A field study on *Saga pedo* (Ensifera, Tettigoniidae, Saginae): Spatial behavior of adult specimens

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Abstract

Despite its large size, the protected predatory bush-cricket *Saga pedo* (Pallas, 1771) is difficult to study in the field. This is mainly due to its strong mimicry, prevalent night activity, and low population density. The aim of this study was to investigate the spatial behavior of some adult individuals through the use of luminescent tags and recording their occurrences at night. The monitored individuals moved considerably during the oviposition period and were found more frequently in small sections of the study area. Two models for count data were implemented to try to explain this behavior. The results indicate that their spatial behavior was predominantly related to the prey availability in the available environment. In addition, predation on the Hymenoptera *Sphex funerarius* Gussakovskij, 1934 is reported for the first time.

Keywords

Count data, Orthoptera, *Sphex funerarius*, UV light, zero-inflated

Introduction

In Europe, *Saga pedo* (Pallas, 1771) is considered the largest insect among Orthoptera (Willemse 1996, Massa et al. 2012, Trizzino et al. 2013). Nevertheless, observing this species in its environment can be difficult, considering its cryptic mimicry and prevailing nocturnal activity (Willemse 1996, Lemonnier-Darcemont et al. 2009, 2016). These traits, combined with the low density that is the result of the species being an obligate predator almost exclusively of other Orthoptera (Kaltenbach 1970, Fontana and Cussigh 1996, Massa et al. 2012, Lemonnier-Darcemont et al. 2016) and the ecology of a parthenogenetic organism (Matthey 1941, 1948, Kaltenbach 1970, Kolics et al. 2012, Lemonnier-Darcemont et al. 2009, 2016), place practical limitations on carrying out field studies. The goal of this study was to investigate the spatial behavior of some adult individuals during the oviposition period by estimating the distances travelled, the relationship between occurrences, and some ecological variables registered in the field.

Marking is commonly used in the field to follow the movements and activities of individual insects and could also be applied to Orthoptera (Gangwere et al. 1964, Hagler and Jackson 2001). Narisu and Schell (1999) described a marking method based on using fluorescent powder that can increase the ease of recapture and reduce the need for manpower and time spent searching. Other authors have successfully experimented with the application of reflective tape on the femur, improving the night re-sighting of specimens (Heller and von Helversen 1990, Hein et al. 2003). A similar marking protocol, but with phosphorescent tags, was also successfully tested on *S. pedo* (Holuša et al. 2013). However, the use of highly visible marks can increase the predation rate. In the present study, I tried to minimize this effect in order to follow the movements of individuals as long as possible during the oviposition period.

S. pedo is included in Appendix II of the Berne Convention and Annex IV of the "Habitats Directive" 92/43/EEC and is classified as LC (Least Concern) in the European IUCN Red List of Threatened Species. However, subpopulations are usually small, and the European population is severely fragmented (Hochkirch et al. 2016).

Methods

Study area.—The study was conducted in NW Italy, in the protected area SCI IT1110030 Oasi xerotermiche della Valle di Susa – Orrido di Chianocco e Foresto. This site hosts vegetation characterized by the presence of Mediterranean and steppic floristic species (Sindaco et al. 2009). Within this site, a survey area of 3745 m^2 was chosen that consisted of a xeric grassland at an altitude of about 625 m above sea level, with southern exposure, a slope of 33%, covered by sparse shrub/tree vegetation and rocks, and surrounded by small wooded areas.

Sampling.—To find the *S. pedo* specimens, the study area was thoroughly inspected on two consecutive sunny days in early August 2019, using a 1.20 m long stick, useful for quickly inspecting the vegetation, as already tested in previous studies on this species (Holuša et al. 2013, Anselmo 2019). Given the aim of the study and the strong cryptic mimicry that characterizes this species, a particular protocol was performed for marking each captured adult specimen. A tag of about 0.8 × 0.8 cm was attached around the right hind femur, consisting of green-yellow adhesive tape fluorescing only under ultraviolet (UV) light and reinforced with a bead of strong glue. A unique identification code has been applied to each tag. The time taken to carry out the marking operations was approximately five minutes per individual, mainly to allow the glue to set. The manipulation and marking protocol were authorized by the Italian Institute for Environmental Protection and Research (ISPRA).

Afterwards, the survey area was covered every four nights from early August to early September (from 10 pm to 1 am), spotting the marked individuals thanks to a powerful blacklight (i.e., Wood's lamp) and the luminescence of the tags, which were visible up to 8 m away. For each recapture, the insect's exact position and behavior were reported.

