Multitasking males and multiplicative females: dynamic signalling and receiver preferences in Cope's grey treefrog

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The 'multitasking hypothesis' for complex signal function predicts performance trade-offs between signal components that negatively covary (e.g. due to energetic or mechanical constraints) and receiver preferences for more extreme values of the negatively covarying components that are difficult to produce simultaneously. We tested these two predictions in Cope's grey treefrogs, *Hyla chrysoscelis*. In a field study, we recorded and analysed 1000 advertisement calls from males calling in breeding choruses to test the prediction that two signal components important in female mating decisions (call rate and call duration) negatively covary under natural conditions. In a laboratory study, we conducted phonotaxis tests with female subjects to test the prediction that females prefer calls with higher overall 'call efforts' (the product of call rate × call duration). Consistent with predictions of the multitasking hypothesis, call rate and call duration were significantly negatively related and females preferred calls produced with higher call efforts, manifested through preferences for greater values of both call rate and call duration. We conducted an additional playback experiment to test the hypothesis that males increase their call effort in competitive situations to maximize their attractiveness to females. Compared to quiet conditions, male subjects increased their call duration and decreased their call rate, but did not alter call effort, in response to a simulated calling neighbour or broadcasts of chorus noise. Together our data have implications for understanding the function of multicomponent signals when signallers must balance performance trade-offs in mate attraction with dynamic signal modifications in other social contexts.

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Several hypotheses have been proposed to explain the function of complex signals (reviewed in Candolin 2003; Hebets & Papaj 2005; Bro-Jørgensen 2010). For example, properties of different signals or multiple signal components may be correlated with either different attributes (the 'multiple messages hypothesis') or the same attributes (the 'redundant signal hypothesis' or 'back-up signal hypothesis') of the signaller or the environment (Møller & Pomiankowski 1993; Johnstone 1996; Hebets & Papaj 2005). Signals or signal components also may be functionally related to environmental transmission media (the 'sensory environment hypothesis') and the sensory system of the receiver (the 'sensory constraint hypothesis') (Candolin 2003; Hebets & Papaj 2005). Hypotheses of complex signal evolution often assume that different signals or signal components are produced by senders independently of each other, processed by receivers independently of each other, or both. However, signals and signal components can interact in complex ways and have a variety of dependent effects on receivers. Hebets & Papaj (2005) suggested that a separate class of 'intersignal interaction hypotheses' may better explain the function of complex signals in such cases (see also Partan & Marler 2005).

In the present study, we tested an intersignal interaction hypothesis termed the 'multitasking hypothesis' (sensu Hebets & Papaj 2005). According to the multitasking hypothesis, a signaller's ability to generate one signal component is constrained by, and thus negatively correlated with, their ability to generate another signal component. In addition, receiver responses may generally favour the joint production of more extreme values of two negatively covarying signal components; signallers capable of producing these presumably more costly or difficult-to-produce signal combinations should be high-quality individuals capable of so-called multitasking (Hebets & Papaj 2005).

Currently, the best evidence supporting a multitasking hypothesis comes from studies of songbirds, in which performance trade-offs impose negative covariance on the trill rate and frequency bandwidth of male songs (Podos 1997; Draganoiu et al.

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2002; Ballentine et al. 2004; Beebee 2004; Illes et al. 2006; Cramer & Price 2007). Female songbirds show directional preferences for male songs with high trill rates and wide frequency bandwidths (Draganoiu et al. 2002; Ballentine et al. 2004). In at least one species (*Melospiza georgiana*), females prefer songs with signal values close to vocal performance limits defined by the upper-bound regression of frequency bandwidth on trill rate, which is a reliable index of male age and size (Ballentine et al. 2004; Ballentine 2009).

Here, we report results from a test of the multitasking hypothesis in Cope's grey treefrog, *Hyla chrysoscelis*. Males of this species produce pulsatile advertisement calls to attract females, and females choose males on the basis of multiple signal components (reviewed in Gerhardt 2001; Gerhardt & Huber 2002). We focused on female mate choice based on three components of male signalling behaviour, including call rate (calls/min), call duration (pulses/call) and the arithmetic product of these two components, which is termed 'call effort' and measured in units of pulses/min (calls/min × pulses/call = pulses/min). We tested two key predictions of the multitasking hypothesis (Hebets & Papaj 2005). First, we tested the prediction that call rate and call duration show the negative covariance indicative of a performance trade-off (e.g. Wells & Taigen 1986). Second, we tested the prediction that females have directional preferences for higher call efforts, manifest through preferences for higher call rates or longer call durations. Finally, because frogs are well known for dynamically adjusting their calling behaviour in competitive social environments (reviewed in Schwartz & Bee, in press), we tested the additional hypothesis that multitasking males adjust their calling performance in ways that maximize their attractiveness to females.

TRADE-OFFS IN CALLING PERFORMANCE

According to the multitasking hypothesis, signal components of interest to receivers should exhibit negative covariance (Hebets & Papaj 2005). We used acoustical and statistical analyses of calls recorded in natural breeding choruses to test the prediction that call rate and call duration are negatively related in Cope's grey treefrogs.

Methods

Subjects

Between May and July of 2006 and 2009, we recorded 20 calls from each of 50 males (1000 calls total). Calls were recorded between 2200 and 0100 hours during active choruses at four field sites within our study area in east-central Minnesota, U.S.A.: Carver Park Reserve (Carver County: 44°55′06″N, 93°23′42″W; $N = 28$ males), Crow-Hassan Park Reserve (Hennepin County: 45°11′30.67″N, 93°39′03.16″W; $N = 10$ males), Tamarack Nature Center (Ramsey County: 45°06′02″N, 93°02′09″W; $N = 3$ males) and Lake Maria State Park (Wright County: 45°01′17″N, 93°30′21″W; $N = 9$ males). The mean pairwise distance between these field sites is 49 km (range 27−75 km). Cope's grey treefrogs in these locales are of the western mitochondrial DNA lineage (Ptacek et al. 1994).

Recordings

We recorded calls using a Sennheiser ME66 microphone and K6 power supply (Sennheiser USA, Old Lyme, CT, U.S.A.) connected to a Marantz PMD670 recorder (D&M Professional, Itasca, IL, U.S.A.) (44.1 kHz sampling rate, 16-bit resolution). The recording tip of the microphone was aimed directly at the focal male and was held approximately 1 m from the frog. We used a quick-reading Miller & Weber thermometer (Avinet Inc., Dryden, NY, U.S.A.) to record the

water and wet-bulb air temperatures at the calling location of each male to the nearest 0.1 °C. The mean (±SD, here and elsewhere) water and wet-bulb air temperatures were 22.5 ± 3.1 °C ($N = 50$, range 15.0−29.0 °C) and 17.3 ± 3.6 °C ($N = 50$, range 10.8−24.0 °C), respectively. We captured a subset of recorded males ($N = 44$) and measured their snout−vent lengths (SVL) to the nearest 0.1 mm using dial callipers and body masses (to the nearest 0.1 g) using Pesola spring scales. The mean SVL and mass were, respectively, 38.8 ± 2.4 mm ($N = 44$, range 33.6–43.5 mm) and 4.3 ± 0.9 g ($N = 44$, range 2.8–5.9 g). Following Baker (1992), we computed an index of body condition (length-independent mass) by dividing the residuals from a linear regression of the cube root of mass on SVL by SVL. Males were released at their calling sites immediately after measurements of temperature, SVL and mass were completed. We did not mark individual males (e.g. by toe clipping). Instead, we attempted to ensure that no male was recorded more than once by making recordings in widely separated areas both within and between nights (and years) at any one particular field site. Populations of grey treefrogs at our field sites are very large, and active choruses are quite dense. We believe the risk of pseudoreplication in our sample of recordings is negligible; we chose to tolerate this level of risk over the possible risk of potential harm resulting from removing toes (Perry et al. 2011).

