

1 **The energetics of social signaling during roost location in Spix's disc-winged**  
2 **bats**

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4 Running title: Energy costs of calling in disc-winged bats

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17 **Summary Statement**

18 Spix's disc-winged bats constantly produce contact calls while searching for roosts, which we

19 show significantly increases an individual's metabolic rate.

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25 **Abstract**

26 Long-term social aggregations are maintained by multiple mechanisms, including the use of  
27 acoustic signals, which may nonetheless entail significant energetic costs. To date, however, no  
28 studies have gauged whether there are significant energetic costs to social call production in bats,  
29 which heavily rely on acoustic communication for a diversity of social tasks. We measure  
30 energetic expenditure during acoustic signaling in Spix’s disc-winged bats (*Thyroptera tricolor*),  
31 a species that commonly uses contact calls to locate the ephemeral furled leaves that they use for  
32 roosting. To determine the cost of sound production, we measured oxygen consumption using  
33 intermittent-flow respirometry methods, with and without social signaling. Our results show that  
34 the emission of contact calls significantly increases oxygen consumption; vocal individuals  
35 spent, on average, 12.42 kJ more during social signaling trials than they spent during silent trials.  
36 Furthermore, production of contact calls during longer periods increased oxygen consumption  
37 for males but not for females. We also found that as resting metabolic rates increased in males,  
38 there was a decreasing probability that they would emit response calls. These results provide  
39 support to the “allocation model”, which predicts that only individuals with lower self-  
40 maintenance costs can afford to spend energy in additional activities. Our results provide a step  
41 forward in our understanding of how physiology modulates behavior, specifically how the costs  
42 of call production and resting metabolic rates may explain the differences in vocal behavior  
43 among individuals.

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45 **Keywords:** allocation model, bats, energetic expenditure, resting metabolic rate, social calls.

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## 48 **Introduction**

49 Many social animals rely on acoustic signals to facilitate social coordination (Kondo and  
50 Watanabe 2009; Fichtel and Manser 2010). In bats, for example, social calls are used to locate  
51 dependent young, mating partners, prompt and coordinate cooperative interactions, and/or  
52 defend and announce the location of resources, including roosts (Chaverri et al. 2018). The latter  
53 is of critical importance given that roosts provide refuge from predators and inclement weather,  
54 and are the main sites where social interactions, such as lactation, grooming, and mating, occur  
55 (Kunz 1982). Thus, the use of social calls during roost finding increases the probability of  
56 engaging in beneficial social interactions while reducing the risks of predation; as such, these  
57 acoustic signals represent a critical component of social living.

58         Despite our growing understanding of the benefits of social signaling, particularly in bats,  
59 we still do not understand its costs in different contexts. Studies in other taxa suggest that  
60 vocalizations that serve a social function increase an individual's risk of being detected by  
61 predators (Magrath et al. 2010) or by potential prey (Deecke et al. 2005), which could reduce  
62 foraging efficiency. Moreover, the production of acoustic signals may also carry significant  
63 metabolic costs. For example, energy expenditure of vocalizing animals could be up to eight  
64 times higher than those of silent ones (Ophir et al. 2010). In bats, echolocation calls produced  
65 during flight carry no additional energetic costs beyond those required to power flight  
66 (Speakman and Racey 1991; Voigt and Lewanzik 2012), yet may entail significant metabolic  
67 costs when produced while roosting, likely due to the contraction of muscles involved in sound  
68 emission (Dechmann et al. 2013). However, despite the costs of sound production, the benefits to  
69 group coordination and roost-finding efficiency are significant, as just a few calls produced by a

70 single roosting bat are enough to maintain group cohesion and decrease the time needed to locate  
71 a new roost site (Sagot et al. 2018).

72         The costs of call production may potentially explain why social calls are not emitted  
73 more frequently, in specific contexts, or by all group members. In moving groups, for example,  
74 members may produce social calls only sporadically (Deecke et al. 2005), and individuals may  
75 become silent altogether when faced with increased levels of predation risk (Abbey-Lee et al.  
76 2016). The energetic costs of sound production may also explain why only some group members  
77 vocalize, as has been observed in bats where lactating females produce significantly fewer calls  
78 compared to non-reproductive and pregnant females (Chaverri and Gillam 2015). These  
79 intraspecific differences suggest that vocalizations involve higher energetic costs and that non-  
80 energetically limited individuals may be able to afford sound production for social  
81 communication.

