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## Curse of the black spot: spotting negatively correlates with fitness in black grouse *Lyrurus tetrix*

Soulsbury Carl D. <sup>1,\*</sup>, Kervinen Matti <sup>2</sup>, Lebigre Christophe <sup>3</sup>

<sup>1</sup> Lincoln Univ, Joseph Banks Labs, Sch Life Sci, Green Lane, Lincoln LN6 7TS, England.

<sup>2</sup> Univ Jyvaskyla, Dept Biol & Environm Sci, POB 35, FI-40014 Jyvaskyla, Finland.

<sup>3</sup> Earth & Life Inst, PI Croix Sud 4, Carnoy Bldg, B-1348 Louvain La Neuve, Belgium.

\* Corresponding author : Carl D. Soulsbury, email address : [csoulsbury@lincoln.ac.uk](mailto:csoulsbury@lincoln.ac.uk)

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### Abstract :

There is growing evidence that achromatic plumage can act as honest indicators of male quality. In some species with areas of white plumage, black melanin spots can be found on parts of the feathers. The functional significance of these spots and the relationship with male quality is yet poorly understood. We investigated the relationship between black melanin spots in an otherwise totally white ornament, the undertail covert, in relation to age, fitness, and covariance with past and present expression of sexual traits, in the lekking black grouse *Lyrurus tetrix*. We found that spots at tips of feathers (tip spots) were negatively related to survival and reproductive success, and covaried negatively with current fighting rate. They also covaried positively with past fighting rate, suggesting high investment in fighting leads to carryover effects on male condition. In contrast, spots found further down the feather (vane spots) were unrelated to fitness and morphological and behavioral trait expression. Our results show that melanin spots can reflect overall male quality, but their adaptive value is dependent on their location on the feather. The exact drivers of melanin spot expression and how these link to male quality are currently unknown.

**Keywords** : achromatic, feathers, fitness, lekking, melanin, oxidative stress

## 34 INTRODUCTION

35 Achromatic plumage patterns (black, white and grey) are found widely across bird species  
36 (McGraw 2006; Stoddard and Prum 2011). There are two different types of melanin  
37 pigments, eumelanin and pheomelanin, with grey and black colorations resulting from a  
38 higher concentration of eumelanin relative to pheomelanin (Haase et al. 1992; McGraw  
39 2006). Alongside melanin-based plumage, white plumage results from the scattering of light  
40 in all directions by unpigmented feather keratin (Prum et al. 1999) and requires neither  
41 pigment nor specialized feather structure. In the past, most research has focused on the  
42 function of brightly colored ornaments, but recent research suggests that achromatic plumage  
43 can be an essential component of sexual signalling in some species (Mennill et al. 2003),  
44 especially being an important part of visual display (see Galván 2008). Moreover, recent  
45 studies have demonstrated that achromatic plumage may also act as a condition-dependent or  
46 as condition-related signals (McGlothlin et al. 2007; Gladbach et al. 2011; Roulin 2015) and  
47 in some cases relates to components of fitness (Doucet et al. 2005; Guindre-Parker et al.  
48 2012). Most work has focussed on black plumage, because it is assumed that white feathers  
49 are cheap to produce (Török et al. 2003). However, white feather patches may also be  
50 condition-dependent (Gustafsson et al. 1995; Török et al. 2003; Hanssen et al. 2006; Blanco  
51 and Fargallo 2013; though see Lehikoinen et al. 2010), and likely to have some production  
52 costs (Vágási et al. 2010). Combined, it means that either the production of black or white  
53 plumage may be condition-dependent or condition-related depending on the context.

54 Our understanding of the condition-dependence of achromatic plumage, hinges on the  
55 control or lack of control over melanin production. Melanin is synthesized endogenously in  
56 the melanocytes that occur primarily in the dermis; the melanocytes migrate into the dermal  
57 pulp of the developing feather germ, where the melanin is packaged into melanosomes which  
58 are transferred to keratinocytes for deposition into developing feathers (Yu et al. 2004;

59 Ducrest et al. 2008). Despite our knowledge of the process of melanin production, we do not  
60 understand how melanin-based pigmentation is regulated, i.e. whether it is caused by  
61 presence/absence of melanocytes, formation or activities of melanosomes, or the transfer of  
62 melanosomes to keratinocytes (Yu et al. 2004). Roulin (2015) distinguished two scenarios  
63 where melanin-based colouration ways could covary with condition: in the first, condition-  
64 related signalling, factors influencing body condition indirectly impact melanin coloration,  
65 whereas in condition-dependent signalling, coloration and condition are causally-linked.  
66 There are a number of potential opportunities for condition-dependence to influence  
67 melanogenesis and the production of depigmented areas (reviewed by Guindre-Parker and  
68 Love 2014), because melanin-based coloration have been associated with physiological and  
69 behavioural functions (Ducrest et al. 2008) and positively and negatively related to fitness  
70 (Roulin et al. 2003; Meunier et al. 2011) in a context-dependent manner (e.g. Roulin et al.  
71 2008a, b).

72         Most work had focussed on the size and shape of melanin or depigmented areas. In  
73 contrast, melanin spotting has received considerable attention but in more limited numbers of  
74 species. There have been extensive investigations in barn owls *Tyto alba* (e.g. Roulin et al.  
75 1998), and to a lesser extent in other species (salmonids *Salmo salar* & *Oncorhynchus mykiss*:  
76 Kittelsen et al. 2009; snow buntings *Plectrophenax nivalis*: Guindre-Parker et al. 2013).  
77 Evidence suggests that spotting is linked to components of fitness such as reproduction  
78 (Guindre-Parker et al. 2013) and survival (Roulin et al. 2010), suggesting a link between the  
79 ability to control melanin production and underlying condition (e.g. Kittelsen et al. 2009).  
80 Melanin spotting, may therefore act as a signal of condition.

