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1 **Assessment of circadian rhythm of activity combined with random regression model as a**
 2 **novel approach to monitoring sheep in an extensive system**

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19
 20 **Abstract**

21 Sensor-based technologies are becoming increasingly available and can be used to automatically
 22 gather long-term data about animal behaviour. With this information, it is possible to assess the
 23 circadian rhythm of activity and monitor its response to internal and external factors. Identifying
 24 irregularities in this rhythm may indicate animal health and welfare issues. The aim of this study
 25 was to collect sensor-based general activity and investigate circadian rhythm of this activity to
 26 identify the changes due to weather influences that act on these parameters throughout the year;
 27 to identify the differences between individuals; and to assess links between general activity and
 28 circadian rhythm of activity with sheep body weight change. In total, 29 Scottish Blackface
 Degree of Functional Coupling (DFC)

29 ewes of different ages and body condition scores were used. The animals were monitored for
30 four consecutive weeks in each of four seasonal periods, in extensive systems on Scottish
31 upland pastures, and without human handling during study periods. Accelerometer-integrated
32 collars were fitted to the animals to collect the motion index continuously every minute. These
33 data were used to calculate the percentage of cyclic behaviour that was harmonic/ synchronized
34 with the environment (over 24 h period), as Degree of Functional Coupling (DFC). The DFC
35 was shown within rolling seven-day periods. Low DFCs indicate low synchronization. Weather
36 data were collected daily. Random regression models were used to assess between-individual
37 variation. During the winter period, the level of the DFC for the activity of nineteen ewes
38 lowered in response to a period of high level of precipitation combined with the low winter
39 temperatures. However, four ewes exhibited a lower level of variation in the DFC values,
40 showing that there were differences between individuals in regard to their response to the
41 precipitation level. The overall mean of the DFC for the general activity was highest in autumn
42 (95.4%, $P < 0.001$), however, it did not differ between summer and spring (respectively 90.2%
43 and 88.1%, $P > 0.05$), but was significantly lower during the winter (81.7%, $P < 0.001$)
44 compared with summer and autumn. Over the spring and summer, variation in DFC was a good
45 estimator of body weight gain. It was concluded that the assessment of circadian rhythms of
46 general activity using the DFC-parameter allows a better understanding of sheep responses to
47 weather influences, compared to the evaluation of general activity alone. The random regression
48 model method was effective in identifying animals that deviated positively or negatively from
49 population responses.

50 **Keywords:** between-individual variation; Degrees of Functional Coupling; phenotypic
51 plasticity; precision livestock; sheep performance; seasonal adaptation.

52 **1. Introduction**

53 In extensive production systems, animals are often kept in semi-natural habitats without
54 close supervision for long periods. The use of a telemetric monitoring system could be very
55 important to ensure health and welfare, and provide management information for the animals.

56 Use of ICT (Information and Communication Technology) to optimise management processes
57 in livestock production has increased in recent years. Major developments can be found in
58 intensive production systems, particularly with the direct use of remotely collected data on
59 behavioural traits, such as the use of activity data to detect oestrus in dairy cows (Firk et al.,
60 2002). Technologies such as accelerometer-based sensors enable us to measure animal
61 behaviour, for example, general activity, and these continuous data can be used to calculate
62 parameters describing the rhythmic structure of behaviour.

63 Biological processes display an endogenous oscillation of about 24 hours, known as
64 circadian rhythms (Foster and Kreitzman, 2005; Koukkari and Sothorn, 2006; Piccione et al.,
65 2005; Refinetti and Menaker, 1992; Wood and Loudon, 2014). Many basic behaviours, such as
66 sleep and activity, are under the control of these rhythms. The rhythms are controlled by genes
67 activating proteins and neurotransmitters (Foster and Kreitzman, 2005; Wood and Loudon,
68 2014). Although these biological processes are endogenous, they are adjusted to the local
69 environment by circadian cues (e.g. light, temperature and humidity) (Foster and Kreitzman,
70 2005; Lincoln and Richardson, 1998). Thus, the circadian rhythm of behavioural variables is an
71 outcome of both endogenous and exogenous motivational factors (Foster and Kreitzman, 2005;
72 Scheibe et al., 1999). Accordingly, Scheibe et al. (1999) developed a parameter called Degree of
73 Functional Coupling (DFC) to characterise the circadian rhythm of behavioural variables.
74 Therefore, DFC expresses the percentage of the measured behaviour that is harmonically
75 synchronized with environmental rhythms, over a 24-hour period.

76 Valuable information may be obtained by looking at the synchrony between behavioural
77 and environmental rhythms. A considerable number of studies has shown a link between
78 environmental influences or health issues with the level of DFC for activity of free-ranging
79 animals (Berger et al., 2003, 1999, Scheibe et al., 1999, 1998), with a strong circadian rhythm
80 of activity known to be a characteristic of healthy and adapted organisms (Berger et al., 2003,
81 1999; Bloch et al., 2013). Thus, the rhythmicity of activity is a variable that has clear potential
82 to be a key indicator of the state of animals. This suggests that the evaluation of the DFC would
83 provide useful information as part of an assessment of animal welfare in farmed livestock. Until

84 recently, continuous 24/7 monitoring of animal activity was not cost-effective. However, the
85 rapid development of new technologies makes it possible to look a new way of using this type
86 of data.

87 The rhythmicity of activity of each animal is unique and may vary according to short- and
88 long-term environmental influences or its physiological status (Berger et al., 2003; Scheibe et
89 al., 1999). Therefore, an in-depth look at the within- and between-individual behavioural
90 rhythmicity is important to understand at an individual level whether there are relationships
91 between the rhythmicity of activity, environmental influences and production traits. However,
92 to fully understand the complex behavioural variation between individuals within a single
93 population is not a simple task, a large number of recent studies have focussed on how to
94 measure these between-individual differences (Dingemanse et al., 2010; Dingemanse and Wolf,
95 2013; Herczeg and Garamszegi, 2012; Nussey et al., 2007; Sih et al., 2004).

96 Many evolutionary ecology studies have used simple linear regression models to
97 understand and identify the population level response to the environment, at both phenotypic
98 and genetic levels. In this model, the coefficient of the linear regression of the phenotype (for
99 example behaviour) with the environmental variable is described as the 'reaction norm'
100 (Pigliucci, 2001). However, a study by Nussey et al. (2007) asserted that little is known about
101 the prevalence, and evolutionary and ecological causes and consequences of the variation in life
102 history phenotypic plasticity (the ability of individuals to adapt their phenotypic traits in
103 response to the environment) in the wild. Considering this, Nussey et al. (2007) outlined an
104 analytical framework using the reaction norm concept and random regression models to access
105 not only the population level responses but also the between-individual variation of labile traits.
106 Labile traits are those that easily alter over an individual lifetime, e.g. those related to
107 physiology or behaviour (Nussey et al., 2007). In order to understand the between-individual
108 variation of activity rhythmicity, this study combines for the first time the DFC analysis for
109 each animal with random regression models.

110 Sheep have seasonal production cycles and often occur in bio-geographic zones with
111 large shifts in grassland quality and availability and light/dark cycles (Lincoln et al., 1990;

112 Lincoln and Richardson, 1998). Extensive systems throughout the world have high levels of
113 variation in environmental and nutritional impacts upon the farmed livestock. Scottish extensive
114 systems pose particularly demands upon sheep, where winter periods of poor grazing nutrition,
115 short day lengths, variable and extreme weather and pregnancy overlap. Poor welfare and high
116 rates of mortality are risks within these systems, and the sheep are often inspected infrequently
117 with limited potential for intervention (Morgan-Davies et al., 2008).

118 The aim of this study was to analyse activity data (through a motion index) and the
119 circadian rhythm of this activity to identify changes due to weather influences that act on these
120 parameters throughout the year; to identify the differences between individuals; and to assess
121 links between general activity and circadian rhythm of activity with sheep body weight gain.
122 The study was carried out with an experimental cohort of animals, kept in a challenging
123 environment, representative of the challenges faced by the wider population of farmed sheep in
124 extensive systems. Our study has the novel approach of addressing the key issue of whether the
125 calculation of the circadian rhythm of general activity using the DFC-parameter better explains
126 sheep behaviour and performance compared to general activity measured through the motion
127 index.

128 **2. Material and Methods**

129 This experiment was conducted at Scotland's Rural College (SRUC) Hill & Mountain
130 Research Centre. The experimental protocol was approved by SRUC's Animal Welfare and
131 Ethical Review Body, the Animal Experiments Committee, and was conducted in accordance
132 with the requirements of the United Kingdom Animals (Scientific Procedures) Act, 1986.

133 *2.1. Animal measurements*

134 The study was done on a group of 29 Scottish Blackface ewes (Supplementary Table 1).
135 The ewes covered a range of ages (two to six years old) with a body condition score (BCS)
136 ranging from 2 to 3 points at the start of the study. The BCS scale ranges from 1 to 5 where 1 is
137 thin and 5 is fat. The ewes were kept on 23.6 ha of semi-natural pasture in the West Highlands

138 of Scotland (SRUC, Hill & Mountain Research Centre, Scotland), with a latitude and longitude
139 of 56.4381 and -4.6684. The annual precipitation is typically 2800 mm. A UK Meteorological
140 Office Weather Station, based on the research farm and a distance of < 2 km away from the
141 field location, measured the precipitation and temperatures every day. Over all seasons the ewes
142 were kept under natural conditions and no formal built shelter was provided. Human contact
143 was avoided, except for the daily visual checks over all seasons and feed provision over the
144 winter. Ewes were closely checked at the start and end of each measurement period. Endpoint
145 criteria for a potential exclusion of an animal from the experiment were in place according to
146 the ethical review body of SRUC, and no sheep fell below body condition score limits. During
147 the winter period, ewes were supplemented with two large round hay bales, one provided on the
148 17th of January and the other on the 5th of February. Energy and protein feed blocks (Rumevite
149 Sheep, Rumenco, Burton-on-Trent, UK) were also provided, at a rate of one per week, each
150 weighing 20 kg. The supplementation and the adjacent grazing was representative of that given
151 to the larger flock from which the experimental cohort were drawn. Overall levels of grass
152 quality and quantity and supplementation provided were judged to be somewhat higher than the
153 larger flock, but the experimental sheep, being within a smaller area had less choice in terms of
154 grazing area and moving to natural shelter features. In spring, summer and autumn, the ewes
155 were exclusively grazed on semi-natural pasture.

