

1 **Effects of ambient temperatures on evolutionary potential of reproductive timing in boreal**
2 **passerines**

3

4 Emma Vatka^{1*}, Markku Orell², Seppo Rytönen², Juha Merilä¹

5

6 ¹ Ecological Genetics Research Unit, Organismal and Evolutionary Biology Research Programme,

7 Faculty Biological & Environmental Sciences, University of Helsinki, P. O. Box 65, FI-00014

8 University of Helsinki, Finland

9 ² Ecology and Genetics Research Unit, University of Oulu, P.O. Box 3000, FI-90014 University of

10 Oulu, Finland

11

12 *Corresponding author: Emma Vatka, e-mail: emma.vatka@helsinki.fi, ORCID ID: orcid.org/0000-

13 0003-2935-8295

14

15 **Abstract**

- 16 1. Many populations need to adapt to changing environmental conditions, such as warming
17 climate. Changing conditions generate directional selection for traits critical for fitness. For
18 evolutionary responses to occur, these traits need to be heritable. However, changes in
19 environmental conditions can alter the amount of heritable variation a population expresses,
20 making predictions about expected responses difficult.
- 21 2. The aim of this study was to evaluate the effects of ambient temperatures on evolutionary
22 potential and strength of natural selection on the timing of reproduction in two passerine
23 birds breeding in boreal forests.
- 24 3. Long-term data on individually marked Willow Tits (*Poecile montanus*, 1975–2018) and
25 Great Tits (*Parus major*, 1969–2018) were analysed with random regression animal models
26 to assess if spring temperatures affect the expressed amount of additive genetic variation
27 (V_A) and heritability (h^2) in the timing of breeding. We assessed if ambient temperatures of
28 different seasons influenced the direction and strength of selection on breeding time. We
29 also evaluated if the strength of selection co-varied with evolutionary potential.
- 30 4. Levels of V_A or h^2 expressed in laying date were unaffected by spring temperatures in both
31 study species. Selection for earlier breeding was found in the Willow Tit, but not in the
32 Great Tit. In the Willow Tit, selection for earlier breeding was more intense when the
33 temperatures of following autumns and winters were low. Different measures of
34 evolutionary potential did not co-vary strongly with the strength of selection in either
35 species.
- 36 5. We conclude that there is no or little evidence that climate warming would either constrain
37 or promote evolutionary potential in timing of breeding through changes in amount of
38 genetic variance expressed in boreal Willow and Great Tits. However, selection on the
39 timing of breeding, a life-history event taking place in springtime, is regulated by

40 temperatures of autumns and winters. Rapid warming of these periods have thus potential to
41 reduce the rate of expected evolutionary response in reproductive timing.

42

43

44 Keywords: breeding time, climate change, heritability, natural selection, quantitative genetics

45

46

47 **Introduction**

48 Many populations are facing a need to adapt to changing environmental conditions, such as to those
49 brought along by the ongoing anthropogenically driven climate change. An evolutionary response is
50 possible, if a trait is subject to directional natural selection, and there is heritable variation in the
51 trait in question (Lynch & Walsh, 1998). However, changes in environmental conditions not only
52 influence the strength of selection, but they can also affect traits' evolutionary potential through
53 changing the amount of heritable variation a population expresses (e.g. Wilson et al., 2006).

54 The amount of genetic variation a population expresses in given trait is not constant, but it may vary
55 according to environmental conditions that prevail (Hoffmann & Merilä, 1999). Phenotypic
56 plasticity refers to genotype's ability to produce different phenotypes in different environmental
57 conditions (Pigliucci, 2001), and inter-individual variation in the amount of phenotypic plasticity
58 can make population to express different amounts of genetic variation in different environmental
59 conditions. Unfavourable conditions may either decrease or increase the amount of genetic variation
60 a population expresses (Hoffmann & Merilä, 1999). For example, poor nutrition often results in
61 lowered heritability of body size in birds (Gebhardt-Henrich & van Noordwijk, 1991; Hoffmann &
62 Merilä, 1999, Merilä & Sheldon, 2000). It is also possible that harsh conditions lead to increased
63 heritability, as often has been observed in insect studies (Hoffmann & Merilä, 1999).

64 The narrow-sense heritability (h^2) of a trait is defined as the proportion of the phenotypic variance
65 (V_P) ascribable to additive genetic variance (V_A) ($h^2 = V_A/V_P$). It indicates the extent to which a
66 trait can be expected to evolve in response to directional selection. However, variation in
67 heritability can be induced either by changes in additive genetic or residual variance (e.g. Merilä &
68 Sheldon, 1999, Wheelwright et al., 2014). Thus, heritability may not be a practical measure to
69 compare evolutionary potential between traits or between populations. Hansen, Pélabon, and Houle
70 (2011) suggest that heritability is not a suitable stand-alone measure of evolutionary potential in the

71 wild, especially when comparing populations or species. However, comparisons of heritabilities of
72 the same trait in different environmental conditions are valid.

73 Changes in the amount of expressed heritable variation may or may not be coupled with the same
74 environmental factors that determine the strength of natural selection. Heritability values can
75 correlate with the strength of natural selection (Merilä, 1997, Wilson et al., 2006), but this is not a
76 universal phenomenon (Ramakers, Culina, Visser, & Gienapp, 2018a). Climate warming has
77 affected phenologies of different organisms in both terrestrial and aquatic environments (e.g.
78 Walther et al., 2002, Parmesan & Yohe, 2003, Root et al., 2003). Different rates of phenological
79 shifts among interacting species can lead to temporal mismatches between e.g. a predator and its
80 prey (Visser & Both, 2005), which can give rise to novel selection pressures. In the temperate zone
81 Dutch Great Tit (*Parus major*) population, availability of caterpillar food during the nestling period
82 that affects natural selection on breeding time is linked with spring temperature (Ramakers,
83 Gienapp, & Visser, 2018b). However, in the boreal zone, the strength of natural selection may be
84 more strongly affected by winter than spring temperatures. For instance, unfavourable spring
85 conditions do not necessarily result in high mortality rates in the following winter if winter
86 conditions are favourable. In fact, winter temperatures in Northern Europe are rising more rapidly
87 than temperatures during other seasons (Ruosteenoja, Jylhä, & Kämäräinen, 2016).