81         The black grouse *Lyrurus tetrix* is a lekking galliform that has been studied  
82 extensively in the context of sexual selection and sexual traits (see Kervinen et al. 2015,

83 2016). Traits include tail (lyre) length and quality (Höglund et al. 1994), the size of  
84 testosterone-dependent red eye combs (Rintamäki et al. 2000), body mass (Lebigre et al.  
85 2013) and blue structural coloration (blue chroma) of breast feathers (Siitari et al. 2007).  
86 Females also prefer mating with males that have high lek attendance (Alatalo et al. 1992),  
87 fight frequently and successfully against other males (Hämäläinen et al. 2012), and occupy  
88 central territories on leks (Hovi et al. 1994). The dominant coloration of black grouse is  
89 eumelanin-based i.e. black, but there are also depigmented patches on the upper and  
90 underside of the wing and the conspicuous white undertail coverts. These undertail coverts  
91 are held open in a fan shape during calling on the lek (Figure 1; Höglund et al. 1994).  
92 However, black grouse tail fans are not always totally white; melanin spotting of various  
93 sizes can be found in different parts of the feathers (Figure 1). Despite being used as  
94 individual identifiers on leks (e.g. Alatalo et al. 1992), the functional significance of these  
95 spots is unknown to date. To address this issue, we therefore tested the relationship between  
96 melanin spotting on a white ornament and (a) its age-dependence, (b) its relationship to two  
97 major components of fitness (mating success, survival) and (c) their interrelationship with  
98 current and past expression of sexual signals in black grouse.

99

## 100 **MATERIALS AND METHODS**

### 101 **Capture and handling**

102 During 2004–2010, we collected longitudinal data describing male mating success and multiple  
103 sexual traits in five study sites in Central Finland (peat bogs with high visibility, ca. 62°15'N;  
104 25°00'E). In addition, males were monitored during the mating season until spring 2014 to  
105 ensure the use of complete cohorts (no ringed males alive) in the analyses. Each study site is a  
106 local main lek with 5–40 territorial males, and as local hunting clubs have refrained from

107 hunting on these areas, the age structure of the study population can reasonably be assumed to  
108 be natural.

109         Measurements of morphological traits were carried out annually in January–March by  
110 catching birds from the winter flocks with oat-baited walk-in traps (for methods, see  
111 Kervinen *et al.* 2012; Lebigre *et al.* 2012). Each captured individual was aged as yearling or  
112 older according to plumage characteristics and individually ringed for future identification  
113 with an aluminium tarsus ring with a unique serial number and three colored tarsus rings. All  
114 captured birds were measured for body mass, lyre (i.e. tail) length, eye comb size and blue  
115 chroma coloration of breast feathers (see Siitari *et al.* 2007; Lebigre *et al.* 2012 for details).  
116 When captured, the grouse holds the tail fan closed and concealed making it impossible to  
117 fully photograph the tail spread manually. Therefore, each feather of the white tail fan was  
118 manually inspected and for each individual, the total number of melanin spots and their  
119 location (at tip or within the main part of the vane) was recorded (Figure 1).

120

### 121 **Mating success, lekking behaviour and survival**

122 Male mating success and lekking behaviour were recorded on the study sites annually during  
123 the mating season (late April – early May, Ludwig *et al.* 2006). During this period, leks were  
124 daily monitored *ca.* from 3:00 to 8:00 a.m. from hides near the lek arenas (for methods, see  
125 Kervinen *et al.* 2012; Lebigre *et al.* 2012). Male behaviour was scan sampled at regular intervals  
126 and categorised as rookooing, hissing (the two main vocal displays), fighting or inactive  
127 (methods and behaviours are described in detail in Höglund *et al.* 1997). Lek attendance  
128 (proportional to the most commonly present male on the same lek), the relative proportion of  
129 each behaviour, and males' territory distances from the lek centre (see Lebigre *et al.* 2012 for  
130 descriptions of all behavioural attributes) were calculated for all individually identifiable males

131 from the records for males. Whenever recording male activity, the spatial location of each male  
132 on the lek was also recorded. Finally, we documented the observed copulations, which are easy  
133 to observe, as males flap their wings conspicuously when mounting females, their location and  
134 the individuals involved. Males captured during winter or recorded at leks were classified as  
135 alive; due to male philopatry for their initial lekking site (Lebigre et al. 2008), males no longer  
136 seen on the study sites were considered dead.

137

### 138 **Data Analysis**

139 We considered the location of melanin spots as being important, as they may reflect different  
140 moments during feather development. We first tested whether there were age-specific  
141 patterns of spot expression by fitting a poisson GLMM with spot number (either vane or tip)  
142 as a dependent variable, age (linear) and age<sup>2</sup> (quadratic) fitted as fixed factors, and age and  
143 individual ring number were fitted as random slopes and random intercepts effect (1+age|id).  
144 We then tested the relationship between spot number and survival and annual mating success  
145 using binomial and Poisson GLMMs, respectively. Survival (alive=1, dead=0) was tested  
146 with age and spot number as fixed effects. Mating success was tested with age, age<sup>2</sup> and  
147 spot number as fixed effects. In both models, age and individual ring number were fitted as  
148 random slopes and random intercepts effect (1+age|id).