156 Sheep activity data were continuously collected by three-axes accelerometers (IceTag
157 Pro, IceRobotics Ltd., Edinburgh, Scotland) integrated into a collar and fitted to the ewes.
158 Activity data were recorded over four consecutive weeks in each season, across a full annual
159 cycle, commencing in the winter. The collars were changed every period and, thus, ewes were
160 wearing different Icetag Pro loggers during different seasons to ensure any systematic
161 differences between collars were not confounded with the animal. The IceTag Pro logger is
162 programmed to record the g-force in three dimensions (IceRobotics Ltd, Product Guide 2010),
163 providing the motion index. Motion index is the average of the magnitude of acceleration on
164 each of the three axes for each minute. A low motion index corresponds to a low activity level

165 and a high motion index corresponds to a high activity level. Weight and condition scores of
166 ewes were taken at the beginning and end of each seasonal measuring period.

167 Twenty-four ewes were measured in each of the four seasons. The ewes used for this
168 study were drawn from a larger flock and kept as a sub-group but in a restricted area. An
169 additional, seven ewes without collars were also included in the monitoring group to ensure
170 replacement when necessary. As some data were missing, the number of collected data sets
171 differed between seasons. In the winter period twenty-four ewes commenced the study with
172 loggers, however, there was missing logger data for one ewe due to technical failure. There
173 were two mortalities during the winter measurement period, and one mortality a few days after
174 the end of measurements (non-experimental period). Three replacement ewes were used in the
175 spring experimental period. Over the summer period, there were missing logger data from five
176 ewes due to technical failures. Over the entire experiment, data from twenty-nine ewes were
177 used, and fifteen ewes had data from all four seasons.

178 Winter measurements resulted in twenty-three data sets from ewes during mid-pregnancy
179 (12th of January to 10th of February), under natural winter conditions, with sunrise between
180 07:57 – 08:46 h and sunset between 16:08 – 17:09 h. Spring measurements were performed on
181 22th of May to 18th of June, after parturition around the 1st of May. Out of the twenty-four ewes,
182 sixteen were rearing lambs (six with twin and ten with single lambs) and eight ewes were
183 without lambs. Ewes and lambs were under natural spring conditions, with sunrise between
184 04:28 – 04:51 h and sunset between 21:39 – 22:11 h. Summer measurements were collected
185 during late lactation (1st to 28th of August). Data were successfully collected from nineteen
186 ewes, of which twelve were rearing lambs (six ewes with twin and six with single lambs). Ewes
187 and lambs were kept under natural summer conditions, with sunrise between 05:20 – 06:14 h
188 and sunset between 20:26 – 21:30 h. Lamb weaning took place at the end of the summer period.
189 Autumn measurements were collected one week after weaning and before mating (5th to 30th of
190 October). Twenty-four data sets were collected from ewes in autumn, with sunrise between
191 07:30 – 08:22 h and sunset between 17:42 – 18:45 h. At the beginning and at the end of each
192 experimental period the ewes were weighed in order to evaluate Body Weight Gain (BWG).

193 2.2. *Data analysis*

194 2.2.1. *Activity circadian and ultradian rhythms*

195 DFC is a parameter, developed by Scheibe et al. (1999), used here to measure the
196 synchrony between circadian rhythm of behaviour and circadian rhythm of the environment
197 (external 24 h period). Thus, DFC expresses the percentage of cyclic behaviour that is
198 harmonically synchronized to the 24 h period. Any analysis of cyclical data involves fitting
199 mathematical functions and these are described in sequence.

200 To carry out the DFC analyses, preliminary analysis using different time intervals (e.g. 1,
201 5, 10, 15, 20 and 30-minute intervals) and different day intervals (e.g. 7 and 10 days) were
202 performed, to identify the appropriate interval that best represented the patterns within the
203 motion index data. Fifteen-minute time intervals and seven-day periods of data were the most
204 appropriate and have been used throughout this paper. Therefore, time series with fifteen-minute
205 intervals were created, by taking the motion index sum of the one minute averages, and then
206 using the method of Scheibe et al. (1999), these data were analysed for its rhythmic
207 components.

208 To reduce the noise component, autocorrelation functions were calculated for each seven-
209 day interval, with a shift by one day and overlap of six days. Thus, the first seven-day interval
210 included the first day to the seventh day of measurement; the second seven-day interval
211 included the second day to the eighth day of measurement and so on. A power spectrum was
212 drawn for each seven-day interval from the autocorrelation functions of the measured data. The
213 periodogram ordinates were calculated for all Fourier frequencies: $\omega=2\pi j/n$, $j=1,\dots,q$ with $q=n/2$
214 (n even) or $q=n/2-1$ (n odd), with n being the number of data points in the sample (Berger et al.,
215 1999; Scheibe et al., 1999). The periodogram ordinates were tested for statistical significance
216 according to the R.A. Fisher test for periodicity (Andel, 1984). The significant ordinates
217 represent the significant periodic components (Supplementary Figure 1, a).

218 Harmonic periods are defined as periods which are synchronized with the day length (24
219 h) in relation to an integral number (1, 2, 3... etc.). Thus, 24 h divided by an integer number

220 gives the harmonic periods (the period lengths of 24, 12, 8, 6, 4.8, 4, 3.4, 3, 2.6, etc. hours are
 221 harmonic). An example of a power spectrum and its significant harmonic periods is shown in
 222 Supplementary Figure 1, b. Here, DFC is expressed as the percentage of the circadian
 223 components and harmonic ultradian components in relation to all rhythmic components of the
 224 spectrum. Therefore, DFC demonstrates the relationship between the total intensity of
 225 significant periods that were harmonic to the circadian period (SI_{Harm}) and the total intensity of
 226 all periods that were significant (SI_{Total}) (Berger et al., 1999; Scheibe et al., 1999):

$$227 \quad \text{DFC (\%)} = (SI_{\text{Harm}} * 100) / SI_{\text{Total}}$$

228 Where, $SI_{\text{Harm}} = \Sigma$ of intensities of significant periods that are harmonic to 24 hours
 229 period;

230 $SI_{\text{Total}} = \Sigma$ of intensities of significant periods.

231 DFC (%) varies from 0% to 100%, where 0% means that only non-harmonic periods were
 232 significant and 100% means that only harmonic periods were significant (Berger et al., 2003,
 233 1999). Low DFC indicates lower synchronization and high DFC higher synchronization to the
 234 24-hour period. DFC was continuously calculated for all ewes, during all experimental days.

235 *2.2.2. Statistical analysis of differences between seasons*

236 The averages of BWG, motion index, DFC, standard deviation (STD) of DFC and STD of
 237 motion index for each season were calculated. Data were checked for normal distribution using
 238 the PROC UNIVARIATE statement of SAS 9.3. The DFC and STD of DFC averages for
 239 seasons were not normally distributed; hence, angular transformations were used. The motion
 240 index and STD of motion index averages for seasons were also not normally distributed; thus,
 241 square root transformations were used. To assist with interpretation of biological meaning,
 242 back-transformed means are presented in the results section. The effect of season was analysed
 243 using the MIXED Procedure of SAS 9.3, with season as repeated measure and ewe as random
 244 effect. The *P* values of the differences of least square means were adjusted for Tukey-Kramer
 245 and the significance level considered was 5%.

246 The general activity patterns of ewes during a 24-hour period in each season were
247 calculated, by taking the overall average of motion index for all ewes for every minute. The
248 threshold to consider the motion index per minute as an activity bout was the value of 1. As
249 there were large differences between seasons in the 24-hour general activity patterns and DFCs
250 averages, the subsequent analyses were done for each of the seasons separately.

251 *2.2.3. Within seasons statistical analyses*

252 *2.2.3.1. Overall links between weather variables, general activity and circadian rhythm* 253 *of activity*

254 Regressions between overall daily averages (of all ewes) of motion index and its STD in
255 relation to daily weather variables were calculated for each season; however, only significant
256 and relevant results are shown in the results section. Each DFC for activity was calculated
257 within moving seven-day intervals, thus, to allow juxtaposition, the moving averages of weather
258 variables were also set up in seven-day intervals. Regressions between overall averages (of all
259 ewes) of DFC and its STD in relation to the moving averages of weather variables were
260 calculated for each season. Only where significant relationships occurred are these described or
261 shown in tables and figures. The regressions were modelled in SAS 9.3 using the REG
262 procedure.

263 *2.2.3.2. Differences between-individual and population response*

264 Using the analytical framework published by Nussey et al. (2007), random regression
265 models were used to assess between-individual variation (random effects) and evaluate the
266 population responses (fixed effects) to environmental parameters. Considering that the
267 population-level response to the environment will depend on individual-level plasticity, the
268 population response was modelled using the data from all animals and then the response from
269 each animal (as a random effect) was compared with the population response and its deviation
270 was calculated. The random regression models were done using the MIXED Procedure with the
271 Restricted Maximum Likelihood (REML) method and COVTEST statement in SAS 9.3. Thus,

272 considering the relationship between trait y (in our case DFC for activity) and environmental
 273 variable e (in our case precipitation and/or temperature), both measured on occasion j , at
 274 individual level y_{ij} , the response of individual I on occasion j was specified as:

$$275 \quad y_{ij} = \underbrace{\mu + \beta e_j}_{\text{Fixed effects}} + \underbrace{p_i + p_{ei}e_j + \varepsilon_{ij}}_{\text{Random effects}}$$

276

277 Where, μ = population mean on e ;

278 β = population mean slope of y on e .

279 p_i = deviation from the population average intercept (response that is independent
 280 of e , representing the individual elevation);

281 p_{ei} = deviation from the population average slope (response that is dependent of
 282 e , representing the individual plasticity).

283 e_j = Environmental variable;

284 ε_{ij} = residual error.