Ecological covariates.—The variables used for the analysis were measured on 15 August 2019 in sunny and windless conditions. To do this, the aerial imagery of the survey area was divided into a 3 × 3 m grid with QGIS (ver. 2.18.25), and the resulting map was used as a cartographic base in a GPS device. Data from the following covariates were collected in each of the resulting 416 grid cells: an estimate of prey abundance obtained from the sum of the number of Orthoptera and Mantodea trapped within a standard sampling surface of 0.16 m^2 (defined in the field by a plastic cylinder) used three times randomly on the ground; visual estimate of the percentage of bare soil; visual estimate of the average height of the grass; and presence/absence of tree or shrub.

Data analysis.—The movements of each insect were analyzed with QGIS (ver. 2.18.25), measuring the linear distances between the occurrences in each survey recorded with a GPS.

To investigate the relationships between the occurrences of the specimens and the ecological covariates, two generalized linear models (GLM) for count data were produced. GLM are commonly used for count data, with family Poisson or negative binomial (P or NB) depending on the distribution of the response variable (Zuur et al. 2009). If the response variable contains more zeros than expected in the frequency distribution, mixture models such as zero-inflated (ZI) or two-part models such as hurdle (H) are used (Zuur et al. 2009). When zero inflation is present, the frequency distribution of the response variable has a large spike at zero, a common situation for ecological count data (Welsh et al. 1996, Cunningham and Lindenmayer 2005, Martin et al. 2005, Wenger and Freeman 2008, Zuur et al. 2009, Lyashevska et al. 2016). Within the ZI models,d zero-inflated Poisson (ZIP) and zero-inflated negative binomial (ZINB) are distinguished, depending on whether there is overdispersion in the count data (Zuur et al. 2009).

The models were performed for two different count data for each grid cell, referring to the whole study period: the number of individuals and the number of occupancy events. All combinations of ecological variables were used to implement the models after checking for the absence of strong correlations between them. The selection of the models was made based on the Akaike Information Criterion (AIC), a widely used evaluation method to compare models (Zuur et al. 2009). All statistical analyses were performed in R (ver. 3.6.3).

Results

Sampling and behavioral observations.—Seven *Saga pedo* adult specimens were captured, marked, and released on 2 and 3 August, 2019. Most individuals were sampled on the first day of research (n = 5). It was possible to spot the individuals on tufts of grass at a height between 5 and 30 cm from the ground (mean = 13 cm, SD = 8 cm), two of which were seen consuming freshly caught prey: *Mantis religiosa* Linnaeus, 1758 and *Sphex funerarius* Gussakovskij, 1934 (Fig. 1). Three *S. pedo* were at the base of the same bush, with a distance of about 50 cm between them.

The recapture was performed every four nights, from 3 August 2019 until 5 September 2019, consisting of nine surveys. It was possible to spot the specimens up to 8 m on tufts of grass, on bushes, and on bare soil at a height between 0 and 98 cm from the ground (mean = 24 cm, SD = 20 cm). However, some individuals