149 We then tested the covariance between spot number and morphological (body mass,  
150 lyre length, blue chroma coloration) and behavioural traits (lek attendance, fighting rate,  
151 distance from lek centre) using Poisson GLMMs; morphological or behavioural traits were  
152 fitted as fixed effects in separate multivariate analyzes. Distance from lek centre was included  
153 as linear and non-linear functions. This is because distance from lek centre is acquired over a  
154 number of years, and high quality but young males may be at the lek periphery and move

155 inwards as they age whereas old males of declining condition may be nearer the lek center  
156 but moving outwards (Kokko et al. 1998; Kervinen et al. 2015). Age has a very strong effect  
157 on the trait expression, particularly between ages 1 and 2, which is likely to exaggerate any  
158 covariance (Kervinen et al. 2015). As a consequence, we tested yearlings and adult birds ( $\geq 2$   
159 years old) separately and included age as a second random effect in the latter analyzes.

160         Lastly, we tested for a lag between past trait expression and current spot number. To  
161 do this, we analyzed the number of spots (vane or tip) and used the traits (morphological or  
162 behavioural) expressed in the previous year. Again, the age-specific increase in trait values  
163 between age 1 and 2 is likely to have an important impact on results, so we analyzed 2 year  
164 old birds against their yearling (1 year old) values separately from older birds ( $\geq 2$  years old).  
165 All models were run in R version 3.2.1 (R Core Team 2015) with GLMMs using the lme4  
166 package (Bates et al. 2015). In total, we have data for 152 individual males and 325 counts of  
167 spots.

168

## 169 **RESULTS**

### 170 **Age and spot numbers**

171 There was a significant negative relationship between vane spots and age and a significant  
172 positive quadratic relationship (GLMM: linear age,  $\beta \pm SE = -0.89 \pm 0.10$ ,  $Z = -9.10$ ,  $P < 0.001$ ;  
173 quadratic age,  $\beta \pm SE = 0.12 \pm 0.02$ ,  $Z = 7.29$ ,  $P < 0.001$ ), which indicated that young and old  
174 individuals had more vane spots than prime/middle age males (Figure 2a). This pattern is  
175 opposite to that of tip spots which were a significantly positively related to male age with a  
176 significant negative quadratic effect (GLMM: linear age,  $\beta \pm SE = 1.91 \pm 0.17$ ,  $Z = 10.82$ ,  
177  $P < 0.001$ ; quadratic age,  $\beta \pm SE = -0.18 \pm 0.02$ ,  $Z = -7.44$ ,  $P < 0.001$ ), which meant that young and

178 old birds had fewer tip spots than prime/middle age males (Figure 2b). There was no  
179 correlation between the number of vane and tip spots ( $r_s=-0.02$ ,  $P=0.645$ ).

180

### 181 **Melanin spotting and fitness**

182 The number of vane spots was unrelated to male survival and annual mating success while  
183 accounting for age (Table 1a and c; Figure 2a). In contrast, individuals with larger numbers  
184 of tip spots had reduced survival and lower annual mating success (Table 1b and d; Figure  
185 2b; Figure 3).

186

### 187 **Melanin spotting and current sexual signals**

188 The number of vane spots and tip spots were unrelated to any other morphological traits in  
189 adults (Table 2). There was a significant negative relationship between vane spots and lek  
190 attendance in adult birds (Table 2; Figure 4a), and a non-significant quadratic relationship  
191 with distance from lek centre suggesting that birds close and further from the lek centre had  
192 fewer vane spots (Table 2). Tip spots were negatively related to fighting rate in adults (Table  
193 2; Figure 4b), and there was a non-significant quadratic relationship with distance from lek  
194 centre suggesting that birds very close and furthest from the lek centre had fewest tip spots  
195 (Table 2; Figure 4c). For yearlings, there was only a weak positive trend for blue chroma to  
196 significantly relate to vane ( $P=0.066$ ) and unrelated to tip spots ( $P=0.122$ ), but not for any  
197 other morphological trait (Table 3). No behavioural traits were associated with vane or tip  
198 spot number in yearlings (Table 3).

199

### 200 **Melanin spotting and past sexual signals**



201 The number of vane spots were unrelated to both morphological and behavioural trait values  
202 from the previous year in >2 year old adults (Table 3). The number of tip spots were also  
203 unrelated to past morphological traits (Table 3), but positively related to fighting rate and  
204 distance from lek centre from the previous year (Table 3; Figure 5a and b).

205 For males which were juveniles in the previous year, there were significant  
206 relationships between blue chroma in the previous year and vane and tip spots, but not for  
207 any other morphological trait (Table 3). Similarly there was no relationship between vane  
208 spots and behavioural traits in the previous year, and a positive trend only for lek attendance  
209 ( $P=0.052$ ) to relate to tip spot number (Table 3).

210

## 211 DISCUSSION

### 212 **Location differences in spotting**

213 The specific location of melanin spots on white undertail covert feathers of black grouse  
214 seems absolutely crucial. Males with more spots at the tips of feathers have lower survival  
215 and reproductive success, whereas spots located in the inner or outer vane are unrelated to  
216 male fitness. At the same time, there was stronger covariance with past and present  
217 behavioural traits in tip spots, when compared to vane spots. It has been previously shown in  
218 other species, that a single plumage characteristic can signal multiple components of quality  
219 or fitness (e.g. Doucet et al. 2003) but in this case we show that the magnitude and direction  
220 of the association of a single signal (white feather melanization), has fine scale difference in  
221 meaning. Such differences in the location of the black spot most probably results from  
222 differences in the timing of the deposition of melanin granules in the feather follicles by the  
223 melanocytes during moult. Indeed, the black spots on the white tail feathers of black grouse

