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Daily and seasonal fluctuation in Tawny Owl vocalization timing

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†This paper is dedicated to the memory of Professor Warren H. Meck, a world-renowned expert and scholar of interval timing and time perception, who died on January 21, 2020.

24 **Abstract**

25 A robust adaptation to environmental changes is vital for survival. Almost all living
26 organisms have a circadian timing system that allows adjusting their physiology to
27 cyclic variations in the surrounding environment. Among vertebrates, many birds are
28 also seasonal species, adapting their physiology to annual changes in photoperiod
29 (amplitude, length and duration). Tawny Owls (*Strix aluco*) are nocturnal birds of prey
30 that use vocalization as their principal mechanism of communication. Diurnal and
31 seasonal changes in vocalization have been described for several vocal species,
32 including songbirds. Comparable studies are lacking for owls. In the present work, we
33 show that male Tawny Owls present a periodic vocalization pattern in the seconds-to-
34 minutes range that is subject to both daily (early vs. late night) and seasonal (spring
35 vs. summer) rhythmicity. These novel theory-generating findings appear to extend the
36 role of the circadian system in regulating temporal events in the seconds-to-minutes
37 range to other species.

38

39 **Introduction**

40 Vocalization is a complex behavior and crucial in language evolution [1]. Birds are
41 the most vocal group of animals other than humans and a few other primates. In
42 songbirds (order Passeriformes, suborder oscines), neural vocal control is
43 achieved by a chain of interconnected brain areas in the fore-, mid-, and hindbrain
44 [2]. Vocal communication is also of vital importance to non-oscine birds (owls,
45 gulls, doves, etc.), which share with oscines a system of vocal control nuclei. In

46 particular, owls (order Strigiformes) rely on acoustic signals for long-distance
47 communication and hence are very vocal in different contexts [3].
48 In songbirds (e.g., zebra finches, canaries), the motor pathway for song proceeds
49 from the high vocal center (HVC) to the nucleus robustus arcopallialis (RA), which
50 then projects directly to vocal motoneurons (XIIIts) and to respiratory premotor
51 neurons in the nucleus retroambiguus (RAm) in the lower medulla. In turn, the
52 RAm projects upon spinal motoneurons that innervate expiratory muscles (which
53 provide the pressure head for vocalization), and upon XIIIts to effect vocal–
54 respiratory coordination [4,5]. In non-oscine birds, the syrinx has fewer muscles
55 than that of oscines and in many cases intrinsic muscles are entirely absent. The
56 non-oscine vocal control pathway has been scarcely investigated [6].
57 Owl vocalizations are simple compared with passerine vocalizations. An adult male
58 canary, for example, has a repertoire of about 2 or 3 dozen different syllable types
59 [7]. On the other hand, the predominant calls of the owls consist of simple notes or
60 syllables that are highly stereotyped [8]. The complex vocal behavior exhibited by
61 oscine songbirds is learnt by imitation of those from older members of their own
62 species. Vocal learning is characterized by its dependence on intact hearing and a
63 specialized forebrain circuitry that innervates vocal and respiratory nuclei of the
64 brainstem. These behavioral and neuroanatomical attributes have not been found
65 in non-vocal learners, which develop species-specific vocalizations in the absence
66 of hearing, and have no known forebrain vocal motor control. In non-vocal learners,
67 the vocal pathway is thought to consist solely of midbrain and brainstem nuclei [9].
68 Besides vocalizations, temporal information is also essential for communication.
69 Biological time mechanisms comprise distinctive processes that span several

70 orders of magnitude, from microseconds to seasonal events [10-12]. Among these
71 temporal orders, almost all living organisms are subjected to the influence of the
72 Earth's rotational 24-h cycle. This rhythmic pattern, with a period close to 24-h, is
73 called circadian rhythm (from the Latin words *circa dies*, around a day). The
74 spectrum of rhythmic events is also subdivided into ultradian rhythms (with periods
75 shorter than 24-h) and infradian rhythms (with periods longer than 24-h). In the
76 seconds-to-minutes range, temporal discrimination, known as interval timing, is
77 critical for fundamental behaviors such as foraging, decision-making and learning
78 [13,14]. In addition, annual/seasonal cycles are fundamental for reproduction,
79 migration and several physiological regulations in many species [15]. Besides the
80 regulation of daily rhythms, the circadian clock is involved in photoperiodic time
81 measurement.