88 The aim of this study was to evaluate the effects of ambient temperatures on evolutionary potential
89 and strength of natural selection on the timing of breeding in two passerine birds breeding in boreal
90 zone forests. We analysed long-term data on individually marked and pedigreed Willow Tit
91 (*Poecile montanus*, 1975–2018) and Great Tit (1969–2018) populations to address the following
92 two questions: First, do spring temperatures affect the expressed amount of additive genetic
93 variation (V_A) and heritability (h^2) in breeding time in two species? Second, how do ambient
94 temperatures of different seasons affect the direction and strength of selection on breeding time, and

- 95 does the strength of selection co-vary with levels of expressed additive genetic variance and
- 96 heritability?

97 **Material and Methods**

98

99 Study species

100 Willow Tits and Great Tits are small hole-nesting passerine birds. The Willow Tit is a boreal forest
101 specialist with a declining population size (Hyvärinen, Juslén, Kemppainen, Uddström, & Liukko,
102 2019), whereas the Great Tit has relatively recently expanded its range to the boreal zone and the
103 population size is growing (Valkama, Vepsäläinen, & Lehikoinen, 2011). Willow Tits breed in
104 cavities they have excavated in decaying stumps, whereas Great Tits accept nest boxes. The timing
105 of breeding is affected by spring temperatures both in the Willow Tit (Vatka, Orell, & Rytönen,
106 2011) and in the Great Tit (Vatka, Rytönen, & Orell, 2014) – both populations show advancing
107 long-term trends in reproductive timing. Annual median egg-laying dates of Willow Tits are
108 strongly correlated with the mean ambient temperature of the period March 27–May 6 ($R^2 = 0.717$;
109 Vatka et al., 2011). For the Great Tit, the corresponding period is 29 March–15 May ($R^2 = 0.721$;
110 Vatka et al., 2014). Spring temperatures also determine the timing of the main food source for
111 nestling provisioning: caterpillars of moths and sawflies that forage in tree canopies (Vatka et al.,
112 2011, 2014). Warming of springs has not yet led to a temporal mismatch between the caterpillar
113 availability and nestlings' food demands in the study populations – in fact, the match with the food
114 peak has improved in the Willow Tit (Vatka et al., 2011, 2014).

115

116 Willow Tits spend their winters in territorial flocks whose social hierarchy is influenced by prior
117 residency (Koivula, Lahti, Orell, & Rytönen, 1993). Willow Tits hoard food for winter (Brodin,
118 Lahti, Lens & Suhonen, 1996) and young from early broods have more time to prepare for the
119 upcoming winter. Because of these, Willow Tit young from relatively early broods have higher
120 recruitment rates than young from late broods, whereas for the Great Tit, recruitment rates are better
121 explained by synchrony with the caterpillar food availability (Pakanen, Orell, Vatka, Rytönen, &

122 Broggi, 2016). Winter survival of boreal Great Tits depends primarily on food supplied by humans
123 (Orell, 1989).

124

125 Study area

126 The Oulu study area in Northern Finland (ca. 65°05'N, 25°33'E) consists of coniferous, deciduous
127 and mixed forests and swamps of varying ages (Orell & Ojanen, 1983a, b; Rytönen & Orell,
128 2001). Similar forest habitats continue outside the study area, and thus the study populations are
129 open. The Willow Tit study area has expanded in size, starting from ca. 2 km² in 1975 (Orell &
130 Ojanen, 1983c) and gradually increasing to ca. 25 km² in 1996. From 1969 onwards, 100–400
131 wooden nest boxes were provided for Great Tits in separate sub-areas of 2–4 km² total in 1969–
132 1997, and since 1998, in four neighbouring sub-areas in total of 8 km² (Orell & Ojanen, 1983a, b;
133 Rytönen & Orell, 2001; Karvonen, Orell, Rytönen, Broggi, & Belda, 2012, Vatka et al., 2014,
134 Pakanen et al., 2016).

135

136 Data collection

137 Long-term monitoring of nesting attempts of Willow Tits (1975–2018) and Great Tits (1969–2018)
138 followed routine procedures (Orell & Ojanen, 1983a, Orell & Koivula, 1988, Orell, Lahti, Koivula,
139 Rytönen, & Welling, 1999). Nests were visited at least weekly and their contents were recorded.
140 The Willow Tit nests were checked through the entrance hole with the aid of a small mirror and a
141 torch, or through a small peak hole cut at the level of the nest, covered afterwards with birch bark.
142 The laying date of the first egg was as a rule calculated from the observed number of eggs in an
143 incomplete clutch (i.e., incubation had not yet started) under the assumption that one egg is laid per
144 day. The data consisted of 3331 and 3903 laying date records of first broods for the Willow Tit and
145 the Great Tit, respectively.

146 Both the parents and young were marked with individually coded aluminium leg rings and parents
147 with unique combinations of plastic colour rings, enabling pedigree construction and identification
148 of individuals that recruited to the study populations. Birds were ringed under a license provided by
149 the Finnish Natural History Museum, University of Helsinki. Parental birds were aged as yearlings
150 or older either by calculating from the ringing date of recruits, or based on the tail feather shape in
151 Willow Tits (Laaksonen & Lehtikoinen, 1976) or on plumage coloration in Great Tits (Svensson,
152 1992).

153 Pruned Willow Tit pedigree consisted of 1950 individuals and reached up to eight generations in
154 depth. It contained the following pair-wise relatedness categories: 0.025 (N=554), 0.05 (460), 0.075
155 (3), 0.1 (1), 0.125 (638), 0.15 (4), 0.175 (4), 0.25 (793), 0.275 (2), 0.3 (5), 0.375 (1), 0.5 (907),
156 0.525 (2), 0.55 (1) and 0.625 (2). The relatedness category 0.5 signifies parent-offspring and full sib
157 pairs, 0.25 grandparent-grandchild and half sib pairs, and 0.125 first cousins *et cetera*. Pruned Great
158 Tit pedigree contained 3187 individuals, with a pedigree depth up to nine generations, with the
159 following pair-wise relatedness categories: 0.025 (N=85), 0.05 (103), 0.125 (196), 0.175 (4), 0.25
160 (303), 0.275 (5), 0.375 (4), 0.5 (589), 0.525 (5), 0.55 (2), 0.625 (1) and 0.75 (2).

161 Data of daily mean ambient temperatures for 1969–2018 were retrieved from the Finnish
162 Meteorological Institute, Oulunsalo observatory that is situated ca. 20 km south from the study area.

