Graellsia, 78(1): e160
enero-junio 2022
ISSN-L: 0367-5041
https://doi.org/10.3989/graellsia.2022.v78.329

A METHOD TO ESTABLISH THE VIABILITY OF THE *AMBLYOCARENUM* TRAPDOOR SPIDER POPULATIONS IN URBAN AREAS: A PILOT STUDY

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ABSTRACT

Amblyocarenum walckenaeri (Lucas, 1846) (Araneae: Mygalomorphae: Nemesiidae) is found in various urban locations in La Safor (Valencia, Spain). We here present a method that might expose the disturbing effect of rapid urban expansion on trapdoor spider populations: counting the year classes of a population as an indicator of its viability. Our results show that most of the sampled urban populations did not include spiderlings. This suggest that *Amblyocarenum* occur in these urban areas as remnant; aging populations that have difficulties to recruit new generations of specimens and are therefore not viable.

Keywords: Urbanization; mygalomorph spiders; ecological indicator; land cover; La Safor.

RESUMEN

Un método para establecer la viabilidad de las poblaciones de la araña trampera *Amblyocarenum* en áreas urbanas: un estudio piloto.

Amblyocarenum walckenaeri (Lucas, 1846) (Araneae: Mygalomorphae: Nemesiidae) se encuentra en varias zonas urbanas de La Safor (Valencia, España). Aquí, presentamos un método que podría exponer el efecto perturbador de la rápida expansión urbana en poblaciones de arañas tramperas: contando las clases de edades de una población como indicador de su viabilidad. Nuestros resultados muestran que la mayoría de las poblaciones urbanas muestreadas no presenta crías. Esto sugiere que *Amblyocarenum* existe en dichas zonas urbanas como remanentes; poblaciones envejecidas que presentan dificultad a la hora de reclutar nuevas generaciones de ejemplares, tornándose inviables.

Palabras clave: Urbanización; arañas migalomorfas; indicador ecológico; uso del suelo; La Safor.

Recibido/*Received*: 26/08/2021; **Aceptado/*Accepted***: 28/12/2021; **Publicado en línea/*Published online***: 16/03/2022

Cómo citar este artículo/*Citation*: Calatayud-Mascarell, A. & Migueles-Miralles, L. 2022. A method to establish the viability of the *Amblyocarenum* trapdoor spider populations in urban areas: a pilot study. *Graellsia*, 78(1): e160. https://doi.org/10.3989/graellsia.2022.v78.329

Introduction

In the last century, the urbanization process has transformed, destructed and fragmented the natural environment on an unprecedented scale (Czech *et al*., 2000). This is particularly alarming in one of the most biodiverse regions of the Mediterranean, the Iberian Peninsula, where especially its coastal habitats have suffered a high rate of human activities, mainly driven by urban development (Gómez-Pina *et al*., 2002). Urbanization is a relatively recent process in the eastern coast of Spain, associated with tourism proliferation (Miralles i Garcia *et al*., 2012). Therefore, this human footprint is novel for native species persisting in coastal areas (Mason *et al*., 2016). Anthropic habitats impose a variety of novel conditions such as reduced predation risk, unusual resources or drastic alterations of the chemosphere by the use of herbicides and pesticides (Gering & Blair, 1999; Ditchkoff *et al*., 2006; Candolin & Heuschele, 2008). These new habitats generate different selective pressures from the natural ones, that are exploited by particular species which are simultaneously found in natural habitats (Iglesias-Carrasco, 2017).

Sedentary species require smaller areas and are less sensitive to habitat fragmentation than species with medium high dispersal abilities (Thomas, 2000). The infraorder Mygalomorphae (tarantulas and trapdoor spiders), and in particular, trapdoor lineages, are typically habitat specialists, extremely sedentary, except for the roaming males, while females remain in their home burrows to live and reproduce several consecutive years (Pérez-Miles & Perafán, 2007). These life-history traits promote geographical isolation and enable mygalomorph spiders to persist in small isolated populations (Bond *et al*., 2001). Longevity increases

