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A European monitoring protocol for the stag beetle, a saproxylic flagship species

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Abstract. 1. Developing protocols for threatened invertebrates is often challenging, because they are not only rare but also elusive. This is the case with the stag beetle (*Lucanus cervus*), a protected and flagship species for the saproxylic beetle fauna in Europe.

2. We applied a standard transect walk at a European scale (8 countries, 29 transects) to test its practicability and reliability as survey design.

3. A total of 533 sightings were recorded throughout the sampling period, but detection probability changed as the season progressed. Considering the observed activity pattern, occupancy models showed that a short period of three consecutive weeks, between the middle of June and the first week of July, resulted in a high probability of detection ($P > 0.7$). As time of the peak of activity varies from year to year and between sites, we propose to extend the sampling period to five weekly surveys.

4. Detailed information on the transect characteristics and the optimal time for surveying were analysed. The data indicate that a weekly transect at dusk provides a reliable method for monitoring this species throughout its

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distributional range. No correlation was found between latitude, longitude and phenology of sightings, however.

5. However, a standard method such as the one presented, allows broadening the scale of monitoring studies, provinding data to evaluate the efficacy of conservation measures.

Key words. dead wood, detection probability, Habitats Directive, *Lucanus cervus*, phenology, transect.

Introduction

Thousands of organisms living in temperate and boreal forests are *saproxylic*, i.e., depending during some part of their life cycle on wounded or decaying woody material from weakened, living or dead trees (Stokland *et al*., 2012; Carpaneto *et al*., 2015). Saproxylic organisms comprise up to a quarter of forest biodiversity (Siitonen, 2001; Schuck *et al*., 2004), and play an essential role in decomposition processes and nutrient cycling (Harmon *et al*., 1986; Müller & Schnell, 2003; Bobiec *et al*., 2005; Stokland *et al*., 2012). In the preceding centuries, management of European forests has led to a substantial decrease in volume and diversity of dead wood, resulting in many saproxylic species becoming regionally extinct or scatteringly distributed (Grove, 2002; Bobiec *et al*., 2005; Nieto & Alexander, 2010; Stokland *et al*., 2012; Carpaneto *et al*., 2015).

Most of the beetles included in Annexes II and IV of the EUs Habitats Directive (92/43/CEE) are saproxylic and their monitoring is mandatory (Articles 11 and 17) for countries belonging to the European Union. Standardised monitoring protocols for assessing their conservation status and population trends across geographical and temporal scales are not available (Schmeller, 2008). Only in some countries national monitoring strategies for saproxylic beetles have been proposed (e.g. Percy *et al*., 2000; Fartmann *et al*., 2001; Schnitter *et al*., 2006; Campanaro *et al*., 2011a; Smith, 2011; Makomaska-Juchiewicz & Baran, 2012; Vrezec *et al*., 2012a; Trizzino *et al*., 2013; Thomaes, 2014) and a critical validation of the proposed protocols is, in many cases, still pending. This lack of standardisation hampers fundamental conservation actions (Balmford *et al*., 2005) and thus establishing a common monitoring strategy for these species throughout European countries is of high importance (Henle *et al*., 2013).

Monitoring can be relatively straightforward for common and easily detectable species. Conversely, many threatened invertebrates are not only rare but also elusive, and developing appropriate monitoring strategies can be challenging (McDonald, 2004). In recent years, monitoring strategies for some large saproxylic beetles have benefited from the development of easy, non-invasive and inexpensive methods (e.g. Gouix & Brustel, 2012; Chiari *et al*., 2013), such as pheromone baits (Larsson & Svensson, 2009; Musa *et al*., 2013). Pheromone baits are not available for the large majority of species including the stag beetle (*Lucanus cervus* Linnaeus, 1758). Even though *L. cervus* is one of the largest European coleopterans (up to 85 mm), detection is not always easy as larvae live in underground decaying wood and adults are active for a short period of the year, mostly at dusk (Harvey *et al*., 2011a).

