls? A comparative study in the katydid genus *Neoconocephalus* (Orthoptera; Tettigoniidae). *J Exp Biol* 206:141–152
- Schul J, von Helversen D, Weber T (1998) Selective phonotaxis in *Tettigonia cantans* and *T. viridissima* in song recognition and discrimination. *J Comp Physiol A* 182:687–694
- Snyder RL, Frederick-Hudson KH, Schul J (2009) Molecular phylogenetics of the genus *Neoconocephalus* (Orthoptera, Tettigoniidae) and the evolution of temperate life histories. *PLoS ONE* 4(9):e7203
- Sokal RR, Rohlf FJ (1997) *Biometry*. W. H. Freeman And Company, New York
- Speaks CE (1999) *Introduction to sound*. Singular Publishing Group Inc., San Diego
- Stearns SC (1989) The evolutionary significance of phenotypic plasticity. *Bioscience* 39:436–445
- Trussel GC, Smith LD (2000) Induced defenses in response to an invading crab predator: an explanation of historical and geographic phenotypic change. *Proc Natl Acad Sci USA* 97:2123–2127
- Van Tienderen PH (1991) Evolution of generalists and specialists in spatially heterogeneous environments. *Evolution* 45(6):1327–1331
- Wagner WE, Hoback WW (1999) Nutritional effects on male calling behaviour in the variable field cricket. *Anim Behav* 57:89–95
- Walker TJ (1957) Specificity in the response of female tree crickets (Orthoptera, Gryllidae, Oecanthinae) to calling songs of the males. *Ann Entomol Soc Am* 50:626–636
- Walker TJ (1975) Effects of temperature on rates in poikilotherm nervous systems: evidence from the calling songs of meadow katydids (Orthoptera: Tettigoniidae: *Orchelimum*) and reanalysis of published data. *J Comp Physiol* 101:57–69
- Walker TJ (2000) Pulse rates in the songs of trilling field crickets (Orthoptera: Gryllidae: *Gryllus*). *Entom Soc Am* 93(3):565–572
- Walker TJ, Greenfield MD (1983) Songs and systematics of Caribbean *Neoconocephalus* (Orthoptera: Tettigoniidae). *Trans Am Entomol Soc* 109:357–389
- Walker TJ, Whitesell JJ, Alexander RD (1973) The robust conehead: two widespread sibling species (Orthoptera: Tettigoniidae: *Neoconocephalus* ‘robustus’). *Ohio J Sci* 73(6):321–330
- Weber T, Thorson J, Huber F (1981) Auditory behaviour of the cricket. I. Dynamics of compensated walking and discrimination paradigms on the Kramer treadmill. *J Comp Physiol* 141:215–232
- West-Eberhard MJ (2003) *Developmental plasticity and evolution*. Oxford University Press, Oxford
- West-Eberhard MJ (2005) Developmental plasticity and the origin of species differences. *Proc Natl Acad Sci USA* 102:6543–6549
- Whitesell JJ (1974) *Geographic variation and dimorphisms in song, development, and color in a katydid: field and laboratory studies* (Tettigoniidae, Orthoptera). Ph D dissertation. University of Florida, Gainesville
- Whitesell JJ, Walker TJ (1978) Photoperiodically determined dimorphic calling songs in a katydid. *Nature* 274:887–888
- Whitman DW, Agrawal AA (2009) What is phenotypic plasticity and why is it important? In: Whitman DW, Ananthakrishnan TN (eds) *Phenotypic plasticity in insects: mechanisms and consequences*. Science publishers, Enfield, pp 1–63
- Yeh PJ, Price TD (2004) Adaptive phenotypic plasticity and the successful colonization of a novel environment. *Am Nat* 164:531–542
- Zar JH (1999) *Biostatistical analysis*. Prentice Hall, Upper Saddle River