285 In each season, the ewes were divided into three groups based on the random regressions
 286 results: 1) fixed effects (data of all ewes modelling the population intercept and slope for
 287 weather variables and estimating the covariance parameter for intercept and slope(s)). A
 288 significant covariance parameter for the intercept implies that there were differences between
 289 individuals that were independent of the weather variables. A significant covariance parameter
 290 for the slope(s) for weather variables implies that there were differences between individuals in
 291 regard to the weather variables and; 2) random effects (data of all ewes modelling the deviation
 292 of each ewe from the population intercept and slope for weather variables). Thereafter, we will
 293 refer to the term individual phenotypic plasticity (as originally used by Nussey et al. (2007)), to
 294 support our division of individual sheep put into groups based upon the responses of their DFC
 295 data to the weather variables.

296 The three groups were: 1) Medium Consistency: represented by ewes that did not present
 297 a significant deviation from the population intercept and slope(s) ($P > 0.05$); 2) Low

298 Consistency: represented by ewes that negatively deviated from the population intercept or
299 slope(s) ($P < 0.05$); and 3) High Consistency: represented by ewes that positively deviated from
300 the population intercept or slope(s) ($P < 0.05$). The entire population could be classified in this
301 way except one ewe, over the winter period. Data for this ewe, for winter only, deviated from
302 the population intercept and slope, but exceptionally the deviations were in different directions,
303 negatively deviated from the intercept (-11.0; $P < 0.001$) and positively deviated from the slope
304 (0.74; $P < 0.03$). In this case, the higher and most significant deviation was used to determine in
305 which group this animal would be and thus, over the winter period this ewe was included in the
306 Low Consistency group. The best random regression models regarding the ewe responses to the
307 weather were fitted for each season. Over the winter, the ewes were divided into these three
308 groups considering the slope for precipitation (Supplementary Tables 2 and 3); over the spring,
309 considering the slope for temperature (Supplementary Tables 5 and 6); over the summer
310 (Supplementary Tables 8 and 9) and autumn (Supplementary Tables 11 and 12), considering the
311 intercept.

312 After placing the ewes within their groups, the overall averages of BWG, DFC and STD
313 of DFC for each group, in each season, were calculated. Data were checked for normal
314 distribution using the PROC UNIVARIATE statement of SAS 9.3. Over the spring, summer
315 and autumn the DFC and STD of DFC averages for the groups were not normally distributed;
316 hence, angular transformations were used. To assist with interpretation of biological meaning,
317 back-transformed means are presented in the results section. The effect of group and the effect
318 of the presence of lambs (over spring and summer) were analysed using the MIXED Procedure
319 of SAS 9.3 and the P values of the differences of least square means were adjusted for Tukey-
320 Kramer. The significance level considered was 5%.

321 *2.2.3.3. Links between production traits, general activity and circadian rhythm of activity*

322 Regressions between BWG, motion index, STD of motion index, DFC and STD of DFC
323 were calculated for each season, using the averages from all ewes. In addition, after the random
324 regression analyses, the same regressions were done for each group within each season. Only

325 where significant and/or relevant relationships occurred are these described or shown in tables
326 and figures. These regressions were modelled in SAS 9.3 using the REG procedure.

327 **3. Results**

328 Overall averages of BWG, motion index, STD of motion index, DFC% for activity and
329 STD of DFC% for each season are shown in Table 1. The overall average of motion index was
330 lower during the autumn compared with the spring. The DFC% for activity was highest and had
331 the highest consistency (as shown by the STD of DFC%) during the autumn whereas it was
332 lowest, with the lowest consistency during the winter. Over the winter, the ewes faced harsh
333 weather that was linked to a breakdown in the circadian rhythm for activity and lowest BWG
334 compared to the other seasons.

335 The general activity patterns of ewes during 24-hour changed over the year,
336 corresponding to different daylight lengths observed between the seasons. Over the winter
337 period, the day length was short and ewes showed activity bouts during both the daylight and
338 dark phases. The general activity of ewes was higher during the daylight, with several activity
339 peaks. During the dark phase, two resting bouts occurred, interrupted by one shorter activity
340 bout with lower intensity (Figure 1, a). The length of the daylight period was longer during the
341 spring and the general activity of ewes was consistently found only during the daylight with no
342 activity bout during the dark phase. Over the spring period, there were several activity peaks, all
343 during daylight (Figure 1, b). Similarly, the daylight period was very long during the summer
344 and the general activity of ewes was higher during daylight, with several activity peaks and
345 there was no activity bout during the dark phase (Figure 1, c). Over the autumn period, when the
346 daylight began to shorten, ewes presented a pattern similar to the winter period (Figure 1, d).

347 As there were large differences between seasons regarding the general activity patterns
348 and DFC% for activity, the results are displayed for each of the seasons separately. Given that
349 the circadian rhythm of each animal is unique it is important to look at the detailed DFC%

350 response to understand whether it could be linked to production traits of each animal. Only
351 when significant relationships occurred are these described or shown in tables and figures.

352 3.1. Winter

353 The ewes experienced a severe and challenging change in weather as the precipitation
354 level reached up to 39 mm per day. The general activity of ewes expressed as motion index
355 decreased showing a linear response in relation to precipitation (Figure 2a). The STD of the
356 motion index also showed a linear reduction in response to increased precipitation (Figure 2b).
357 However, the r-squares of these relationships were poor. The overall daily average of DFC% for
358 activity presented a strong linear regression with the moving averages for precipitation (Figure
359 2c). Accordingly, the DFC% of the ewes decreased with increasing precipitation, descending to
360 lower values with higher levels of precipitation. In addition, the variation of DFC% increased
361 along with the precipitation increase, followed by a plateau, with a quadratic relationship
362 providing the best relationship, suggesting the beginning of an adaptation pattern (Figure 2d).

363 The DFC% values of most ewes started to drop dramatically from the 13th and 14th days
364 (Figure 3, b and c) when the precipitation level started to increase (Figure 3a). The differences
365 between animals over the winter were mostly in regard to their different slope for precipitation
366 (Supplementary Tables 2 and 3). For ewes in the Medium Consistency group (did not deviate
367 from the population intercept and slope for precipitation) the DFC% continued to decrease until
368 the 17th day and from this day started to show an adaptation pattern represented by increasing of
369 DFC% levels, even with high levels of precipitation. Ewes in the High Consistency group
370 presented a reduced variation of the DFC% response. In the Low Consistency group, ewes
371 showed a greater decrease in the DFC% response; thereafter, the DFC% level of two ewes
372 started to rise, but the ewe W134 did not show a recovering DFC% and died on the 20th day.
373 Ewe G58 in the Low Consistency group showed deviation from the population slope and
374 intercept. The positive deviation of the slope for precipitation showed that this ewe had a good
375 capacity to cope with the precipitation level, however, it had a high negative deviation from the
376 population intercept. Overall averages within groups are shown in Table 2.

377 Over the winter period, the population, in general, showed reductions in the DFC%
378 response, as well as low ranges of BWG. However, there were no significant regressions
379 between BWG and DFC% for activity (Supplementary Table 4). Considering the entire
380 population (all ewes), there was no significant regression between BWG and motion index.

381 3.2. *Spring*

382 The between-individual variation of the DFC% response was well explained by the
383 weather variables (Figure 4 and detailed statistical results in Supplementary Tables 5 and 6).
384 Three ewes deviated from the population quadratic slope for precipitation and nine ewes
385 deviated from the population slope for temperature, suggesting that temperature was the main
386 effect for between-individual variation in the DFC% responses.

387 Regressions between BWG, DFC%, STD of DFC% and the presence and number of
388 lambs are shown in Supplementary Table 7. Considering the entire population (all ewes), there
389 was a significant negative multiple regression for BWG with STD of DFC% and the number of
390 lambs; and a negative linear regression between STD of DFC% and BWG (Figure 5a). To gain
391 a better understanding of this relationship and study the between-individual differences in the
392 DFC% responses, linear regressions are also displayed within groups. The BWG of ewes in the
393 Medium Consistency group was linked to lamb effect, with BWG decreasing with the
394 increasing number of lambs. The BWG of ewes in the Low Consistency group was linked to the
395 variation in the DFC% response, as there was a strong negative relationship between BWG and
396 STD of DFC%, with BWG decreasing with the increasing STD of DFC% (Figure 5b). In
397 addition, the number of lambs was not related to the high variation in the DFC% response
398 shown by ewes in the Low Consistency group. Within the High Consistency group, the
399 between-individual variation in the DFC% and its STD seems to be linked to the presence and
400 number of lambs (Supplementary Figure 2). Considering the entire population (all ewes), there
401 was no significant regression between BWG and motion index.

402 3.3. *Summer*

403 The regressions between the overall daily averages of DFC% for activity parameters and
404 weather variables are shown in Supplementary Figure 3 and 4. Besides, these relations between
405 the population DFC% and weather variables, the differences between-individual in the DFC%
406 responses were regarding unknown factors that were independent of weather (Figure 6 and
407 detailed statistical results in Supplementary Tables 8 and 9).

408 DFC% for activity was related to ewe performance; the regressions between BWG,
409 number of lambs, DFC% and STD of DFC% considering all ewes and within groups are
410 displayed in Supplementary Table 10. Considering all ewes, there was a negative linear
411 relationship between BWG and STD of DFC%, indicating that ewes with lower variation in the
412 DFC% response showed higher BWG (Figure 7a). The same negative linear relationship was
413 present within the Medium Consistency group, suggesting that there is a close relationship
414 between BWG and STD of DFC% (Figure 7b). In addition, considering all ewes, the DFC% and
415 STD of DFC% were well explained by the BWG and number of lambs. Regarding the Low
416 Consistency group, just two ewes negatively deviated over the summer period. Therefore, it was
417 not possible to conduct a regression analysis, but it is relevant to mention that the ewe OR227,
418 with the highest breakdown in activity rhythmicity, had the lowest BWG of the flock (-0.114
419 kg/day). Considering the entire population (all ewes), there was no significant regression
420 between BWG and the motion index.

421 3.4. Autumn

422 The overall daily averages of DFC% for activity parameters showed a quadratic
423 regression with the moving average of temperature (Supplementary Figure 5). Likewise in the
424 summer period, over the autumn there were no differences between individual responses with
425 temperature; and these differences between individuals were regarding an unknown factor that
426 occurred within each animal (Figure 8 and detailed statistical results in Supplementary Tables
427 11 and 12). The ewe OR227 in the Low Consistency group repeatedly showed a high
428 breakdown in the DFC% response, reaching low values. During the autumn period, there were
429 no significant regressions between BWG and DFC% for activity (Supplementary Table 13).