82 Biological timing is essential in birds. Among vertebrate species, birds have highly
83 sophisticated photoperiodic mechanisms that detect changes in daylength to adapt
84 to seasonal environmental variations [16]. In the circadian range, robust nocturnal
85 elevation in vocal activity has been previously documented in several bird species
86 [17-19]. Most of the research related to circadian rhythmicity in vocalization has
87 been performed in songbirds [19, 20]. However, both circadian and ultradian
88 rhythmicity has previously been described in owls. For example, studies in the barn
89 owl (*Tyto alba*) showed that nestlings depend on the daily regulation of stress
90 hormones [21]. Moreover, ultradian rhythms in sleep-wakefulness were found in
91 barn owlets [22].

92 The Tawny Owl (*Strix aluco*) is a crepuscular predator - with little activity during the
93 day - that attacks vertebrate prey from perches in trees [23]. Tawny Owls are

94 extensively distributed throughout the Eurasian continent, from Britain in Western
95 Europe and northwest Africa to East and South Asia [24]. This work investigates
96 whether the vocalization pattern of male Tawny Owls (*Strix aluco*), from hundreds
97 of milliseconds to seconds range, is subjected to daily (early vs. late night) and
98 seasonal (spring vs. summer) rhythmicity. Our study addresses the general
99 hypothesis that, similar to songbirds, owl's vocalization pattern may also present
100 daily and seasonal variations. We present exploratory evidence indicating that
101 phase regularity (i.e., Interval timing consistency between calls) presents both daily
102 and seasonal variations.

103

104 **Materials and Methods**

105 **Experimental design**

106 Territorial calls (also referred as hooting) of 30 male Tawny Owls (*Strix aluco*) in
107 their natural environment were recorded in several European countries along
108 different seasons and recording times (see Table 1). Most recordings were
109 obtained from www.xeno-canto.org (Xeno-canto, XC), a non-profit website set up
110 to share recordings of bird sounds worldwide [25], which has already been used for
111 research purposes [26,27]. Only high quality recordings were chosen. Personal
112 recordings were obtained by the corresponding author. Each recording contained
113 calls of one focal owl, which serve both in territorial defense and mate choice. The
114 recordings were made during comparable, favorable meteorological conditions
115 (without strong winds or precipitation). The present study was purely observational
116 and non-invasive, therefore no special permits were required.

117

118 **Statistical analysis**

119 Digital sonogram analysis from audio files was performed by using Adobe Audition
120 software (San Jose, California, USA). Male Tawny owls presented a distinctive
121 pattern of two vocalizations (Call1-Call2) that was repeated over time (Fig 1). The
122 time interval between these two vocalizations was defined as T1, and the time
123 interval between each repetition event was named T2, as shown in Figs 1A and
124 1B. Mean times for Call1, Call2, T1 and T2 were calculated for each owl by taking
125 the average times of recorded calls, with a minimum of 6 calls per owl (Fig 1B). We
126 analyzed temporal variation (daily and seasonal) in these parameters. Tawny Owls
127 have nocturnal or crepuscular habits and, in order to assess daily variations in
128 vocalization, owls were divided into two groups: early owls (calls emitted between
129 17:00 and 0:00 h, n=17) and late owls (calls emitted between 0:00 and 6:00 h,
130 n=12). Owl #5 was excluded from daily analysis because recording time is
131 unknown. For seasonal analysis, owls were also divided into two groups: spring
132 owls (recorded between February and May, n=15) and summer owls (recorded
133 between June and September, n=12). Owls #20, #21 and #25 were excluded from
134 seasonal analysis because they were not recorded in the above-mentioned
135 months.