224 are located on the closed pennaceous portions of the feather and individual pennaceous barb  
225 ridges grow helically around the follicular collar towards the rachis (Prum and Williamson  
226 2001). This means that melanin is being deposited at the very start of the barb's growth in the  
227 case of tip spots, and later on for vane spots. In black grouse, the onset of moult occurs  
228 directly after the breeding season (Ginn and Melville 1983) and it has been shown that in a  
229 closely related species (the blue grouse; *Dendragapus obscurus*), the under tail coverts are  
230 moulted at the same time as the rectrices, right at the end of moulting (Zwickel and Bendell  
231 2004). Moulting in birds has an energetic cost and specific nutrient requirements (Bryant  
232 1997). Depletion of certain resources such as of calcium and cysteine may impact  
233 melanization (Stewart and Westneat 2013). For example, glutathione is used as a cysteine  
234 reservoir during moult (Murphy and King 1990), and depletion of glutathione can lead to  
235 increased melanogenesis (Galván and Alonso-Alvarez 2008). In addition, melanin production  
236 is tightly linked to production of corticosterone (Ducrest et al. 2008). Increased amounts of  
237 spotting may therefore reflect greater physiological stress during moult. We unfortunately can  
238 only speculate as to how and why the melanin spots occur because we lack detailed  
239 physiological measurements in our study system, and addressing this issue will be a key  
240 future research question.

241

#### 242 **Age and covariance with other sexual traits**

243 There was a strong age-dependence of melanin spotting suggesting that the factors driving  
244 production of these spots are linked to other age-specific processes. Indeed, vane spots were  
245 highest in young and older birds, whereas tip spots number increased with age and within-  
246 individual changes were highest at older ages. In barn owls, the number of spots in males  
247 declined during young age classes, but not at older ages (Dreiss & Roulin 2010). Many

248 species show maximal trait expression during prime-age (e.g. Vanpe et al. 2007), and in  
249 grouse other sexual signals are highest during prime-age (Kervinen et al. 2015). This suggests  
250 that potential drivers of spot production are linked to age-specific changes in condition,  
251 signalling, regulation or behaviour.

252 Males of many species have multiple signals of quality. These multiple signals can be  
253 used for a number of purposes such as backup signals or multiple messages for mate choice  
254 (Candolin 2003; Bro-Jørgensen 2010) and often multiple traits covary positively, even if they  
255 have different temporal integration with male quality (Badyaev 2004). Yet, contrary to these  
256 expectations age-specific patterns of spotting did not strongly covary with other traits This  
257 suggests that tail spots are signalling different components of the condition that are unlinked  
258 to the development of other sexual ornaments. This result may confirm past results  
259 suggesting that multiple cues can provide different information on male quality (Møller &  
260 Pomiankowski 1993, Johnstone 1997, Siitari et al. 2007, Kervinen et al. 2016.). Other studies  
261 comparing numbers of black spots have found mixed results. Snow buntings showed a  
262 significant negative covariance between total spotting and three current plumage traits (S.L.  
263 Guidre-Parker pers. comm.), whereas in barn owls, tail length and body mass (Roulin 2006),  
264 but not wing feather traits (Roulin et al. 2013) are related to spotting. We also found limited  
265 relationships with previous morphological traits. Only in yearling males were there any  
266 significant patterns, with lighter yearling males having more tip spots in the following year,  
267 again suggesting that tip spots are linked to males' past condition.

268 Moulting in black grouse typically occurs during June-September (exceptionally May;  
269 Ginn and Melville 1983). This means there is a short temporal separation between past  
270 behavioural traits and spot production. Unsurprisingly, the relationship between past traits  
271 and tip spots is stronger. We found a positive covariance with past fighting rates and tips  
272 spots and a negative covariance between current fighting rate and tip spots and current lek

273 attendance and vane spots. Fighting is a costly activity and high investment leads to large  
274 losses in body mass (Hämäläinen et al. 2012). As is widespread in ecology (Harrison et al.  
275 2011), it seems that there is a carry-over effect from high investment in lekking (fighting and  
276 lek attendance) from one year to the next; high effort in one year leads to increased spotting  
277 the following year. Since the relationship between spotting and behavioural traits is negative  
278 in the current year, this suggest that there is some decline in condition between years which  
279 spotting is correlating with.

280           In black grouse and many other species, males nearest to the lek centre are the best  
281 quality and have the highest mating success (Hovi et al. 1994; Bro-Jørgensen, and Durant  
282 2003; Stein and Uy 2006); hence, males close to the lek centre are of high quality. Males  
283 move closer to the lek centre as they reach prime-age and move further from the lek centre  
284 post-prime (Kervinen et al. 2015). There was a positive relationship between distance from  
285 lek centre and tip spots in the previous year and a trend for a negative quadratic relationship  
286 in the current year. This means that those males closest to the lek centre had few tip spots  
287 both in the past and the current year. Again, this suggests that spotting is correlating  
288 somehow with male condition or quality.

289

### 290 **Fitness effect and spotting**

291 There is typically a strong positive relationship between sexually selected traits and male  
292 fitness (Andersson 1994). We found that male black grouse with a large number of tip spots  
293 had lower annual mating success and were less likely to survive to the next year. This  
294 suggests that the melanisation at the tip spots themselves are honestly signalling some  
295 negative component of males current and past condition (Galván and Alonso-Alvarez 2009;  
296 Hõrak et al. 2010). In black grouse, reproductive effort leads to significant loss of body mass

297 (Lebigre et al. 2013), particularly in prime-aged males (Kervinen et al. 2012, 2015). This  
298 high reproductive effort is likely to generate high levels of oxidative stress and potentially  
299 deplete key antioxidants such as glutathione (Anderson 1988), which in turn can lead to  
300 greater melanin expression (Galván and Alonso-Alvarez 2008). Lekking may also be  
301 stressful, and high corticosterone levels would further link to melanin production (Ducrest et  
302 al. 2008).