82         Here, we aim to estimate the energetic cost of social calling in roosting bats to understand  
83 patterns of inter-individual differences in vocal behavior. We focus on Spix's disc-winged bat,  
84 *Thyroptera tricolor*, a small insectivorous species that roosts in the developing tubular leaves of  
85 plants in the order Zingiberales (Vonhof and Fenton 2004) in groups of approximately 5  
86 individuals (Vonhof et al. 2004; Sagot et al. 2018). This species is known to use a call-and-  
87 response contact calling system for maintaining very stable group composition (Chaverri 2010)  
88 despite moving among roost-sites on a daily basis. Spix's disc-winged bats produce two different  
89 types of social calls: the "inquiry" calls that are emitted by flying individuals and "response"  
90 calls that are emitted by roosting individuals in response to inquiry calls to guide and attract their  
91 conspecifics to the roosts (Chaverri et al. 2010). In this species, the rates of response call  
92 production are relatively consistent within, but vary widely among individuals (Chaverri and

93 Gillam 2015). Furthermore, social groups are composed by a combination of vocal and non-  
94 vocal bats in the context of response calling, and thus around 50% of individuals produce  
95 response calls upon hearing inquiry calls from group and non-group members, whereas the rest  
96 never vocalize (Chaverri and Gillam 2015; Sagot et al. 2018).

97 We simulate vocal exchanges in *T. tricolor* to gauge the energetic costs of response call  
98 production. If individuals actively respond to the inquiry calls of their conspecifics, we expect  
99 metabolic rates to increase significantly; specifically, oxygen consumption should increase when  
100 bats vocalize for longer periods of time, as studies in a number of taxa demonstrate that  
101 vocalizations increase energy expenditure (Ryan 1988; Oberweger and Goller 2001; Ophir et al.  
102 2010). We also test whether resting metabolic rates (RMR), i.e. those that reflect the metabolic  
103 rate of an individual during its inactive period (McNab 1997), correlate with response call  
104 production. Previous studies suggest that levels of activity or aggressiveness, which are traits that  
105 allow us to distinguish among animal personalities, are either positively or negatively influenced  
106 by resting metabolic rates (Careau et al. 2008). In the first case, termed the “performance  
107 model”, animals with greater levels of activity or aggression require larger organs to sustain  
108 these traits, and thus have higher-than-average maintenance costs (Daan et al. 1990). In contrast,  
109 the “allocation model” predicts a negative relationship between RMR and activity or  
110 aggressiveness because when food is limited, only individuals with lower self-maintenance costs  
111 can afford to spend energy in additional activities (Careau et al. 2008). While we have no a priori  
112 expectation regarding which model, performance or allocation, may predict response calling  
113 rates in *T. tricolor*, we test this to increase our understanding of the factors that may explain  
114 vocal personalities in the context of social communication.

115

## 116 **Methods**

117 We collected data on metabolic rates for 38 individuals (18 adult females, 10 adult males, 3  
118 subadult females, 4 subadult males and 3 juvenile males) from 11 social groups (i.e., individuals  
119 using the same roost at the same time) at Barú Biological Station in Southwestern Costa Rica, in  
120 July 2017. To find groups, we searched *Heliconia* spp., *Calathea* spp. and *Musa* spp. furled  
121 leaves, commonly used by *T. tricolor* as roosting sites (Vonhof and Fenton 2004). Once we  
122 located a roost, we captured all group members and placed them inside a cloth holding bag to  
123 bring them to the laboratory. Back in the laboratory, we weighted all the individuals and  
124 measured their forearm lengths (as a measure of body length). We also sexed, aged, and  
125 determined the reproductive condition for all bats captured.

126 For each individual, we were interested in two parameters: 1) Resting Metabolic Rate  
127 (RMR), and 2) metabolic rate while producing response calls. The animals were placed singly  
128 inside a tubular structure made of transparent plastic; there they remained safely attached to the  
129 interior's smooth surface. The tube and bat were then placed inside a metabolic chamber and let  
130 to acclimate for 30 min. We measured the bats' oxygen consumption using the methods  
131 described below, resting and while listening/responding to conspecific inquiry calls. All  
132 measurements were made in a silent room at ambient humidity (70%) and temperature (27°C)  
133 during daytime hours. At the end of the experiments, we provided mealworms (*Tenebrio*  
134 *molitor*) and water *ad libitum* to all individuals before releasing them in the same area where  
135 they were originally captured.

136 *Thyroptera tricolor* bats only produce response calls after an inquiry call has been  
137 emitted, and do so primarily during the day (Chaverri et al. 2010); thus, we broadcasted  
138 previously recorded inquiry calls to elicit response calling from the bats within the chamber.

139 These inquiry calls were previously collected from five individuals flying within a large flight  
140 cage (3 x 4 x 9 m) for a total of 1 minute; none of these individuals were later included in our  
141 respirometry experiments. A total of 67 inquiry calls were identified in the 1-min recording, and  
142 the playback was continuously run for 10 minutes through an UltraSoundGate Player to a  
143 broadband loudspeaker (Ultrasonic Omnidirectional Dynamic Speaker Vifa, Avisoft  
144 Bioacoustics) placed inside the chamber. We recorded response calls produced by the individuals  
145 inside the chamber with an Avisoft condenser microphone (CM16, Avisoft Bioacoustics, Berlin,  
146 Germany) through Avisoft's UltraSoundGate 116Hm onto a laptop computer running Avisoft-  
147 Recorder software (sampling rate 384 kHz, 16-bit resolution), placed also inside the chamber.  
148 We also video-recorded each of the trials to estimate the effect of movement (i.e. how long the  
149 bats were actively moving during the trials) for better interpretation of the metabolic rate results.