136 While the two vocalizations often contain distinct temporal and spectral profiles, the
137 spectral differences were absent in Owls #6, #25, #31. Therefore, an unbiased
138 classification of calls was done using an unsupervised DBSCAN clustering method
139 utilizing Python's sklearn package. The choice of this method over other classifiers

140 was twofold. First, it does not require the number of clusters to be specified *a priori*.
141 In addition to intervals T1 and T2, breaks in the typical vocal pattern may lead to
142 other groupings based on temporal properties. This allows the detection of subtler
143 complexities in vocalization structure. Second, it can detect arbitrarily shaped
144 clusters. As the variance of the model parameters may not be equal, this allows the
145 model to be run with minimal preprocessing or distortion of vocalization data.
146 Classification was done using all calls with two model features related to the
147 temporal components of the vocalization: duration of call (in seconds) and time
148 between calls (also in seconds). All call data was used allowing classifications to
149 be validated against calls with available spectral data. The model parameters for
150 maximum distance between samples (Eps) and the minimum number of samples
151 in a neighborhood (MinPts) were set to 0.9053 and 7, respectively, and the
152 distance metric was Euclidean. Parameterization was done in accordance with
153 previous research [28]. In short, MinPts was first set using the heuristic $\text{MinPts} \approx$
154 $\ln(n)$, rounded to the nearest whole number, where n is the number of samples.
155 Eps is then calculated by first finding the distances between *k-nearest neighbors*,
156 with k set to MinPts. These values are then sorted in descending order and the
157 point of maximum curvature is assigned to Eps.
158 Daily (early vs. late night) and seasonal (spring vs. summer) data were evaluated
159 using a two-tailed t-test. When equality of variances was not met, Welch's
160 corrections were applied.
161 Statistical analyses were performed using Graphpad Prism (GraphPad Software
162 Inc., CA, USA). In all cases, the alpha level was set at 0.05.
163

164 **Table 1. Mean values for *Strix aluco* vocalization patterns analyzed in the**
 165 **present study.**