430 Considering the entire population (all ewes), there were no significant regression between BWG
431 and the motion index.

432 **4. Discussion**

433 The general activity patterns of ewes during 24-hours were different between seasons and
434 this variation is believed to be mainly in response to the daylight length. Diurnal animals exhibit
435 higher activity during daylight and the activity pattern is often related to feeding, especially for
436 herbivorous animals (Bloch et al., 2013; Piccione et al., 2010; Umstätter et al., 2008). Umstätter
437 et al. (2008) observed that extensively managed ewes in Scotland were active for over 60.5% of
438 the daytime and spent 59.9% of the active time grazing and only 0.6% walking, without grazing.
439 Thus, Umstätter et al. (2008) stated that based on activity data it is possible to define two
440 behaviour categories, active (mainly grazing) and inactive (mainly recumbent).

441 Our study recorded additional activity after midnight with decreasing day length in
442 autumn and winter. Langbein et al. (1996) also registered high proportions of nocturnal activity
443 with decreasing day length in autumn and Champion et al. (1994) observed an eating bout over
444 midnight. Considering that most activity behaviour is linked with feeding, it is reasonable to
445 presume that ewes in our study spent time grazing overnight during the autumn and winter
446 period. Longer periods of daylight, as observed during the spring and summer, are believed to
447 allow adequate time for grazing. Accordingly, days with shorter daylight lengths, such as
448 observed over the winter and autumn recording periods, were probably not enough to allow
449 sufficient grazing, and associated rumination time, to satisfy nutritional needs and, thus, some
450 grazing overnight was required.

451 Motion index data is thus important to understand the daily oscillation of activity
452 behaviour and its variation between seasons, however, our results showed that activity
453 behaviour alone had very poor or non-significant regression with the weather variables. In
454 addition, we further analysed the motion index data using random regression models to
455 understand the differences between animals regarding activity behaviour in response to the

456 weather variables. However, the weather had little or no effect on the between-individual
457 differences and over all seasons these differences were regarding the individual reaction norm
458 elevation (Nussey et al., 2007). Our results showed that examining data in a different way, by
459 analysing the circadian rhythm through the calculation of DFC% for activity and its variation
460 (STD of DFC%) enables a better understanding of animal responses with respect to the weather
461 influences compared to using motion index data alone. In addition, the random regression model
462 method was effective in identifying animals whose DFC% deviated from population responses.
463 Considering the new proposal of this study, a deviation from the DFC% intercept is a way of
464 measuring a temporary distress within the individual. Further, this methodology also linked this
465 disturbance to weather events and changes, when some animals deviated from the population
466 slope for weather variables.

467 In this study, the weather variables influenced the activity rhythmicity of ewes over all
468 seasons. But, the links between weather and rhythmicity were different between seasons. Over
469 the winter period, the largest decrease in the DFC% response of ewes was linked with high
470 precipitation level, during which weather events there was also lower temperatures. The
471 quadratic response between DFC% in relation to moving average of daily precipitation over the
472 spring period suggest that sheep were less affected by rain when the temperatures were higher in
473 spring. Our result is in line with Warren and Mysterud (1991) who studied sheep on a
474 coniferous forest range in southern Norway and observed that sheep activity patterns were
475 rhythmic and consistent throughout the summer season, although were affected by both weather
476 conditions and day length. These authors also observed that cold ($< 10^{\circ}\text{C}$), wet weather or foggy
477 conditions reduced the overall activity of the flock. Other studies of extensively managed sheep
478 have shown that they reduce grazing during heavy winter rainfall, but are less affected by rain in
479 the summer months (Champion et al., 1994; Hunter, 1995).

480 Abrupt environmental changes, such as periods of high precipitation levels with low
481 temperatures faced by ewes over the winter, may have a direct effect on the activity rhythmicity.
482 This lack of rhythmicity may persist until the end of the environmental disturbance, the animal

483 adaptation or its failure to adapt adequately. The animal failure may result in poor welfare and
484 potentially death. However, the degree of these negative effects can clearly differ between
485 individuals and we postulate that this difference is an outcome of the animal's ability to cope
486 with short-term environmental change, its individual phenotypic plasticity (deviation from the
487 population slope for the weather variable). Analysing the individual DFC response combined
488 with the use of random regression models to classify the differences between each animal's
489 ways of coping with the challenging environment was a novel approach of our study. We
490 believe this has proved to be successful.

491 Knowledge of individual plasticity is important because the population level response to
492 the environment will depend on individual levels of phenotypic plasticity (Dingemanse et al.,
493 2010; Nussey et al., 2007; Wilson, 1998). We found weather variables influenced ewe DFCs for
494 activity over all seasons. However, only during the winter and spring was there the association
495 between the weather and the variation between animals. During the summer and autumn
496 periods, between-individual variation in DFC% data was not related to temperature or
497 precipitation. These findings suggest that the inconsistent rhythmicity of ewes in the Low
498 Consistency group over the summer and autumn periods were linked with others factors that we
499 did not measure. However, it can be assumed that these were particular factors within these
500 animals.

501 When animals are experiencing an environmental disturbance, we typically found the
502 DFC% for activity is low. Over the winter period, all ewes had breakdowns in the circadian
503 rhythm of activity, even ewes in the High Consistency group. However, the ewes in the High
504 Consistency group showed less variation of the DFC% response, evidencing that their plastic
505 response was positive and enabled them to cope better with higher levels of precipitation and
506 consequently showing a faster adaptation. On the other hand, ewes in the Low Consistency
507 group showed a stronger decrease in DFC% reaching values lower than 30%, as well as a more
508 difficult recovery of the DFC% response, suggesting an adaptation problem. Some of these
509 ewes had a maladaptive plasticity, with a greater lack of circadian rhythmicity and an indication

510 of poorer welfare, and consequently a great risk of poor health and death. A lack of circadian
511 rhythmicity caused by an environmental change might be temporary and not affect animal
512 health, however, it also might be harmful, as the long-term circadian rhythm disorganization
513 acts as stress factor that may have consequences to animal health. Dwyer and Bornett (2004)
514 noted that chronic stress induces alterations in behaviour and its circadian rhythms, particularly
515 for activity and feeding. These authors also observed that chronic stress reduce immune
516 function. Martino et al. (2008) have noted that circadian organization is critical for normal
517 health and longevity, whereas circadian rhythm disorganization produces profound
518 cardiovascular and renal disease.

519 The winter recording period had the highest level of nocturnal activity after midnight, the
520 lowest overall DFC%, the greatest overall variation through STD of DFC%, and a clear
521 reduction in DFC% linked to high rainfall and cold temperature. The Low Consistency group
522 during this period had a high level of mortality. Ewe mortality and welfare are seen as industry-
523 wide issues during this period (Morgan-Davies et al., 2008). The highest level of nocturnal
524 activity after midnight over the winter was probably an attempt to obtain heat rewards, by
525 feeding. Shorter daylight lengths were probably not enough to allow sufficient grazing, and
526 associated rumination time, to satisfy nutritional needs. When energy intake is restricted,
527 metabolic rate is correspondingly reduced to correct for the energy imbalance, a process called
528 diet-induced thermogenesis (Stock, 1999). The reduction in metabolic rate is accompanied by a
529 lowering of body temperature and this is observed primarily during the inactive phase (Stock,
530 1999). Thus, while exposed to a cold environment, animals would try to obtain heat rewards, by
531 feeding overnight, to suppress lowering of body temperature attempting to regulate body
532 temperature at the normothermic level (Piccione et al., 2002). In addition, the energy costs of
533 cold exposure must eventually be reflected in poor animal performance (Slee, 1971), as
534 observed in this study over the winter period. However, this does not demonstrate the ewes were
535 in thermal distress but it may be an attempt to prevent it.

536 In a review, Dwyer and Bornett (2004) state that in hot weather, sheep adjust their diurnal
537 patterns to the coolest times of the day, but when temperatures are low, wind speed is high
538 and/or it is raining, sheep make use of shelter; a mechanism that help animals to deal with
539 thermal extremes and decrease thermoregulation risk. However, these authors note that cold-
540 exposed sheep, but lacking in shelter, may experience thermal distress. According to Nikkhah
541 (2012), eating time is a major external cue and a feasible life strategy that affects production and
542 health physiology. In the Low Consistency group, over the winter, ewes showed a greater
543 decrease in the DFC% response. Thereafter, the DFC% level of two ewes started to rise but ewe
544 W134 did not show a recovering DFC% and died on the 20th day. DFC% combined with
545 random regression model appeared to predict the death of this ewe and showed the link with the
546 harsh weather. Ewe G58 in the Low Consistency group showed deviation from the population
547 slope and intercept. The positive deviation of the slope for precipitation showed that this ewe
548 had a good capacity to cope with the precipitation level, however, it had a high negative
549 deviation from the population intercept. DFC% combined with random regression model
550 detected that its death was linked to a factor that was independent of the weather and occurred
551 within this animal. The death of this ewe was regarded as an accident as it caught its leg in the
552 feeding ring. Ewe B252 in the Low Consistency group also died a few days after the end of
553 measurements. Looking at the ewe mortality in the Low Consistency group, three of four ewes
554 in this group died over the winter period, while there were no deaths for the other 19 ewes in the
555 Medium and High Consistency groups. The probability of the actual distribution of
556 deaths/survivals amongst Low and combined Medium/High Consistency groups is $P = 0.002$.
557 Thus, the probability of death in the Low Consistency group is higher than by chance alone
558 further indicating that this approach may have a value for monitoring livestock in extensive
559 systems. The flock from which this study flock was taken had a study conducted in Morgan-
560 Davies et al. (2008) when the annual ewe mortality was reported as 8.2%, similar to that found
561 in our study group. These levels of mortality highlight the need for improved husbandry, where
562 new technology may play a part.