150

### 151 **Metabolic rate measurements**

152 We measured O<sub>2</sub> consumption (VO<sub>2</sub>) of each individual using an intermittent-flow-through  
153 respirometry. This set-up consisted of short-term trials (10 min) of closed respirometry followed  
154 by a flushing interval of 10 min that allowed the saturated air to be pumped out of the chamber  
155 and replaced by new air, avoiding CO<sub>2</sub> accumulation. This method was used instead of a flow-  
156 through respirometry since it was not possible to measure flow rate. We placed each bat into a  
157 2L acrylic chamber lined with paper to reduce sound disturbance (i.e., reduction of echo  
158 interference from playback). Air was pumped into the chamber using a standard fish tank pump  
159 and then drawn out and passed through a column of indicating Drierite TM connected to the  
160 ML206 gas analyzer fed from a damped, micro-vacuum pump (200 mL/min; ADInstruments,  
161 Bella Vista, NSW, Australia). Since we did not dry the air going into the chamber, we measured

162 relative humidity of incurrent air with an electronic hygrometer, and mathematically scrubbed  
163 water vapor to provide a  $\dot{V}O_2$  corrected to standard temperature pressure dry (STPD). We  
164 recorded the voltage outputs of the gas analyzer and thermocouple at a sampling frequency of 10  
165 Hz using a PowerLab ML750 A/D converter (ADInstruments) and LabChart software  
166 (ADInstruments). For each bat we recorded  $O_2$  consumption for 10-min intervals of closed  
167 respirometry with and without sound broadcast. We calculated the whole individual metabolic  
168 rate ( $O_2$  ml  $h^{-1}$ ) using equation (4.9) of Lighton (2008), correcting for ambient pressure and  
169 standard temperature afterwards.

$$\dot{V}O_2 = \frac{(V \text{ chamber} - V_{H_2O}) - [FiO_2 - FE_{O_2}]}{1 - FE_{O_2} * 1 - RQ}$$

170 where V chamber is the volume of the chamber calculated by subtracting an approximation of  
171 the volume of the bat (mass multiply by 0.98) to the actual volume of the chamber (2L),  $V_{H_2O}$  is  
172 the water vapor in the chamber ;  $FiO_2$  and  $FE_{O_2}$  are the fractional concentration of  $O_2$  at the start  
173 and end of the experiment respectively. RQ is the respiratory quotient.

174 We converted oxygen consumption rate  $\dot{V}O_2$  into energy expenditure in kJ by utilizing  
175 the oxy-joules equivalents ( $MR_{kj}$  in  $kJ \text{ hr}^{-1}$ ) according to the following equation from Lighton  
176 (2008):

$$MR_{kj} = \dot{V}O_2 \times [16 + 5.164 (RER)]RER = \frac{VCO_2}{VO_2}$$

177 where RER is the respiratory exchange ratio ( $VCO_2/VO_2$ ). We assumed a RER of 0.77,  
178 previously reported for insectivorous bats (Speakman et al. 1989b).

179 All sampling protocols followed guidelines approved by the American Society of  
180 Mammalogists for capture, handling and care of mammals (Sikes 2016) and the ASAB/ABS  
181 Guidelines for the use of animals in research. This study was conducted in accordance with the



182 ethical standards for animal welfare of the Costa Rican Ministry of Environment and Energy,  
183 Sistema Nacional de Áreas de Conservación, permit no. SINAC-ACOPAC-RES-INV-008-2017.

184

## 185 **Data Analyses**

186 We compared metabolic variables (i.e., RMR and energy expenditure during trials with sound)  
187 among age categories using a one-factor ANOVA and Tukey comparisons at an alpha level of  
188 0.10. We found significant differences in RMRs between juveniles and adults, but not between  
189 adults and subadults ( $F_{2,35}=2.95$ ,  $P = 0.01$ ). Therefore, we merged data for the latter but  
190 eliminated juveniles from further analyses. Our sample size for subsequent tests was 21 females  
191 and 14 males.

192 To determine if males and females differed in the amount of time spent producing  
193 response calls or moving, we conducted two separate Mann-Whitney U-tests, as the data were  
194 non-normally distributed. We also ran a Chi-square test to determine if the proportion of vocal  
195 (i.e., an individual that produced at least one response call) vs. non-vocal bats differed between  
196 males and females. We then determined if males and females differed in resting metabolic rate  
197 and metabolic rate while producing response calls with two separate independent samples t-tests.  
198 We analyze data separately for males and females as previous studies have shown that the  
199 strength and direction of selection on resting metabolic rates may differ according to sex (Burton  
200 et al. 2011).

