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1 Running head: *Season and syntax reversion in Great Tits*

2 **Biological conclusions about importance of order in mobbing calls vary**
3 **with the reproductive context in Great Tits *Parus major***

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10

11 Currently, there is considerable debate surrounding the presence of some human language
12 specific characteristics in non-human animals, such as the use of compositional syntax (i.e.,
13 meaning of a sequence determined both by meaning of its individual parts and in the way they
14 are combined). Compositional syntax has been investigated in mobbing calls of two closely
15 related tit species, the Japanese Tit *Parus minor* and the Great Tit *Parus major*, but with one
16 contrasting result: hearing calls in the reversed order diminished the behavioural responses of
17 Japanese Tits, however only partially those of Great Tits. This difference may have been due
18 to an external factor such as the season in which the experiment was undertaken, as the Japanese
19 Tits were tested in winter and Great Tits in spring. Here, we studied the responses of Great Tits
20 towards natural and reversed mobbing sequences during spring and winter by investigating two
21 behaviours: approaching and vigilance behaviours. We found that sensitivity to syntax reversal
22 was impacted by the season. The birds were vigilant but less likely to approach reversed calls
23 in winter. However the opposite occurred in spring, with the birds scanning less but still
24 approaching. This study suggests that the perception of combinatorial calls in Great Tits is
25 influenced by the season, emphasizing the importance of context in studies investigating
26 complex cognitive processing in animals.

27 **Keywords:** Acoustic communication, Alarm call, Compositionality, Mobbing Behaviour,

28 *Paridae*

29 Animal communication is one of the most prolific subjects in animal behaviour studies
30 (Bradbury & Vehrencamp 2011), with one major question being the extent to which we can
31 compare it to human language. Some defining characteristics of human language are
32 classically presented as intentionality, referentiality, vocal learning, and syntax (Hauser *et al.*
33 2002). Numerous comparative studies conducted in animal systems have focused on the first
34 three criteria - intentionality (Seyfarth & Cheney 2003, Graham *et al.* 2019), referentiality
35 (e.g., in Siberian Jays *Perisoreus infaustus*, Griesser 2008), and vocal learning (e.g., song in
36 oscines, Wilbrecht & Nottebohm 2003, Tyack 2019) - and have revealed that these abilities
37 are present in diverse animal species, emphasizing the importance of studying animal
38 communication in our understanding of the evolution of language (Fishbein *et al.* 2019).
39 Indeed, finding such characteristics in our closest relatives may indicate a gradual emergence
40 of language features (Searcy 2019). In contrast, the presence of analogous traits in distant
41 species suggests convergence: the independent rise of the same evolutionary strategy (Searcy
42 2019).

43 Birds are one great example of distant taxa possessing characteristics similar to human
44 language, especially regarding complex combinatorial rules (i.e., a set of principles by which
45 meaning-bearing units can be combined into well-formed complexes, Zuberbühler 2019a).
46 However, previous findings have focused on sequences lacking direct semantic content, with
47 many combinatorial forms being phonocoding (i.e., sound combinations whose individual
48 parts do not possess proper meaning, Engesser & Townsend 2019). Numerous examples of
49 such combinatorial rules can be found in bird songs. For instance, while information about
50 male quality is extracted from the global song, the individual syllables that make up the song
51 do not carry specific information (Catchpole & Slater 1995).

52 Recently, studies have begun to discover various forms of semantic combinatoriality
53 in animals, with some recent research focusing on compositional syntax (Engesser &

54 Townsend 2019, Suzuki *et al.* 2019b, Zuberbühler 2019b). This is observed when, in
55 opposition to phonocoding, the meaning of the whole depends on the meanings of the
56 component parts, and the way they are organized (Hurford 2011). Cases where animals use
57 such compositional structuring have been reported in birds when mobbing predators
58 (Engesser *et al.* 2016, Suzuki *et al.* 2016, Suzuki *et al.* 2017).