563 Berger et al. (1999) observed similar results to those in our study, in research with
564 Przewalski horse. Przewalski horses showed lowered DFCs in periods of adaptation after
565 translocation from a zoo to a semi-reserve. Lowered DFCs of Przewalski horse were also
566 observed during the period of hunting and shooting in the surrounding area of the semi-reserve
567 (Scheibe et al., 1999). These findings show that not only weather variables can cause circadian
568 rhythm disorganization, but also other stressful environmental disturbances. Thus, other
569 environmental data sets with daily observations could potentially be used to evaluate the
570 differences between individual rhythmicity and the way that animals cope with their
571 environment. Moreover, health problems, injury, immobilization, social interactions and even
572 some normal physiological changes such as parturition may also be the cause of a lack of
573 behavioural circadian rhythmicity (Berger et al., 2003, 1999; Scheibe et al., 1999, 1998). Our
574 study has confirmed the potential value of the chronobiological approach to analysing activity
575 behaviour.

576 The relationship between BWG and the variation of DFC% for activity was noteworthy
577 over the spring and summer periods. Over the spring period, the BWG of the flock was well
578 explained by a combination of presence and number of lambs and by the variation of DFC% for
579 activity. However, clustering ewes into groups allowed a better understanding of these
580 relationships. The BWG of the ewes in the Medium Consistency group showed a significant
581 regression with the number of lambs, with an r-square similar to the entire flock regression.
582 Thus, the influence of the lambs for the entire flock was mostly driven by the ewes in the
583 Medium Consistency group. It can be assumed that the differences between-individual BWG for
584 the Medium Consistency group was related to the metabolic rate of the ewes. The spring period
585 measurements occurred soon after parturition and ewes with lambs were in early lactation, and
586 under higher metabolic demand for milk production, and thus showing lower BWG. Likewise,
587 the influence of the variation of the DFC% for the entire flock was mostly driven by the ewes in
588 the Low Consistency group. The BWG of the ewes in the Low Consistency group showed a
589 significant regression with the STD of DFC%, with a higher r-square than the one found for the

590 regression considering the entire flock. Thus, the differences between-individual BWG for the
591 Low Consistency group was related only to the variation of the DFC%.

592 During the summer period, there were no significant regressions between BWG and
593 number and presence of lambs. Ewes with lambs were in late lactation, with reduced milk yield
594 and then BWG will be less directly influenced by the presence and number of lambs.
595 Nevertheless, BWG was associated with the variation of DFC% for activity. Ewes with a lower
596 variation of DFC% showed higher BWG. Regarding the Low Consistency group, just two ewes
597 negatively deviated over the summer period. Therefore, it was not possible to display a
598 regression analysis, but it is important to mention that the ewe OR227, with the highest
599 breakdown in activity rhythmicity, showed the lowest BWG of the flock (-0.114 kg/day).
600 Autumn measurements were made after weaning, a period typical of rapid weight gain with no
601 reproduction demand and it is clear to see that almost all ewes were consistent in the DFC%
602 response. Considering that motion index had no significant regression with BWG or presence of
603 lambs, the variation in the DFC% response was a better estimator of BWG than the use of
604 simple motion index.

605 Analysing the DFC% for activity and the differences between individuals using the
606 random regression model enabled the detection not only of environmental disturbances but
607 showed that it is also possible to detect animals with particular issues. In addition, clustering
608 ewes into groups using random regression models allowed a better understanding of the
609 relationships between BWG, the presence of lambs and STD of DFC% over the spring period.
610 The new framework proposed by this study, combining DFC analyses and random regression
611 model could be used in practice to understand whether the differences between animals were in
612 response to the environment or whether the difference was shown in relation to a particular
613 problem by a particular animal linked to animal performance. Further, the measurement of
614 individual DFCs may have potential to help the genetic selection of healthy and adapted animals
615 and to provide a monitoring tool for health and welfare issues.

616 The current study was conducted with raw data that was created and saved on a memory
617 card, and post-processed. With further technological developments, animal-based sensors may
618 increasingly be part of a real-time decision support system for researchers, veterinarians or
619 farmers with circadian rhythm analysis providing a different perspective to that from the only
620 short-term, real-time visualisation of data. Additionally, studies of circadian rhythmicity could
621 be a useful contributor to the better understanding of biology and to evaluate animal husbandry
622 systems considering aspects of animal welfare.

623 **5. Conclusions**

624 This study confirmed the strong dynamic created by the seasons and by the
625 production/physiological cycle in sheep in high latitude systems. The analysis of the circadian
626 rhythm of activity using the DFC-parameter enables a better understanding of sheep responses
627 to weather and environmental influences, compared to the use of a simple quantitative activity
628 parameter, such as motion index. The random regression model method was effective in
629 identifying animals that deviated from population responses regarding the weather influence or
630 to a particular problem. Over the spring and summer periods, the variation in the DFC response
631 was a better estimator of BWG than the use of a simple motion index. Clustering ewes into
632 groups using random regression models allowed a better understanding of the relationships
633 between BWG, the presence of lambs and STD of DFC% over the spring period.

634 The combination of circadian rhythm analysis and the clustering of individuals into
635 groups based on their regression response to environmental variables provides a considerable
636 potential to glean information relevant to group and individual animal management. With
637 increasing availability of such data captured through automated telemetric systems, this work
638 shows that these approaches may enhance the quality and meaningfulness of data coming from
639 automated sensors.

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740 *Endocrinol.* 222, R39-59. doi:10.1530/JOE-14-0141
- 741

742 **Table 1** Body Weight Gain (BWG), Motion Index (MI), Degree of Functional Coupling (DFC)
 743 for activity, MI and DFC standard deviation (STD) for each season.

	Seasons				SEM ²	P ³
	Winter mid- pregnancy	Spring ¹ after parturition	Summer ¹ late lactation	Autumn after weaning		
BWG, kg/day	0.05 ^b	0.18 ^a	0.20 ^a	0.20 ^a	0.03	< 0.001
MI	3895 ^{ab}	4596 ^a	3422 ^{ab}	2907 ^b	374	< 0.01
STD of MI	1866 ^a	1122 ^b	818 ^{bc}	564 ^c	224	< .0001
DFC, %	81.66 ^c	88.06 ^{bc}	90.22 ^{ab}	95.38 ^a	1.65	< .0001
STD of DFC, %	17.39 ^a	9.77 ^b	8.90 ^{bc}	5.39 ^c	1.28	< .0001

744 ¹ Fixed effect of lamb was not significant ($P > 0.05$). ² Maximum standard error of the mean. ³ P value for
 745 the fixed effect of season. ^{a,b,c} Means with different superscripts along the same line are significantly
 746 different with $P < 0.05$ for differences of least squares means.

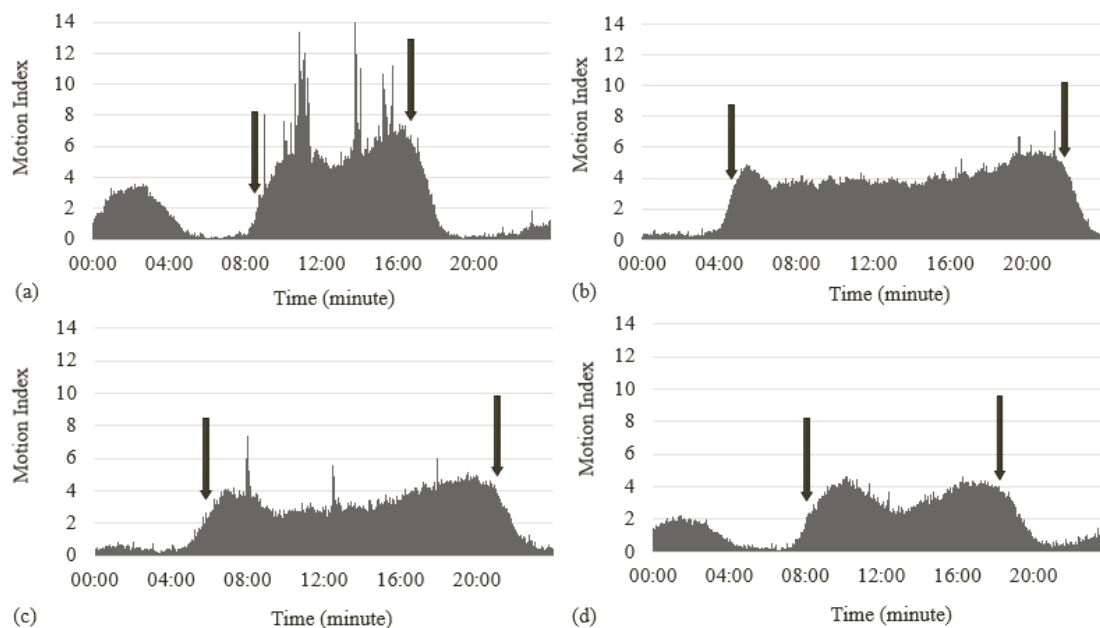
747

748 **Table 2** Overall averages of Degree of Functional Coupling (DFC) for activity, DFC standard
 749 deviation (STD) and Body Weight Gain (BWG) for each group within the seasons.