Acoustical analyses

In total, we measured or calculated 16 acoustic properties for each of the 1000 calls in our recordings using Raven v1.3 (Cornell Lab of Ornithology, Ithaca, NY, U.S.A.). Full results of these acoustical analyses are reported in the Supplementary Material. Here, we focus on three properties: call duration (pulses/call), call rate (calls/min) and call effort (pulses/min). We counted the number of pulses per call directly from Raven's waveform display. We used cursors in the waveform display to measure call duration (in seconds) and the subsequent intercall interval (also in seconds) so that we could calculate an instantaneous call rate for each call as the inverse of the instantaneous call period (where call period (s)=call duration (s) + intercall interval (s)). We computed an instantaneous call rate for each call (as opposed to computing one estimate of call rate over the total time required for 20 full call periods) so that we could also estimate within-individual variation in call rate (see Supplementary Material). We adjusted values of call duration (pulses/call) and call rate (calls/min) to 20 °C (Value₂₀) according to the equation:

$$Value_{20} = Value_T - m(T - 20) \tag{1}$$

where T represents the recorded temperature, $Value_T$ represents the value of the measured signal property and m represents the slope of the regression of $Value_T$ on T using all 1000 calls (Platz & Forester 1988). For each male, the most appropriate temperature was selected based on his calling position at the time the recording was made; we used water temperature for males calling from the surface of (and in contact with) the water and wet-bulb air temperature for males calling from emergent vegetation with their bodies out of the water. We computed an estimate of instantaneous call effort (pulses/min at 20 °C) for each call as the product of its temperature-adjusted instantaneous call rate (calls/min at 20 °C) and call duration (pulses/call at 20 °C). We averaged the 20 temperature-adjusted values of each call property for each male prior to statistical analysis unless indicated otherwise.

Statistical analyses

We tested the prediction of the multitasking hypothesis that call duration and call rate negatively covary using linear regression on mean values for each male ($N = 50$). We also used linear regression to examine the relationships between call effort and both call rate and call duration. For each individual, we also computed a Pearson

product-moment correlation to assess the relationship between instantaneous call rate and call duration across the 20 calls recorded for that individual. We assessed the effects of male body size on mean call rate, call duration and call effort via separate multiple linear regressions with SVL and mass specified as predictor variables ($N = 44$). We fitted additional linear regression models assessing the relationships between each call property and body condition ($N = 44$); condition was not included in the analyses with body size due to issues of multicollinearity. For all analyses, we used $\alpha = 0.05$ as a significance criterion. Our data met the assumptions of parametric statistics. All statistical analyses were conducted with SPSS v.17.0.2 (Armonk, NY, U.S.A.).

RESULTS

Mean call rate ranged from 6.1 to 19.9 calls/min ($\overline{X} = 11.4 \pm 2.9$ calls/min; $N = 50$) and mean call duration ranged from 22 to 38 pulses ($\overline{X} = 30 \pm 4$ pulses; $N = 50$). The mean call effort was 333.9 ± 65.9 pulses/min (range 226.9–523.7 pulses/min; $N = 50$). Descriptions of within-individual and among-individual variation (coefficients of variation) are reported in the Supplementary Material for these and other call properties. As illustrated with two exemplars in Fig. 1a, males that called at faster rates tended to produce shorter calls. Across all 50 individuals, call rate and call duration were significantly negatively related
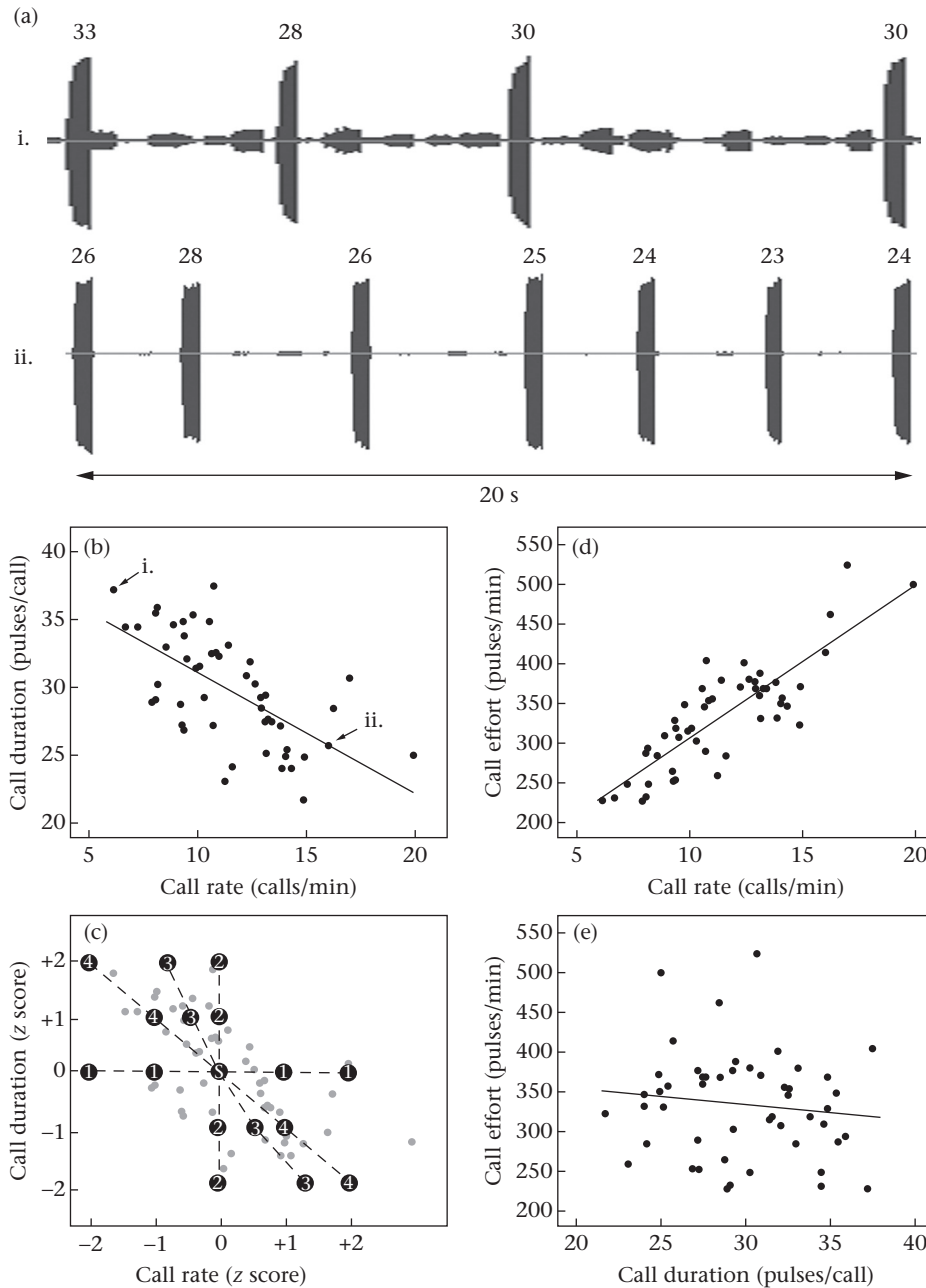
Figure 1. Relationships among call components for males calling in natural choruses ($N = 50$). (a) Two representative sound clips of different males (i and ii) recorded in different choruses on different nights; note that the male calling at the relatively faster rate (ii) produced relatively shorter calls. (b) Scatterplot showing a negative relationship between call duration and call rate; note that mean values for males i and ii from panel (a) are also depicted in the scatterplot. (c) Data for individual means from (b) (small grey circles) depicting values for the acoustic stimuli used in female choice experiments (large black circles), expressed in terms of z scores (± 1 SD or ± 2 SD) away from the population average. Numbers 1–4 refer to the experiment in which the stimulus having the indicated combination of call rate and call duration was used (see Table 1). The 'S' indicates the values used for the standard call. (d, e) Scatterplots showing the relationships between call effort and (d) call rate and (e) call duration.