303         Other studies have shown that the expression of a larger melanin ornaments may  
304 honestly signal underlying physiological costs, e.g. reduced survival (Moore et al. 2015),  
305 possibly because of the costs of melanin production. For the white feathers of black grouse,  
306 we suggest the reverse. It appears that the white feathers themselves either have production  
307 costs (Vágási et al. 2010) or that the ability to suppress melanin production is condition-  
308 dependent. Similar to our results, spotting seemed to be negative for males of other species;  
309 male barn owls with larger melanin spots had reduced survival (Roulin et al. 2010) and male  
310 snow buntings with more spotting on white feathers had lower fledgling success (Guidre-  
311 Parker et al. 2013).

312         We do not know whether tip spots themselves are sexually-selected. The tail fan and  
313 its intactness has been shown to have a role in female choice in black grouse possibly as a  
314 secondary cue in less dominant males (Höglund et al. 1994). The location of tip spots makes  
315 them more visible during display. As they additionally seem to honestly indicate some  
316 underlying male quality/condition, they have the potential to be used by females during mate  
317 choice.

318

319 **Conclusions**

320 Our results demonstrate that spots at the tip of the undertail coverts of male black grouse  
321 were negatively correlated to male quality. Males with more tip spots had reduce  
322 performance during lekking, lower mating success and lower subsequent survival. Though tip  
323 spots clearly indicated some negative relationship with condition, we have no clear idea why.  
324 The melanin spotting provides an important system to further advance our understanding of the  
325 mechanistic basis for the control of pigment production and achromatic plumage.

326

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496

497 FIGURE LEGENDS

498 Figure 1: Photographs showing male black grouse holding their fan open during lek display,  
499 with examples of both tip and vane spots.

500

501 Figure 2: Mean $\pm$ SE number age specific variation in the number of (a) vane and (b) tip spots.  
502 Number of spots for individuals that died (open boxes) or survived to the following year  
503 (filled boxes) are shown.

504

505 Figure 3: The relationship between annual mating success and number of tip spots across all  
506 age classes is shown.

507

508 Figure 4: The relationship (a) current fighting rate the number of vane spots and (b) the current  
509 fighting rate and(c) distance from lek centre and number of tip spots

510

511 Figure 5: The relationship between number of tip spots (a) fighting rate and (b) distance from  
512 lek centre in the previous year.

513

514 Table 1: GLMM results for male survival and annual mating success in relation to age and  
 515 the number of either vane or tip spots.

516

<b>Model</b>	<b>Parameter</b>	<b><math>\beta \pm SE</math></b>	<b>Z</b>	<b>P</b>
(a) Survival	Age	-0.55±0.21	-2.62	0.009
	Vane spots	0.05±0.03	1.51	0.131
(b) Survival	Age	-0.46±0.18	-2.49	0.013
	Tip spots	-0.08±0.04	-2.05	0.041
(c) Annual Mating Success	Age (linear)	4.87±0.62	7.82	<0.001
	Age (quadratic)	-0.66±0.10	-6.86	<0.001
	Vane spots	0.05±0.04	1.28	0.200
(d) Annual Mating Success	Age (linear)	5.17±1.10	8.78	<0.001
	Age (quadratic)	-0.68±0.09	-7.44	<0.001
	Tip spots	-0.07±0.03	-2.07	0.038



517

518 Table 2: GLMM outputs for the relationship between vane and tip spots and other sexually  
 519 selected traits in juvenile and adult black grouse males.

520

Model	Parameter	Juveniles			Older		
		$\beta \pm SE$	Z	P	$\beta \pm SE$	Z	P
Vane spots	Body mass	0.00±0.00	-0.01	0.989	0.00±0.00	0.69	0.491
	Lyre length	0.00±0.00	-0.32	0.752	0.02±0.01	1.68	0.093
	Blue chroma	2.81±1.53	1.84	0.066	3.54±2.64	1.34	0.181
	Red eye combs	0.08±0.05	1.55	0.121	0.08±0.07	1.09	0.277
Vane spots	Lek attendance	0.58±0.54	1.09	0.276	-1.32±0.65	-2.03	0.042
	Fighting rate	-0.06±0.72	-0.08	0.938	0.02±0.04	0.04	0.971
	Distance from lek centre (linear)	0.04±0.02	1.60	0.110	0.04±0.02	1.62	0.105
	Distance from lek centre (quadratic)	0.00±0.00	-1.51	0.130	0.00±0.00	-2.14	0.032
Tip spots	Body mass	0.00±0.00	1.23	0.218	0.00±0.00	-0.06	0.949
	Lyre length	-0.02±0.02	-0.89	0.372	0.01±0.01	0.76	0.445
	Blue chroma	9.92±6.43	1.55	0.122	-4.51±3.68	-1.23	0.220
	Red eye combs	0.23±0.22	1.06	0.291	0.18±0.10	1.71	0.087
Tip spots	Lek attendance	-0.19±3.39	-0.06	0.955	-0.65±0.80	-0.81	0.418
	Fighting rate	0.26±4.55	0.06	0.954	-2.31±1.02	-2.27	0.023
	Distance from lek centre (linear)	-0.03±0.14	-0.22	0.826	0.07±0.04	1.65	0.099
	Distance from lek centre (quadratic)	0.00±0.00	0.36	0.721	0.00±0.00	-2.60	0.009

521 Table 3: GLMM outputs for the relationship between vane and tip spots and past sexually  
 522 selected traits in adult black grouse males. Individuals can express traits as 2 years old  
 523 expressed past sexual signals as juveniles (1 year olds) so are analysed separately from older  
 524 bird (>2 years old).