59 Mobbing - when a prey moves towards and harasses a predator to chase it away
60 (Carlson *et al.* 2018) - is a suitable behaviour to study compositional syntax. Indeed, prey
61 produce calls to recruit mobbers from a variety of species (Hurd 1996, Randler & Vollmer
62 2013, Dutour *et al.* 2016). In such risky situations (Curio & Regelman 1986), where callers
63 and receivers have a level of shared interest, and communication should be clear and
64 unambiguous to co-ordinate a response that combines several behaviours, it has been
65 suggested that compositional syntax could emerge (Griesser *et al.* 2018). Experimental studies
66 in the Japanese Tit *Parus minor* provide the strongest case of compositional syntax to date
67 (Suzuki *et al.* 2016, Suzuki *et al.* 2017, but see also Engesser *et al.* 2016 on Southern Pied
68 Babblers *Turdoides bicolor*). Indeed, Suzuki and colleagues demonstrated that the mobbing
69 sequences of Japanese Tits are made through the concatenation of two types of notes -
70 Frequency modulated elements (FME) and D notes (FME-D calls), respectively triggering a
71 vigilance and an approach behaviour - and that order has importance (Suzuki *et al.* 2016).
72 Debates about these results have been profuse (Bolhuis *et al.* 2018a, 2018b, Suzuki *et al.*
73 2018, Townsend *et al.* 2018), and there is a need for broader data on potential
74 compositionality in birds.

75 Dutour *et al.* (2019b) replicated the same experiment on a French population of Great
76 Tits *Parus major*, during the breeding season. This species is the closest relative of the
77 Japanese Tit (Päckert *et al.* 2005, Johansson *et al.* 2013) and produces similarly structured
78 mobbing calls (FME-D calls, Fig. 1). For both species, the FME motif appears to be used in

79 vigilance situations and serves as an alarm signal to which receivers respond by scanning for
80 danger (Suzuki *et al.* 2016, Dutour *et al.* 2019b). The D notes are produced within foraging
81 flocks and by nest mates to recruit social partners and elicit an approach by receivers
82 (recruitment calls, Suzuki *et al.* 2016, Dutour *et al.* 2019b). Therefore, these acoustically
83 distinct calls fulfil the first characteristic of compositionality (i.e., each different part has
84 meaning when produced in isolation). In addition, Great Tits showed excitement signs (wing
85 flicking and body swinging) only toward the combined FME-D calls, and never toward the
86 isolated parts, indicating a meaning more complex than the simple sum of the isolated parts
87 (Salis *et al.* 2020). The second characteristic of compositional structuring reflects the
88 importance of organization, tested through measuring the Great Tits' response when the
89 sequence is reversed (D-FME, demonstrated in the Japanese Tit, Suzuki *et al.* 2016). Here
90 however, almost as many Great Tits approached the speaker to natural sequence (FME-D)
91 than to reversed sequence (D-FME, Dutour *et al.* 2019b). Therefore, the conclusion on syntax
92 use in Great Tits suggested it may not be as clear-cut as for the Japanese Tit and may globally
93 weaken a generalization of syntax results to other species in the genus.

94 One crucial distinction between the studies of Japanese Tits and Great Tits, however,
95 is that the response of Japanese Tits to the inverted sequence was tested during the non-
96 breeding season in winter flocks (Suzuki *et al.* 2016), while these tests were done in a
97 reproductive context at the onset of the breeding season for the Great Tits (Dutour *et al.*
98 2019b). The behaviour of Parids is known to vary markedly between seasons. When breeding,
99 they show enhanced individual territoriality and aggressive behaviour (Shedd 1983, Krams
100 and Krama 2002, Samplonius 2018). In contrast, they join other individuals and form flocks
101 in winter (Hinde 1952, Marra *et al.* 2015). In addition, Great tits approached more
102 heterospecific calls in winter than in the breeding season (Dutour *et al.* 2019a). The attention
103 paid to the same auditory stimuli therefore seems to be dependent on the reproductive state of

104 the birds. We can hypothesize that increased aggressiveness toward intruders in spring may
105 directly affect the behavioural response recorded during syntax-related studies (i.e., approach
106 and scanning, Suzuki *et al.* 2016). In addition, sensibility to conspecific D notes may increase
107 in spring, as pairs use them frequently, and fledglings use similar notes when calling their
108 parents (M.D. pers.obs). In the same way, an increased response toward heterospecific calls
109 (even towards unknown ones, Dutour *et al.* 2017b) in winter potentially indicates an increased
110 attention towards new calls. As a consequence, different biological conclusions could result
111 from the same experiment done in different parts of the life cycle, even if the general syntax-
112 processing ability of the bird remain the same. On a more global scale, determining whether
113 context can impact conclusions on syntax use in Great Tits could be of value in influencing
114 future studies investigating complex language features in animals.

115 Here, we used field playback experiments to examine whether season has an impact on
116 sensitivity to syntax reversion in Great Tits, allowing us to understand the differences found
117 in the two studies investigating the use of syntax in the family Paridae. Specifically, we
118 recorded the behavioural response of Great tits to either naturally ordered calls, reversed calls,
119 or to a control (background noise) in two seasons: in spring, when territoriality is increased,
120 compared to winter, when individuals join other species in flocks.