($r^2 = 0.40$ $F_{1,48} = 32.27$, $P < 0.001$; Fig. 1b). We found a significant positive relationship between call rate and call effort ($r^2 = 0.70$, $F_{1,48} = 113.40$, $P < 0.001$; Fig. 1d), but no relationship between call duration and call effort ($r^2 = 0.02$, $F_{1,48} = 0.79$, $P = 0.377$; Fig. 1e). Across the 20 calls recorded within individuals, the correlations between call rate and call duration ranged widely ($-0.65 < r < 0.57$) and reached statistical significance for only 5 of 50 individuals (one significantly negative and four significantly positive). The mean correlation coefficient was $r = 0.11 \pm 0.26$ ($N = 50$ correlations). There were no significant relationships between calling behaviour and body size (SVL and mass) or condition (see Supplementary Material).

FEMALE PREFERENCES

A prediction of the multitasking hypothesis is that receivers prefer signallers that overcome signalling constraints to produce signals with more extreme values of negatively covarying components (Hebets & Papaj 2005). In the case of Cope's grey treefrogs, in which call rate and call duration are inversely related, a multitasking hypothesis predicts females should have directional preferences for callers with high overall call efforts (the product of call rate and call duration). Males potentially achieve high call efforts by calling at relatively higher call rates, producing relatively longer calls, or both. To the extent that female preferences really are multiplicative, and hence based on the product of these two interacting components, the multitasking hypothesis further predicts that females should exhibit weak or no preferences between callers differing in call rate and call duration so long as their call efforts are the same. We tested these predictions using two-stimulus choice tests that examined female preferences for call rate, call duration and call effort. We replicated all choice tests in quiet and in the presence of artificial chorus noise to assess the stability of preferences under both optimal and more naturalistic listening conditions.

Methods

Subjects

We conducted phonotaxis tests with gravid females between May and July 2012. Our procedures for collecting, handling and testing females closely followed protocols described in detail in our earlier studies of this species (Bee 2007, 2008a, 2008b, 2010; Bee & Schwartz 2009; Vélez & Bee 2010, 2011); readers are referred to these previous studies for methodological details not reported here. Briefly, subjects were collected in amplexus from ponds located at the field sites described above and returned to the laboratory where they were maintained at approximately 2 °C to prevent egg deposition until the time of testing. At least 30 min prior to testing, we placed subjects in an incubator to permit their body temperature to reach 20 ± 1 °C, the temperature at which all tests were conducted. Females were tested and returned to their location of collection within 2–3 days.

Experimental design

We conducted 32 two-stimulus choice tests. In each test, we presented females ($N = 32$/test) with two synthetic calls differing in some combination of call rate, call duration and call effort (Table 1). Our tests were organized into four separate experiments; each experiment comprised four separate tests and was replicated twice, once in quiet and once in noise (4 experiments × 4 tests/ experiment × 2 replicates = 32 separate tests). In all tests, one stimulus was a 'standard call' with values of call rate (11 calls/min), call duration (30 pulses/call) and call effort (330 pulses/min) near the population averages reported above. Properties of the 'alternative call' stimulus were based on the fact that call rate, call duration or call effort is constrained if values of the other two properties are specified by the experimenter (Table 1). As illustrated in Fig. 1c, we chose values of call rate and call duration for the alternative calls that sampled the bivariate distribution of these two call properties.

In experiments 1–3, one property of the alternative call was systematically manipulated across the four tests (± 1 SD and ± 2 SD relative to the population average); a second property was held constant at the population average; the third property was free to vary across the four tests given the specified values of the other two properties (Table 1). In experiment 1 (Table 1, Fig. 1c), we held call duration constant and tested the hypothesis that females prefer calls produced at higher rates (and thus higher call efforts also). In experiment 2 (Table 1, Fig. 1c), we held call rate constant and tested the hypothesis that females prefer calls with longer durations (and

Table 1
Experimental design and stimulus call properties used in two-stimulus choice tests to measure female preference functions

Experiment	Manipulated property	Constant property	Freely varying property	Alternative call property values		
				Call rate (calls/min)	Call duration (pulses/call)	Call effort (pulses/min)
1	Call rate	Call duration	Call effort	5	30	150
				8	30	240
				14	30	420
				17	30	510
2	Call duration	Call rate	Call effort	11	22	242
				11	26	286
				11	34	374
				11	38	418
3	Call duration	Call effort	Call rate	8.68	22	330
				9.71	26	330
				12.69	34	330
				15.00	38	330
4	Call duration and call rate	—	Call effort	5	38	190
				8	34	272
				14	26	364
				17	22	374

Females were given a choice between a standard call (30 pulses/call, 11 calls/min, 330 pulses/min) and an alternative call that differed in two or more properties. Manipulated call properties deviated from values in the standard call by ± 1 or ± 2 SD based on estimates of these population parameters. Constant call properties matched those of the standard call. Freely varying call properties varied according to the values of constant and manipulated properties. All four experiments were replicated in quiet and noisy conditions.

thus higher call efforts also). In experiment 3 (Table 1, Fig. 1c), we held call effort constant and asked whether females prefer longer calls produced at slower rates, or shorter calls produced at faster rates. In this third experiment, we manipulated call duration by ± 1 and ± 2 SD while maintaining corresponding values of call rate within the range of natural variation among individual means. In a fourth experiment (experiment 4; Table 1, Fig. 1c), we simultaneously and orthogonally manipulated the rate and duration of the alternative call and allowed its corresponding call effort to vary accordingly. Across all four experiments, the alternative call stimuli had call rates ranging between 5 and 17 calls/min, call durations ranging between 22 and 38 pulses/call and call efforts ranging between 150 and 510 pulses/min (Table 1). These values approximate the ranges of variation in individual means we recorded in natural choruses (call rate (6.1–19.9 calls/min); call duration (22–38 pulses); call effort (226.9–523.7 pulses/min)).

Acoustic stimuli

The standard and alternative calls were composed of synthetic pulses (44.1 kHz sampling rate, 16-bit resolution) designed using MATLAB® 7.6.0 (MathWorks, Natick, MA, U.S.A.) to have temporal and spectral properties close to the average values of calls recorded in local populations at 20 °C (see Supplementary Material; M. A. Bee, unpublished data). Each pulse was 10 ms in duration and was created by adding two phase-locked sinusoids with frequencies (and relative amplitudes) of 1.25 kHz (-9 dB) and 2.5 kHz (0 dB). The pulse amplitude envelope was shaped to have an inverse exponential rise time of 3.6 ms and an exponential fall time of 6.4 ms. Pulses were separated by 10 ms interpulse intervals (20 ms pulse period; 50 pulses/s). We shaped the call amplitude envelope using a linear rise over the first 60 ms of the call. The standard call had 30 pulses, which equals the population average number of pulses in calls recorded in choruses, as reported above ($\overline{X} = 30 \pm 4$ pulses/call). The standard call repeated with a period of 5.45 s and, thus, had a call rate of 11 calls/min, which approximates the population average call rate reported above ($\overline{X} = 11.4 \pm 2.9$ calls/min). The call rates and durations of alternative calls are specified in Table 1. During a choice test, sequences of standard calls and alternative calls were presented from separate channels of a stereo sound file in which each channel was created by repeating calls having the appropriate call duration and period (reciprocal of call rate) until the sound file just exceeded 6 min in length. Whether the very first call broadcast was the standard call or an alternative call was counterbalanced across subjects; we found no effect on subject preferences related to which call started the sequence of alternating calls. In experiment 2 (constant call rate; Table 1), the standard and alternative calls alternated in time such that each was preceded and followed by an equivalent silent interval over the duration of a test. This temporal arrangement of strict alternation also applied to the first three calls in the call sequences broadcast in all other experiments; however, because call rate varied across conditions and between the standard and alternative calls in these other experiments, subsequent calls drifted in and out of phase according to their designated call rates and durations.