	Group			SEM ¹	P ²
	High	Medium	Low		
	Consistency	Consistency	Consistency		
Winter					
DFC%	93.1 ^a	82.7 ^b	66.5 ^c	3.32	< .0001
STD of DFC%	8.6 ^c	16.4 ^b	30.1 ^a	2.65	< 0.001
BWG, kg	0	0.08	-0.07	0.08	0.14
Spring³					
DFC%	96.42 ^a	90.35 ^b	76.98 ^c	2.09	< .0001
STD of DFC%	5.69 ^b	8.43 ^b	14.29 ^a	1.49	< 0.001
BWG, kg	0.246	0.223	0.111	0.052	0.13
Summer³					
DFC%	97.65 ^a	91.87 ^b	69.30 ^c	2.98	< .0001
STD of DFC%	4.81 ^c	8.50 ^b	22.00 ^a	2.82	< 0.01
BWG, kg	0.146	0.218	0.086	0.072	0.16
Autumn					
DFC%	99.84 ^a	96.52 ^b	87.56 ^c	0.78	< .0001
STD of DFC%	0.72 ^c	4.89 ^b	11.75 ^a	1.73	< .0001
BWG, kg	0.099	0.221	0.167	0.089	0.43

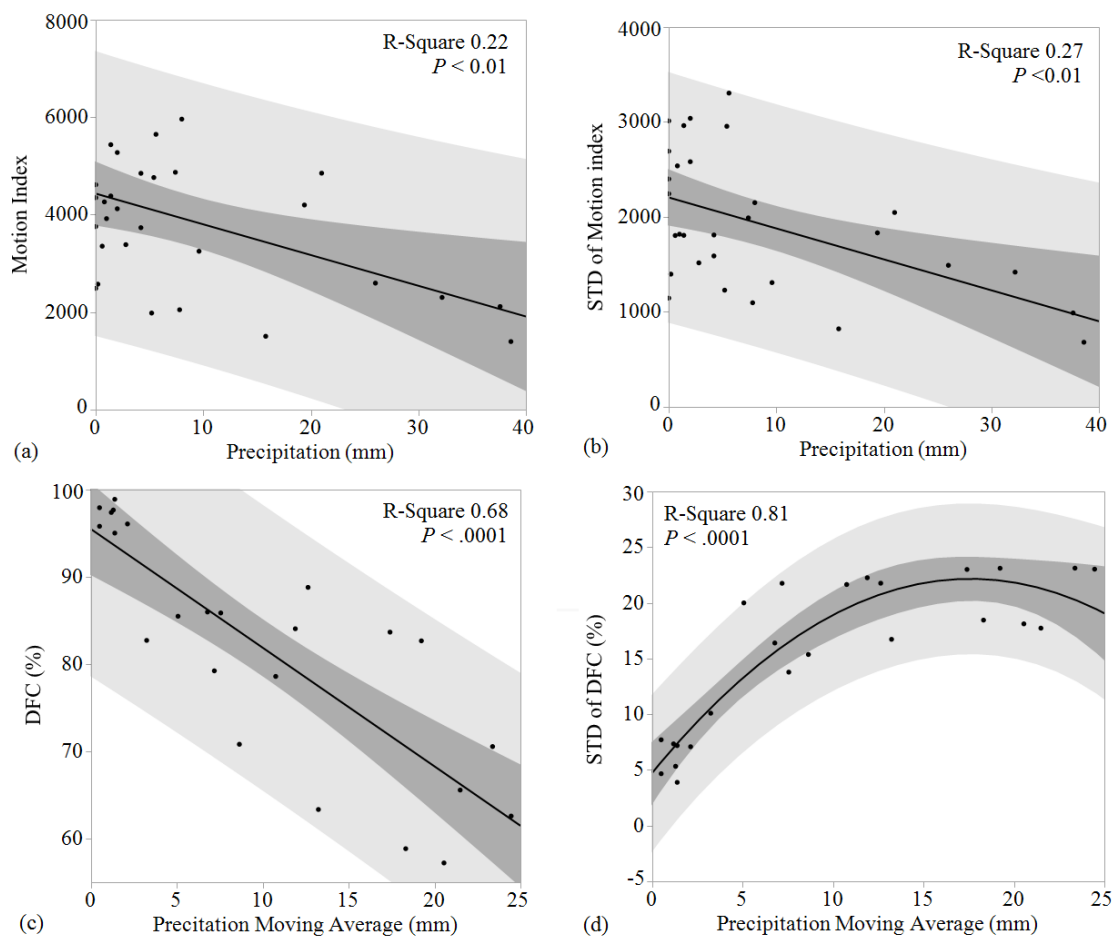
750 ¹Maximum standard error of the mean. ²P value for the fixed effect of season. ³Fixed effect of lamb was
 751 not significant ($P > 0.05$). ^{a,b,c} Means with different superscripts along the same line are significantly
 752 different with $P < 0.05$ for differences of least squares means.

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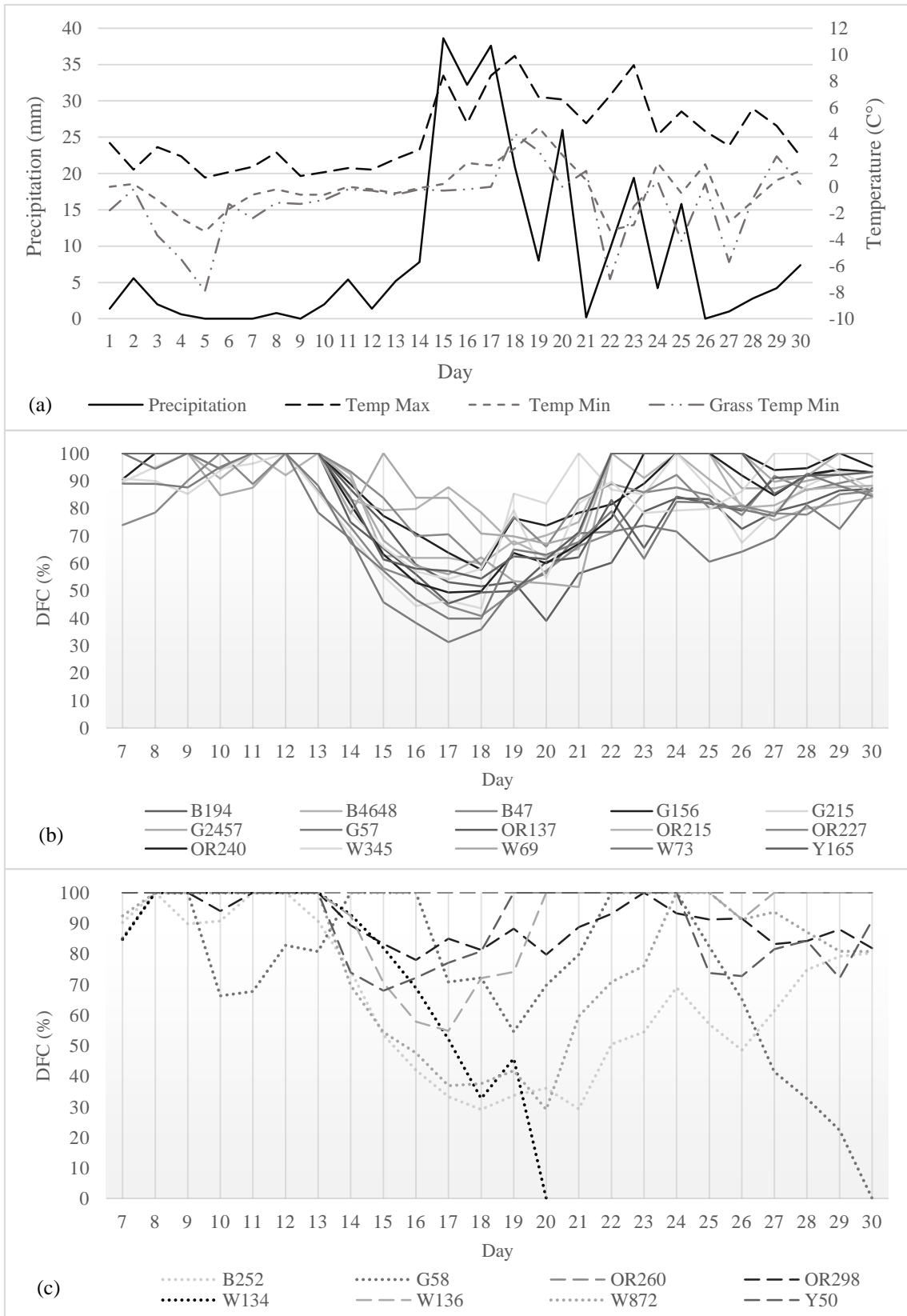
754

755 **Figure 1.** General activity patterns of ewes during 24-hour over the seasons. One bar per minute and
 756 every minute value is an overall average for motion index of all ewes. A low motion index corresponds to
 757 a low activity level and a high motion index corresponds to a high activity level. Arrows show average
 758 time of sunrise and sunset. (a) Winter period: Jan/Feb, 30 days, 23 ewes, sunrise was between 07:57 –
 759 08:46 h and sunset between 16:08 – 17:09 h. (b) Spring period: May/Jun, 28 days, 24 ewes, sunrise was
 760 between 04:28 - 04:51 h and sunset between 21:39 - 22:11 h. (c) Summer period: August, 28 days, 19
 761 ewes, sunrise was between 05:20 - 06:14 h and sunset between 20:26 - 21:30 h. (d) Autumn period:
 762 October, 26 days, 24 ewes, sunrise was between 07:30 - 08:22 h and sunset between 17:42 - 18:45 h.



763

764 **Figure 2.** General activity (motion index) and Degree of Functional Coupling (DFC%) for the activity of
 765 ewes in response to precipitation level over the winter period. The dark grey area represents 95%
 766 confidence limits and the light grey area represents 95% prediction limits. (a) Linear regression between
 767 the overall daily average of motion index and daily precipitation. (b) Linear regression between the
 768 overall daily average of standard deviation (STD) of motion index and daily precipitation. (c) Linear
 769 regression between DFC% for activity and moving averages for precipitation (DFC% was calculated
 770 within moving seven-day intervals, thus, to allow juxtaposition, the moving average for precipitation was
 771 also set up in seven-day intervals. Each DFC% value is an overall average of all ewes). (d) Quadratic
 772 regression between the standard deviation of DFC% for activity and moving average for precipitation.