163

164 Quantitative genetic analyses

165 Random regression animal models were used to estimate how additive genetic variances and
166 heritabilities were related to spring temperatures. The function ‘MCMCglmm’ (library
167 ‘MCMCglmm’; Hadfield, 2010) was used to fit models in R 3.5.0 (R Core Team, 2018). Laying
168 date (y) of the individual i in year j was modelled as:

169 $y_{ij} = \alpha_y + \beta_1 age_{ij} + \beta_2 T_j + year_j + a_i + b_i T_j + A_i + B_i T_j + e_{y,ijl},$ (1)

170 where α_y is the intercept and β_1 and β_2 are regression coefficients for fixed effects female's age
 171 (age_{ij}) and spring temperature (T_j), respectively. Spring temperature was the mean temperature of
 172 periods March 27–May 6 or 29 March–15 May for the Willow and the Great Tits, respectively. For
 173 the main analysis, temperatures were not individually centred, but for comparison results from
 174 analysis with individually centred temperatures are given in Appendix 1. Year ($year_j$) was used as
 175 a block random factor with estimated variance of $year_j \sim N(0, \sigma_{year}^2)$. a_i and b_i are female specific
 176 random intercepts and slopes of permanent environmental effect, and A_i and B_i are random
 177 intercepts and slopes of the additive genetic component. Permanent environmental and additive
 178 genetic variances were estimated using two 2×2 variance-covariance matrices:

179 $P = \begin{bmatrix} \sigma_a^2 & \sigma_{a,b} \\ \sigma_{a,b} & \sigma_b^2 \end{bmatrix}$ (2)

180 $G = \begin{bmatrix} \sigma_A^2 & \sigma_{A,B} \\ \sigma_{A,B} & \sigma_B^2 \end{bmatrix}$ (3)

181 $e_{y,ijl}$ (eqn. 1) is the residual term. Possible heteroscedasticity of residual variance across spring
 182 temperatures was considered by estimating the residual variance for each equal-interval group l of
 183 spring temperatures as $e_{y,ijl} \sim N(0, \sigma_{e,l}^2)$ (Ramakers et al. 2018a). The number of groups n was
 184 decided upon from four alternatives ($n = 4, 6, 8$ or 10) based on model comparison using DIC
 185 values (Ramakers, Visser, & Gienapp, 2020). For both species, $n = 10$ was selected.

186 We used a wide normal distribution as a prior for fixed factors as a default. For the residual
 187 variance, we used inverse-Wishart prior with $V = \text{diag}(n)$ and $\text{nu} = 0.002$. For other variance
 188 components, parameter-expanded priors ($V = \text{diag}(x)$, $\text{nu} = x$, $\text{alpha.mu} = 0$, $\text{alpha.V} =$
 189 $\text{diag}(x) * 1000$) were used. A total of 10 100 000 MCMC iterations were run for each species,

190 including a burn-in period of 100 000 iterations. The remaining 10 000 000 iterations were sampled
191 with a thinning interval of 10 000, leading to sample sizes of 1000 saved iterations.

192 Additive genetic variances (V_A) and heritabilities (h^2) were estimated for each documented spring
193 temperature value for both species. The method described in the Appendix of Hadfield, Wilson,
194 Garant, Sheldon, and Kruuk (2010) was applied to create confidence intervals for these estimates.
195 This involved calculation of the above-mentioned estimates for each of the saved 1000 iterations to
196 create distributions of estimated values, of which median values are reported along with 95% HPD
197 intervals using function ‘HPDinterval’ in library ‘coda’ (Plummer, Best, Cowles, & Vines 2006).
198 V_A for each spring temperature value T_j were derived using the **G** matrix as

$$199 \quad V_{A_j} = \sigma_A^2 + 2\sigma_{A,B}T_j + \sigma_B^2T_j^2 \quad (4)$$

200 and V_{PE} similarly using the **P** matrix. Temperature-dependent heritability was calculated as

$$201 \quad h_j^2 = \frac{V_{A_j}}{V_{A_j} + V_{PE_j} + \sigma_{year}^2 + \sigma_{e,l}^2}, \quad (5)$$

202 where $\sigma_{e,l}^2$ is the error variance component in the corresponding temperature group l . Temperature-
203 dependent V_A and h^2 values were plotted against spring temperatures.

204

205 Selection on the breeding time

206 We studied which temperature periods (*viz.* spring, autumn and winter) affect selection on breeding
207 time. These periods were selected *a priori* to present different potential mechanisms of selection.
208 Spring temperatures (the mean temperature of periods March 27–May 6 or 29 March–15 May for
209 the Willow and the Great Tit, respectively) may affect the temporal match-mismatch with food
210 availability during the nestling period (Vatka et al., 2011), which in turn may affect breeding
211 success and thus selection on the timing of breeding. Temperatures of the following autumn (the
212 mean temperature of August–October) coincide with the period of intensive food hoarding in the

213 Willow Tit (Haftorn, 1956), and winter temperatures (the mean temperature of December–
214 February) represent the coldest time of the year. These may affect survival, and thus, recruitment
215 rates of the young.

216 We used generalized linear mixed effect models fit with function ‘glmer’ in library ‘lme4’ (Bates,
217 Mächler, Bolker, & Walker, 2015) with Poisson error structure. In these models, the response
218 variable was **annual proxy of individual fitness**, measured as the number of recruits a female
219 produced annually, including recruits from the first brood and a potential re-nesting attempt or
220 second brood. The number of recruits (rather than the number of fledglings or eggs produced) was
221 used as a proxy of fitness, because the definition of fitness refers to the relative contribution of a
222 phenotype to the future generations (e.g. Krebs, 2009) and thus, only offspring that make it to
223 express the trait (the timing of breeding) count. However, for comparison we present an analysis
224 with the number of fledglings as a proxy of individual fitness in Appendix 1. **Annual proxy of**
225 **individual fitness** was regressed against the annually centred timing of breeding of the female’s first
226 brood in interaction with temperature (centred temperatures of the three periods were tested one at a
227 time). A significant interaction term would tell that ambient temperatures affect selection on the
228 timing of breeding. Female identity and year were used as block random factors.

229 These analyses were restricted to years 1991–2017 and 1999–2017 for the Willow and the Great
230 Tit, respectively. These restrictions were applied because the Willow Tit study area was small and
231 fragmented before 1991 (Lampila, Orell, Belda, & Koivula, 2006), and the Great Tit study area
232 reached its current extent in 1998. Thus, early years were omitted in order to acquire selection
233 gradients that are comparable between years. In 1998, a large number of Great Tit nests were
234 experimentally destroyed during the incubation stage, and therefore this year was also excluded
235 from the analysis.