Owl	Source Information	Sound Recordist*	Country	Month	Time	Call1	Call2	T1	T2
						(s)	(s)	(s)	(s)
1	https://www.xeno-canto.org/358345	Xeira, A.	Germany	Feb	19:30	1.041	1.262	5.063	18.154
2	Personal recording	Peryer, G.	England	May	00:00	1.016	1.572	3.453	27.591
3	https://www.xeno-canto.org/302231	M, D.	England	March	05:24	0.810	1.149	6.287	20.809
4	Personal recording	Peryer, G.	England	May	00:00	1.004	1.258	5.768	17.484
5	https://www.xeno-canto.org/374099	Rossi, C.	Spain	May	-	0.919	1.209	6.958	15.808
6	https://www.xeno-canto.org/171690	Knychata, A.	Poland	March	22:30	1.027	1.309	5.985	20.941
7	https://www.xeno-canto.org/333455	Buhl, J.	Germany	June	05:00	1.142	1.404	5.552	23.063
8	https://www.xeno-canto.org/342461	Buhl, J.	Germany	June	04:00	1.368	1.622	5.516	39.291
9	https://www.xeno-canto.org/176220	Szczypinki, P.	Poland	April	22:00	0.860	1.070	6.084	12.670
10	https://www.xeno-canto.org/310463	Knychata, A.	Poland	April	22:30	1.095	1.244	5.731	15.014
11	https://www.xeno-canto.org/323831	Buhl, J.	Germany	June	04:00	1.437	1.587	5.573	22.986
12	https://www.xeno-canto.org/323832	Buhl, J.	Germany	June	04:30	1.228	1.420	5.380	23.168
13	https://www.xeno-canto.org/329497	Buhl, J.	Germany	June	21:30	1.389	1.642	5.441	21.591
14	https://www.xeno-canto.org/324439	Buhl, J.	Germany	June	04:00	1.147	1.333	5.368	27.426
15	https://www.xeno-canto.org/324428	Buhl, J.	Germany	June	04:00	1.163	1.326	5.289	23.797
16	https://www.xeno-canto.org/333452	Buhl, J.	Germany	June	04:00	1.338	1.574	5.689	26.668
17	https://www.xeno-canto.org/310638	Aberg, P.	Sweden	March	21:00	0.926	1.039	5.925	14.408
18	https://www.xeno-canto.org/310628	Aberg, P.	Sweden	March	19:00	0.866	1.207	6.989	17.247
19	https://www.xeno-canto.org/240986	Yablonovska-Grishchenko, E.	Ukraine	April	05:00	0.872	1.088	6.436	26.026
20	https://www.xeno-canto.org/208101	Melichar, D.	Czech Rep.	Nov	19:00	0.887	1.267	4.842	22.020
21	https://www.xeno-canto.org/115886	Ryberg, E.A.	Norway	Dec	17:00	0.998	1.247	5.895	16.034
22	https://www.xeno-canto.org/54025	Dragonetti, M.	Italy	Sept	22:00	0.814	1.110	5.166	14.968
23	https://www.xeno-canto.org/393976	Livon	Estonia	May	21:00	0.868	1.035	4.946	16.278
24	https://www.xeno-canto.org/346106	Buhl, J.	Germany	June	04:00	1.467	1.743	5.682	28.239
25	https://www.xeno-canto.org/298881	Brookes, C.	England	Jan	17:30	0.855	1.187	7.525	22.295
26	https://www.xeno-canto.org/293574	van Bruggen, J.	France	July	23:00	0.831	1.587	4.183	15.678
27	https://www.xeno-canto.org/198251	Matacz, L.	Poland	April	21:20	1.030	1.732	5.640	15.839
28	https://www.xeno-canto.org/165382	Tumiel, T.	Poland	April	00:30	1.173	1.443	4.656	19.745
29	https://www.xeno-canto.org/143531	Szczypinki, P.	Poland	April	21:00	1.600	1.754	4.555	12.319
30	https://www.xeno-canto.org/328946	Buhl, J.	Germany	June	04:00	1.221	1.263	5.371	31.524

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167

168 **Fig 1. Vocalization pattern in male Tawny Owls. (A)** Sonogram depicting the

169 vocalization events of a single owl along time. Call1, Call2, T1 and T2 are shown.

170 The ~ 200 msec vocal bout that precedes the second call (bottom arrow) was not

171 present in all animals, and was not included in Call2. (B). Schematic representation

172 of temporal vocalization events in a single owl, showing this pattern of two

173 vocalizations (Call1-Call2) that is repeated as a function of time. The time interval

174 between these two vocalizations is called T1, and the time interval between each

175 repetition event is called T2. Mean values for each parameter were calculated per
176 owl.

177

178 **Results**

179 Our findings indicate that the temporal structure of Tawny Owl vocalizations - in the
180 seconds-to-minutes range of interval timing - presents both daily and seasonal
181 variation.

182 Table 1 displays the mean times for the parameters Call1, Call2, T1 and T2 (Fig 1
183 and Methods) of all 30 owls evaluated. In total, Call2 was longer than Call1,
184 presenting a mean time of 1.36 ± 0.22 sec (mean \pm S.D.), while mean time for
185 Call1 was 1.08 ± 0.22 sec. ($t_{29}=9.768$, $p<0.0001$, two-tailed paired t-test). Interval
186 times T1 and T2 presented an average of 5.57 ± 0.82 sec and 20.95 ± 6.12 sec,
187 respectively ($t_{29}=13.51$, $p<0.0001$, two-tailed paired t-test). Moreover, T2 intervals
188 presented higher variability compared to T1. In this sense, the coefficient of
189 variation (CV) for the 30 owls evaluated was higher for T2 than T1 ($t_{29}=7.211$,
190 $p<0.0001$, two-tailed paired t-test, S1 Fig). An analysis of the geographical
191 distribution of all data evaluated indicated no significant effect of region in all four
192 parameters analyzed (S2 Fig).