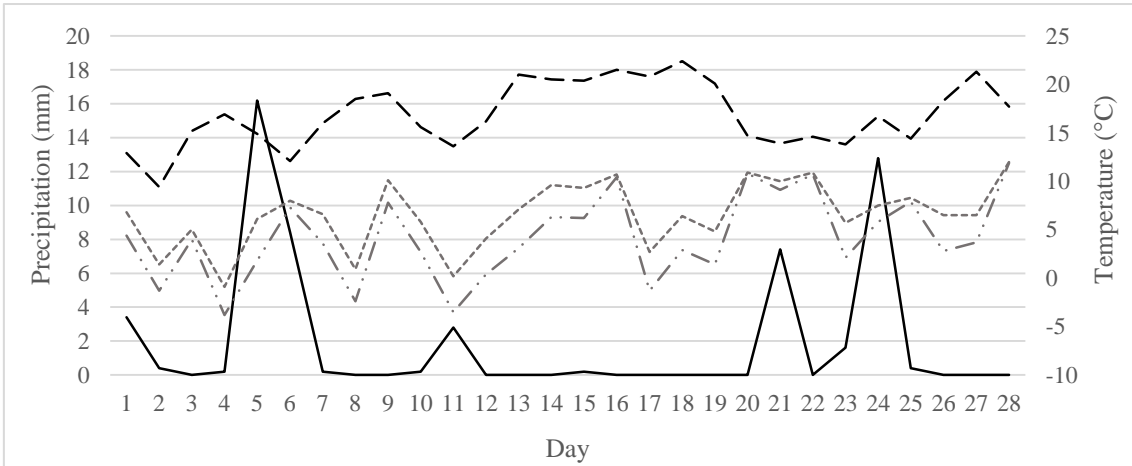
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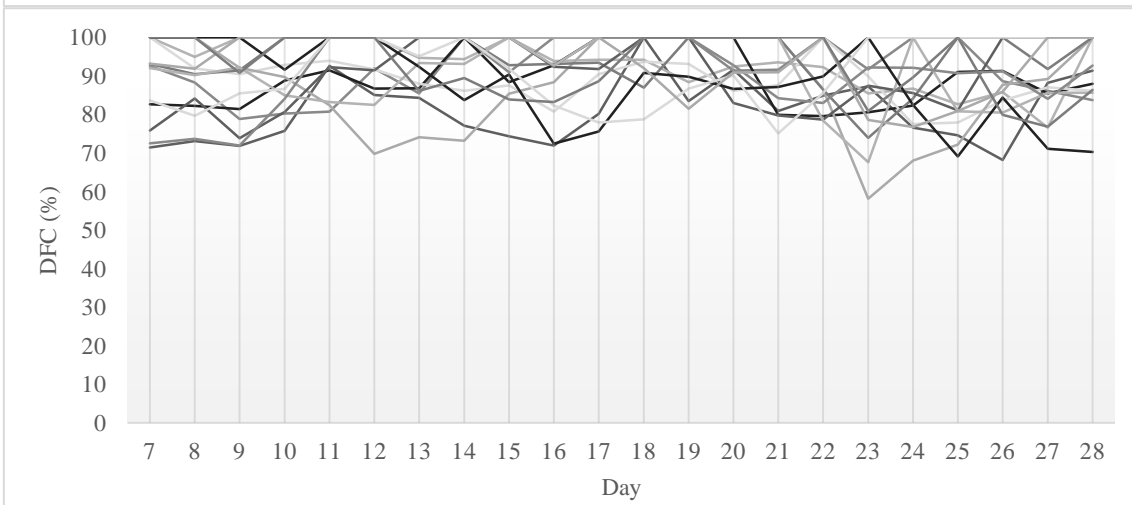
776 **Figure 3.** Degrees of Functional Coupling (DFCs) for the activity of ewes over the winter period. DFC%
 777 was calculated for moving seven-day intervals, thus, the graphic starts at the 7th day. (a) Absolute values
 778 for weather variables over the winter period. (b) DFC% response of 15 ewes that did not present deviation

779 from the population intercept and slope over the winter period (Medium Consistency group). DFC% of
780 these ewes declined until the 17th day and from this day started to show an adaptation pattern represented
781 by an increase of DFC% levels, even with high levels of precipitation. (c) DFC% response of eight ewes
782 that presented deviation from the population slope over the winter period. The four ewes represented by
783 dashed lines showed a reduced decrease of DFC% (High Consistency group) whereas the four ewes
784 represented by dotted lines showed a greater decrease of DFC% (Low Consistency group). The ewes
785 W134 and G58 died on 20th and 30th, respectively. The ewe G58 also showed deviation from the
786 population intercept.



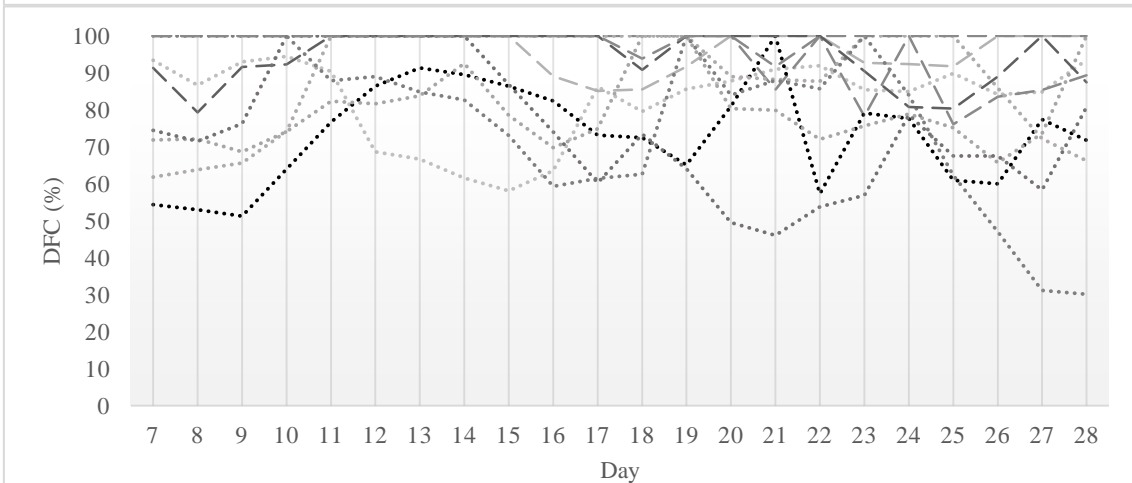
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(a) — Precipitation - - - Temp Max Temp Min - · - · - Grass Temp Min



788

(b) — B164 — B4648 — B47 — G215 — G2456 — G30 — G57
 — OR137 — OR215 — OR240 — W136 — W73 — W872 — Y50



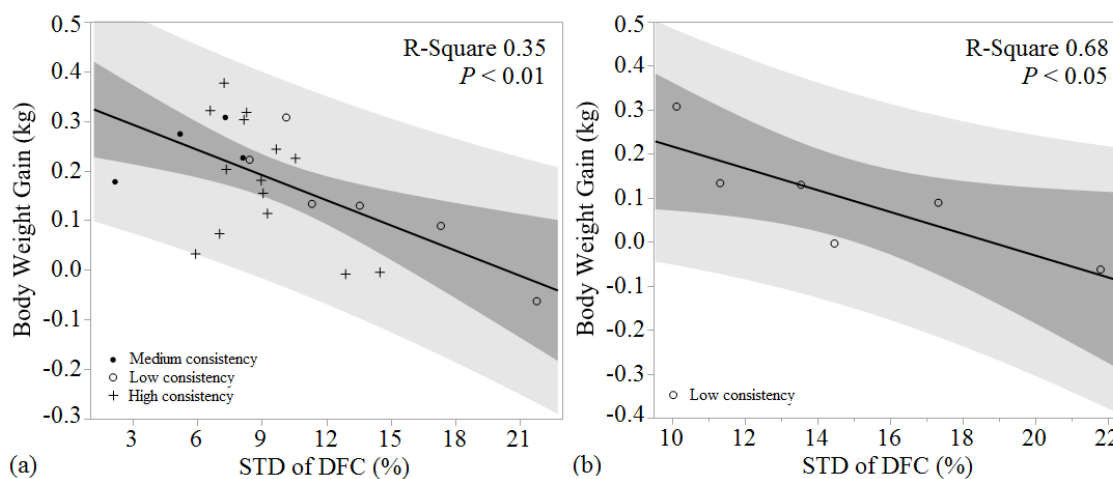
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(c) B360 - - - G172 - - - G2457 OR227 OR260
 OR298 W300 - - - W345 W69 - - - Y165

790 **Figure 4.** Degrees of Functional Coupling (DFCs) for the activity of ewes over the spring period. DFC%
 791 was calculated for moving seven-day intervals, thus, the graphic starts at the 7th day. (a) Absolute daily
 792 values for weather variables over the spring period. (b) DFC% response of 14 ewes that did not present

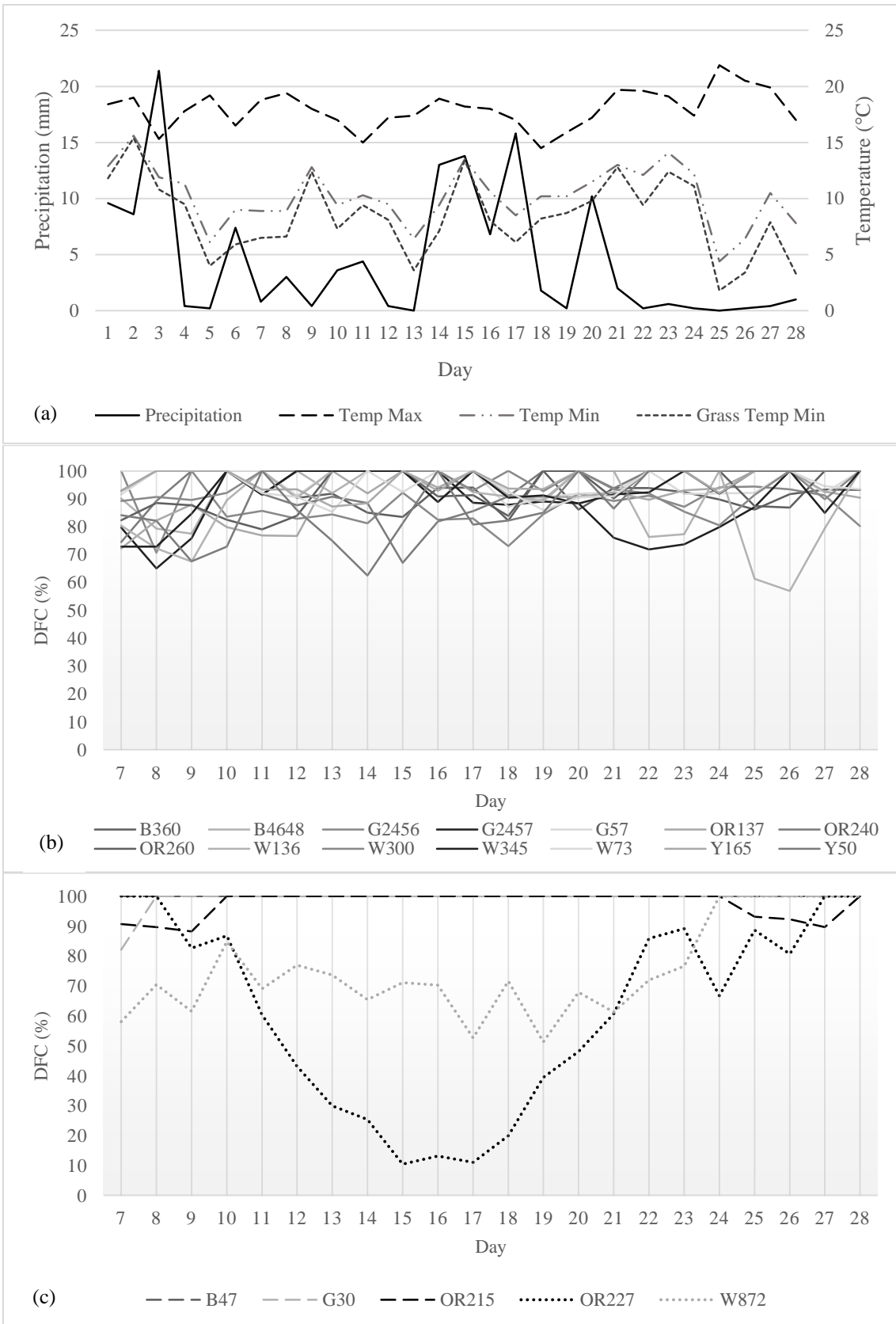
793 deviation from the population intercept and slopes over the spring period (Medium Consistency group).
 794 (c) DFC% response of 10 ewes that presented deviation from the population temperature or squared
 795 precipitation slope over the spring period. Four ewes represented by dashed lines presented higher
 796 consistency of DFC% (High Consistency group), whereas six ewes represented by dotted lines showed
 797 the lower consistency of DFC% (Low Consistency group). Ewe OR260 showed deviation from the
 798 population slope for squared precipitation. Ewes B360 and OR227 showed deviation from both
 799 temperature and squared precipitation slopes. The others ewes showed deviation from the population
 800 slope for temperature.