121

122 **METHODS**

123 **Study sites and experimental design**

124 Data were collected at the start of the breeding season when individual territories are
125 established (February/ March 2018, territories are established in February and defended until
126 June in the study population) and during the winter when tits often form mixed-species flocks
127 (November 2018/ January 2019, Hinde 1952), in the north of Lyon, France.

128 The ecological context of the two seasons is different. In spring, Great Tits invest
129 mainly in breeding (i.e., defending a territory, building a nest, rearing a brood of nestlings,
130 Cramp *et al.* 1993). The defence of the nest against any intruder is intense and well
131 documented (Hinde 1952, Hollander *et al.* 2008). As winter arrives, Great Tits join small
132 conspecific groups and often heterospecific flocks (Carlson *et al.* 2020). Even if some
133 dominance relationship remains (Oberski & Wilson 1991), flocks can combine, and space is
134 divided into undefended overlapping home ranges rather than discrete territories (Ekman
135 1989). Flocks are thought to be an efficient behavioural response toward predator and
136 foraging pressures being different in winter (Sridhar *et al.* 2009). Indeed, increased food
137 storage to counteract the loss of food supply increases predation risk (increase of body mass
138 being negatively correlated with manoeuvrability, Gosler *et al.* 1995), while a decrease in
139 individual territory defence whilst not breeding allows birds to gather without strong
140 competition (Morse 1970).

141 In order to examine whether the response of Great Tits was dependent upon season,
142 we conducted three playback types. First, we examined whether tits hearing the combined
143 motifs in the natural order (FME-D calls, natural mobbing call sequence) displayed a
144 combination of the behaviours classically monitored in mobbing studies (approach and scan,
145 see below for more details), and we tested whether this was also the case when the combined
146 motifs were presented in the reverse order (D-FME calls, artificially reversed mobbing call
147 sequence). Finally, we also performed control tests for which the playback contained only
148 background noise (hereafter referred as BN tests). We conducted these tests on 120 adult
149 Great Tits (20 individuals for each call sequence, at two different seasons).

150

151 **Field test procedure**

152 Each test was conducted by two field assistants. One was assigned to the soundtrack
153 preparation and playback operation, while the other was assigned to the observation of the
154 focal bird. After the localization of a focal bird, the loudspeaker was placed 30 m away from
155 the bird at the bottom of a tree. Both field assistants were positioned opposite each other at
156 vantage points at least 15 m from the loudspeaker and the focal bird to avoid any perturbation
157 during the test. Prior to playback, the baseline behaviour of the focal bird was observed for at
158 least a one-minute pre-trial period. The playback was started when there was no other
159 passerine observed near the focal individual. If the bird showed alarm behaviour before the
160 test (e.g., gave mobbing calls), or if other birds also responded to the playback, the test was
161 abandoned. The sequence was broadcast using a remotely controlled Shopinnov 20 W
162 loudspeaker with an amplitude of $\sim 80 \pm 3$ dB(A) (measured at 1 m from the loudspeaker
163 using Lutron SL-4001, Taipei, Taiwan; C weighting, slow settings, re: 20 μ Pa), as this is the
164 natural amplitude of Great Tit mobbing calls (from 77 to 80 dB, Templeton *et al.* 2016). The
165 background noise observed in the field (mean \pm sd = 46.1 \pm 2.1 dB(C), n = 20 measurements)
166 associated to the spherical spread and excess attenuation of sound energy radiating from a
167 source in a deciduous forest (Lengagne & Slater 2002) allowed us to estimate that the
168 broadcast signal reached background noise level at 60-70 m distance.

169 Subsequently, during one minute of playback treatment, two behavioural variables
170 were recorded, respectively, (1) the vigilance effort with the number of horizontal scans (we
171 counted the number of obvious movements that birds made with their heads from left to right
172 or right to left ($\sim 180^\circ$ turn, Suzuki *et al.* 2016) and (2) approach within 15 m of the
173 loudspeaker (Dutour *et al.* 2017b). Scanning for threats is a stereotyped agitation behaviour
174 (Curio 1975, 1978) commonly measured in studies of mobbing response of tits (e.g., Suzuki
175 *et al.* 2016, Carlson *et al.* 2017, Suzuki *et al.* 2017). Both dense vegetation and the small size

176 of these passerine birds prevented a detailed measurement of scanning behaviour using video-
177 recording. These behaviours were thus directly counted an observer using binoculars and
178 reported on a digital audio recorder. To minimize the measurement errors and to make
179 scanning as objective a behaviour as is possible, we considered an individual to scan only if
180 the head movements of the bird from right to left or left to right were obvious. These methods
181 were unable to completely eliminate the potential for unconscious observer bias and we
182 acknowledge that future studies using similar techniques should ideally employ fully blinded
183 designs.