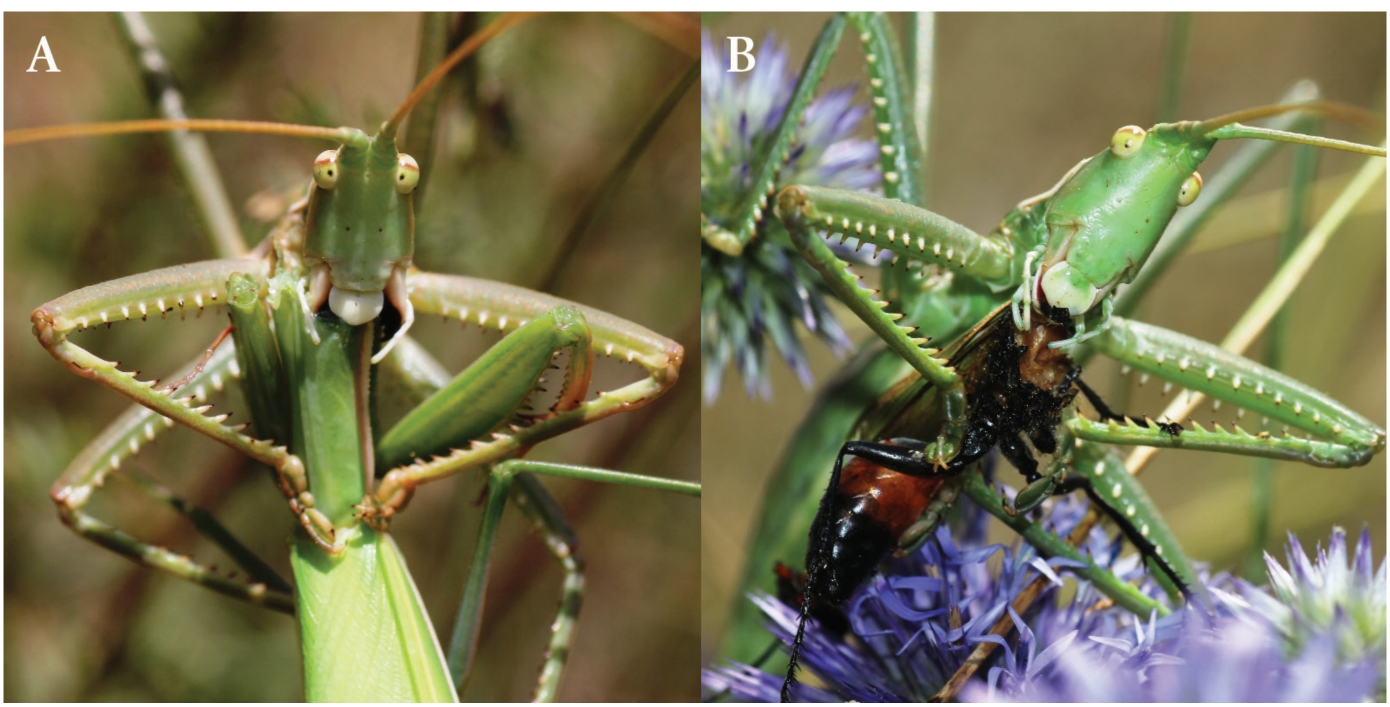

Fig. 1. Behavioral observations. **A.** Predation on *Mantis religiosa*; **B.** Predation on *Sphex funerarius*.

Fig. 2. Behavioral observations. **A.** Oviposition (7 August 2019, 23:05 pm); **B.** Ovipositor cleaning (11 August 2019, 22:30 pm).

were not recaptured in every survey (Table 1). In the recapture surveys, the individuals were apparently motionless, and in 3 cases, it was possible to observe oviposition (Fig. 2A). One individual was observed cleaning the ovipositor, behavior that suggests recent laying of the eggs (Lemonnier-Darcemont et al. 2016) (Fig. 2B). On some occasions, 2–3 individuals were found simultaneously, with less than 100 cm between them.

Table 1. Recapture events of marked individuals during the monitoring period.

ID	Survey date								
	3.VIII	7.VIII	11.VIII	15.VIII	19.VIII	23.VIII	27.VIII	1.IX	5.IX
A01	•		•	•	•	•	•	•	•
A02	•	•	•	•	•	•	•	•	•
A03	•	•		•	•	•		•	
A04	•	•	•	•			•	•	•
A05	•	•		•	•	•		•	
B01	•	•	•	•	•	•		•	
B02	•			•	•	•	•	•	•

Data analysis.—The individual distance travelled during the whole study period has wide variability, between 18 and 201 m (mean = 96 m, SD = 63 m), which is the sum of the linear distances measured between the occurrences for each individual (Fig. 3).

To better understand the use of space made by the species, some models were implemented based on the ecological covariates collected. All the count data show strong positive skewness, with zero-inflation in frequency distribution (Fig. 4A); this was confirmed using the vcdExtra package in R (Friendly and Meyer 2016). Therefore, the ZIP and ZINB models were chosen as implemented in the countreg package in R (Zeileis et al. 2008). The models that best explain the two different types of count data are ZIP models with only the covariate of the abundance of the potential prey (Table 2). The hanging rootograms (Kleiber and Zeileis 2016) in Fig. 4B show the models fitting for the two-count data: departures from expected counts are smaller, and the zero-count bin is always well fitted. However, there were some small deviations from the observed data, particularly in the highest counts.