201 To test if more vocal bats (i.e., bats that vocalized for longer periods of time) had higher  
202 metabolic rates, we conducted a linear model with energy expenditure in kilojoules (kJ) as the  
203 response variable, sex as a fixed factor, and as regressors, we selected the time the bats spent 1)  
204 moving, 2) producing response calls, 3) or other types of calls (echolocation, distress, and other

205 calls of unknown function). We also included 4) mass as an additional regressor in the model.  
206 We retained the strongest explanatory variables using backward elimination and ran analyses  
207 separately for males and females. We also determined which was the variable with the greatest  
208 explanatory value based on the CP Mallows. We generated Q-Q and predicted vs. residual plots  
209 to test for normality and homogeneity of variances, respectively; both tests show that all  
210 assumptions of the model were met. In our results, we include the estimates of the multiple  
211 regression and those of simple regressions for explanatory variables kept in the model, to verify  
212 if independent effects on O<sub>2</sub> consumption are positive or negative.

213 To determine if RMR is related with the time bats spend producing response calls, we  
214 conducted a generalized linear model with time spent producing response calls as the response  
215 variable, and energy expenditure (kJ) and sex (and their interaction) as fixed factors. The  
216 dependent variable was non-normally distributed (Shapiro-Wilks tests = all p-values < 0.001)  
217 and could be modeled best by a negative binomial distribution (p-value = 0.17).

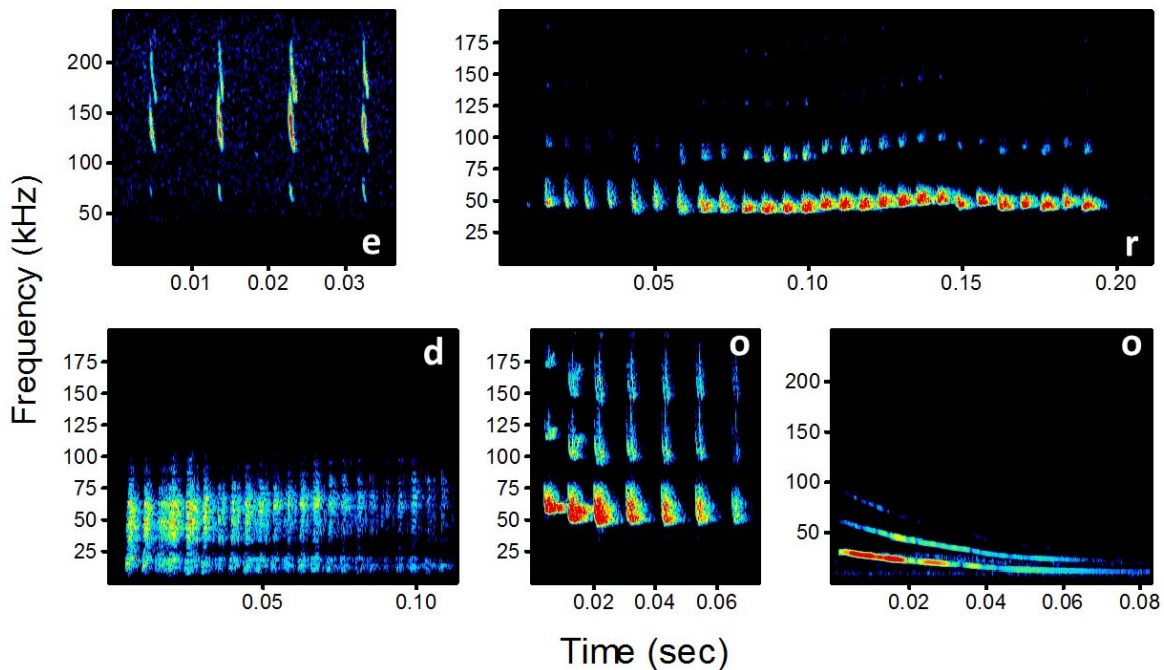
218 Finally, we tested if sex and the propensity to produce response calls or not had an effect  
219 on the difference in energy expenditure during resting trials and during trials with sound through  
220 a general linear model. The difference in energy expenditure was estimated as the amount of kJ  
221 consumed during trials with sound minus the amount of kJ consumed during trials without  
222 sound. Bats were categorized as being vocal if they produced at least one response call during  
223 our trials with sound.

224

## 225 **Results**

226 Bats were non-vocal during the 10-minute trials in which we measured the resting metabolic  
227 rates, i.e., those for which no sounds were broadcast. However, for trials in which we broadcast

228 inquiry calls, bats vocalized for an average of 27.47 seconds (SD = 37.17); many individuals (n =  
229 11) were non-vocal while the rest produced various types of vocalizations for up to 125.42 s.  
230 Animals produced three distinct calls with known functions and in decreasing order of  
231 frequency: response calls, which accounted for 61% of the time spent vocalizing; echolocation  
232 calls, which accounted for 21%; and distress calls (10% of the time). In some occasions, bats  
233 produced other calls with unknown function, which accounted for 8% of the time spent  
234 vocalizing (Fig. 1). There was no difference in the time spent vocalizing between males and  
235 females for any of the call types analyzed ( $P > 0.13$ ), nor was there a difference in the proportion  
236 of vocal vs. non-vocal individuals between males and females (vocal females = 38%, vocal  
237 males = 43%;  $P = 0.77$ ). Time spent moving was also not significantly different between males  
238 and females ( $P = 0.72$ ).  
239



240

241 **Fig. 1.** Sonograms depicting exemplars of call types recorded during our 10-min respirometry

242 sessions: echolocation (e), response (r), distress (d), other (o).