193 As not all owls emit a short vocalization prior to Call2 (Fig 1A) and occasional
194 breaks in the vocalization pattern lead to repeated call types, an unsupervised
195 clustering algorithm was run on interval times. This allowed for unbiased
196 classification of intervals that may be distinct from T1 and T2 distributions (S3 Fig).

197 The data were found to have four discrete clusters: initial calls, T1 calls, T2 calls
198 and long T2 calls.

199 As short prior vocalizations are unique to Call2, the accuracy of the classifier was
200 assessed by comparing the number of intervals identified as T1 against known T1
201 intervals that were followed by an identifiable Call2. The classifier showed
202 exceptional accuracy by correctly identifying all 286 T1 intervals in owls that
203 emitted short vocalizations prior to Call2 and only 9 of 325 as T1 intervals when it
204 was not followed by a Call2, giving a classification accuracy of 98.5%. Interestingly,
205 despite the difference in spectral composition, there was no significant difference
206 between the duration of T2 calls with or without short vocalizations prior
207 ($t_{29}=-0.404$, $p=0.689$, two-tailed unpaired t-test).

208 Daily and seasonal analysis was performed for the parameters Call1, Call2, T1 and
209 T2. For that purpose, owls were classified as early vs. late and spring vs. summer
210 (see Methods). Call1 presented significant time of day and seasonal effects. As
211 shown in Fig 2, Call1 duration was shorter in early vs. late owls ($t_{27}=2.481$,
212 $p=0.0196$, two-tailed unpaired t-test, Fig 2A). A similar effect was observed in
213 spring vs. summer owls for both Call1 ($t_{25}=3.464$, $p=0.0019$, two-tailed unpaired t-
214 test, Fig 2B) and Call2 ($t_{25}=2.749$, $p=0.0109$, two-tailed unpaired t-test, Fig 2E). On
215 the other hand, there were no time of day differences in Call2 duration ($t_{27}=1.047$,
216 $p=0.3043$, two-tailed unpaired t-test, Fig 2D). Cluster heat maps were generated
217 from these data to visually represent the increase or attenuation in call duration
218 across the different groups. Fig 2C and F display heat maps containing the mean
219 value per owl (colored square) for Call1 and Call2, respectively. Color rank for
220 Call1 is clearly different for early vs. late owls, as well as for spring vs. summer

221 owls, while Call2 color rank is visibly different only in spring vs. summer owls.
222 Additionally, the coefficient of variation (CV) for Call1, calculated as the ratio
223 between the standard deviation and the mean, was significantly increased in
224 summer vs. spring owls ($t_{25}=3.063$, $p=0.0052$, two-tailed unpaired t-test).

225

226 **Fig 2. Daily and seasonal variation in call duration.** (A) Call1 duration in early
227 vs. late owls ($p=0.0197$), (B) Call1 duration in spring vs. summer owls ($p=0.0019$),
228 (C) Heat map for Call1. (D) Call2 duration in early vs. late owls ($p=0.3043$). (E)
229 Call2 duration in spring vs. summer owls ($p=0.0109$), (F) Heat map for Call2. Data
230 from scatter dot plots represent the mean value for each owl. In heat maps, each
231 row corresponds to the mean value per owl, and the columns represent the
232 different groups ($n=17$ for early owls, $n=12$ for late owls, $n=15$ for spring owls, and
233 $n=12$ for summer owls). ** $p<0.01$, * $p<0.05$, two-tailed Student t-test.