Discussion

Saga pedo individuals regularly move for hunting, shelter, or laying eggs (Lemmonier-Darcemont et al. 2016). Holuša et al. (2013), in a study done in the Czech Republic, reported daily distances for adults between 0.5 and 2 m, with peaks at 14 and 37.5 m. Richard (2010) found a mean of 2.87 m in a favorable habitat in France. Thus, as confirmed by the results of the present case study, *S. pedo* individuals show a remarkable capacity for movement. To make a comparison, if we divide the mean distance traveled by individuals by the 33 days of the entire study period, we can estimate a mean daily shift of 2.9 m. Moreover, it cannot be ruled out that the tags applied to individuals limited the displacements to some extent. Without marking, individuals might be expected to move more easily and thus cover more distances.

Following the movements of the small sample of monitored *S. pedo*, it can be said that the spatial behavior seems related to specific ecological conditions. As evidenced by the models, the marked individuals mostly frequented the areas particularly rich in Orthoptera and Manotidea prey. The two types of modelled count data express this evidence quite differently: the number of different individuals that occupied the cells shows that some of these are especially attractive, particularly those characterized by high availability of prey. The number of occupancy events indicates a greater likelihood of observing the species in these cells rather than in those with scarce availability of food resources. Furthermore, individuals were sometimes spotted simultaneously on these sites, within a short distance of each other. This suggests that the species constantly moves to hunt and is able to identify the best places to do so. Therefore, in the