243

244           Animals consumed an average of 7.80 ml O<sub>2</sub> h<sup>-1</sup> during trials when no sounds were  
245 emitted (Table 1). Females had a significantly greater energy expenditure (kJ) during periods of  
246 inactivity than males ( $t = 2.57$ ,  $p = 0.01$ ). During the experiments with sound, bats consumed an  
247 average of 16.93 ml O<sub>2</sub> h<sup>-1</sup>. There was not a significant difference in energy expenditure (kJ)  
248 between males and females during trials with sound ( $t = 1.06$ ,  $p = 0.29$ ).

249

250 **Table 1.** Whole Animal Metabolic Rate (ml O<sub>2</sub> h<sup>-1</sup>) during trials when inquiry calls were  
251 broadcasted (sound) or when bats were resting (no sound).

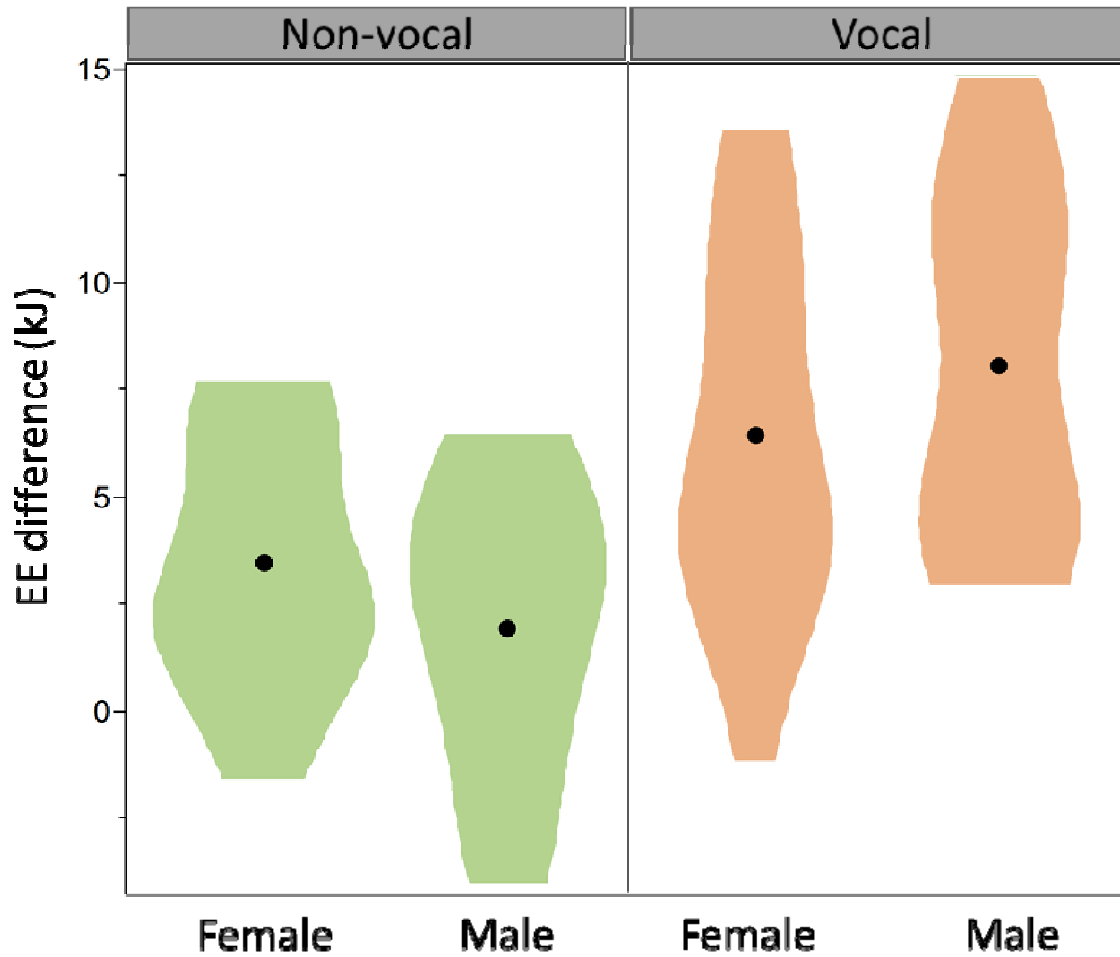
Sex	Weight (g)	Sound		No sound	
		Range	Mean±SD	Range	Mean±SD
Female	4.50 ±0.40	8.37-38.89	17.94±7.40	3.38-16.29	8.86±3.62
Male	4.10±0.36	4.80-30.13	15.42±6.74	2.30-10.95	6.22±2.33
All	4.32±0.43	4.80-38.89	16.93±7.15	2.30-16.29	7.80±3.39

252

253

254           The results of our general linear model, where we tested if sex and being vocal had an  
255 effect on the difference in energy expenditure during trials with sound compared to resting trials,  
256 show that the latter factor (being vocal) had a significant effect ( $F_{1,31} = 11.70$ ,  $p < 0.01$ ). The  
257 average increase in energy expenditure for vocal bats during trials with sound was 12.42 kJ  
258 ( $\pm 1.48$ ), whereas the increase for silent bats was 4.48 ( $\pm 1.78$ ; Fig. 2). Although the difference in  
259 energy expenditure for vocal vs. non-vocal individuals was greater for males than for females,  
260 neither sex nor the interaction between sex and vocal behavior was significant ( $p > 0.25$ ).