234

235 Owls also displayed significant time of day and seasonal effects in T2, as shown in
236 Fig 3. Calls from late owls presented longer T2 intervals compared to calls from
237 early owls ($t_{27}=4.849$, $p<0.0001$, two-tailed unpaired t-test, Fig 3A). T2 also
238 exhibited a seasonal variation, with longer values in summer vs. spring owls
239 ($t_{25}=3.228$, $p=0.0035$, two-tailed unpaired t-test, Fig 3B). These differences in T2
240 can also be observed in the cluster heat map shown in Fig 3C. There were no
241 effects of time of day or season in T1 values ($t_{24}=0.3124$, $p=0.7574$ for time of day,
242 Fig 3D; $t_{19}=1.032$, $p=0.3147$ for season, Fig 3E, two-tailed unpaired t-test with
243 Welch's correction; cluster heat map in Fig 3F). However, both standard deviation
244 (SD) and coefficient of variation (CV) for T1 were significantly increased in early vs.

245 late owls ($t_{24}=2.718$, $p=0.0134$ and $t_{24}=2.646$, $p=0.0166$, respectively, two-tailed
246 unpaired t-test with Welch's correction).
247 With the duration of T2 intervals falling into two distinct groups (i.e. T2 and long
248 T2), the existence of a multiplicative relationship between the mean time and
249 standard deviation of these populations was assessed. Due to the sparseness of
250 long T2 calls, a Bayesian approach for estimating the population's distribution was
251 used. Both the mean and standard deviation of long T2 calls were found to not be
252 significantly different than twice that of the T2 call distribution (Fig 4). Some
253 examples of distribution of T2 and long T2 interval times around the median are
254 shown in S4 Fig.

255

256 **Fig 3. Daily and seasonal variation in timing between calls.** (A) T2 duration in
257 early vs. late owls ($p<0.0001$), (B) T2 duration in spring vs. summer owls
258 ($p=0.0037$), (C) Heat map for T2, (D) T1 duration in early vs. late owls ($p=0.7574$),
259 (E) T1 duration in spring vs. summer owls ($p=0.3147$), (F) Heat map for T1. Data
260 from scatter dot plots represent the mean value for each owl. In heat maps, each
261 row corresponds to the mean value per owl, and the columns represent the
262 different groups ($n=17$ for early owls, $n=12$ for late owls, $n=15$ for spring owls, and
263 $n=12$ for summer owls). *** $p<0.0001$, ** $p<0.01$, two-tailed Student t-test.

264

265 **Fig 4. Distribution of T2 and Long T2 intervals.** (A) Histogram and individual
266 Gaussian fits for T2 and Long T2 intervals. (B) Scatter plot of the mean and 95%
267 CI of the credible parameter space for the Long T2 intervals' Gaussian Fit

268 normalized to those of the T2 interval fit. Solid red line indicates the value that
269 would be twice that of the T2 fit.

270

271 **Discussion.**

272 Vocal communication has an important biological function in male owls and is used
273 for attracting females as well as establishing territory. Previous analysis of the
274 frequency and temporal components of the calls made by male Tawny Owls has
275 suggested a relationship with the health and fitness of the owl as indexed by
276 parasite burden [29]. A number of factors are known to influence the vocal activity
277 pattern of nocturnal birds. One factor is the time of the year, with calling rate
278 varying along the breeding cycle because of the territorial/mating functions of calls
279 [30-32]. Another factor is the time of day, with most owl species being more vocally
280 active during dusk and dawn [17]. Thus, circadian changes in vocal production
281 have been displayed in non-oscine birds such as the domestic Japanese quail
282 (*Coturnix coturnix japonica*) [18]. Moreover, melatonin - the major timekeeping
283 hormone in vertebrates - affects the temporal pattern of vocal signatures in both
284 the oscine bird zebra finch (*Taeniopygia guttata*) and the non-oscine Japanese
285 quail [19]. Melatonin is also relevant for vocal communication in the midshipman
286 fish. Nocturnal fish vocalizations follow both daily and circadian rhythmicity under a
287 light/dark cycle and constant dark conditions, respectively, and are rescued by
288 melatonin under constant light [33]. These studies in birds and fish show daily
289 and/or seasonal variations on comparable time scales as used in the present work.
290 However, such investigations have not been previously researched in owls.