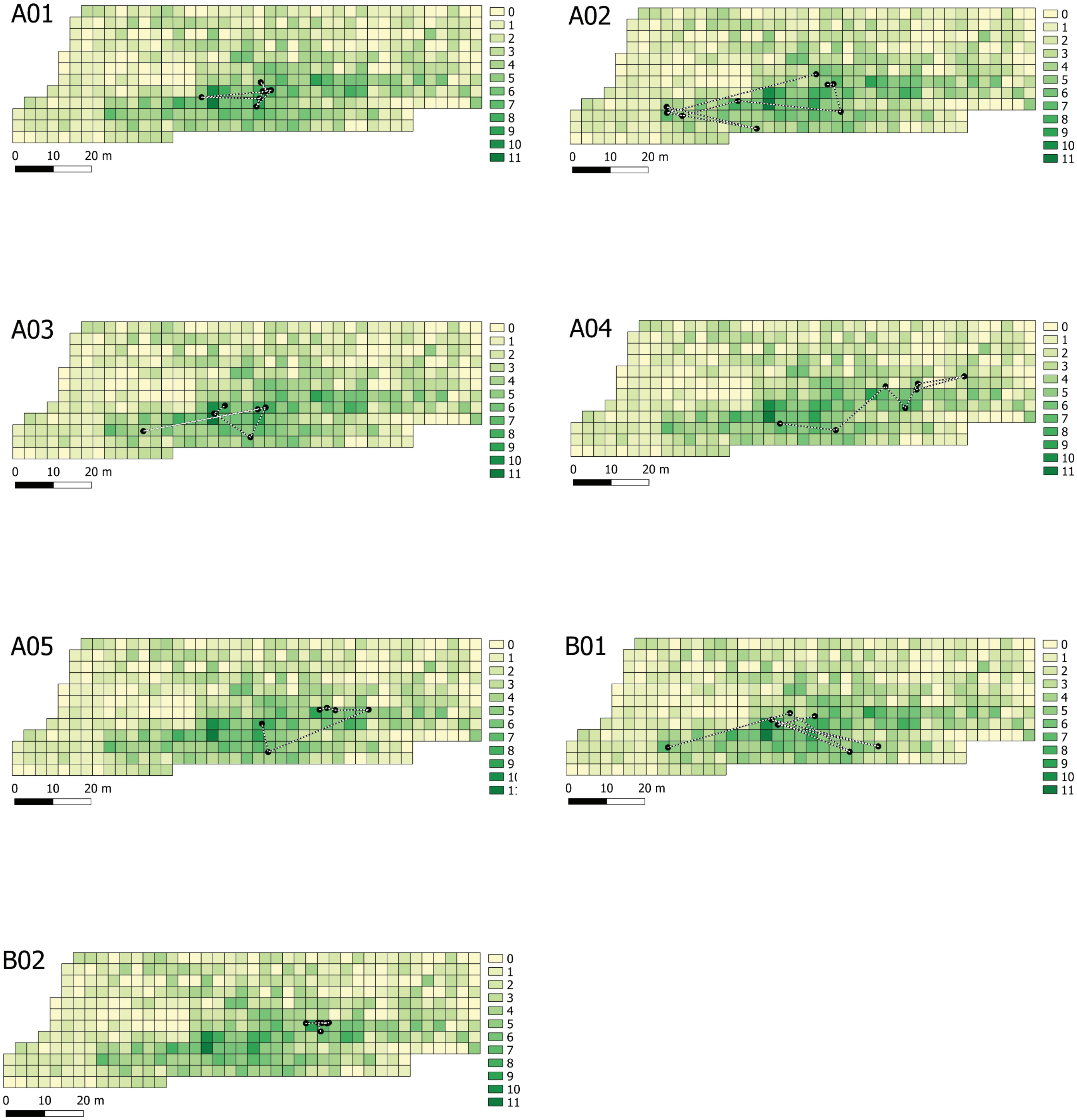

Fig. 3. Displacements of individuals monitored (lines and points) and abundance of prey in the grid cells (green scale).

case of discontinuous prey density, the possibility of observing more than one individual at a short distance becomes possible, and this could at least partially explain other aggregation events reported by Kaltenbach (1970), Fontana and Cussigh (1996), Carrière (2004), and Anselmo (2019).

The marking method was fairly efficient; in most cases, it was possible to spot individuals from a considerable distance. However, not all individuals were recaptured in every survey. This could be due to the difficulty encountered in spotting the tags at ground level, when individuals were among the bottom of the grass, or because they may have moved away from the study area; it is also possible that some individuals died due to predation or a natural decrease in abundance.

Predation on *Sphex funerarius* represents the first such observation made in nature and indicates that *S. pedo* can feed on other orders of insects in addition to Orthoptera and Mantodea, as was believed until recently (Kaltenbach 1970, Fontana and Cussigh 1996, Massa et al. 2012, Lemonnier-Darcemont et al. 2016). Conservation of *S. pedo* should include efforts to maintain high availability and diversity of prey through high biodiversity in grasslands.

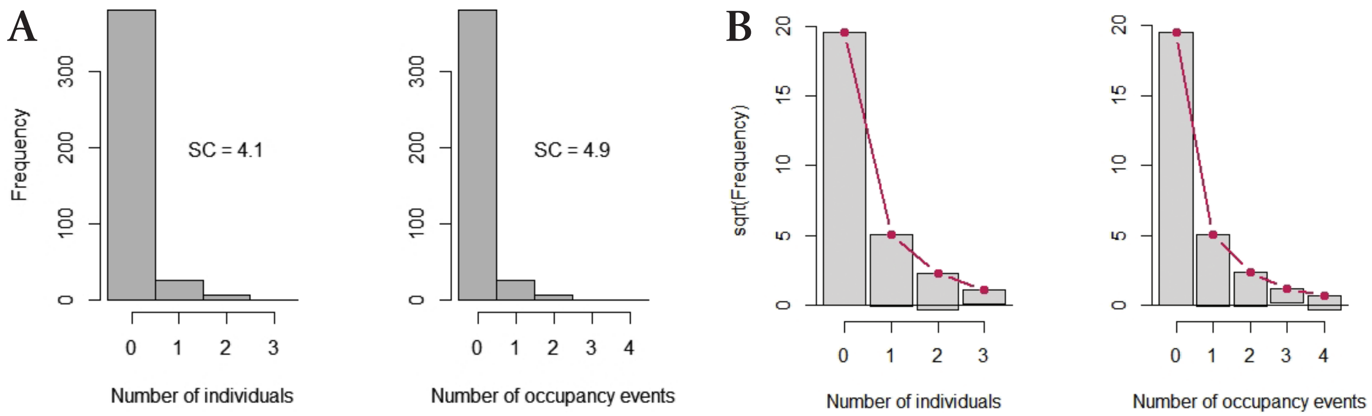

Fig. 4. A. Frequency distribution of count data in the grid cells (SC =skewness coefficient). **B.** Hanging rootograms of best models. Expected counts are shown by the thick line and observed counts are shown as bars. Bar not reaching the zero line indicates that the model over predicts the count; bar exceeding the zero line indicates that the model under predicts the count.

Table 2. Results of the models. The best supported covariate combination for each model is shown with the corresponding AIC score. Letters correspond to the following covariates: A = abundance of potential prey; B = percentage of bare soil; C = average height of the grass; D = presence/absence of tree or shrub.

Model	Number of individuals		Number of occupancy events	
	Covariate	AIC	Covariate	AIC
ZIP	A	171.6	A	176.9
ZINB	A + B + C + D	171.9	A	178.8

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