For more than 20 years, non-invasive monitoring methods for the stag beetle have been developed and tested (Table 1). Many of these methods, however, present difficulties in their application and standardisation. For example, acoustic detection (Harvey *et al*., 2011b) requires sophisticated equipment and has only been tested under controlled conditions. Traps employed for the monitoring of stag beetles captured few specimens when compared to local population size (Brustel & Clary, 2000; Krenn *et al*., 2002; Vrezec & Kapla, 2007; Fremlin & Hendriks, 2011; Harvey *et al*., 2011b; Jansson, 2011; Vrezec *et al*., 2012a) and attempts to use baits have failed to detect this species (Harvey *et al*., 2011b; Chiari *et al*., 2014; Chris Georgiadis, Arno Thomaes, John Smit, personal communication). Furthermore, the number of dead specimens, frequently used as a proxy of population abundance, depends not only on the population size but also on the number of travelling cars and/or local predators, which are unlikely to be present in comparable densities in

Table 1. Summary of the survey methods developed for the stag beetle in Europe.

Survey method	References
Counts of roadkill	Álvarez Laó and Álvarez Laó (1995), Hawes (2005), Thomaes (2008), Harvey *et al*. (2011b)
Counts of predation remains	Kervyn (2006), Schnitter and Malchau (2006), Campanaro *et al*. (2011b)
Counts of living adults	Chiari *et al*. (2014)
Counts of living adults including data from citizen science	Moretti and Sprecher-Uebersax (2004), Schnitter and Malchau (2006), Smit and Krekels (2006), Vrezec *et al*. (2012a), Mason *et al*. (2015)
Trapping of adults	Brustel and Clary (2000), Krenn *et al*. (2002), Kervyn (2006), Schnitter and Malchau (2006), Vrezec and Kapla (2007), Fremlin and Hendriks (2011), Harvey *et al*. (2011b), Jansson (2011), Vrezec *et al*. (2012a,b)
Acoustic detection of larvae	Harvey *et al*. (2011b)
Surveys of trunks	Vrezec *et al*. (2012a)

different sites. Counting flying specimens at dusk, using a transect walk, has been carried out since 1994 (Proyecto CiervoVolante, 1995), and several variations in this method have been implemented (Sprecher-Uebersax & Durrer, 1998; Kervyn, 2006; Vrezec & Kapla, 2007; Campanaro & Bardiani, 2012; Chiari *et al*., 2014). In Slovenia (Vrezec *et al*., 2012a) and Italy (Chiari *et al*., 2014), different methods were tested simultaneously and the transect walk proved to be the most successful method for stag beetle detection and population estimates.

This study is a direct result of a call for international collaboration for the conservation of *L. cervus* (Harvey *et al*., 2011b), an approach that has gained momentum only in recent years (Ranius *et al*., 2005; Harvey *et al*., 2011a). In particular, the goals of our study were (i) to apply a standard transect walk at European scale to test the practicability and reliability of the survey design, (ii) to investigate if detection probability may be affected by date and/or survey covariates, and (iii) to describe the phenology of the stag beetle across the study areas based on standardised counts.

Materials and methods

Study areas

We conducted this study in eight European countries and for each country, one to three areas, where the presence of stag beetle was already known were selected (Table 2). Study areas spanned from northern to southern Europe (Fig. 1) and included a broad representation of habitats where stag beetles occur, such as urban parks, agricultural landscapes and woodlands (Table 2).

Field survey

In 2012, one to three transect walks, each 500-m long and 10-m wide, were carried out within each study area, following landscape features such as roads, paths and forest edges, making a total of 29 transects (Table 3). Weekly surveys were performed during the flight period of adults: 21 May to 25 August, for a total of 14 weeks, hereafter called the season (Table 4). They were carried out only on days with suitable weather (no rain or strong wind, and temperature above 13 °C), and preferably on the same day of the week. Surveyors walked the transects at a constant speed of 0.28 m/s, so that the total duration was 30 minutes. All specimens observed within the transect were recorded, taking note of time (expressed as 5 minutes intervals), sex (male, female and unknown), height of flight (0, <2 m and >2 m) and relative position. This latter information was obtained dividing the total width of the transect (10 m) in three parts: left (3 m), centre (4 m) and right (3 m), see Appendix S1. Stag beetles walking on the ground within the transect area were also recorded.

Table 2. Characteristics of study areas surveyed for stag beetle.