184 All trials were conducted between 06:00 and 13:00 h during calm and dry weather. On
185 average 10 tests were done each day. Selection of all focal animals was based on location,
186 ensuring successively tested animals were always separated by more than 100 m to minimize
187 pseudoreplication risk and ensure that tested birds had not heard a previous test, given that the
188 active space of a sound emitted at 80 dBA with a 46 dB background noise level was less than
189 100 m (Brenowitz 1982, Lengagne & Slater 2002).

190 In addition, we never returned to the same forest path, the size of the research area was
191 large (i.e. $\sim 43 \text{ km}^2$), and population density in this area is high (M.D. unpubl. data). No bird
192 was seen following us. Hence, although birds were not individually ringed, the probability of
193 testing the same individual twice was low and we are confident that our observations were
194 performed on different individuals.

195

196 **Playback stimuli and materials**

197 We used mobbing calls produced by three Great Tits, previously recorded in response to
198 intraspecific mobbing calls (Dutour *et al.* 2017b). Calls were recorded with a Fostex FR2LE
199 digital recorder (Tokyo, Japan) connected to a Sennheiser ME67-K6 microphone (Wedemark,
200 Germany; see Dutour *et al.* 2017b for more details). We also used mobbing calls obtained

201 from the Xeno Canto online database (www.xeno-canto.org) recorded in different European
202 countries ($N = 7$). By controlling several factors when constructing our playbacks (see below),
203 we homogenised our stimuli and reduced potential variation between different geographical
204 areas. We recorded or selected good quality (sampling rate: 44.1 kHz; sample size: 16-bits)
205 recordings, and all playback files were saved as .wav files.

206 From these recording files, we built 20 unique soundtracks of natural mobbing calls
207 (i.e., 20 soundtracks with one individual per soundtrack) using Avisoft-SASLab software
208 (Avisoft Bioacoustics, Glienicke, Germany, one sample of spectrogram and audio file
209 available in Sup.Mat.). These soundtracks were then used to construct the D-FME (i.e.
210 reversed) calls by reversing the motifs order of the original FME-D calls of Great Tits. The
211 same donor individuals were therefore used for both the natural and reversed call sequences.
212 Within each soundtrack, calls were repeated at a rate of 26 calls per minute (characteristics
213 within the range of the natural repetition rates, Dutour *et al.* 2019b). We used a series of five
214 to eight D motifs to construct playback (mean \pm se = 7.11 ± 0.06). In each track, D notes were
215 placed 82 ± 40 ms before FME (no difference with time between FME and D notes in the
216 natural sequence; t -test = 1.79, $P > 0.05$). All playback sequences were arranged to control the
217 ratio of the D notes over the FME notes in a mobbing call sequence, as note composition is
218 known to alter Great Tits' response (Kalb & Randler 2019). We also constructed 20 control
219 soundtracks using sections where no birds were calling in the same recordings as natural
220 mobbing calls (BN). In order to avoid pseudoreplication (Kroodsma *et al.* 1989), each
221 soundtrack was played back only once.

222

223 **Statistical analysis**

224 All analyses were carried out in R Studio 1.2.5033 (R core development team 2018)
225 using generalized linear mixed models (GLMM, *glmer* package *lme4*). Because the same

226 soundtracks were used in both seasons, soundtrack ID was included as a random effect. To
227 analyse the number of horizontal scans, we used a Poisson error distribution, and a log-link
228 function as no overdispersion was detected (*overdisp.glmer*, package *RVAideMemoire*). We
229 also included the actual time that the bird had been seen during the one-minute trial as offset
230 (57.48 ± 6.36 sec, mean \pm sd) and discarded three individuals (all in the spring-natural order
231 call sequence) for which the time was under 20 sec (i.e., sample size for each treatment is 20,
232 except for the spring-natural call sequences with $n = 17$). Approaching behaviour
233 (approaching vs. not approaching, where 1 = response, 0 = no response) was analysed using a
234 binomial distribution for the error term.