236

237 Covariance of measures of evolutionary potential and selection

238 To estimate annual selection gradients, we used models with Poisson error structure fit by
239 ‘MCMCglmm’. Annual proxy of individual fitness (measured as the number of recruits a female
240 produced annually) was regressed against the standardized timing of breeding (centred to a mean of
241 zero and scaled to a variance of one) in annual subsets of data. Standardisation was done before
242 creating annual subsets. Regression coefficients β_j were interpreted as directional selection
243 gradients (Morrissey & Goudie, 2016).

244 We examined covariance of different measures of evolutionary potential (i.e., expressed additive
245 genetic variation and heritability) and selection. As explained above, V_{A_j} and h_j^2 were calculated
246 and β_j were retrieved for each of the 1000 saved iterations. We calculated Pearson’s correlation
247 coefficients r between each k^{th} set of values of V_{A_j} or h_j^2 and β_j , reporting their mean and HPD
248 intervals. In similar fashion, we calculated Pearson’s correlation coefficients r between measures of
249 evolutionary potential and $|\beta_j|$ to infer the covariance between evolutionary potential and the
250 strength of selection (regardless of its direction).

251

252 **Results**

253

254 Effects of spring temperatures on additive genetic variance and heritability

255 The amount of additive genetic variance (V_A) expressed in laying date tended to be higher for Great
256 Tits than for Willow Tits (Fig. 1a, b), but in both species V_A was independent of spring temperatures
257 (Fig. 1a, b). In fact, the variance attributable to random slopes of the additive genetic component σ_B^2
258 was low in both species (Table S1, Table S2). For results with analysis using individually centred
259 temperatures, please see Tables S3 and S4.

260 Heritability estimates (h^2) of laying date were somewhat low both in the Willow Tit (h^2 range =
261 0.132–0.232; Table S5) and the Great Tit (h^2 range = 0.228–0.411; Table S6), and independent of
262 spring temperatures (Fig. 1c, d). There was some variability in heritability estimates especially in
263 the Great Tit (Fig. 1d), owing to variation in estimated error variances between different
264 temperature groups (Table S2).

265

266 Effects of ambient temperatures on the strength of selection

267 Early broods produced more recruits than late broods and thus, there was a significant selection
268 pressure for earlier breeding in the Willow Tit (Table 1). However, for the Great Tit, the main effect
269 of the timing of breeding on recruitment rate was nonsignificant and thus no significant overall
270 selection for earlier breeding was found (Table 2). The strength of directional selection on the
271 timing of breeding was independent of spring temperatures in both species (Table 1, Table 2). In the
272 Willow Tit, selection for earlier breeding was more intense when the temperatures of the following
273 autumn or winter were cold, indicated by significant interaction terms Timing*Temperature (Table
274 1). Winter temperatures had also a significantly negative main effect, meaning that recruitment rates

275 are lower in warm winters (Table 1). In the Great Tit, autumn or winter temperatures did not affect
276 the strength of directional selection on reproductive timing (Table 2).

277

278 Covariance of measures of evolutionary potential and selection

279 There was year-to-year variation in the selection gradients β on breeding time, ranging from -1.208
280 to 0.246 in the Willow Tit ($\bar{\beta} = -0.408$) and from -0.634 to 1.497 in the Great Tit ($\bar{\beta} = 0.006$; Table
281 S7). Neither the levels of expressed additive genetic variance nor heritability were correlated with
282 annual selection gradients, or with the strength of selection in either species (Table 3).

283 **Discussion**

284 We did not detect any significant changes in the expression of additive genetic variance or
285 heritability with warming spring temperatures. As in the case of our results, V_A in timing of
286 breeding did not change with warming springs in a UK Great Tit population (Husby et al., 2010).
287 Similarly, Ramakers, Gienapp, and Visser (2018) found little genotype-by-environment interaction
288 in the timing of breeding related to spring temperatures in a Dutch Great Tit population. However,
289 inconsistent effects of environmental conditions on expression of additive genetic variance have
290 been reported in other traits (Hoffmann & Merilä, 1999).

291 Heritability values of breeding time were rather low in our study populations, which is typical of
292 avian life-history traits (e.g. Merilä & Sheldon, 2000, McCleery et al., 2004). The observed
293 variation in heritability estimates was mostly due to variation in residual variance estimates between
294 different temperature groups, rather than due to variation in the additive genetic variance
295 component. Studies from other Great Tit populations have reported heritability values on breeding
296 time that are somewhat lower than the values we report here (h^2 range: 0.228–0.411; an overall
297 average = 0.315, Table S4). McCleery et al. (2004) reported a heritability of 0.159 (SE=0.059) for
298 Wytham Woods population, whereas Gienapp, Calus, Laine, and Visser (2019) estimated
299 heritability of 0.24 (SE=0.07) based on a social pedigree and 0.17 (SE=0.06) based on kinships
300 inferred from genetic markers for Hoge Veluwe population. To our knowledge, no other heritability
301 estimates of the timing of breeding are available for the Willow Tit. That the additive genetic
302 variance and heritability were lower in the Willow than in the Great Tit makes sense in the light that
303 the timing of breeding in Willow Tits seem to be a subject for directional selection more often than
304 in Great Tits, and because directional selection is expected to erode genetic variation. Whatever the
305 ultimate reason for the low heritability, low heritabilities translate to low rates of expected
306 evolutionary change for given intensity of selection (Falconer & Mackay, 1996). As the
307 heritabilities in both species did not show any trend in relation to spring temperatures, warming of

308 springs are not expected to neither accelerate nor slow down evolutionary responses as far as the
309 effect of heritability values is concerned.

310 As noted also in earlier studies (e.g. Sheldon, Kruuk, & Merilä, 2003, Husby, Visser, & Kruuk,
311 2011, Visser et al., 2015), the strength of directional selection on breeding time was quite variable
312 among years. However, in spite of the fact that boreal Willow and Great Tits show advancing long-
313 term trends in their timing of breeding (Vatka et al., 2011, 2014), we did not detect statistically
314 significant directional selection for earlier breeding in the Oulu Great Tit population. This indicates
315 that the observed change in the timing of breeding in the Great Tit results most likely from
316 phenotypic plasticity, and hence, is not a genetically based evolutionary response (cf. Radchuk et
317 al., 2019). In contrast, we found significant selection for earlier breeding in the Willow Tit, which
318 in combination with low but significant heritability of breeding time should promote evolutionary
319 response towards earlier breeding. However, to what degree the advanced breeding time in the
320 Willow Tit reflects genetic *vs.* plastic changes remains to be investigated.