Country	Study area	No. of transects	Habitat	Main tree species
Belgium	Beersel	2	Gardens, wooded strips and forest remnants in suburban areas	*Robinia pseudoacacia*, *Carpinus betulus*, *Acer pseudoplatanus*, *Quercus robur*, *Fagus sylvatica*
	Overijse	2	Gardens and wooded strips	*Ulmus* sp., *C. betulus*
Germany	Mittelmosel	2*	Village areas with gardens	*Prunus* sp., *Malus* sp.
Italy	Varese	2	Woodland	*Q. robur*
	Bosco Fontana	2†	Woodland	*C. betulus*, *Q. robur*
	Colline moreniche	2†	Agricultural landscape with forest remnants	*Q. pubescens*, *Ostrya carpinifolia*, *R. pseudoacacia*
Poland	Przemków	2†	Woodland (140 years old)	*Q. petraea*
	Janików	2	Woodland (100 years old)	*Q. robur*, *Pinus sylvestris*
	Krzyżowiec	2†	Woodland (120 years old)	*Q. petraea*
Slovenia	Ljubljana	2†	Urban forest (woodland)	*Q. robur*, *Q. rubra*, *F. sylvatica*, *C. betulus*, *Picea abies*
	Vipava Valley	2†	Agricultural landscape with forest remnants	*Q. pubescens*, *Ostrya carpinifolia*, *Fraxinus ornus*
Spain	Guadarrama Range	2†	Rangeland with forest remnants	*Q. pyrenaica*, *Fraxinus angustifolia*
Switzerland	Ruetihard	1†	Woodland	*F. sylvatica*
United Kingdom	Bentley, Suffolk	3	Roadside trees, orchards, village gardens, horse pasture, hedges and hedge line trees in an agricultural landscape	*Q. robur*. *Acer campestre*, *Fraxinus excelsior*
	Surrey	1	Gardens in suburban areas	*Q. robur*, *F. sylvatica*, *Crataegus* sp., *Cupressus* sp.

*One transect of which included in the reduced data set.
†Transects included in the reduced data set.

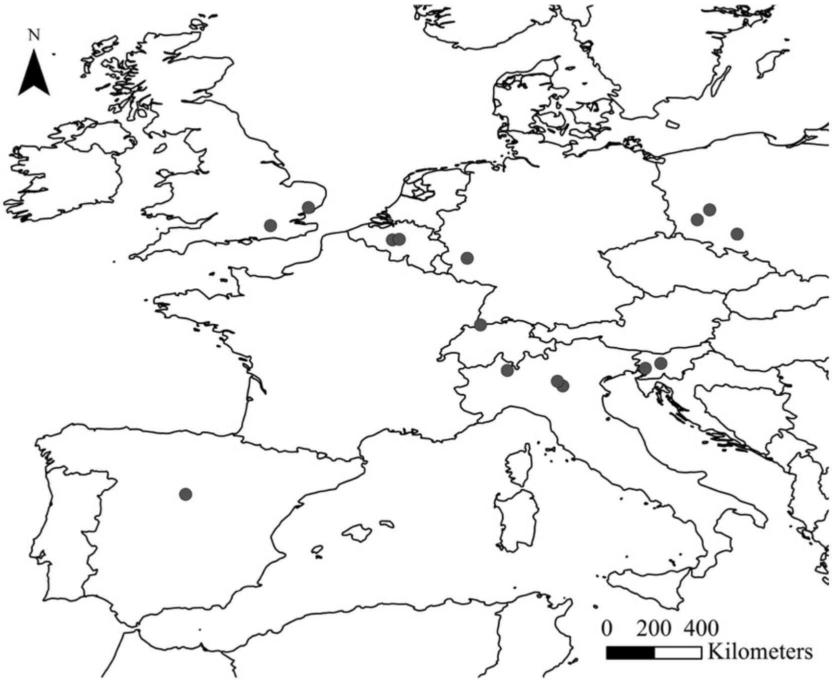

Fig. 1. Study areas are spread across the majority of the stag beetle distribution range.

Table 3. Summary of stag beetle sightings in Europe.

Country	No. of study areas	Flying			Not flying		
		♂	♀	Uncertain	♂	♀	Uncertain
Belgium	2	16	0	4	42	14	0
Germany	1	4	6	1	1	0	0
Italy	3	114	4	28	19	15	0
Poland	3	24	29	14	16	20	0
Slovenia	2	31	1	1	3	1	0
Spain	1	26	3	0	5	2	0
Switzerland	1	49	2	0	0	7	0
United Kingdom	2	20	3	0	5	3	0

Survey covariates

We registered three weather covariates at the beginning and at the end of the survey: temperature (T, as °C), relative humidity (RH, as percentage %) and wind speed (WS, ranked according to the five categories described in Appendix S1). These were used to calculate the mean between initial and final values for each transect walk. A further covariate was the time to sunset (TS), the number of minutes between the beginning the transect walk and the sunset time. Finally, two covariates were related to the moon: the visibility of the moon (VM, as the percentage of moon surface visible each night regardless of the moon phase, data downloaded at www.eurometeo.com) and the moon phase (MP, as ranked values correspondent to the four moon phases: full, waning, new and crescent).