261



262

263 **Fig. 2.** Difference in energy expenditure, measured as the difference in kJ during trials with  
264 sound minus kJ during trials without sound, for vocal and non-vocal males and females.

265

266 For females, our multiple linear regression model indicates that energy expenditure (kJ)  
267 was significantly and positively influenced by the time spent producing calls, such as  
268 echolocation and distress; however, time spent producing response calls did not affect their  
269 energy expenditure, nor did time spent moving and body mass (Table 2, Fig. 3). In the model for  
270 males, there was a significant and also positive effect of time spent producing response calls on  
271 energy expenditure; thus, males that produced more response calls had greater energy

272 expenditure. Time spent moving also contributed to an increase in energy expenditure in males  
273 (Table 2, Fig. 3).

274

275 **Table 2.** Results of the multiple regression analysis with energy expenditure (kJ) as the response  
276 variable and several explanatory variables. We include the estimates of the multiple regression  
277 (MR) as well as those of the simple linear regression (SR). The CP-Mallows indicates the  
278 explanatory power of the variables included in the model.

Sex	R <sup>2</sup>	Explanatory	Estimate		P-value	CP- Mallows
			MR	SR		
Female	0.49	All calls except response (s)	0.31	0.31	<b>&lt;0.001</b>	18.13
Male	0.67	Response (s)	0.06	0.09	<b>0.04</b>	6.17
		Move (min)	0.80	1.15	<b>0.02</b>	7.26