321 Spring temperatures do not seem to influence selection on the timing of breeding in either of the
322 two study species. Yet, spring temperatures affect the synchrony between the breeding time and the
323 timing of caterpillar food availability in the Willow Tit: synchrony is better in warm years (Vatka et
324 al., 2011). In contrast, Great Tits advance their breeding time at the same rate as the timing of the
325 caterpillar food peak advances with rising spring temperatures, and thus, spring temperatures do not
326 affect the level of synchrony in the Oulu Great Tit population (Vatka et al., 2014). Hence, one
327 would expect that spring temperatures would influence selection on the timing of breeding in the
328 Willow Tit. However, a previous study suggests that the synchrony with the caterpillar food peak is
329 not the most relevant selection mechanism in the Willow Tit – instead, timing of breeding in
330 relation to conspecifics affected recruitment rate (Pakanen et al., 2016). Visser et al. (2015) also
331 found a similar lack of association between the level of synchrony with the food availability and
332 selection on the timing of breeding in migratory Pied Flycatchers (*Ficedula hypoleuca*).

333 Autumn and winter temperatures were found to affect the strength of selection on the timing of
334 breeding in the Willow Tit. This is an understandable outcome when considering the species'
335 ecology. Willow Tits spend their winters in flocks whose social hierarchy is affected by prior
336 residency (Koivula et al., 1993). They hoard food in autumn (Brodin et al., 1996) and young from
337 early broods have more time to prepare for the winter. Thus, young from early broods are in a better
338 position to survive than young from late broods. This seems to be particularly so when autumns and
339 winters are cold. Autumn temperatures likely affect food hoarding, inducing a selection pressure on
340 breeding time. Cold winters are associated with higher overall recruitment rate, but selection for
341 earlier breeding is more intense when winters are cold. Rapid warming of autumns and winters
342 appear to reduce the strength of directional selection for earlier breeding. This in turn can reduce the
343 rate of expected evolutionary change, unless some other important ultimate factor(s) comes into
344 play.

345 That warm winters are associated with lower recruit production in the Willow Tit can be explained
346 by several mechanisms, yet they are so far only speculative explanations. Temperatures fluctuating
347 above and below the freezing point can first melt the snow and then create an ice shield covering
348 the food hoards. Warm winters can cause food hoards to decay (Sechley, Strickland, & Norris,
349 2015), and thus food availability can decrease drastically. It is also possible that wet weathers (rain
350 coming down as water instead of snow) affect the thermoregulation of birds when plumage gets
351 wet. Winter rain can decrease foraging efficiency during short days, followed by cold and long
352 nights. These mechanisms would likely affect all birds similarly, regardless of their social status or
353 birth date.

354 In order to understand how environmental changes affect natural selection, one needs to identify the
355 life stage when selection kicks in. In the boreal zone, (pre-)winter conditions seem to play an
356 important role for the selection on breeding time. The situation may be different in temperate zone
357 populations where winters are not equally harsh. Visser et al. (2015) found that in a long-distance

358 migratory bird, temperatures at the time of arrival to the breeding grounds affected selection on
359 timing of reproduction. They suggested that environmental conditions with a lag of up to two years
360 can affect selection on a phenological trait (Visser et al., 2015). We also found that environmental
361 conditions affect selection on timing of breeding with a temporal lag, although in our case the lag
362 was not more than about six months. It is important to recognize that evolution of the timing of
363 breeding, a life history event that takes place in spring, can be regulated by environmental
364 conditions outside this period. In other words, one needs to consider the whole lifespan of
365 individuals when trying to assess factors influencing a certain life-history trait.

366 Environmental coupling of heritability and selection appears to be rare in wild populations
367 (Ramakers et al., 2018, but see Merilä, 1997, Wilson et al., 2006, Husby et al., 2011). In accordance
368 with this, we did not find any strong correlations between the different measures of evolutionary
369 potential and the selection or its strength. In this perspective, it might be worth emphasizing that it
370 may be relevant to consider separately on the one hand factors that affect the expressed amount of
371 heritable variation, and on the other hand factors that affect selection on the trait in question – these
372 might be totally distinct factors, influenced differently by changes in environmental conditions. If
373 this is a common, or a general pattern, it will be difficult to predict how populations respond to
374 selection brought by, for instance, changing climatic conditions.

375

376 Conclusions

377 The results suggest that climate warming will neither constrain nor promote evolutionary potential
378 in boreal populations of Willow and Great Tits. However, environmental conditions can affect
379 selection on breeding time with a temporal lag: evolution of a life history event that takes place in
380 springtime appears to be regulated by conditions prevailing in following autumns and winters in the
381 Willow Tit. Hence, in the light of the findings of this study, rapid warming of these periods can be

382 expected to reduce the strength of directional selection for earlier breeding, and thereby also the
383 expected evolutionary response to selection.

384 **Acknowledgements**

385 We acknowledge all the people who have participated in collecting the parid breeding data,
386 especially M Ojanen, K Koivula, K Lahti, K Kumpulainen, P Welling, M Leppäjärvi, N Verboven,
387 J Broggi, E Belda, S Lampila, J Karvonen, C Westerduin, J Ollinmäki, J Laukkala, numerous
388 graduate students and staff of the Biodiversity Unit, University of Oulu. We thank Finnish
389 Meteorological Institute for providing the data of ambient temperatures and M Morrissey, P
390 Gienapp and M Kivikoski for valuable discussions. EV was funded by the Ella and Georg
391 Ehrnrooth Foundation. Kvantum Institute, University of Oulu provided funding for data collection.
392 Authors have no conflicts of interests.

393

394 **Authors' contributions**

395 EV and JM conceived the ideas; MO, SR and EV collected the data; EV analysed the data and led
396 the writing of the manuscript. All authors contributed critically to the drafts and gave final approval
397 for publication.

398

399 **Data Availability Statement**

400 Data available from the Dryad Digital Repository XXXX.

401

402 **References**

403

404 Bates, D., Mächler, M., Bolker, B. M., & Walker, S. C. (2015). Fitting linear mixed-effect models
405 using lme4. *Journal of Statistical Software*, 67, 1–48. doi: 10.18637/jss.v067.i01

406 Brodin, A., Lahti, K., Lens, L., & Suhonen, J. (1996). A northern population of willow tits *Parus*
407 *montanus* did not store more food than southern ones. *Ornis Fennica*, 73, 114–8.