291 There are several indications that interval timing is present in songbirds but, again,
292 there is no information on the subject in owls. Indeed, it has been proposed that
293 the syllable can be used as a reliable time marker in order to predict song
294 completion [34]. Calls in non-oscine birds are also temporally structured. In
295 particular, male Tawny Owl calls present an organized structure of two clear
296 vocalizations repeated over time, with a mean fundamental frequency below 1 kHz
297 [35]. Call duration and silent intervals between these male Tawny Owl
298 vocalizations fit in the seconds-to-minutes range of interval timing. Here we found a
299 remarkable diurnal (early vs. late night) and seasonal (spring vs. summer) variation
300 in parameters Call1 and T2, as well as seasonal variation in parameter Call2.
301 Specifically, the time interval between each 2-call repetition event (T2) was around
302 18 sec in early owls and around 26 sec in late owls. These data indicate that time
303 of day regulates the timing between calls. In a similar way, seasons influence the
304 length of T2, being around 18 sec for spring owls and 25 sec. for summer owls. T1,
305 on the other hand, remained mostly invariant around 5 sec.
306 This is the first description of such changes in non-oscine birds. In songbirds,
307 syllables, intervals, motor control, and respiratory pathways have been well
308 described [36]. Moreover, a clear circadian variation in song and calling behavior
309 was found in zebra finches, controlled by pineal melatonin signaling [37]. Because
310 we have previously demonstrated a role for melatonin in the circadian-interval
311 timing interaction in other models [38], it is tempting to speculate that the pineal
312 gland might influence the temporal allocation and short-length duration of owl
313 vocalizations. Although this is beyond of the scope of the present study, the
314 biological function of daily and seasonal variation in Tawny Owl vocalizations may

315 be related to the breeding and territorial behaviors, ensuring the maximal survival
316 of their offspring [16].

317 Some limitations of the present study must be considered. First, since only high-
318 quality recordings were analyzed, our dataset is not widely distributed over the
319 seasons (e.g., the summer season presents fewer recordings from July to
320 September compared to June). Second, recordings were not performed under
321 standardized or laboratory conditions. However, any fluctuations in recording
322 distance or vegetation that may induce differences in reverberation times are
323 relatively small for the fundamental frequencies recorded (close to 1 KHz) [39, 40],
324 and therefore do not affect the parameters evaluated in the scale of interval timing.

325 Finally, although we have no information related to the age of the animals
326 evaluated, the parameters of Tawny Owl calls make individuals traceable over
327 years [8], indicating vocal consistency within members of this species. Despite the
328 listed methodological limitations, a clear variation in the temporal structure of calls
329 can be identified.

330 Given that a robust relationship between the circadian oscillator and events in the
331 seconds-to-minutes range has been previously established [41-44], these results
332 contribute to the expansion of the role of the circadian system in regulating the
333 shorter-duration temporal events mediated by the interaction of “time cells” in the
334 cerebellum, striatum, and hippocampus [11, 45, 46]. Moreover, these novel
335 findings take the lead in establishing a fundamental relation between interval timing
336 in the seconds-to-minutes range and daily photoperiod as a function of the
337 annual/seasonal cycle [47]. We provide a foundation for future studies of owl
338 vocalizations under standardized conditions which may examine the observed

339 temporal patterns in greater detail. Collaborating with contributors to existing
340 databases of nature recordings may lead the way to more powerful field research
341 to further understand temporal determinants of behavior.

342

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346

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