Table 4. Correspondence between the sampling weeks as referred in the current study and the calendar days.

Sampling week number	Calendar days 2012
1	21 May–27 May
2	28 May–3 June
3	4 June–10 June
4	11 June–17 June
5	18 June–24 June
6	25 June–1 July
7	2 July–8 July
8	9 July–15 July
9	16 July–22 July
10	23 July–29 July
11	30 July–5 August
12	6 August–12 August
13	13 August–19 August
14	20 August–26 August

Data analyses

We extracted from the total data set (TD, $N = 29$) a reduced data set (RD), which contained only those transects ($N = 16$) with more than five sightings of stag beetles and which had no missing data (Table 2).

Survey design

The reliability of the survey design was assessed analysing the homogeneity of the distribution of sightings, in terms of relative position (left, centre and right), height of flight and sex.

We tested the observed frequency distribution of sightings with the chi-square test, applying the Holm adjusted *P*-value to correct for simultaneous inference. We carried out the analysis using R version 3.0.3 (R Development Core Team, 2014).

Detection probability and survey covariates

We applied single species, single season open and closed occupancy models (MacKenzie *et al.*, 2006; Kendall *et al.*, 2013) to evaluate if detection probability (P) on a particular survey was a function of the date and/or the survey covariates. We tested, on the TD, models with detection probability constant (P, i.e., detection probability constant between surveys), full time dependent (P_t, the detection probability changes between surveys) or constrained time dependent (P_1, P_2, P_3, i.e., the detection probability within the constrained time intervals is different from detection probabilities of time intervals before and after that constrain). Considering the observed phenology, we derived time-constrained models from sighting distribution as follows: (i) P from weeks 4 to 8, (ii) P from weeks 4 to 7, and (iii) P from weeks 5 to 7. For open models, we calculated entry probability (e), i.e., the emergence and/or immigration of new individuals, and departure probability (d), i.e., the death and/or emigration of individuals. We modelled both e and d probabilities as constant over time or time dependent (P_t). We also modelled the magnitude of the effect of each survey covariate as constant ($P_{\text{survey covariate}}$) or time dependent ($P_{t+\text{survey covariate}}$). We tested the relationship between P and survey covariates on first- (T, RH, WS, TS, VM, MP) and second-order (Survey Covariate+Survey Covariate^2) polynomial models, hypothesising single, additive and multiplicative effects. For the survey covariates analyses, we excluded transects with missing values in more than three surveys (i.e. Belgium, United Kingdom and Colline Moreniche study area in Italy) and we considered only flying males.

Models were ranked according to their AIC (Akaike's information criterion) value, only candidate models were considered (ΔAIC < 2) (Burnham & Anderson, 2002). Analyses were performed using program PRESENCE 8.3 (Hines & MacKenzie, 2004).

Phenology

To verify and describe the relationship between the peak of activity and geographical variables, we performed the Spearman's correlation between the week in which the median number of sightings was obtained and three geographical variables (longitude, latitude and altitude). In addition, we ran generalised linear models (GLMs), with the week of peak activity as dependent variable and longitude, latitude or altitude and their quadratic terms as independent variables, using a Poisson error distribution and a log-link function, using the GLM package. All the analyses were performed on the RD using R version 3.0.3 (R Development Core Team, 2014).

Results

In total, we sighted 533 stag beetles in 29 transects, most specimens (N = 183) were recorded in Italy, while the lowest number (N = 13) was recorded in Germany (Fig. 2; Tables 2 and 3).