525

Model	Parameter	2 years old			Older		
		$\beta \pm SE$	Z	P	$\beta \pm SE$	Z	P
Vane spots	Body mass	0.00±0.00	-0.45	0.650	-0.00±0.00	-1.55	0.122
	Lyre length	0.00±0.01	-0.14	0.890	-0.01±0.01	-0.78	0.433
	Blue chroma	11.00±2.81	3.92	<0.001	7.45±5.19	1.44	0.151
	Red eye combs	-0.08±0.09	-0.86	0.388	0.00±0.11	0.08	0.935
	Lek attendance	-0.19±0.61	-0.31	0.758	-0.35±0.73	-0.48	0.629
	Fighting rate	0.48±0.72	0.67	0.506	1.08±0.84	1.29	0.196
	Distance from lek centre	0.00±0.01	-0.15	0.883	0.00±0.01	0.30	0.762
Tip spots	Body mass	-0.01±0.00	-3.73	<0.001	0.00±0.00	-0.58	0.562
	Lyre length	0.01±0.01	1.68	0.093	-0.03±0.02	-1.61	0.107
	Blue chroma	18.55±3.36	5.53	<0.001	9.59±6.32	1.52	0.129
	Lek attendance	1.76±0.91	1.94	0.052	1.88±1.51	1.25	0.211
	Fighting rate	1.04±0.85	1.22	0.221	2.89±1.09	2.66	0.007
	Distance form lek centre	-0.01±0.02	-0.80	0.424	0.05±0.02	2.47	0.014

526



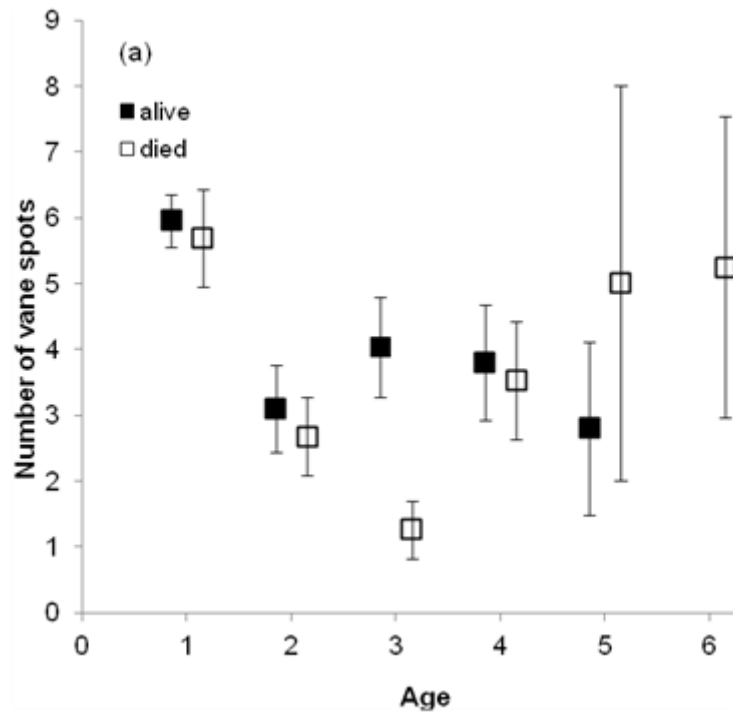
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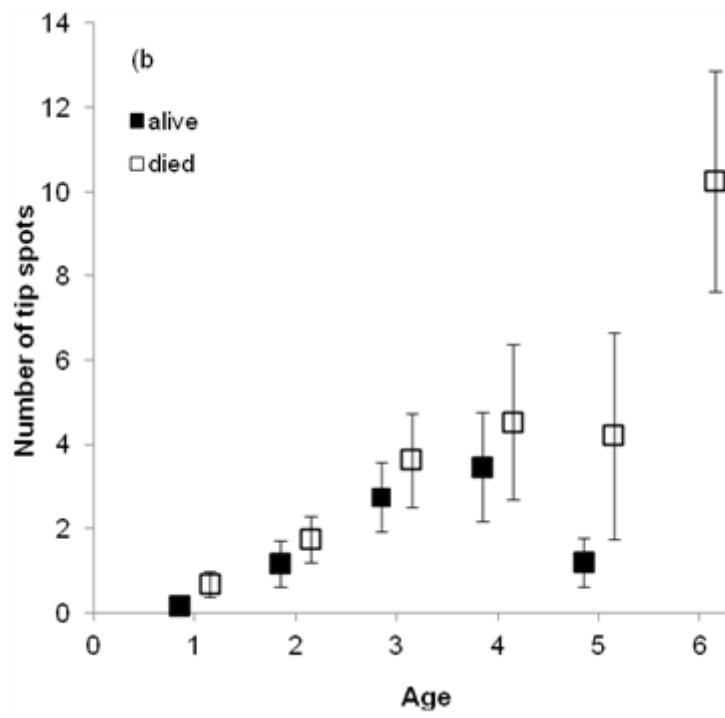
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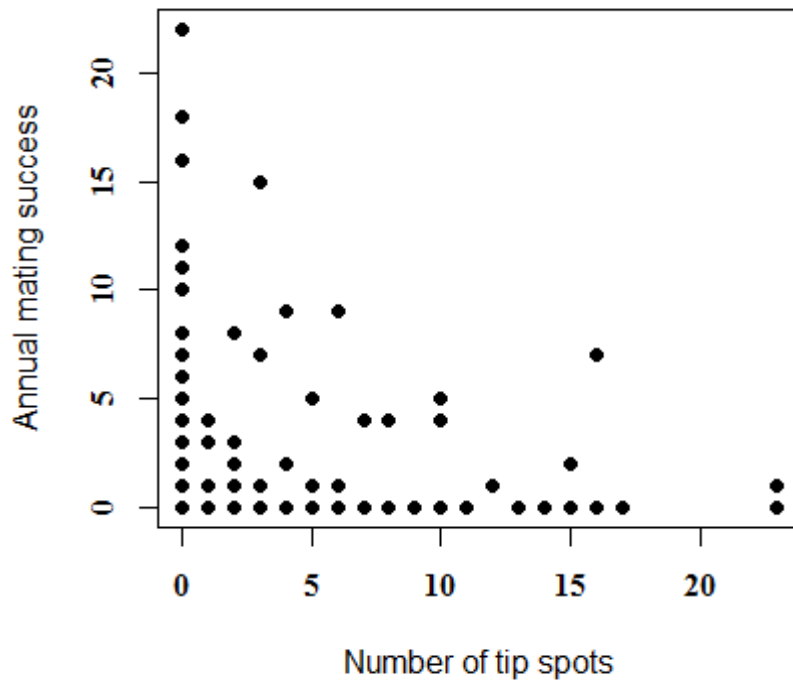