235 For both behavioural variables, the same three steps of analysis were carried out: (1)
236 we first ran a model with the three treatments (BN, Natural, and Reverse) at each season. We
237 then extracted the estimated marginal means and pairwise comparisons with related effect
238 sizes (package *emmeans*, function *emmeans*, and package *multcomp*, function *multcomp::cld*).
239 We only focused on five comparisons: in each season, the BN treatment versus the two other
240 treatments, and the difference between the BN treatment in spring and in winter. We therefore
241 used Bonferroni correction with $\alpha = 0.05/5 = 0.01$. (2) Secondly, we discarded the BN
242 tests and ran a model only with Natural and Reversed treatments in order to test the
243 significance of the interaction term (Season* Treatment, *Anova* function, package *car*). This
244 term indicates whether the difference between the two treatments is different between
245 seasons. (3) Finally, we reported the effect sizes of the difference between Natural and
246 Reversed treatments for the two seasons (two comparisons: $\alpha = 0.025$). The pairwise
247 comparisons and the related effect sizes of the differences were assessed using odds ratio
248 (hereafter OR, *odds.ratio*, package *questionr*) for the approach behaviour, and with the
249 exponential effect size for the scanning behaviour (package *emmeans* and *multcompview*).

250 Finally, we also verified that approaching behaviour was not correlated to scanning
251 behaviour using Wilcoxon tests for each season and each playback type. As revealed by these
252 analyses, no association between these two behaviours was detected (all $P > 0.05$).

253

254 **RESULTS**

255 **Scanning Behaviour**

256 In both seasons, Great Tits scanned less toward the control treatment (BN =
257 background noise) than toward the two other call sequences (all $P < 0.001$, Fig. 2, Table 1a),
258 except for the reversed call sequences in spring ($P = 0.89$, Table 1a). There was no significant
259 difference in scan number between seasons for BN soundtracks ($P = 0.03$, $\alpha = 0.01$, Table 1a).

260 The interaction between treatment and season was statistically significant when the
261 control tests were discarded (Table 1b), with a substantially larger difference between
262 acoustic treatments in spring than in winter. Indeed, Great Tits produced 2.29 more scans
263 toward natural calls than toward reversed calls in spring (Table 1c, Fig. 2), while there was no
264 such difference between treatments in winter (ratio of 1.3, Table 1c, Fig. 2). The treatment
265 term was also statistically significant, indicating an overall reduced response to reversed calls
266 ($P < 0.001$, Table 1b).

267

268 **Approaching behaviour**

269 Respectively 50% and 65% of Great Tits approached natural calls in spring and
270 winter. In contrast, only 5% of Great tits in spring, and none of them in winter, approached
271 toward the controls (Fig 3). The difference between natural calls and controls was statistically
272 significant in both seasons (Table 1a). In contrast, no statically significant difference in
273 probability of approach was detected between reversed calls and controls (35% in spring, 25%
274 in winter, Fig 3, Table 1a). There was almost no approach exhibited to control playbacks and

275 so the difference between controls in spring and in winter was not significant ($P = 0.49$, Table
276 1a).

277 The interaction between treatment and season was not statistically significant in our
278 model ($P = 0.25$, Fig 3, Table 1b), but the treatment term was statistically significant (more
279 approach to the natural treatment, $P = 0.02$, Fig 3). Nonetheless, even though no interaction
280 was detected, we found a statistically significant difference between reversed and natural
281 playback in winter (OR = 5.57, Table 1c), but not in spring (OR = 1.86), indicating that Great
282 Tits reduced their response toward reversed calls in winter, but approached both treatments
283 similarly in spring.

284

285 **DISCUSSION**

286 Great Tits reduced their overall behavioural response toward reversed calls compared
287 to natural ones. However, season affected the strength of that effect. Great Tits strongly
288 reduced their scanning behaviour when hearing reversed calls compared to natural calls in
289 spring, but such a reduction was not statistically significant in winter. In contrast, for the
290 approach behaviour, the difference between natural and reversed call sequences was
291 statistically significant in winter, but not in spring. The compositional syntax hypothesis
292 requires a decrease in behavioural responses (i.e., scanning and approaching) toward reversed
293 call sequences. Here, we demonstrate that testing birds in different seasons would have
294 resulted in different biological conclusions about syntax-use ability in tits.

295 For the approach behaviour, the difference between natural and reversed calls was
296 significant in winter, but not in spring. Importantly however, the interaction in our model was
297 not statistically significant. This may be due to the response variable being binary, thus
298 limiting power to detect effects with the available sample sizes. Nonetheless, the fact that this

299 difference was strong in spring but not in winter is some evidence of an effect of season on
300 the response to different treatments by Great Tits. The absence of variation in the probability
301 of approach in spring is consistent with the results of Dutour *et al.* (2019b) in which the same
302 protocol was used, and could therefore explain the difference with the study of Suzuki *et al.*
303 (2016), in which birds were tested in winter. These results therefore suggest that season does
304 affect Great Tit behavioural responses to natural and reversed playback.