All tests were replicated in quiet and in the presence of an artificial noise designed to approximate the long-term frequency spectrum of grey treefrog breeding choruses. To create this 'chorus-shaped noise', we used MATLAB® 7.6.0 to filter white noise two different times to produce a low-frequency band and a high-frequency band. The centre frequencies of these bands were based on the spectra of noise that we previously recorded in 14 different choruses of grey treefrogs (Swanson et al. 2007; Vélez & Bee 2010). The low-frequency band was created using a band-pass finite impulse response (FIR) filter of order 300, with

passband frequencies of 1200 and 1300 Hz, stopband frequencies of 1000 and 1500 Hz, passband ripple of 0.1 Hz and stopband attenuation of 60 dB. The high-frequency band was created using a band-pass FIR filter of order 150, with passband frequencies of 2400 and 2600 Hz, stopband frequencies of 2000 and 3000 Hz, passband ripple of 0.1 Hz and stopband attenuation of 60 dB. The amplitude of the low-frequency band was adjusted to -6 dB in relation to the high-frequency band before adding the two noise bands together.

Apparatus

Tests were conducted under infrared (IR) lighting in a temperature-controlled ($20° \pm 1$ °C), hemi-anechoic sound chamber (2.8 × 2.3 × 2.1 m, L × W × H; Industrial Acoustics Company, IAC, Bronx, NY, U.S.A.). Acoustic insulation covered with dark grey perforated metal covered the interior ceiling and walls of the sound chamber to reduce reverberation (IAC's Planarchoic™ panel system). The floor of the sound chamber was covered in dark grey, low-pile carpet. In the centre of the chamber was a 2 m diameter circular test arena constructed with walls made from hardware mesh covered in acoustically transparent but visually opaque black cloth. In the centre of the test arena on the floor was an acoustically transparent release cage (9 cm diameter, 2 cm height). The lid of the cage could be removed to release subjects into the arena via a rope-and-pulley system operated from outside the sound chamber. Test stimuli were broadcasted from two Mod1 Orb speakers (Orb Audio, New York, NY, U.S.A.) separated by 2 m and 180° around the outside perimeter of the test arena and facing inward towards the centre of the arena. A third Mod1 Orb speaker suspended from the ceiling directly above the centre of the test arena was used to broadcast the noise. Sounds were broadcast from a Dell Optiplex 980 PC computer (Dell Computer Corporation, Round Rock, TX, U.S.A.) located outside of the chamber using Adobe Audition 1.5 (Adobe Systems Inc., San Jose, CA, U.S.A.) interfaced with an M-Audio FireWire 410 multichannel soundcard (M-Audio, Irwindale, CA, U.S.A.) and HTD DMA-1275 amplifier (Home Theater Direct, Inc., Plano, TX, U.S.A.). We calibrated the sound pressure levels (SPL re 20 µPa) of calls and noise to 85 dB SPL (LCF_{max}) and 70 dB SPL (LC_{eq}), respectively, using a Brüel & Kjær Type 2250 sound level meter (Brüel & Kjær, Norcross, GA, U.S.A.). For calibration, the microphone of the sound level meter was placed in the centre of the arena at the same position as a subject's head at the start of a test. We chose these levels because they approximate natural sound levels recorded in chorus environments (Gerhardt 1975; Swanson et al. 2007). Noise levels were uniform (± 2 dB) across the entire floor of the test arena, and the frequency response of the playback system was flat (± 3 dB) between 500 Hz and 4000 Hz. We periodically changed both the absolute and relative positions of playback speakers around the perimeter of the circular test arena to eliminate any potentially confounding effects of directional bias. Sounds were calibrated at the start of each testing day and after any speaker movements.

Protocol

We designed our testing procedures so that we maintained statistically independent samples of subjects tested in the presence and absence of the chorus-shaped noise; that is, no subject underwent the same choice tests in both the presence and absence of noise. To achieve this, we used a within-subjects design that assigned each subject to one of four different testing blocks (Table 2), each of which consisted of the four experiments described in Table 1. Two of the four experiments in each block were conducted in quiet and the other two experiments were conducted in the presence of the chorus-shaped noise. Across testing blocks, we varied which experiments were conducted in quiet and in noise (Table 2). This procedure allowed us to test the

Table 2
Blocked testing design used for female choice experiments

Block	N/experiment	Quiet replicate	Noise replicate
A	16	Experiment 1	Experiment 2
		Experiment 3	Experiment 4
B	16	Experiment 2	Experiment 1
		Experiment 4	Experiment 3
C	16	Experiment 1	Experiment 2
		Experiment 4	Experiment 3
D	16	Experiment 2	Experiment 1
		Experiment 3	Experiment 4

Each block consisted of all four experiments. Two experiments were conducted in the presence of chorus-shaped noise and two experiments were conducted in quiet ($N = 16$ subjects/experiment within each block). Responses from a total of 32 subjects were recorded for each experiment in each noise condition. No subjects were tested in the same experiment in both quiet and in noise.

effects of noise using a between-subjects design with independent samples of subjects. For each subject, we randomized the order of the four experiments as well as the order of the four tests within each experiment.

At the start of each test, a female was placed in the release cage at the centre of the test arena where she was permitted to acclimate in quiet for 1 min. After this 1 min acclimation, we exposed females in noise conditions to 30 s of chorus noise before commencing stimulus presentation. In quiet conditions, subjects experienced 30 additional seconds without noise before stimulus presentations began. After 15 s of stimulus presentation, we released the female from the cage. Female choice was determined on the basis of phonotaxis, which we scored by viewing a monitor located outside the chamber showing a view of the complete test arena as seen through an overhead CCTV camera (Panasonic WV-BP334) mounted from the centre of the sound chamber's ceiling. A response was scored when a female entered a response zone within 15 cm of a speaker. Subjects were given a 3–5 min time-out period between consecutive tests. For inclusion in the final data set we required that a subject make a choice in all four tests within a given experiment. If a female failed to make a choice within 5 min, she was removed from the experiment and replaced with a new female. We then restarted the experiment in progress with the new female. In our experience, females that do not respond within 5 min do not respond in subsequent tests either. In total, we examined the responses of 91 females; 27 females stopped responding during an experiment and were replaced.

Statistical analyses

We separately analysed the results of tests conducted under quiet conditions and in the presence of chorus-shaped noise using two-tailed binomial tests of the null hypothesis that equal proportions (0.5) of females would chose the standard call in each test. We interpreted a significant result as indicating a preference for one of the two stimuli. We compared female choices in the presence and absence of noise using chi-square tests conducted on the numbers of females that selected the standard and alternative calls in each condition. We used an experiment-wide α level of 0.05 as a significance criterion.

Results

We first consider responses in tests conducted in quiet (Fig. 2a–d). When call effort was allowed to vary (experiments 1 and 2), the proportions of females choosing higher call rates (experiment 1; Fig. 2a) and longer calls (experiment 2; Fig. 2b) were significantly greater than expected by chance across all choices tested. In all eight tests across these two experiments, significant preferences for higher call rates (experiment 1) and longer calls (experiment 2) were also preferences favouring the stimulus with the higher call effort (Fig. 2a, b).

In contrast, when we held call effort constant (experiment 3, Fig. 2c), preferences were nonsignificant in three of four tests. However, significantly more females chose the standard call over an alternative with a shorter call duration (−1 SD, 26 pulses/call) and faster call rate (+0.56 SD, 12.69 calls/min). While this 2.6:1 preference for the relatively longer (by 1 SD) standard call produced at a relatively slower rate (by 0.56 SD) was statistically significant, we would note that there was also a nonsignificant 2.2:1 preference in the opposite direction favouring a shorter call (−2 SD, 22 pulses/call) produced at a faster rate (+1.3 SD, 15 calls/min) (see Fig. 2c). Hence, we do not regard the single significant difference in this experiment to be particularly compelling evidence of any strong, overall pattern of preference.

In the final experiment, we varied call rate and call duration orthogonally (in terms of SD units) and allowed call effort to vary (experiment 4; Fig. 2d). Significantly more females than expected by chance chose the standard call (and thus discriminated against the alternative call) when the alternative had slower call rates (−2 SD and −1 SD) but longer call durations (+1 SD and +2 SD). There were no significant preferences when the alternative had a call rate above average (+1 SD and +2 SD) and call duration below average (−1 SD and −2 SD). Because call effort was allowed to vary in this experiment, we would point out that females discriminated against the two alternatives with the lowest call efforts (equivalent to −0.9 SD and −2.1 SD; Fig. 2d) in favour of the standard call, but showed no discrimination between the standard call and alternatives with somewhat higher call efforts (equivalent to +0.5 SD and +0.7 SD; Fig. 2d).