Survey design

Most of the specimens were observed laterally with respect to the observer; this pattern was significant for all categories considered (Table 5; Fig. 3a, b). Among flying individuals, most were observed above 2 m (Fig. 3b), a result significant only for males (Table 5; Fig. 3b, c). For females, the same numbers were observed walking and

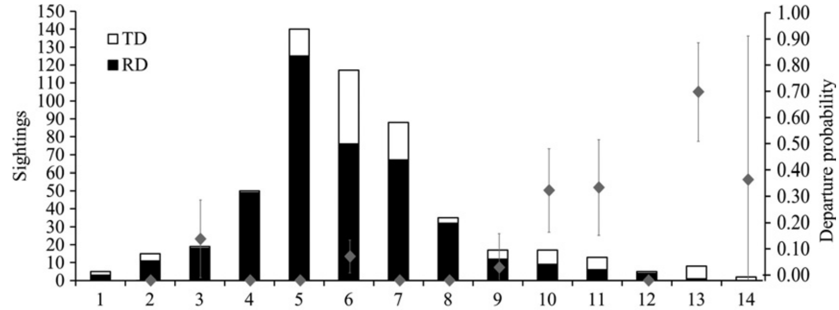

Fig. 2. Weekly distribution of stag beetle sightings in Europe obtained using transect walks at dusk. Sightings from the total data set (TD) not included in the reduced data set (RD) are presented in white. Grey dots indicate stag beetle departure probability according to the occupancy models and vertical bars the standard error. TD, total data set; RD, reduced data set.

Table 5. Chi-square test results of stag beetle sightings in relation to sex, part of the transect and flight height.

	χ^2	d.f.	*P*	Holm adjusted *P*-value
Sex	302.944	2	2.20 e^{-16}	2.64 e^{-15}
Part of the transect				
Total	131.088	2	2.20 e^{-16}	2.64 e^{-15}
Female	39.664	2	2.44 e^{-9}	1.46 e^{-8}
Male	63.889	2	1.34 e^{-14}	1.34 e^{-13}
Unknown	42.036	2	7.45 e^{-10}	5.22 e^{-9}
Flight height				
Total	49.9133	2	1.5 e^{-11}	1.16 e^{-10}
Female	13.517	2	1.16 e^{-3}	1.67 e^{-3}
Male	58.329	2	2.16 e^{-13}	1.94 e^{-12}
Unknown	24.571	2	4.62 e^{-6}	1.85 e^{-5}

flying (pooling the two height categories: <2 m and >2 m) (Fig. 3c).

The number of individuals of unidentified sex was highest for those flying above 2 m, whereas we successfully sexed all individuals observed at ground level.

Detection probability and survey covariate effect

The plausible models for stag beetle detection were open, indicating that most individuals are present, and thus available for detection, in a limited time interval within the whole sampling season (Table 6). In fact, the top-rank model (Table 6a) strongly supported the P_3 hypothesis with the highest detection probability from weeks 5 to 7 ($\widehat{P}$ = 0.87, SE = 0.05), whereas during the other weeks it decreased ($\widehat{P}$ = 0.60, SE = 0.06). Conditional entry probability, i.e., the probability that individuals enter the study area, for instance through emergence and colonisation, was 0.26 (SE = 0.05) and remained constant across the season. In contrast, the departure probability, i.e., the probability that individuals depart from the study areas, for instance through emigration or death, increased during the season (Fig. 2).

Considering only flying males, in the top-rank model, the detection probability was time dependent ($_t$), i.e., the detection probability varied during the single surveys, which lasted 30 minutes and showed a constant second-order polynomial relationship with time to sunset