305 Seasonal effects on responses to different syntactic forms has previously been
306 demonstrated in birds. For example, a similar result was found by Clucas *et al.* (2004)
307 investigating the vocal response of Carolina chickadees *Poecile carolinensis* when confronted
308 with familiar (AAADDD) and unfamiliar order calls (ADADAD): birds discriminated the
309 familiar from the unfamiliar calls in winter, but not in summer. Both Carolina Chickadees and
310 Great Tits live in mixed species flocks during winter (Hinde 1952), which are thought to
311 increase both foraging efficiency and defence against predators (Sridhar *et al.* 2009).
312 Vigilance toward heterospecific and potentially unfamiliar calls may therefore increase in
313 winter, as contact with heterospecifics with respect to food and alarm calls is common (Morse
314 1970, Clay *et al.* 2012). Corroborating this hypothesis, Great Tits increased their response
315 toward heterospecifics in winter compared to spring (Dutour *et al.* 2019a). We can therefore
316 hypothesize that in winter, birds are highly vigilant and more efficient in discriminating
317 unfamiliar calls, consequently leading to high scanning behaviour and low approach
318 behaviour toward artificial sequences. In contrast, during the breeding season, the response of
319 Great Tits is probably linked to strong competition and reproductive investment expressed
320 through increased aggression (Wingfield & Ramenofsky 1985). This leads to a more risk
321 prone behaviour: approaching toward any calls similar to recruitment calls (i.e., calls with D
322 notes, Kalb *et al.* 2019) without necessarily scanning. Further research is clearly required to
323 test this hypothesis and to understand why the response to reversed ordering of the mobbing

324 call sequence varies according to the season. In particular, comparisons of the response to the
325 isolated recruitment call (FME notes) and alarm call (D notes) according to the season is still
326 lacking. Such experimental analysis is required to fully appreciate the underlying reason for
327 scanning and approaching behaviours varying according to the season. Moreover, testing such
328 a hypothesis could help understand whether during the breeding season, Great Tits are indeed
329 more sensitive to recruitment calls, irrespective of the syntactic form.

330 Our findings reveal that in spring, we could not conclude that Great Tits reduced their
331 approach behaviour when hearing reversed calls. On the opposite, the reduced scanning
332 response was statistically significant in spring, but not in winter. Consequently, the same
333 experiment done in different seasons would not have led to the same biological conclusions.
334 Indeed, approaching and scanning more toward natural mobbing call sequences than toward
335 reversed call sequences is an essential prerequisite for compositional syntax in birds (Suzuki
336 *et al.* 2016). Concluding whether Great Tits do truly use syntax is consequently difficult: it is
337 unlikely that high mental ability such as syntax use vary with the seasons; but we show that
338 the external proof of it (the bird's behaviour) does. This may reflect a change of priorities in
339 birds between seasons (heterospecific co-operation in winter versus high territorial defence in
340 spring). In fact, such a problem may be common in language-related studies of animal
341 behaviour (Graham *et al.* 2019). For example, deciphering whether chimpanzees *Pan*
342 *troglydtes* possess a zero or second order intentionality level (i.e., when the sender does or
343 does not intend to signal and attribute mental state to the receiver; Townsend *et al.* 2017) is
344 dependent on aspects of predator stimuli (Crockford *et al.* 2012, Schel *et al.* 2013, Graham *et*
345 *al.* 2019).

346 Even if the effect sizes of the differences varied between the two seasons, we found an
347 overall reduced response in both scanning and approaching behaviours in response to the
348 reversed mobbing calls. This is consistent with previous studies of bird syntax (Suzuki *et al.*

349 2016, 2017, Dutour *et al.* 2019b). Discussion of whether this reduced response does support
350 the presence of compositional syntax in birds has been intense (see Bolhuis *et al.* 2018a,
351 2018b, Suzuki *et al.* 2018). Indeed, active use of compositional syntax by birds or simpler
352 mechanisms such as the masking hypothesis (D notes put in the first place acoustically hiding
353 the FME notes, Dutour *et al.* 2019b, Dutour *et al.* 2020) are still to be tested. We advocate for
354 the monitoring of other fine behavioural cues such as wing flicking, tail flit or calling in order
355 to provide a more reliable overview of the bird's internal state, (Cully & Ligon 1976, Carlson
356 *et al.* 2017, Salis *et al.* 2020). Other lines of inquiry, for example in neurobiology, may
357 complete field data and allow a broader view of birds' abilities (Marler 2004). Experiments on
358 Black-capped Chickadees *Poecile atricapillus* brains have shown, for example, that the neural
359 response (i.e., protein expression important for processing and categorizing conspecific
360 vocalizations) increased with the level of threat conveyed in the calls (Avey *et al.* 2011).
361 Consequently, going one step further by studying neural responses to different syntactic forms
362 and thus, the neural mechanisms underlying the production and perception of call
363 combinations could be investigated (Suzuki *et al.* 2019a).