408 Falconer, D. S., & Mackay, T. F. C. (1996). *Introduction to Quantitative Genetics*. Longman Group
409 Limited, Essex.

410 Gebhardt-Henrich, S. G., & van Noordwijk, A. J. (1991). Nestling growth in the Great Tit I.
411 Heritability estimates under different environmental conditions. *Journal of Evolutionary*
412 *Biology*, 3, 341–362. doi: 10.1046/j.1420-9101.1991.4030341.x

413 Gienapp, P., Calus, M. P. L., Laine, V. N., & Visser, M. E. (2019). Genomic selection on breeding
414 time in a wild bird population. *Evolution Letters*, 3, 142–151. doi: 10.1002/evl3.103

415 Hadfield, J. (2010). MCMC methods for multi-response generalized linear mixed models: the
416 MCMCglmm R package. *Journal of Statistical Software*, 33, 1–22. doi: 10.18637/jss.v033.i02

417 Hadfield, J. D., Wilson, A. J., Garant, D., Sheldon, B. C., & Kruuk, L. E. B. (2010). The misuse of
418 BLUP in ecology and evolution. *The American Naturalist*, 175, 116–125. doi:
419 10.1086/648604

420 Haftorn, S. (1956). Contribution to the food biology of tits especially about storing of surplus food
421 Part III The Willow Tit (*Parus atricapillus* L.). Det Kgl Norske Videnskabers Selskabs
422 Skrifter 1956 Nr 3, 1-79.

423 Hansen, T. F., Pélabon, C., & Houle, D. (2011). Heritability is not evolvability. *Evolutionary*
424 *Biology*, 38, 258–277. doi: 10.1007/s11692-011-9127-6

425 Hoffmann, A. A., & Merilä, J. (1999). Heritable variation and evolution under favourable and
426 unfavourable conditions. *Trends in Ecology & Evolution*, 14, 96–101. doi: 10.1016/S0169-
427 5347(99)01595-5

428 Houle, D. (1992). Comparing evolvability and variability of quantitative traits. *Genetics*, 130, 195–
429 204.

430 Husby, A., Nussey, D. H., Visser, M. E., Wilson, A. J., Sheldon, B. C., & Kruuk, L. E. B. (2010).
431 Contrasting patterns of phenotypic plasticity in reproductive traits in two great tit (*Parus*
432 *major*) populations. *Evolution*, 64, 2221–2237. doi: 10.1111/j.1558-5646.2010.00991.x

433 Husby, A., Visser, M. E., & Kruuk, L. E. B. (2011). Speeding up microevolution: the effects of
434 increasing temperature on selection and genetic variance in a wild bird population. *PLoS*
435 *Biology*, 9, e1000585. doi: 10.1371/journal.pbio.1000585

436 Hyvärinen, E., Juslén, A., Kemppainen, E., Uddström, A. & Liukko, U.-M. (eds.) (2019). The 2019
437 Red List of Finnish Species. Ympäristöministeriö & Suomen ympäristökeskus. Helsinki. 704
438 p.

439 Karvonen, J., Orell, M., Rytönen, S., Broggi, J. & Belda, E. (2012). Population dynamics of an
440 expanding passerine at the distribution margin. *Journal of Avian Biology*, 43, 102–108. doi:
441 10.1111/j.1600-048X.2011.05376.x

442 Koivula, K., Lahti, K., Orell, M., & Rytönen, S. (1993). Prior residency as a key determinant of
443 social dominance in the willow tit (*Parus montanus*). *Behavioral Ecology and Sociobiology*,
444 33, 283–287. doi: 10.1007/BF02027126

445 Krebs, C. J. (2009). *Ecology. The Experimental Analysis of Distribution and Abundance*. 6th ed.
446 Pearson.

447 Laaksonen, M., & Lehtikoinen, E. (1976). Age determination of willow and crested tit *Parus*
448 *montanus* and *P. cristatus*. *Ornis Fennica*, 53, 9–14.

449 Lampila, S., Orell, M., Belda, E., & Koivula, K. (2006). Importance of adult survival, local
450 recruitment and immigration in a declining boreal forest passerine, the willow tit *Parus*
451 *montanus*. *Oecologia*, 148, 405–413. doi: 10.1007/s00442-006-0386-3

452 Lynch, M., & Walsh, B. (1998). *Genetics and Analysis of Quantitative Traits*. Sinauer, Sunderland,
453 MA.

454 McCleery, R. H., Pettifor, R. A., Armbruster, P., Meyer, K., Sheldon, B. C., & Perrins, C. M.
455 (2004). Components of variance underlying fitness in a natural population of the great tit
456 *Parus major*. *The American Naturalist*, 164, E62–E72. doi: 10.1086/422660

457 Morrissey, M. B., & Goudie, I. G. J. (2016). Analytical results for directional and quadratic
458 selection gradients for log-linear models of fitness functions. *bioRxiv.org preprint*. doi:
459 10.1101/040618

460 Orell, M., & Koivula, K. (1988). Cost of reproduction: parental survival and production of recruits
461 in the Willow Tit *Parus montanus*. *Oecologia*, 77, 423–432. doi: 10.1007/BF00378054.

462 Orell, M., & Ojanen, M. (1983a). Timing and length of the breeding season of the great tit *Parus*
463 *major* and the willow tit *P. montanus* near Oulu, Northern Finland. *Ardea*, 71, 183–198.

464 Orell, M., & Ojanen, M. (1983b). Effect of habitat, date of laying and density on clutch size of the
465 Great Tit *Parus major* in northern Finland. *Holarctic Ecology*, 6, 413–423.

466 Orell, M., & Ojanen, M. (1983c). Breeding biology and population dynamics of the willow tit
467 *Parus montanus*. *Annales Zoologici Fennici*, 20, 99–114.

468 Orell, M. (1989). Population fluctuations and survival of Great Tits *Parus major* dependent on food
469 supplied by man in winter. *Ibis*, 131, 112–127. doi: 10.1111/j.1474-919X.1989.tb02750.x

470 Orell, M., Lahti, K., Koivula, K., Rytkönen, S., & Welling, P. (1999). Immigration and gene flow in
471 a northern willow tit (*Parus montanus*) population. *Journal of Evolutionary Biology*, 12, 283–
472 295. doi: 10.1046/j.1420-9101.1999.00030.x

473 Pakanen, V.-M., Orell, M., Vatka, E., Rytkönen, S., & Broggi, J. (2016). Different ultimate factors
474 define timing of breeding in two related species. *PLoS ONE*, 11(9), e0162643. doi:
475 10.1371/journal.pone.0162643

476 Parmesan, C., & Yohe, G. (2003). A globally coherent fingerprint of climate change impacts across
477 natural systems. *Nature*, 421, 37–42. doi: 10.1038/nature01286

478 Pigliucci, M. (2001). *Phenotypic Plasticity: Beyond Nature and Nurture*. Baltimore, U.S.A: John
479 Hopkins University Press.