801



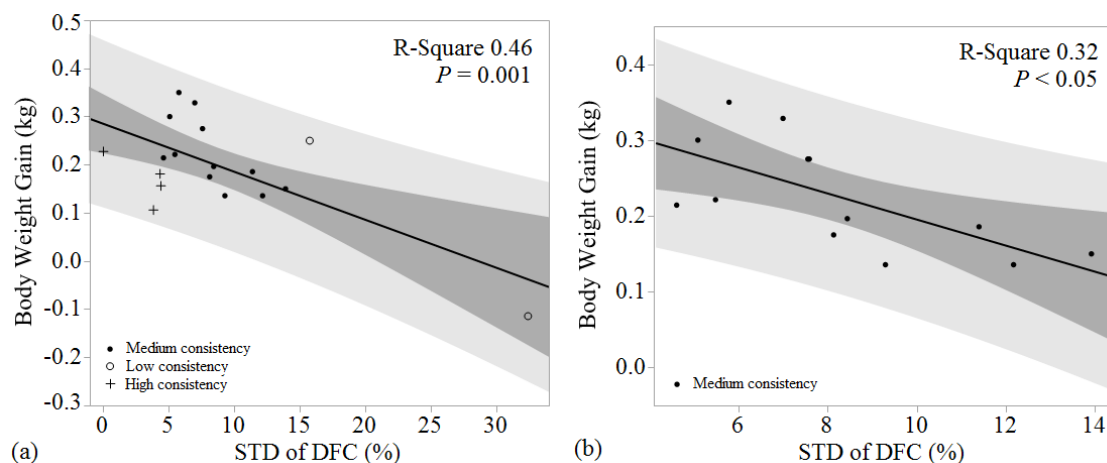
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803 **Figure 5.** Body weight gain (BWG) in relation to the variation of the Degrees of Functional Coupling
 804 (DFC) for activity, over the spring period. The dark grey area represents 95% of confidence limits and the
 805 light grey area represents 95% of prediction limits. Each DFC% value is an overall average for each ewe,
 806 over the spring period. (a) Linear regression for BWG in relation to the standard deviation (STD) of
 807 DFC% for all ewes. (b) Linear regression for BWG in relation to STD of DFC% for the Low Consistency
 808 group.

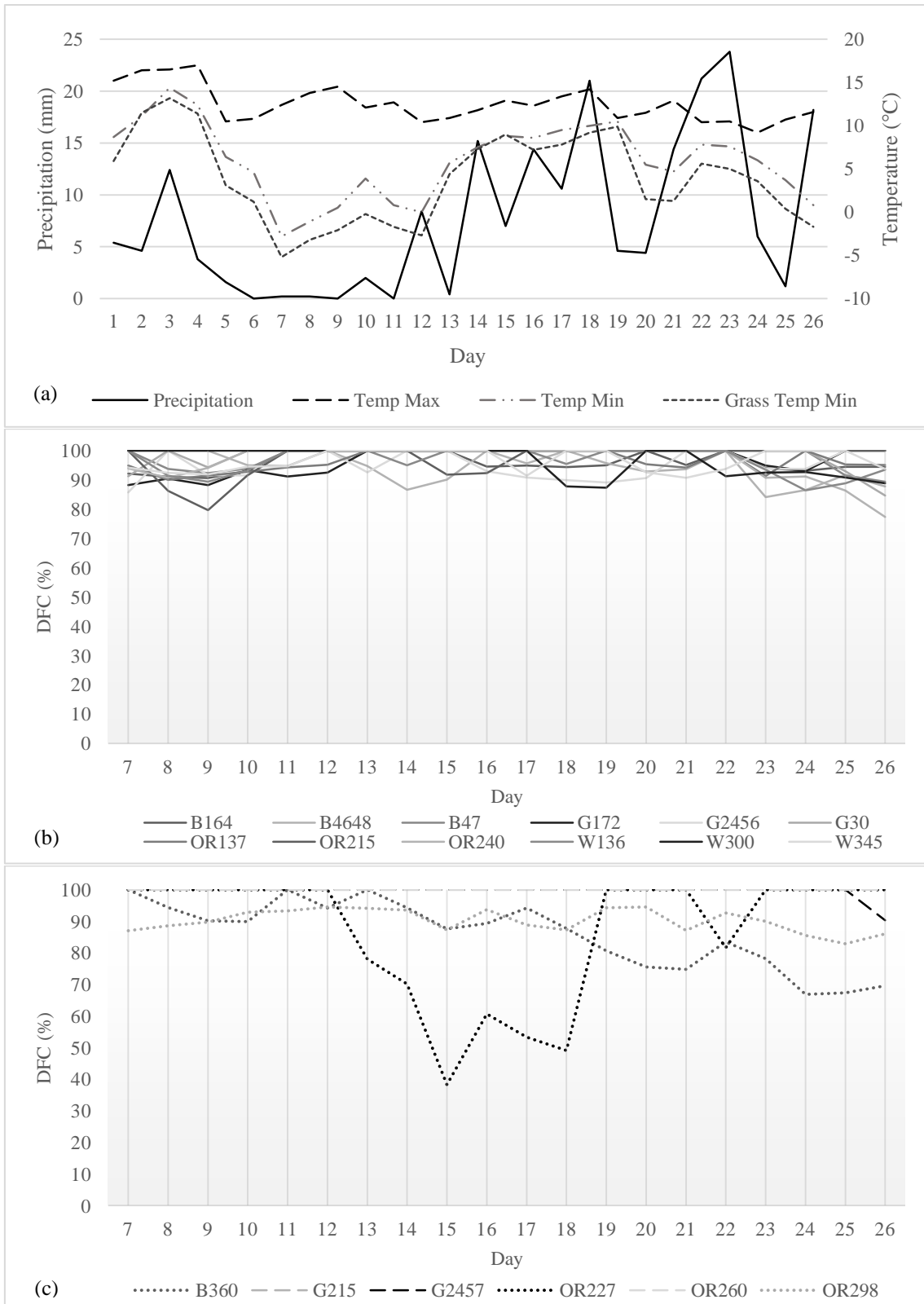


812 **Figure 6.** Degrees of Functional Coupling (DFCs) for the activity of ewes over the summer period.
 813 DFC% was calculated for moving seven-day intervals, thus, the graphic starts at the 7th day. (a) Absolute
 814 values for the weather over the summer period. (b) DFC% response of 14 ewes that did not present

815 deviation from the population intercept and slope over the summer period (Medium Consistency group).
 816 (c) DFC% response of five ewes that showed a deviation from the population intercept over the summer
 817 period. The three ewes represented by dashed lines presented higher consistency of DFC% (High
 818 Consistency group), whereas the two ewes represented by dotted lines showed the lower consistency of
 819 DFC% (Low Consistency group).
 820



821 (a) 822 **Figure 7.** Body weight gain (BWG) in relation to the variation of the Degrees of Functional Coupling
 823 (DFC) for activity, over the summer period. The dark grey area represents 95% of confidence limits and
 824 the light grey area represents 95% of prediction limits. Each DFC% value is an overall average for each
 825 ewe, over the spring period. (a) Linear regression for BWG in relation to the standard deviation (STD) of
 826 DFC% for all ewes. (b) Linear regression for BWG in relation to STD of DFC% for the Medium
 827 Consistency group.



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830

831 **Figure 8.** Degrees of Functional Coupling (DFCs) for the activity of ewes over the autumn period. DFC%
 832 was calculated for moving seven-day intervals, thus, the graphic starts at the 7th day. (a) Absolute values
 833 for weather over the autumn period. (b) DFC% response of 18 ewes that did not present deviation from
 834 the population intercept and slope over the autumn period (Medium Consistency group). (c) DFC%

835 response of six ewes that presented deviation from the population intercept over the autumn period. These
836 ewes differed from the population by an individual level that is independent of the temperature. The three
837 ewes represented by dashed lines presented higher consistency of DFC% (High Consistency group),
838 whereas the three ewes represented by dotted lines showed the lower consistency of DFC% (Low
839 Consistency group).