364

365 In conclusion, context can affect the behavioural cues used to assess syntax
366 understanding by Great Tits, and thus affect biological conclusions about syntax use in this
367 species. Thorough work on the impact of context on animal behaviour could turn out to be of
368 critical importance and directly impact future studies investigating high cognitive processing
369 in non-human animals.

370

371

372

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380

381

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536 **SUPPORTING INFORMATION**

537 **Figure 1.** Spectrograms of call treatment played to Great Tits. (a) FME-D calls and (b) D-
538 FME calls, which are a reversed combination of FME and D calls. These calls were digitally
539 edited using AvisoftSASLab Pro software.

540 **Table 1.** Overview of the playback stimuli used for the study. XC is the acronym for xeno-
541 canto.

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543 TABLE

544 **Table 1.** Details of the pairwise comparisons (a, c) and model output (b). (a) Refers to the
 545 comparisons with the control (BN), (b) to the model in which control tests were discarded,
 546 and (c) to the comparison between natural and reversed calls in the two seasons studied. For
 547 scanning behaviour, the ratio (effect size) indicates the strength of the difference between the
 548 first and the second term (e.g., Great Tits scanned on average 1.44 times more toward
 549 background noise in spring than in winter). The same reasoning applies for the odds ratio
 550 (OR) for the approach behaviour. A GLMM with a Poisson error distribution was used for the
 551 number of scans (~180° horizontal head turns) during 1min. A GLMM with a binomial error
 552 distribution was used for approach behaviour. The pairwise comparisons were extracted from
 553 the model and *P*-values were corrected for multiple comparisons (Bonferroni correction: $\alpha =$
 554 0.01 for (a), $\alpha = 0.05$ for (b), and $\alpha = 0.025$ for (c) due to correction for multiple
 555 comparisons). se, standard error; *z*, *z* statistic; *P*, *p*-value (bold= statistically significant); χ^2 ,
 556 Chi-squared test; df, degree of freedom; 95%CI, 95% confidence intervals.

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(a) Differences with control BN	SCAN				APPROACH			
	Ratio	se	<i>z</i>	<i>P</i>	OR	95%CI	<i>z</i>	<i>P</i>
BN Winter- BN Spring	1.44	0.24	2.2	0.03	3.15	[0.12; 82.17]	0.69	0.49
BN Winter- Natural Winter	3.13	0.53	6.8	< 0.001	73.8	[3.89; 1401.64]	2.86	0.004
BN Winter- Reversed Winter	2.41	0.41	5.11	< 0.001	14.54	[0.75; 283.38]	1.77	0.08
BN Spring - Natural Spring	2.34	0.36	5.46	< 0.001	19	[2.12; 170.39]	2.63	0.008
BN Spring- Reversed Spring	1.02	0.17	0.13	0.89	10.23	[1.12; 93.35]	2.06	0.04
(b) Model discarding BN tests	χ^2	df	<i>P</i>		χ^2	df	<i>P</i>	
Season (Spring or Winter)	3.52	1	0.06		0.05	1	0.82	
Treatment (Natural or Reversed)	18.26	1	< 0.001		5.64	1	0.02	
Season* Treatment	11.49	1	< 0.001		1.33	1	0.25	
(c) Difference Natural-Reversed calls	Ratio	se	<i>z</i>	<i>P</i>	OR	95%CI	<i>z</i>	<i>P</i>
Natural Winter- Reversed Winter	1.3	0.18	1.92	0.05	5.57	[1.42; 21.86]	2.46	0.01
Natural Spring- Reversed Spring	2.29	0.35	5.45	< 0.001	1.86	[0.52; 6.61]	0.96	0.33