480 Plummer, M., Best, N., Cowles, K., & Vines, K. (2006) CODA: Convergence Diagnosis and Output
481 Analysis for MCMC. *R News*, 6, 7-11.

482 R Core Team (2018). *R: A language and environment for statistical computing*. R Foundation for
483 Statistical Computing, Vienna, Austria. URL <https://www.R-project.org/>.

484 Radchuk, V., Reed, T., Teplitsky, C, van de Pol, M., Charmantier, A., Hassall, C., ... Kramer-
485 Schadt, S. (2019). Adaptive responses of animals to climate change are most likely
486 insufficient. *Nature Communications*, 10, 3109. doi: 10.1038/s41467-019-10924-4

487 Ramakers, J. J. C., Culina, A., Visser, M. E., & Gienapp, P. (2018a). Environmental coupling of
488 heritability and selection is rare and of minor evolutionary significance in wild populations.
489 *Nature Ecology & Evolution*, 2, 1093–1103. doi: 10.1038/s41559-018-0577-4

490 Ramakers, J. J. C., Gienapp, P., & Visser, M. E. (2018b). Phenological mismatch drives selection
491 on elevation, but not on slope, of breeding time plasticity in a wild songbird. *Evolution*, 73,
492 175–187. doi: 10.1111/evo.13660

493 Ramakers, J. J. C., Visser, M. E., & Gienapp, P. (2020). Quantifying individual variation in reaction
494 norms: Mind the residual. *Journal of Evolutionary Biology*, 33, 352–365. doi:
495 10.1111/jeb.13571

496 Root, T. L., Price, J. T., Hall, K. R., Schneider, S. H., Rosenzweig C., & Pounds, J. A. (2003).
497 Fingerprints of global warming on wild animals and plants. *Nature*, 421, 57–60. doi:
498 10.1038/nature01333

499 Ruosteenoja, K., Jylhä, K., & Kämäräinen, M. (2016). Climate projections for Finland under the
500 RCP forcing scenarios. *Geophysica*, 51, 17–50.

501 Rytönen, S., & Orell, M. (2001). Great tits (*Parus major*) lay too many eggs: experimental
502 evidence in mid-boreal habitats. *Oikos*, 93, 439–450. doi: 10.1034/j.1600-
503 0706.2001.930309.x

504 Sechley, T. H., Strickland, D., & Norris, D. R. (2015). Linking the availability of cached food to
505 climate change: an experimental test of the hoard-rot hypothesis. *Canadian Journal of*
506 *Zoology*, 93, 411–419. doi: 10.1139/cjz-2015-0016

507 Sheldon, B. C., Kruuk, L. E. B., & Merilä, J. (2003). Natural selection and inheritance of breeding
508 time and clutch size in the collared flycatcher. *Evolution*, 57, 406–420. doi: 10.1111/j.0014-
509 3820.2003.tb00274.x

510 Svensson, L. (1992). Identification guide to European passerines, 4th edn. British Trust for
511 Ornithology.

512 Valkama, J., Vepsäläinen, V., & Lehikoinen, A. (2011). The Third Finnish Breeding Bird Atlas. –
513 Finnish Museum of Natural History and Ministry of Environment.
514 <<http://atlas3.lintuatlas.fi/english>> (cited 11.3.2019) ISBN 978-952-10-7145-4

- 515 Vatka, E., Orell, M., & Rytönen, S. (2011). Warming climate advances breeding and improves
516 synchrony of food demand and food availability in a boreal passerine. *Global Change*
517 *Biology*, 17, 3002–3009. doi: 10.1111/j.1365-2486.2011.02430.x
- 518 Vatka, E., Rytönen, S., & Orell, M. (2014). Does the temporal mismatch hypothesis match in
519 boreal populations? *Oecologia*, 176, 595–605. doi: 10.1007/s00442-014-3022-7
- 520 Visser, M. E., & Both, C. (2005). Shifts in phenology due to global climate change: the need for a
521 yardstick. *Proceedings of the Royal Society B*, 272, 2561–2569. doi: 10.1098/rspb.2005.3356
- 522 Visser, M. E., Gienapp, P., Husby, A., Morrissey, M., de la Hera, I., Pulido, F., & Both, C. (2015).
523 Effects of spring temperatures on the strength of selection on timing of reproduction in a long-
524 distance migratory bird. *PLoS Biology*, 13, e1002120. doi: 10.1371/journal.pbio.1002120
- 525 Wilson, A. J., Pemberton, J. M., Pilkington, J. G., Coltman, D. W., Mifsud, D. V., Clutton-Brock,
526 T. H., & Kruuk, L. E. B. (2006). Environmental coupling of selection and heritability limits
527 evolution. *PLoS Biology*, 4, e216. doi: 10.1371/journal.pbio.0040216