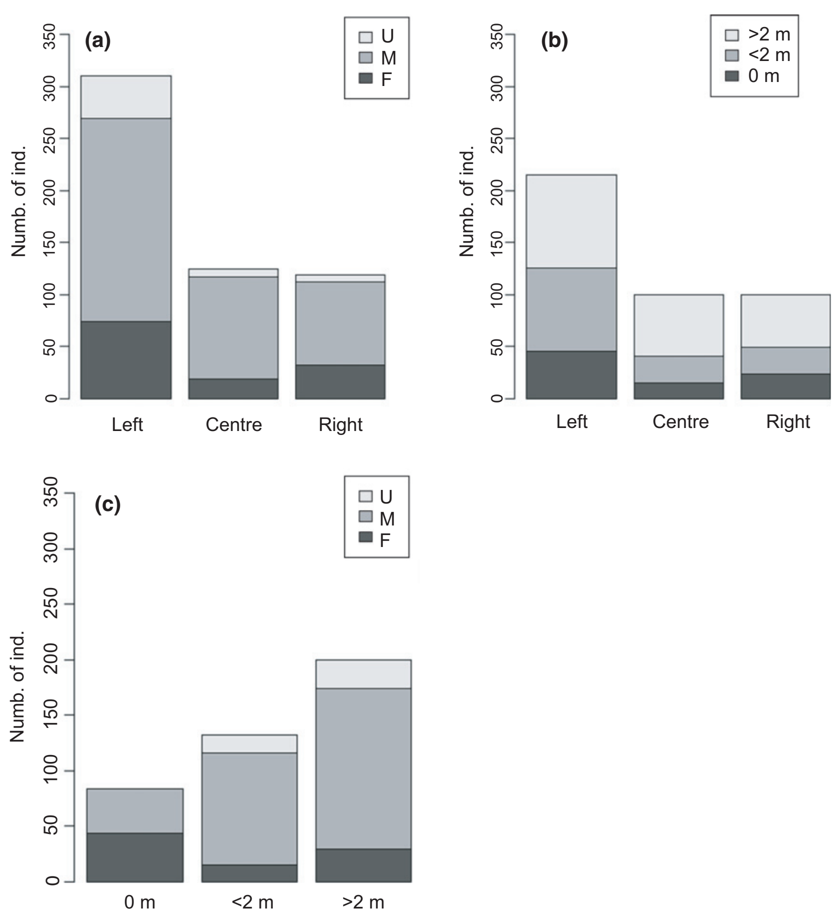

Fig. 3. Frequency of stag beetle sightings in relation to: (a) sex and part of the transect, (b) height of flight and part of the transect, and (c) sex and height of flight. M, males; F, females; U, unknown.

Table 6. Results of the plausible occupancy models (ΔAIC < 2) of the European stag beetle survey for (*a*) the total data set (TD) and (*b*) for the effect of survey covariates on flying males.

Model	K	-2Log (L)	ΔAIC	w
(a) TD				
ψ, e, d_t, p_3	17	380.46	0.00	0.47
(b) flying males				
ψ, e, d, $p_{t\ \mathrm{TS+TS}^2}$	19	213.79	0.00	0.19
ψ, e, d, P	4	263.82	0.49	0.15
ψ, e, d, p_{TS}	5	260.74	0.61	0.14
ψ, e, d, $p_{\mathrm{TS+TS}^2}$	6	258.01	0.95	0.12

Occupancy (ψ), detection (P), entry (e) and departure (d) probabilities are considered. Constraints: $_t$, full time dependence, $_3$, constrained time dependence (hypothesis where P from weeks 5 to 7 is different from the P observed during the other weeks); TS, time to sunset; TS + TS^2, second-order polynomial relationship with TS. K represents the number of parameters in the model, w the Akaike weight and -2Log (L) is twice the negative log-likelihood value. Akaike's information criteria (ΔAIC) were calculated for each model (Burnham & Anderson, 2002).

($w_{\mathrm{TS+TS}}2 = 0.31$), i.e., the detection probability increased from the start of the survey, reached a peak and declined thereafter (Table 6b; Fig. 4).

Phenology

The week of peak activity ranged from week 2 at sites in Germany (Appendix S2b) to week 6 at sites in both Slovenia and the United Kingdom (Appendix S2e, h). The number of stag beetle sightings increased rapidly from week 1 to a maximum in week 5, declining afterwards, though less rapidly, but extended over a longer period from weeks 6 to 14 (Fig. 2). We recorded the lowest number of sightings during the first and last weeks of the study.

We did not simultaneously include geographical variables in any analysis due to significant correlations of longitude with both latitude ($r_s = 0.726$, $n = 16$, $P < 0.001$) and altitude ($r_s = -0.553$, $n = 16$, $P = 0.026$). We did not find any significant correlation between week of peak activity and either longitude ($r_s = 0.468$, $n = 16$, $P = 0.068$), latitude ($r_s = 0.152$, $n = 16$, $P = 0.575$) or altitude ($r_s = -0.280$, $n = 16$, $P = 0.293$). The GLMs yielded no significant influence of any of the geographical variables or their quadratic terms (all $P > 0.327$) on the median week of peak activity. Therefore, we pooled all data from the RD for a 'European' phenology (Fig. 2).

Discussion

The standardised sampling protocol proposed in the present article, applied at the pan-European scale, allowed us to record a large number of sightings. The results showed that this method is a practicable and reliable sampling protocol, applicable at any suitable stag beetle location, especially for flying individuals.