We observed broadly similar patterns in replicate tests conducted in quiet (Fig. 2a–d) and in the presence of chorus-shaped noise (Fig. 2e–h). There was a trend for relatively fewer females tested in noise to choose a call that was preferred by females tested in quiet. Qualitative comparisons of the outcomes of the two binomial tests conducted for the two replicates of each choice test indicated that four choice tests yielded different statistical outcomes in quiet and noise (cf. Fig. 2a–d, e–h). In all cases, outcomes that were statistically significant in quiet were nonsignificant in noise. In experiment 2 (cf. Fig. 2b, f), for example, females no longer discriminated between calls of average length and those that were shorter or longer by 1 SD. In experiment 3 (cf. Fig. 2c, g) there were no significant preferences in choice tests conducted in noise, whereas one of four choice tests was significant in quiet. In experiment 4 (cf. Fig. 2d, h) females no longer discriminated in noise against calls that were longer than the standard call by +1 SD, but produced at a slower rate (by −1 SD). These trends notwithstanding, the numbers of females choosing the standard and alternative calls in most choice tests did not differ statistically between tests conducted in quiet and in noise (X^2s < 3.69, Ps > 0.050). The single exception was in experiment 1 (cf. Fig. 2a, e); compared with quiet, significantly fewer females tested in noise chose the alternative with a call rate that was +2 SD faster than that of the standard call ($X^2 = 5.1$, $P = 0.02$). Despite this difference, preferences for the alternative call were significant in both conditions (binomial tests: Ps < 0.05).

DYNAMIC SHIFTS IN CALLING PERFORMANCE

Previous studies have shown call rate and call duration to be dynamic properties of advertisement calls in frogs and insects (Gerhardt 1991; Ryan & Keddy-Hector 1992). In many instances,
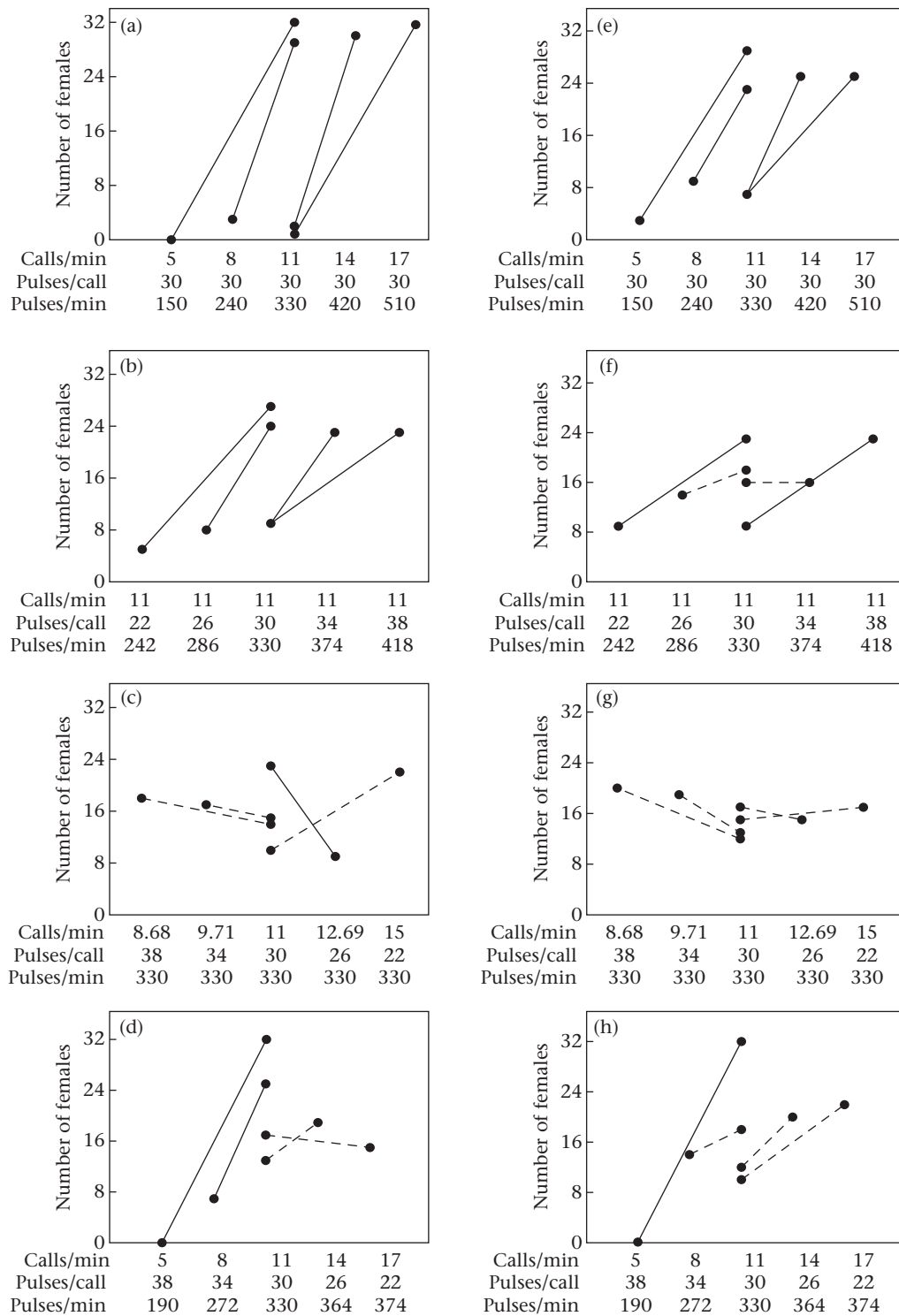
Figure 2. Preferences of females in two-stimulus choice tests replicated in quiet (a–d) and in the presence of chorus-shaped noise (e–h). Two points connected by a line depict results from a single choice test ($N = 32$ per test) and show the numbers of females choosing each call. Call property values are depicted below each panel. In (a, b) and (e, f), either call rate or call duration was held constant, the other property was manipulated and call effort was allowed to vary. In (c) and (g), call effort was fixed, call duration was manipulated and call rate varied according to these other two properties. In (d) and (h), call rate and call duration were orthogonally manipulated and call effort was allowed to vary accordingly. Middle values along the X axis indicate population means. Values to the left and right indicate ±1 or ±2 SD for the manipulated property. Solid lines indicate statistical significance (two-tailed binomial test: $P < 0.05$); dashed lines indicate nonsignificance ($P \geq 0.05$).

males dynamically alter one or both call properties depending on the local social environment (reviewed in: Gerhardt & Huber 2002; Schwartz & Bee, in press). If male grey treefrogs alter their calls to become more attractive to females in more competitive social

environments, our results (Fig. 2) indicate they should shift to producing longer calls at faster rates so as to maximally increase their overall call effort. We conducted a playback experiment under controlled acoustic conditions to test the hypothesis that males

increase their call effort in acoustically competitive environments, thereby making their calls more attractive to females.

Methods

Subjects

We used a within-subjects design to record 20 calls from each of 20 males in each of three randomly ordered test conditions (total of 60 calls per male) simulating three different acoustic environments: (1) under quiet conditions, (2) in response to playback of a simulated neighbouring male and (3) in response to broadcasts of chorus-shaped noise. Male subjects for this experiment were collected in amplexus during the 2011 breeding season and handled prior to testing similarly to how females were handled prior to the female choice experiments described in the previous section. At least 30 min prior to testing, we placed focal males in a temperature-controlled incubator to allow their body temperature to reach $20 \pm 1\ ^{\circ}C$, the temperature at which our experiment was conducted. Males were tested and returned to their location of collection within 2–3 days.