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559 **FIGURES**

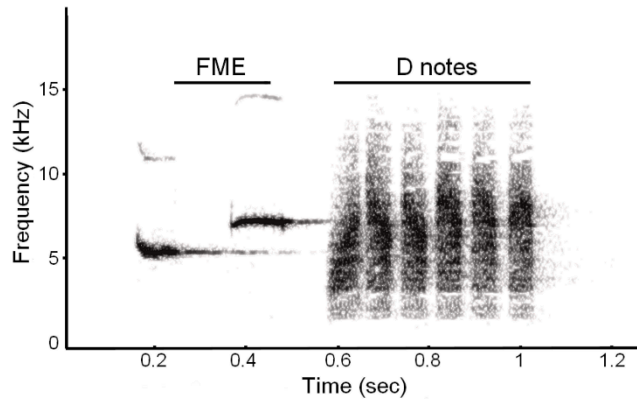
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566 **Figure 1.** Sonogram of a Great Tit mobbing call, comprising two distinct calls: (i) alert call
567 (composed of Frequency Modulated Elements, FME) and (ii) recruitment call (a string of
568 notes with broadband frequencies, D notes). This mobbing call was produced with Avisoft-
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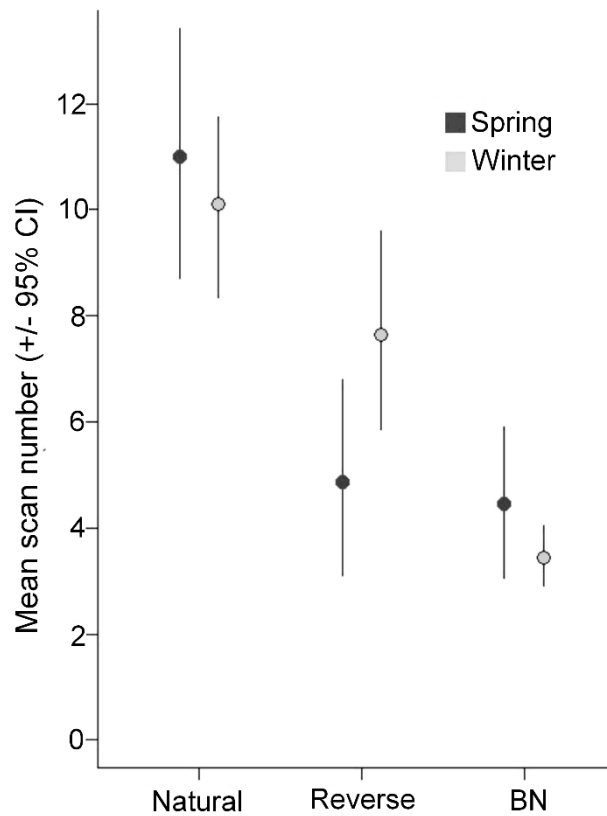


Figure 2. Mean number of scans (horizontal head movement of $\sim 180^\circ$) produced in 1min by Great Tits as a function of the call sequence used and the Season ($\pm 95\%$ boot-strapped Confidence Intervals). Great Tit mobbing calls in their natural order (FME-D) are compared to reversed calls (D-FME) and to Background Noise (BN). Statistical inference was made using confidence intervals: difference is statistically significant when less than half of the CI overlap (Cumming & Finch 2005, Cumming 2007).

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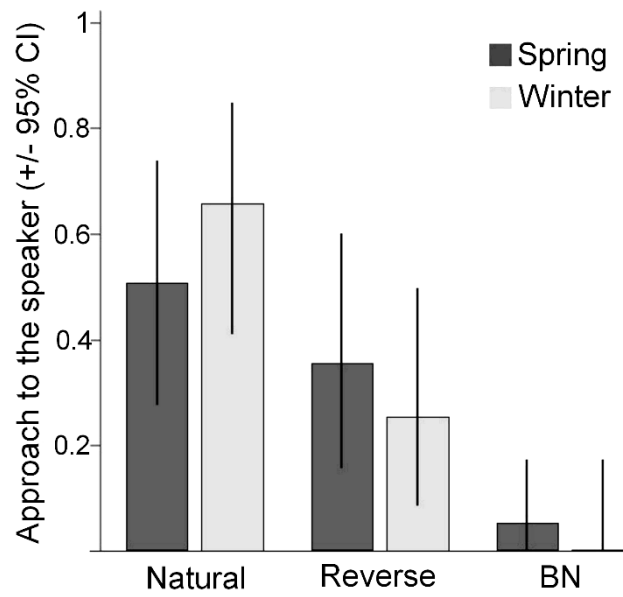
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Figure 3. Percentage of individuals (\pm 95% CI) approaching the speaker as a function of the call sequence and the season. Great Tit mobbing calls in their natural order (FME-D) are compared to reversed calls (D-FME) and to Background Noise (BN). Statistical inference can be made using the confidence intervals: the difference is statistically significant when less than half of the CI overlap (Cumming & Finch 2005, Cumming 2007).