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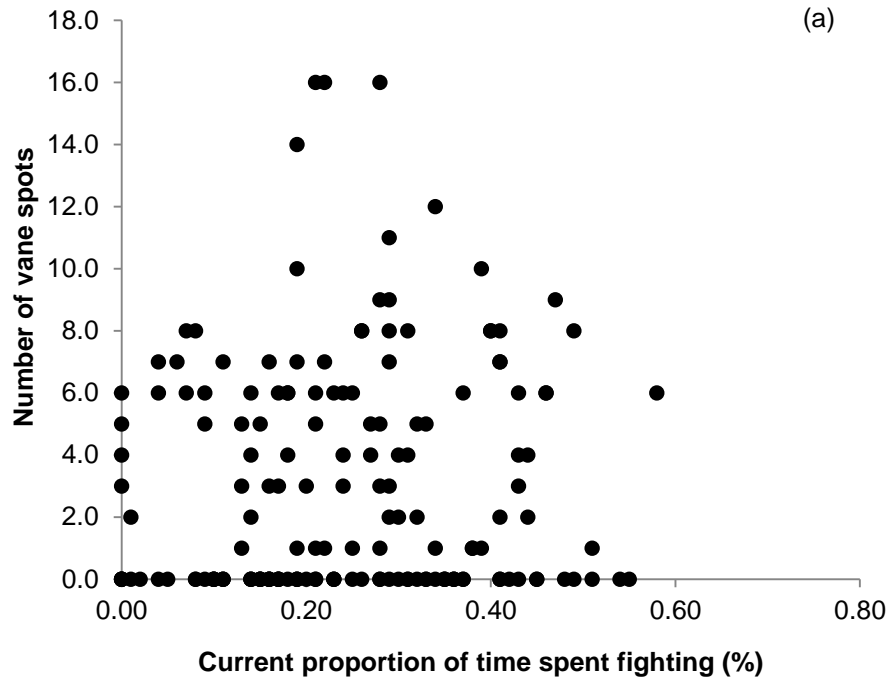
Figure 2



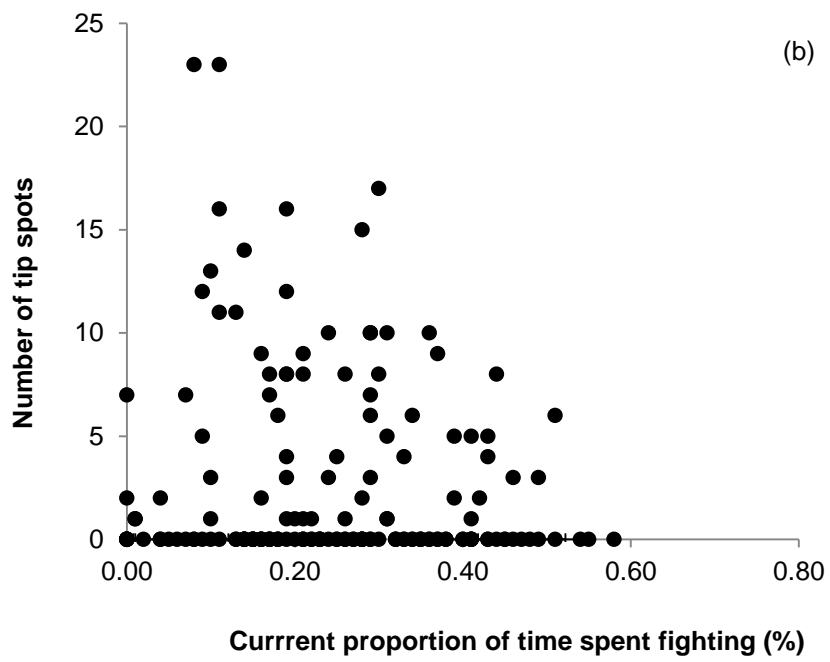
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Figure 3