Apparatus

We conducted playback tests using a hemi-anechoic sound chamber and 2 m diameter test arena similar to those described above (chamber size: $3.0 \times 2.8 \times 2.2$ m; Industrial Acoustics Company). In the centre of the arena on the floor was an acoustically transparent, plastic dish (10 cm in diameter, 2.8 cm deep) in which the subject was held during the tests. Two speakers (Mod1 Orb) were used to broadcast acoustic stimuli. One speaker, located behind the arena wall at a distance of 1 m from the subject, was used to broadcast calls mimicking a nearby calling neighbour. A second speaker for broadcasting artificial chorus-shaped noise was suspended from the ceiling in the centre of the arena directly above the subject. Stimuli were broadcast using the same hardware and software described above for our female choice experiments. Prior to testing each subject, we calibrated the neighbour and chorus noise stimuli to 85 dB SPL (LCF_{max}) and 70 dB SPL (LC_{eq}), respectively, using a Larson Davis System 824 sound level meter (Larson Davis Inc, Provo, UT, U.S.A.) with the microphone placed at the same position as the subject's head during tests. An IR-sensitive video camera centred above the arena allowed us to monitor activity during testing from outside the chamber without disturbing the subject.

Acoustic stimuli

The 'neighbour stimulus' involved repeatedly broadcasting a call similar to the standard call used in our experiments investigating female preferences described in the previous section, with the following differences. It was created at a sampling rate of 20 kHz using custom-made software (courtesy J. J. Schwartz); it had spectral peaks (with relative amplitudes) of 1.3 kHz (-6 dB) and 2.6 kHz (0 dB); it had 11 ms pulse durations (4 ms inverse exponential rise, 7 ms exponential fall) and interpulse intervals (22 ms pulse period, 45.5 pulses/s). All of these values are near the population averages (see Supplementary Material). We created 20 unique exemplars of the neighbour stimulus (one for each subject) using MATLAB$^{®}$ 7.6.0. The exemplars differed in the lengths of the silent intercall interval between the offset and onset of two consecutive calls. Each consecutive intercall interval in each exemplar of the neighbour stimulus was drawn randomly from the actual distribution of temperature-corrected intercall intervals recorded in natural breeding choruses ($\overline{X} = 6.1 \pm 2.3$ s; see Supplementary Material); hence, subjects could not predict the exact timing of each subsequent call. Exemplars had an average call rate of 9.9 ± 2.6 calls/min, which approximates the average call rates of males in natural choruses that we observed

($\overline{X} = 11.4 \pm 2.9$ calls/min). Our 'chorus noise stimulus' was the same stimulus as the chorus-shaped noise used in the female choice experiments described in the previous section.

Protocol

At the beginning of a test, we placed the subject in the dish in the centre of the arena. We permitted the male to acclimate for at least 1 min before the start of the test. We recorded the calls of each subject in each of the three acoustic environments (quiet, neighbour, chorus noise), and subjects were given a 2–3 min time-out between tests during which they were returned to the incubator. We recorded 20 calls of each male in each test condition using a Sennheiser ME66 microphone and K6 power supply mounted on a tripod. The recording tip of the microphone was positioned 50 cm from the subject. All calls were recorded with a Marantz PMD670 digital recorder located outside the chamber (44.1 kHz sampling rate, 16-bit resolution). In the neighbour and chorus noise tests, we permitted the male to produce approximately 10 spontaneous calls in quiet prior to commencing broadcasts of the test stimulus and recordings of response calls. For tests conducted under quiet conditions, we began recording after the male had produced 10 spontaneous calls. For each subject, we used the waveform display of Adobe Audition 1.5 to determine its call rate (calls/min, measured over 20 complete, consecutive call periods) and mean call duration (number of pulses per call, averaged over 20 consecutive calls) in each of the three tests. We computed call effort (pulses/min) as the product of call rate (calls/min) and call duration (pulses/call).

Statistical analyses

We tested the hypothesis that males dynamically adjust calling behaviour in different acoustic environments using a repeated measures MANOVA with call rate, call duration and call effort as the dependent variables and acoustic environment as the within-subjects factor. Because this analysis revealed significant differences in male calling behaviour across acoustic environments, we used univariate repeated measures ANOVAs to compare each dependent variable separately; significant ANOVAs were followed by post-hoc, pairwise comparisons (Fisher's LSD tests). Our data met the assumptions of parametric statistics. We report Greenhouse–Geisser adjusted significance values in cases where the additional assumption of sphericity in repeated measures tests was violated (Mauchly's test: $P < 0.05$).

Results

Repeated measures MANOVA revealed significant differences in calling behaviour across our three acoustic environments (Wilks' $\lambda = 0.30$, $F_{6,72} = 10.11$, $P < 0.001$). Subsequent univariate analyses revealed significant differences in call rate ($F_{2,38} = 3.61$, $P = 0.037$; Fig. 3a) and call duration ($F_{2,38} = 42.80$, $P < 0.001$; Fig. 3b). Post hoc comparisons revealed that call rates were significantly slower (by 17%) in chorus noise compared with quiet conditions (Fig. 3a). Males also called at slower rates (by 15%) in chorus noise than in the neighbour condition, but this difference was not quite significant (Fig. 3a). We also observed no significant difference between call rates produced in the neighbour and quiet conditions (Fig. 3a). Post hoc comparisons indicated that males produced significantly longer calls during playbacks of both the neighbour stimulus and the chorus noise stimulus compared with the quiet condition (Fig. 3b). On average, individual males exposed to noise produced calls that were 16% longer than calls produced in quiet. Calls produced during broadcasts of a neighbouring male were 6% longer than calls produced under quiet conditions. Males also produced calls in the presence of noise that were significantly longer (by 10%)
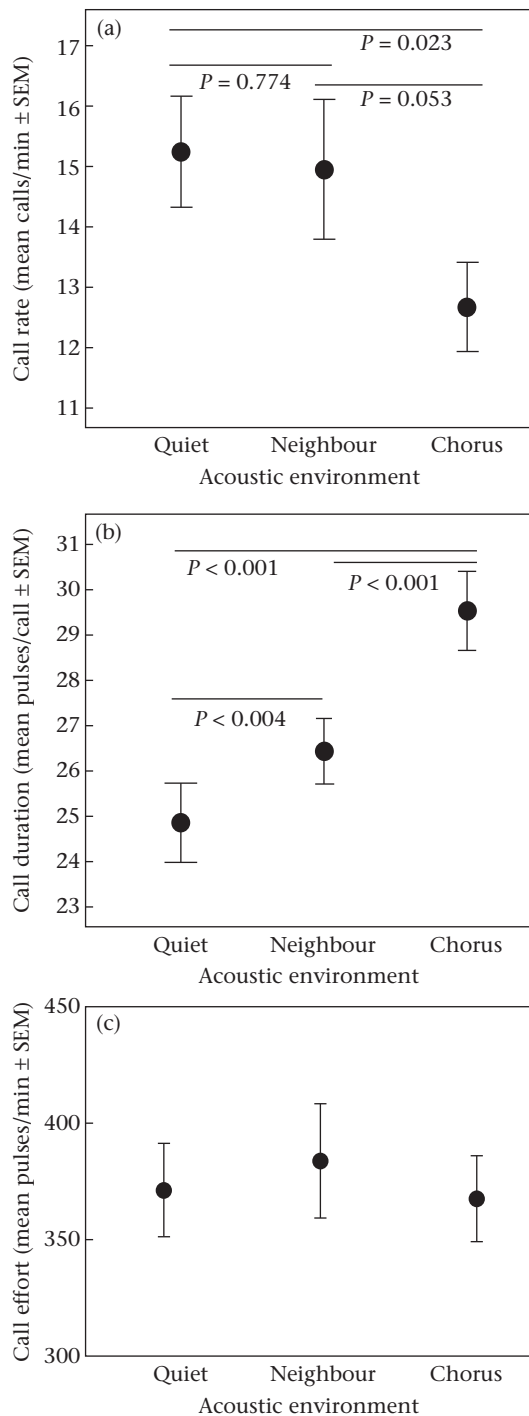
Figure 3. Dynamic shifts in male calling behaviour included changes in call rate and call duration, but not call effort. Points and whiskers depict mean values and standard errors, respectively, calculated across all males ($N = 20$). P values from pairwise post hoc tests comparing calling behaviour in quiet conditions and during broadcasts of a simulated neighbouring male and chorus noise are presented for cases where univariate ANOVAs (reported in the main text) revealed significant differences in calling behaviour.