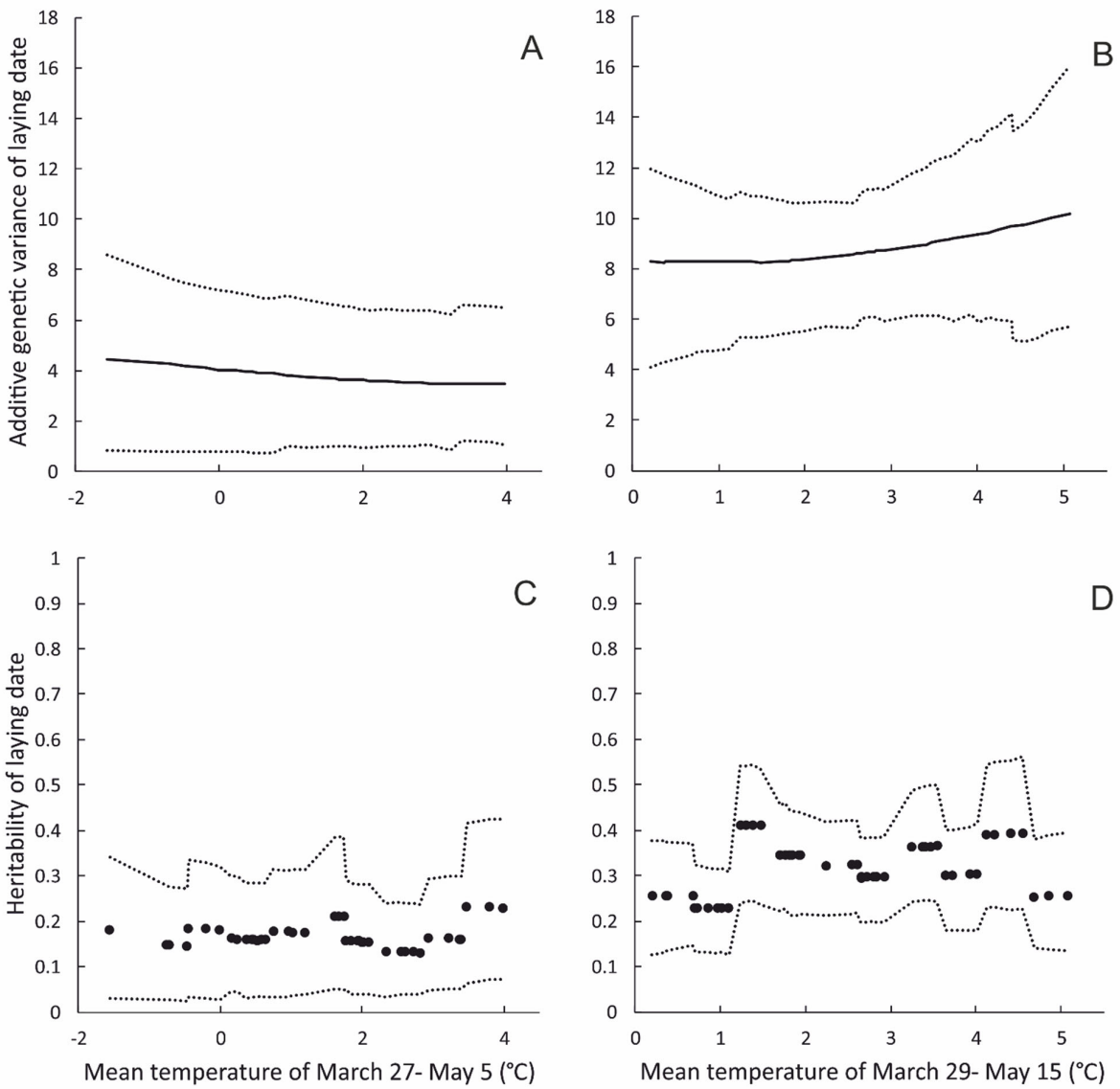
528 **Figure legends**

529

530 Figure 1. Additive genetic variances (A-B) and heritabilities (C-D) of breeding time in relation to
531 spring temperatures in the Willow Tit (A, C) and the Great Tit (B, D).

532

533 **Figures**



534

535 **Figure 1.**

536 **Tables**

537

538 Table 1. Parameters of generalized linear mixed effect models explaining annual proxy of
 539 individual fitness (measured as the number of recruits produced) in the Willow Tit. Explanatory
 540 variables are centred. A significant interaction term (Timing*Temperature) indicates that selection
 541 on the timing of breeding is influenced by ambient temperatures. Statistically significant ($p < 0.050$)
 542 parameter estimates are depicted in boldface.

Model	Fixed effects	Estimate	SE	z value	p
Spring temperature	Intercept	-1.534	0.112	-13.686	< 0.001
	Timing of breeding	-0.067	0.010	-6.510	< 0.001
	Temperature	-0.042	0.079	-0.534	0.594
	Timing*Temperature	0.003	0.007	0.470	0.638
Autumn temperature	Intercept	-1.544	0.113	-13.657	< 0.001
	Timing of breeding	-0.071	0.010	-6.861	< 0.001
	Temperature	-0.040	0.108	-0.373	0.709
	Timing*Temperature	0.026	0.012	2.159	0.031
Winter temperature	Intercept	-1.550	0.102	-15.130	< 0.001
	Timing of breeding	-0.066	0.010	-6.434	< 0.001
	Temperature	-0.086	0.036	-2.351	0.019
	Timing*Temperature	0.009	0.004	2.169	0.030

543

544

545 Table 2. Parameters of generalized linear mixed effect models explaining annual proxy of
 546 individual fitness (measured as the number of recruits produced) in the Great Tit. Explanatory
 547 variables are centred. A significant interaction term (Timing*Temperature) would indicate that
 548 selection on the timing of breeding is influenced by ambient temperatures. Statistically significant
 549 ($p < 0.050$) parameter estimates are depicted in boldface.

Model	Fixed effects	Estimate	SE	z value	p
Spring temperature	Intercept	-1.927	0.125	-15.414	< 0.001
	Timing of breeding	-0.008	0.012	-0.654	0.513
	Temperature	0.026	0.086	0.306	0.760
	Timing*Temperature	0.015	0.010	1.507	0.132
Autumn temperature	Intercept	-1.933	0.123	-15.691	< 0.001
	Timing of breeding	-0.004	0.012	-0.350	0.726
	Temperature	0.140	0.135	1.033	0.302
	Timing*Temperature	0.020	0.016	1.287	0.198
Winter temperature	Intercept	-1.947	0.122	-16.000	< 0.001
	Timing of breeding	-0.004	0.012	-0.370	0.712
	Temperature	-0.063	0.036	-1.740	0.082
	Timing*Temperature	0.007	0.005	1.410	0.159

550

551

552

553 Table 3. Pearson's correlation coefficients r (and their 95% HPD intervals) between different
 554 measures of evolutionary potential and selection in the Willow Tit and the Great Tit.

Species	Measure of evolutionary potential	Annual selection gradients		Strength of selection	
		r	95% HPD interval	r	95% HPD interval
The Willow Tit	Additive genetic variance V_A	-0.082	[-0.364, 0.233]	0.051	[-0.252, 0.377]
	Heritability h^2	-0.097	[-0.387, 0.201]	0.043	[-0.286, 0.387]
The Great Tit	Additive genetic variance V_A	0.043	[-0.384, 0.355]	-0.133	[-0.441, 0.277]
	Heritability h^2	-0.038	[-0.328, 0.237]	-0.120	[-0.427, 0.189]

555

556