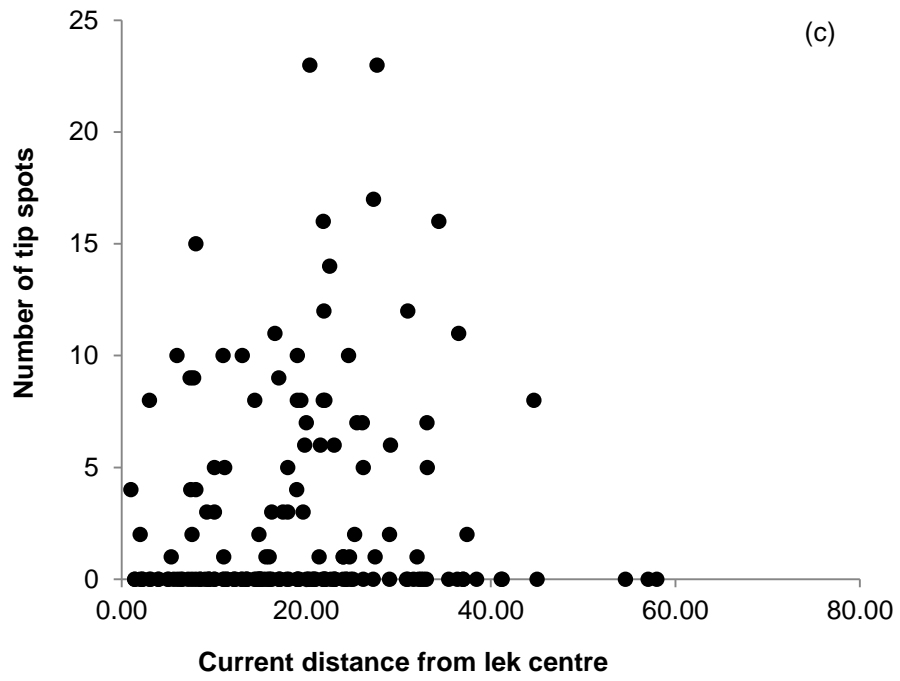


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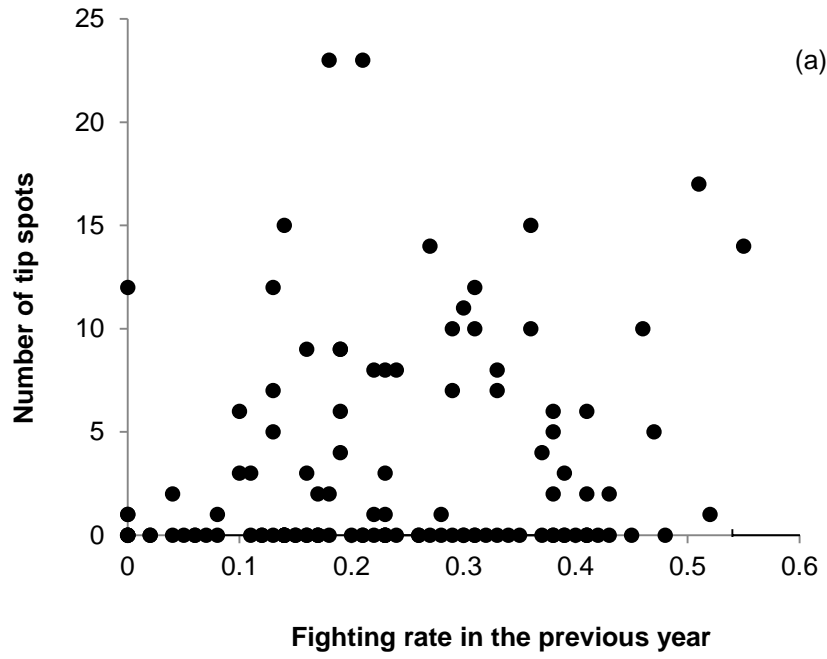


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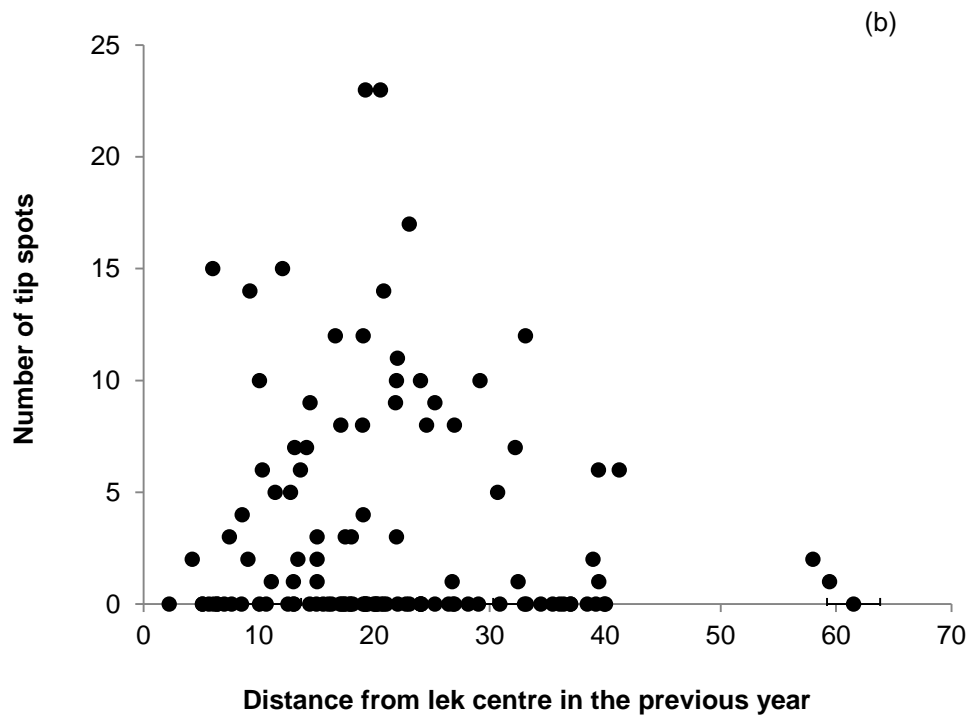
Figure 4

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Figure 5