It appears that the width of the transect was appropriate, because the distribution of sightings was not biased towards its central part. In accordance with other authors (Klausnitzer, 1982; Klausnitzer & Sprecher-Uebersax, 2008; Chiari *et al.*, 2014), flying males were most commonly recorded, being 70% of total sightings. Despite the strong sexual dimorphism of this species (Franciscolo, 1997; Solano *et al.*, 2016), establishing the sex of flying individuals was not easy, especially for those sighted above a height of 2 m and because of the occurrence of small males. In relation to the total number of flying individuals recorded, those for which the sex could not be determined represented only 13%. The described sampling protocol for transect walks also proved to be suitable for detecting females, even if they represented only 20% of total sightings. Surprisingly, females were observed walking and flying in similar proportions. This does not follow the characteristic searching behaviour usually reported in literature for this sex (Harvey & Gange, 2003; Rink & Sinsch, 2007). Identification problems might also occur in European areas where more than a single species of large stag beetles are recorded (e.g. in central Italy, where *L. cervus* and *L. tetraodon* are sympatric in several sites: Franciscolo, 1997; Solano *et al.*, 2016; in southern France, where *L. cervus* and *L. pontbrianti* co-occur: Boucher, 2014; in

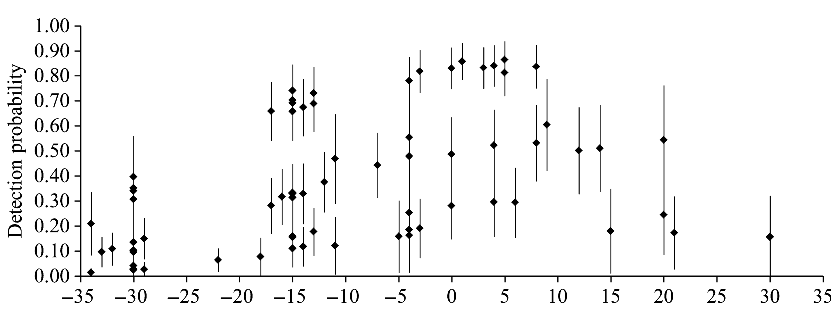

Fig. 4. Detection probability of flying stag beetle males in relation to time to sunset. Black dots indicate detection probability and vertical bars the standard error.

Mediterranean Spain, where *L. cervus* and *L. barbarossa* co-occur; in Greece and the surrounding of the Black Sea, where *L. cervus* and *L. ibericus* co-occur).

Detection probability and survey covariate effect

In our study, the stag beetles were observed during the entire sampling season. The detection probability, however, varied and was highest at weeks 5 to 7, corresponding to the period between 18 June and 8 July. Therefore, if sampling effort is a constraint, the monitoring period could be shortened to these 3 weeks.

The top model strongly supported the hypothesis of seasonality in adult stag beetle with constant entry and fluctuating departure probabilities. The constant entry probability could be explained by the biology of the species, where the emergence of adults is continuous from May to August (Harvey *et al.*, 2011a). However, it should be taken into account that colonisation from nearby reproductive sites, up to 2500 m distant (Rink & Sinsch, 2007), can occur, balancing a possible uneven rate of emergence during the season. The departure probability was low at the beginning of the season, but subsequently rose starting from the ninth week of sampling (i.e. 16 July), which can be explained by increasing mortality towards the end of the season. This result supports the suggestion that the monitoring should be concentrated on the period between 18 June and 8 July, i.e., before mid-July when the mortality of beetles starts to increase.

The detection probability obtained is encouraging when compared with other insect monitoring programmes. In the Swiss Biodiversity Monitoring programme for butterflies, the average detection probability per survey was 0.50 and ranged from 0.17 to 0.81 (Kéry & Plattner, 2007). For the cobblestone tiger beetle, *Cicindela marginipennis* (Dejean, 1831), the detection probability calculated for habitat surveys was 0.67 (Hudgins *et al.*, 2012) and for the longhorn beetle *Morimus asper* (Sulzer, 1776) it was 0.53 (Chiari *et al.*, 2013). In comparison with the above-mentioned studies and other approaches previously tested for the monitoring of the stag beetle, the transect walk is, at present, the best candidate sampling method to be applied in future monitoring programmes.

Among the survey covariates analysed in this study, the second-order polynomial relationship with TS was the factor which mostly influenced the detection of flying stag beetle. Detection probabilities were above the value of 0.70 starting from 15 minutes before the sunset to 8 minutes after, reaching the maximum value ($P = 0.86$) 1 minute after the sunset (Fig. 4). As in previous studies (Harvey *et al.*, 2011a; Smith, 2011; Vrezec *et al.*, 2012a,b; Chiari *et al.*, 2014), we found the highest stag beetle flying activity at dusk. Therefore, to improve the probability of detecting flying stag beetles, it is suggested that the transect walk should start 15 minutes before sunset.