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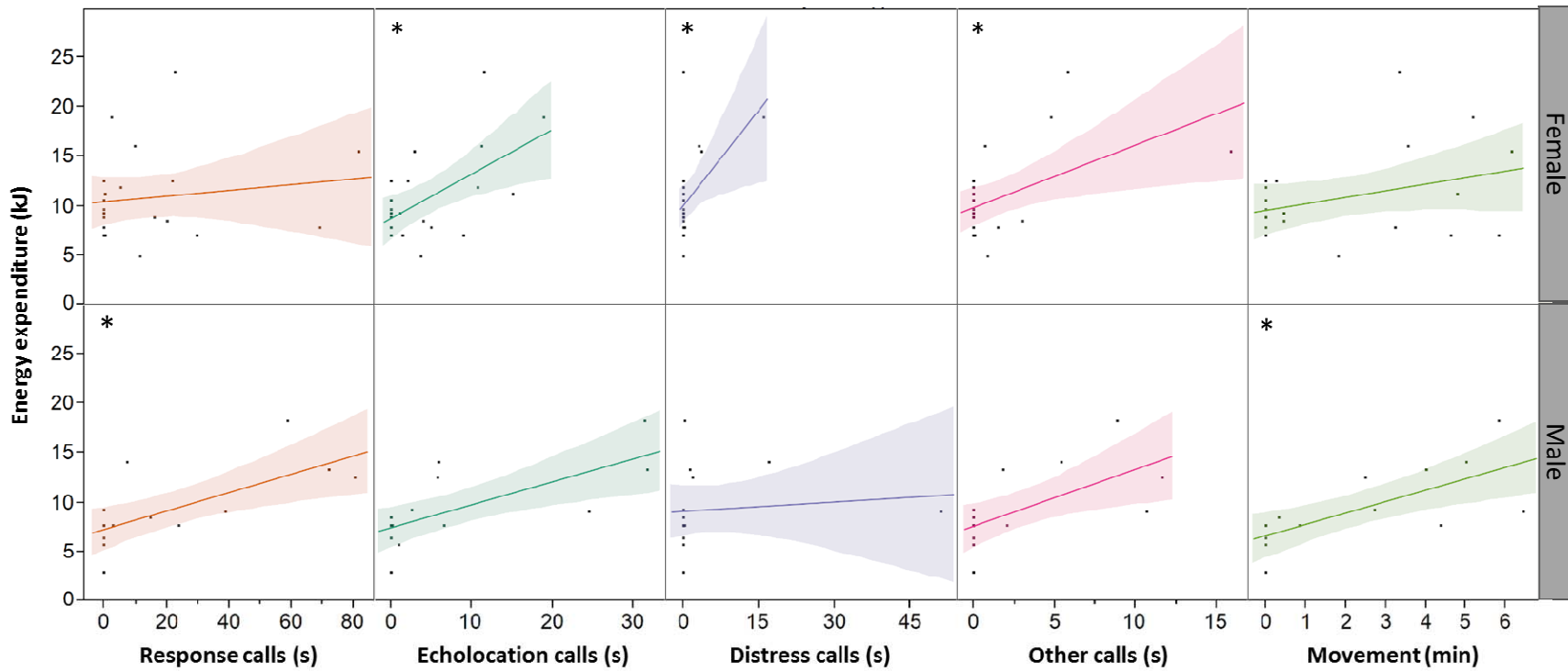
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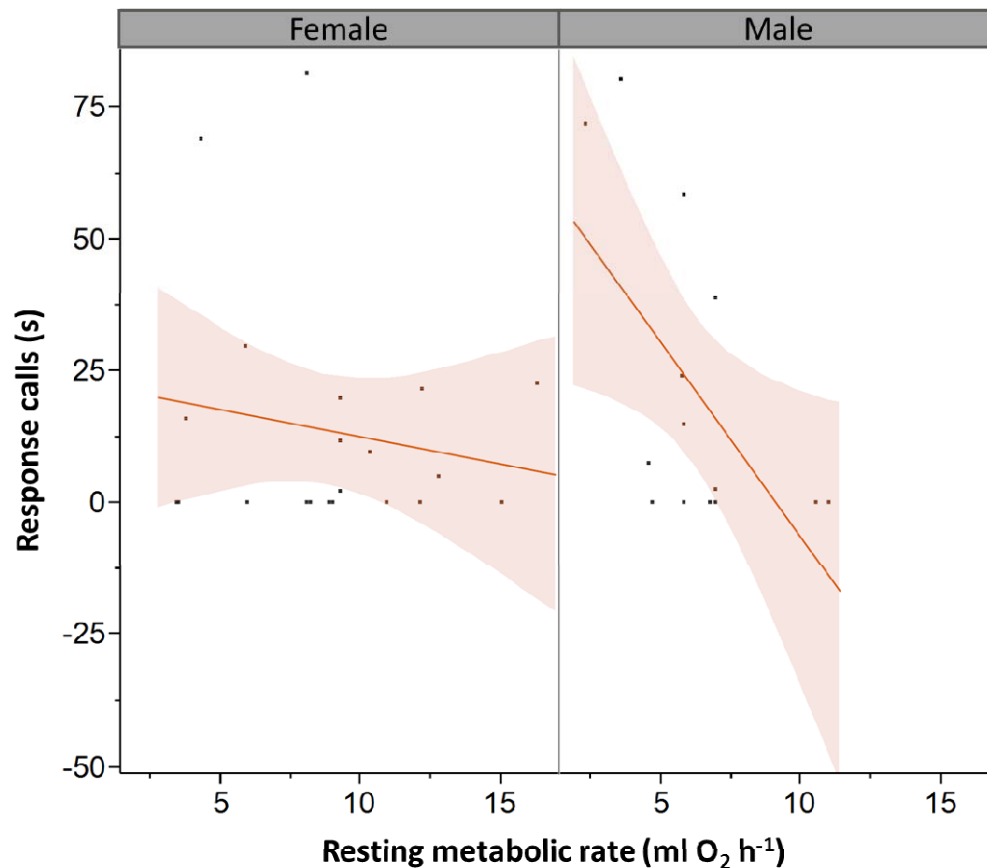
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289 **Fig. 3.** Scatter plots showing the relationship between time invested in various activities (emission of several types of calls and  
290 movement) and energy expenditure. Upper plots show the results for females and lower plots results for males. Asterisks indicate  
291 significant relationships according to the multiple regression analysis (see table 2).

292 Time spent producing response calls was significantly influenced by the interaction  
293 between sex and RMR ( $F_{1,31}=5.05$ ,  $p$ -value = 0.03), according to our generalized linear model.  
294 When performing the model separately for males and females, the relationship between RMR  
295 and time producing response calls for females was non-significant ( $p$ -value = 0.65), whereas for  
296 males the relationship was negative and significant ( $p$ -value = 0.003; Fig. 4). Thus, as RMR  
297 decreases in males, there is an increasing probability that they will emit response calls for longer  
298 periods of time.



299  
300 **Fig. 4.** Relationship between RMR and the time spent producing response calls (in seconds) for  
301 males and females. The shaded area around the trendline shows the 95% confidence interval.

302

303



304 **DISCUSSION**

305 Our results demonstrate that the production of social calls that are used to indicate the position of  
306 a roost site increases the energetic expenditure of bats. By producing even just one response call  
307 upon hearing an inquiry call, individuals significantly increased their resting metabolic rate. For  
308 males, the time spent producing response calls had a positive effect on energy expenditure,  
309 which did not occur in females, despite the fact that males and females produced a similar  
310 number of response calls and both were vocal in similar proportions. Vocal females increased  
311 their metabolic rate, on average, 1.4-fold when producing response calls, whereas males  
312 experienced a 3.2-fold increase.