371 pulses/min under quiet conditions, 384 pulses/min in response to a single calling neighbour (a 3.5% increase over quiet) and 367 pulses/min in response to chorus noise (a 1.1% decrease from quiet). Hence, despite dynamic shifts in behaviour to produce longer calls at slower rates, overall call effort remained relatively constant (Fig. 3c).

GENERAL DISCUSSION

This study yielded three main findings. First, analyses of calls recorded in natural choruses revealed negative covariance between the signal components of call rate and call duration across individuals. Second, laboratory choice tests showed that while females preferred higher values of both call rate and call duration, these preferences were actually based on call effort, a property dependent on the interaction between both components. These two findings are consistent with predictions of a multitasking hypothesis and indicate that female preferences are multiplicative in the sense that they are based on the product of two call components (call rate × call duration = call effort). Finally, laboratory playback tests showed that individual males dynamically adjusted call rate and call duration in response to changes in the local competitive social environments (simulated acoustically), but did not increase their overall call effort. Hence, dynamic shifts in calling behaviour did not appear to be targeted towards increasing the attractiveness of a male's calls to females. Taken together, findings from this study add to the growing number of studies that examine how females respond to negative covariation in the expressions of multiple signal components used in mate choice (e.g. Klump & Gerhardt 1987; Saino et al. 2003; Ballentine et al. 2004; Pryke & Andersson 2005; Murai et al. 2009). We next discuss each of our three main findings in detail.

Multitasking Males

Acoustic signalling in anurans is an energetically expensive aerobic activity (Taigen & Wells 1985; Wells & Taigen 1986; Prestwich et al. 1989; 1994; Grafe 1997; reviewed in Wells 2001, 2007). Much of the work on calling energetics in frogs has investigated vocal behaviour in the sister-species of *Hyla chrysoscelis*, the tetraploid eastern grey treefrog, *Hyla versicolor* (Taigen & Wells 1985; Wells & Taigen 1986; Wells et al. 1995; Grafe 1997; McLister 2001). Together, these two closely related species represent a cryptic species complex, with the diploid *H. chrysoscelis* being an ancestor of the allotetraploid *H. versicolor* (Ptacek et al. 1994; Holloway et al. 2006). The negative covariance reported here between call rate and call duration is similar to that reported previously for *H. versicolor* (Wells & Taigen 1986; Grafe 1997; Schwartz et al. 2002; Reichert & Gerhardt 2012). Our results thus extend this previous work by showing that the trade-off between these two properties was likely present in an ancestral species.

In *H. versicolor*, the trade-off between call rate and call duration appears due, in part, to a performance limitation imposed by the high energetic costs of calling (Wells & Taigen 1986). In this species there are strong, direct relationships between metabolic expenditure during calling (measured as oxygen consumption, VO_2) and call rate ($r^2 = 0.76$: Taigen & Wells 1985; $r^2 = 0.88$: Grafe 1997) and call effort ($r^2 = 0.85$: Wells 2001; see also McLister 2001); relationships with call duration are not as strong ($r^2 = 0.23$: Taigen & Wells 1985; Wells & Taigen 1986; $r^2 = 0.26$: Grafe 1997). We here assume the patterns of metabolic costs associated with calling behaviours are broadly similar between *H. chrysoscelis* and *H. versicolor*. Given this assumption, our results for *H. chrysoscelis* (Fig. 1b) are consistent with the interpretation that maximizing call

than calls produced in the presence of a single neighbouring male (Fig. 3b). Although males adjusted call rates and call durations in response to stimulation by the calls of a neighbour and chorus noise, there was no difference in call effort across the three acoustic environments ($F_{2,38} = 0.17$, $P = 0.835$). Males produced averages of

effort is energetically constrained by a performance limitation that forces males to trade off call rate and call duration. This interpretation is consistent with predictions of the multitasking hypothesis (Hebets & Papaj 2005).

We found no relationships between call effort and condition, SVL or mass. Furthermore, we found no evidence of strong relationships between other signal components (call rate, call duration) and these aspects of signaller phenotype. Therefore, these correlative data do not support the idea that individual differences in size or physical condition underlie individual differences in call rate, call duration and call effort. Nevertheless, results from previous manipulative studies suggest quality-based judgements are possible in grey treefrogs. For example, Welch et al. (1998) showed that females of H. versicolor that were artificially mated with males producing longer calls had offspring with higher fitness. Sires that produced longer calls also had greater call efforts than sires with shorter calls, although this difference in call effort was not statistically significant (Welch et al. 1998). Schwartz & Rahmeyer (2006) investigated the relationship between locomotor performance (endurance swimming) and calling behaviour in H. versicolor. Males with relatively higher call efforts had a greater capacity for sustained swimming, suggesting these males were in better condition. Call duration, on the other hand, was not a significant predictor of swimming performance. Together, these studies suggest males with higher call efforts are potentially higher-quality males, a result consistent with the multitasking hypothesis (Hebets & Papaj 2005) and indicator models of sexual selection more generally (Andersson 1994).

The negative covariance we found between call rate and call duration likely reflects not only performance constraints but also individual differences in behaviour associated with differences in the local social environments of recorded males. We did not quantify properties of the local social environment at the times our recordings were made. However, Wells & Taigen (1986) previously reported that isolated males of H. versicolor called at faster rates and produced shorter calls compared with males calling in denser parts of the chorus. Interestingly, call effort and the estimated aerobic costs of calling were largely independent of chorus density. Schwartz et al. (2002) later corroborated the trade-off between call rate and call duration in experimental choruses of H. versicolor created in an enclosed laboratory pond. As chorus size was experimentally reduced, males dynamically shifted towards producing shorter calls at faster rates. Although a trade-off exists at the population level, individual males are sometimes capable of breaking the trade-off under select conditions in choruses, such as when a female approaches and touches a male to solicit mating or when two males compete directly over a calling site (Fellers 1979; Schwartz et al. 2001; Reichert & Gerhardt 2012). We return to issues of dynamic signalling in a subsequent section. What this study and previous studies of H. versicolor confirm is that females of both species must make mating decisions in choruses in which calling males face a performance trade-off between call rate and call duration.

Multiplicative Females

Consistent with the multitasking hypothesis, the results from our female choice tests suggest call rate and call duration interact to produce the call feature most salient to attracting females, namely call effort. Consistent with previous studies of H. chrysoscelis (Gerhardt et al. 1996; Bee 2008a) and H. versicolor (e.g. Klump & Gerhardt 1987; Sullivan & Hinshaw 1992; Gerhardt et al. 1996, 2000; Schwartz et al. 2001; Gerhardt & Brooks 2009), females preferred both higher call rates (Fig. 2a, e) and longer calls (Fig. 2b, f) when call effort was permitted to covary with these two signal

components. These preferences for higher call rates and longer calls were also preferences favouring higher call efforts. However, when we held call effort constant at the population average, most preferences for higher call rates and longer call durations were either abolished or attenuated (Fig. 2c, g). Moreover, when call effort was held constant (experiment 3), females generally failed to discriminate behaviourally between differences in call rate and call duration that had elicited robust discrimination when call effort was allowed to vary (experiments 1, 2 and 4). These results suggest females attend more to overall call effort (i.e. the arithmetic product of call rate and call duration) than to either of the single components of call rate and call duration in making mate choice decisions. Hence, positive female responses for higher call rates and longer calls appear to represent a more generalized preference for males producing the greatest acoustic output (i.e. higher call effort), rather than preferences for specific call properties (Ryan & Keddy-Hector 1992; see also Rosenthal & Evans 1998 for an analogous finding in fish).