According to our models, the detection probability was not influenced by the other survey covariates: wind speed (WS), visibility of the moon (VM) and moon phase (MP).

Phenology

This study is the first to report on the phenology of the stag beetle based on a standardised sampling protocol in several European countries and across a range of habitat types. The resulting phenology was not found to be influenced by longitude, latitude or altitude.

Phenological data on stag beetles have been reported before from single countries using different sampling methods (Fig. 5). The 'European' phenology described here, with most observations close to 21 June (Fig. 2), is consistent with most of the previous studies (Fig. 5). The phenology of adult stag beetles is rather concentrated in a narrow time window, suggesting that a limited number of weekly surveys are sufficient for monitoring the species. This must be considered because the number of surveys is the most cost-relevant factor and has to be optimised for cost-effectiveness. The highest stag beetle detection probability corresponded to only three surveys carried out at

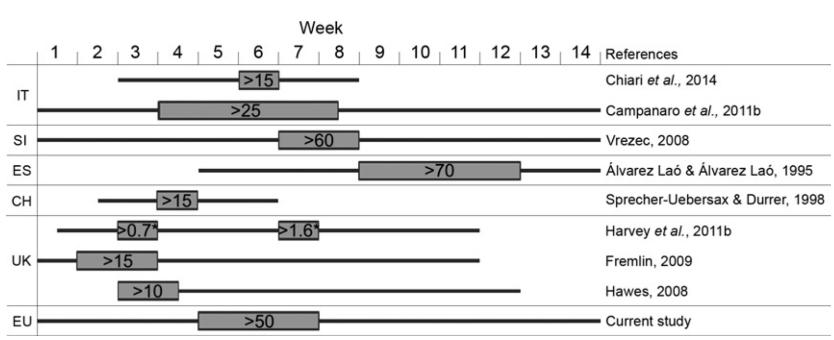

Fig. 5. Comparison between the flying activity of the stag beetle observed in this study (EU) and that observed in different European countries (IT, Italy; SI, Slovenia; ES, Spain; CH, Switzerland; UK, United Kingdom) according to literature. Narrow grey line = observation of at least one individual during the season. Thick grey line = maximum individuals (numbers in black) sighted according to the reference indicated. *The value is reported from the authors as mean number of individuals per day.

the beginning of the season (weeks 5 to 7: between the mid-June and the first week of July).

Transect walk at dusk: implication for conservation and future strategies

Our results provide an important step towards a large scale monitoring programme, supported by a reliable sampling protocol, which allows the optimisation of sampling effort. The high probability of detection, especially in the peak period, leads us to suggest that a low number of person/hours is sufficient to monitor the species across all of its distribution range, and that the high effort suggested by Campanaro and Bardiani (2012), which requires two surveyors for 60 minutes, is unnecessary. Neither are the 12–13 surveys per season suggested by Proyecto Ciervo Volante (1995) and Trizzino *et al*. (2013). Our study, in fact, demonstrated that one survey per week for 3 weeks is sufficient to obtain the higher values of detection probability. The peak of activity may vary yearly (Vrezec *et al*., 2012b; C. Hawes, unpublished) and among sites, and thus cannot be relied on. These considerations lead us to suggest that a sampling effort of five weekly surveys (between the beginning of June and the beginning of July) might be an appropriate compromise between costs and benefits for an experimental European sampling programme. Our standard sampling protocol is already being applied in ongoing conservation programmes of several European countries (Vrezec *et al*., 2012b; Thomaes, 2014; Mason *et al*., 2015).

The weekly transect walk at dusk provides the best candidate monitoring protocol at present to obtain homogeneous and reliable data at a European scale, from both specialists and citizens, to fulfil the Habitats Directive requirements. Moreover, the European survey presented here could be taken as an example of a practical and successful application of the 'network' concept of Natura 2000.

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Supporting Information

Additional Supporting Information may be found in the online version of this article under the DOI reference: doi: 10.1111/icad.12194:

Appendix S1. Surveyors field recording sheet.

Appendix S2. Weekly distribution of stag beetle sightings obtained using transect walks at dusk for each country: Sightings of the total data set (TD) not included in the reduced data set (RD) are presented in white.

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