313 Vocal communication can be observed in every major taxonomic group and in virtually  
314 every environment, and it is energetically demanding for many species (Ryan 1988; Prestwich  
315 1994; Oberweger and Goller 2001). Birds, for instance, increase their metabolic rate at least 2.5-  
316 fold when producing courtship calls, while ectotherms such as insects and amphibians can  
317 exhibit an 8-fold increase (Ophir et al. 2010). This is because sound production elevates  
318 muscular activity (Prestwich 1994; Gillooly and Ophir 2010), and increases the vibration  
319 frequency of the muscles that produce the sounds, elevating metabolic rates (Skoglund 1961;  
320 Martin 1971; Elemans et al. 2004). In *T. tricolor*, both males and females significantly increased  
321 their metabolic rates while producing response calls, suggesting that energetically compromised  
322 bats cannot afford extra energy expenditures in functions that are not part of their normal daily  
323 maintenance activities. This might help us explain why many individuals are non-vocal  
324 (Chaverri and Gillam 2015; Sagot et al. 2018). Furthermore, in males but not in females, the  
325 increase in metabolic rate was proportional to the time spent vocalizing, suggesting that males  
326 that are energetically limited cannot produce response calls, or can only vocalize for short

327 periods of time. This is possibly the reason why, in our study, only a small proportion of males  
328 produced vocalizations during relatively large amounts of time. This has also been found in other  
329 species such as bottlenose dolphins and birds, in which oxygen consumption increases with song  
330 duration and call rate (Oberweger and Goller 2001; Franz 2003; Noren et al. 2013).

331         Although sound production can be energetically demanding, in some species this activity  
332 does not increase an individual's metabolic rate (Ilany et al. 2013). For example, male hyraxes  
333 (*Procavia capensis*) that sing more, counterintuitively conserve more energy. Likewise,  
334 echolocating bats do not significantly increase their energy expenditure during flight (Speakman  
335 et al. 1989a; Voigt and Lewanzik 2012). However, even when vocalizations are not energetically  
336 demanding, they can still be considered a handicap (Gil and Gahr 2002). This is because  
337 producing these signals requires time, learning and specialized structures, and it can increase the  
338 chances of being detected by prey and predators (Koren and Geffen 2009; Charlton et al. 2011;  
339 Wyman et al. 2012).

340         We also found that differences in RMRs may predict the time spent producing response  
341 calls in males. Specifically, we found that males with lower RMRs emit response calls during  
342 longer periods of time. These results confirm that levels of activity, in our case measured through  
343 the time spent vocalizing, are negatively influenced by RMR, which provides support for the  
344 allocation model. This model predicts that only individuals with lower self-maintenance costs  
345 can afford to invest part of their daily energy budget in additional activities (Careau et al. 2008).  
346 Despite our results, the most common trend in vertebrates is for RMR to positively influence  
347 activity, thus supporting the performance model; however, males often exhibit the opposite  
348 trend, which might indicate that they produce signals with enough energy to experience a trade-  
349 off between RMR and activity (Stoddard and Salazar 2011). This latter argument might explain

350 the differences in energetic expenditure during response calling observed for males and females  
351 in our study. For instance, response calls in males might not only play a role in cooperative  
352 signaling of roost location (Chaverri and Gillam 2010), but may also function for mate attraction;  
353 if so, males could be under strong selection to produce high quality/energy calls as an honest  
354 signal of their body size and condition (Schuchmann and Siemers 2010). Thus, the physiological  
355 explanation for the differences in energetic costs of social signaling between males and females  
356 could be hormonal, as several studies demonstrate that male sexual hormones significantly alter  
357 the relationship between resting metabolic rates and signal quality or levels of activity (Wikelski  
358 et al. 1999; Lynn et al. 2000; Buchanan et al. 2001). Future studies should try to confirm the link  
359 between acoustic features of social calls like maximum energy, metabolic cost and mating  
360 success, in addition to addressing the potential role of response calls for mate attraction in *T.*  
361 *tricolor*.

362 In conclusion, our study demonstrates for the first time that social calls increase energetic  
363 expenditure in bats. Given that bats depend so strongly on acoustic signals for modulating  
364 multiple social activities (Gillam and Fenton 2016; Chaverri et al. 2018), our findings suggest  
365 that energetic trade-offs may be of particular importance to understanding communication in this  
366 group of mammals. The results of our study will surely extrapolate to various other species in  
367 diverse contexts; however, it is the differences among systems that seem most fascinating. In our  
368 case, we have addressed the costs of acoustic signaling during contact calling, but further studies  
369 could reveal interesting tradeoffs for signals such as those employed between mothers and  
370 offspring, or between males and females in the context of mate attraction, among others. Finally,  
371 our results provide a step forward in our understanding of how physiology modulates behavior.  
372 For example, many studies demonstrate that there is a link between resting metabolic rates and

373 various personality traits (Careau et al. 2008; Careau and Garland 2015). Incorporating  
374 physiological trade-offs to studies of animal personalities in the context of communication may  
375 allow us to understand many aspects of social aggregations, including social roles and  
376 communication networks.

377

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383

### 384 **Competing interests**

385 The authors declare no competing or financial interests.

386

### 387 **Data availability**

388 The data supporting this article are available from the Figshare Digital Repository:

389 <https://doi.org/10.6084/m9.figshare.13003805.v1>.

390

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