Our results in the equal call effort experiment (experiment 3) differ in important ways from those reported in some previous studies of grey treefrogs. In a study of H. chrysoscelis from Missouri, U.S.A., Gerhardt et al. (1996) gave females a choice between a 12-pulse call and a 24-pulse call produced at half the rate of the 12-pulse call, so that the call efforts of the two stimuli were equal. In contrast to females in Minnesota, females in Missouri significantly preferred the longer call alternative produced at the slower rate. Two nonmutually exclusive explanations might account for the differences between our results and those of Gerhardt et al. (1996).

First, H. chrysoscelis populations can be assigned to one of two different genetic lineages (e.g. eastern and western; Ptacek et al. 1994; Holloway et al. 2006). Recent studies have documented lineage differences in female preferences for some call properties (Gerhardt 2005; Gerhardt et al. 2007; Bee 2010; Schrode et al. 2012). The present study was conducted with frogs of the western genetic lineage, whereas subjects in the study by Gerhardt et al. (1996) were of the eastern genetic lineage. Hence genetic differences between lineages might have contributed to the differences between our results and those of Gerhardt et al. (1996).

Second, methodological differences and nonlinear preferences might also have contributed to differences in our results. Gerhardt (1994) reported a mean call duration for Missouri H. chrysoscelis of 30 ± 6 pulses, which we calculated from his reported mean temperature-corrected ($20\,^{\circ}$C) call duration (in seconds) and pulse rate (in pulses/s). We would note that Gerhardt's (1994) reported average for Missouri males was similar to that reported here for Minnesota males (30 ± 4 pulses). However, Gerhardt et al. (1996) tested only two call durations (12 and 24 pulses) and both were below the average population duration. Hence, Gerhardt et al.'s (1996) long call was -1 SD below the average duration in Missouri populations, and their short call was -3 SD below average. Female preferences across a diversity of taxa, including grey treefrogs, are commonly nonlinear across the range of natural variation in trait values (Gerhardt et al. 1996, 2000; Schwartz et al. 2001; Chenoweth & Blows 2005; Rundle et al. 2005; Wagner & Basolo 2007; Bee 2008a). It is possible that nonlinearities in female preferences might account for apparent differences between results reported by Gerhardt et al. (1996) and in the present study.

Our results with western-lineage H. chrysoscelis also contrast with those from some previous studies of the tetraploid H. versicolor. Klump & Gerhardt (1987) found that tetraploid females from Missouri (northwestern lineage; Ptacek et al. 1994; Holloway et al. 2006) preferred longer calls produced at slower rates over shorter calls produced at faster rates even when call effort was nearly equal, a result replicated by Gerhardt et al. (1996) and Schwartz et al. (2001) in Missouri populations using stimuli

with equal call efforts. However, in two studies of *H. versicolor* in Maine and New York, U.S.A. (eastern lineage of Ptacek et al. 1994; northeastern lineage of Holloway et al. 2006), Sullivan & Hinshaw (1992) and Schwartz et al. (2008), respectively, failed to replicate this same basic result using stimuli with equal call effort. Together, these previous studies and ours on distinct but closely related genetic lineages and species highlight the potential for evolutionary lability in patterns of female preferences for multicomponent signals among close relatives, similar to instances reported in other taxa (e.g. Boughman et al. 2005; Rodriguez et al. 2006)

Mismatch between Dynamic Signalling and Receiver Preferences?

In response to hearing the calls of a simulated neighbour and the sounds of a chorus, males of *H. chrysoscelis* dynamically shifted to producing longer calls at slower rates but did not significantly increase their call effort (Fig. 3). These results corroborate and extend our earlier study of this species showing that males increase call duration and decrease call rate while maintaining a constant call effort and call amplitude in response to chorus noise broadcast at different sound pressure levels (Love & Bee 2010).

Previous studies of socially mediated plasticity in calling behaviour in *H. versicolor* have reported that males calling in more competitive social environments produce longer calls at slower rates (Wells & Taigen 1986; Schwartz et al. 2002). Males actively lengthened their calls and reduced their call rate in response to playbacks (Wells & Taigen 1986) and experimental manipulations of chorus size (Schwartz et al. 2002). Importantly, these dynamic shifts in calling behaviour in *H. versicolor* were not associated with increases in energy expenditure. As noted earlier, in some populations, females of this species prefer longer calls produced at slower rates over shorter calls produced at faster rates when call effort is constant. Hence, in competitive situations, dynamic changes in calling behaviour may allow males of *H. versicolor* to maximize their attractiveness to females while mitigating any energetic costs associated with producing longer, more attractive calls by decreasing their call rate.

In contrast to this previous work on *H. versicolor*, our results on evoked calling in *H. chrysoscelis*, when considered in light of results from our female preference tests, do not allow us to draw the same conclusion that males modify their calls to become more attractive to females while conserving energy. We found little evidence to suggest that long calls produced at slow rates were more attractive to females than short calls produced at fast rates when call effort was held constant (Fig. 2c, g). If females of *H. chrysoscelis* generally prefer higher call efforts and do not find longer calls at slower rates any more attractive than shorter calls produced at faster rates, then why do males dynamically shift towards producing longer calls at slower rates in socially competitive environments? Several hypotheses are worth testing in future studies.

First, males may lengthen their calls to maximize the number of pulses free from acoustic interference by other males (the 'interference risk hypothesis'; Schwartz et al. 2001, 2008). Schwartz et al. (2001) found that male *H. versicolor* with fewer overlapped pulses had higher mating success in an artificial pond. However, a follow-up series of choice tests failed to support the hypothesis that acoustic interference increases the strength of female preference for longer calls (Schwartz et al. 2008). Second, longer calls may increase the likelihood of signal detection in the presence of high levels of chorus noise (the 'call detection hypothesis'; Schwartz et al., in press). Longer sounds are commonly detected at lower thresholds due to temporal summation in the auditory system (reviewed in Brumm & Slabbekoorn 2005). Schwartz et al. (in press) tested this hypothesis in *H. versicolor*, but found no evidence that thresholds in the presence of chorus noise varied as a function of

pulse number, suggesting that both short and long calls were equally likely to be detected in noisy environments. Additional comparative research on *H. chrysoscelis* would be useful to evaluate the generality of the negative results reported by Schwartz et al. (2008) for the interference risk hypothesis and Schwartz et al. (in press) for the call detection hypothesis.

Finally, it is possible that females are not always (or even usually) the intended receivers of the dynamic shifts in calling behaviour that occur in competitive social environments. Males of both grey treefrog species produce exceptionally long calls, often at high rates, in response to being directly approached and touched by a female (Fellers 1979; Wells & Taigen 1986; Schwartz et al. 2001; H. C. Gerhardt & G. M. Klump, unpublished data, cited in Reichert & Gerhardt 2012; M. A. Bee, unpublished data). Fellers (1979) suggested such female-induced changes in calling might function to provide females with enhanced cues for source localization in dense vegetation. However, these dynamic responses to females are made presumably only after females have selected the male as a mate, not necessarily to attract them in the first place. The dynamic changes in calling reported here may be targeted more to other males in the chorus. Graded changes in calling behaviour associated with male–male interactions are common among frogs (e.g. Schwartz 1989; Wagner 1989; Grafe 1995; Owen & Gordon 2005). In their recent study of close-range male–male interactions over possession of calling sites, Reichert & Gerhardt (2012) showed that males of *H. versicolor* momentarily exceed performance limits determined at the population level by increasing both call rate and call duration (and hence, call effort). Thus, it is possible that males, not females, are the intended receivers of the dynamic changes in call rate and duration observed in real and artificial choruses and in response to playbacks. If so, males that adjust calling behaviour without altering overall call effort might effectually engage in signalling interactions with nearby males without compromising the attractiveness of their calling behaviour to females. Such an explanation based on multiple receivers would add another dimension to the multitasking hypothesis for the function of multicomponent signals.

Supplementary Material

Supplementary material for this article is available, in the online version, at http://dx.doi.org/10.1016/j.anbehav.2013.05.016.

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