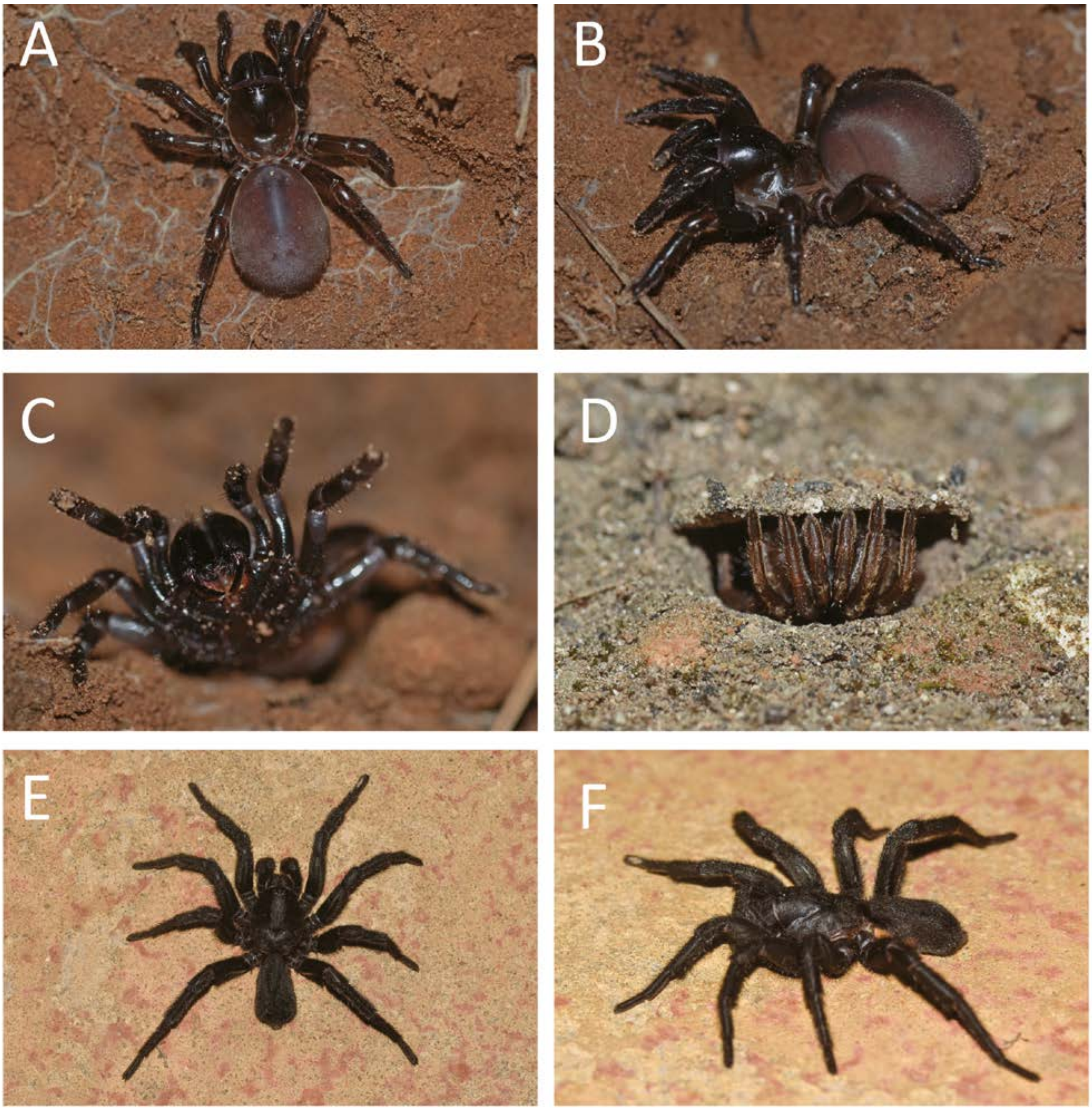

Fig. 1.– *Amblyocarenum walckenaeri*, living female photographed in the field and dead male photographed "ex situ". A) adult female dorsal view; B) adult female lateral view; C) adult female threatening position; D) spider opening the trapdoor; E) adult male dorsal view; F) adult male lateral view.

Fig. 1.– *Amblyocarenum walckenaeri*, hembra viva fotografiada en el campo y macho muerto fotografiado "ex situ". A) hembra adulta, vista dorsal; B) hembra adulta, vista lateral; C) hembra adulta, posición amenazante; D) araña abriendo la trampilla; E) macho adulto, vista dorsal; F) macho adulto, vista lateral.

the probability of mygalomorphs to temporarily persist as remnant populations. However, aging populations that can no longer recruit are, therefore, not viable in the long term (Mason *et al*., 2016).

Trapdoor spiders are represented in the Iberian Peninsula by the genera *Nemesia* Audouin, 1826, *Iberesia* Decae & Cardoso, 2006 and *Amblyocarenum* Simon, 1892, in the family Nemesiidae and *Ummidia* Thorell, 1875 in the family Halonoproctidae (World Spider Catalog, 2021). *Amblyocarenum* differs from *Nemesia* and *Iberesia* by the absence of a clasper-spur on the male tibia I, the absence of a cheliceral rastellum in both sexes, the relatively wide eye-formation, relatively strong development of the spinnerets and the presence of a variable, but generally procurved fovea. *Amblyocarenum* differs from *Ummidia* by the absence of a saddle depression in the dorsal tibia III, dense lateral fields of short curved spines on the distal anterior legs and palps in females, and the presence of densely scopulated tarsi and metatarsi (Nentwig et al., 2021). The spiders of the genus *Amblyocarenum* are medium sized mygalomorphs, living in silk-lined burrows. Females persist their whole life in their retreat, assuming a threatening position if extracted from the burrow (Fig. 1A-C). Burrows are closed with a thin hinged trapdoor, which the spider opens slightly pending the passage of prey (Fig. 1D) (Decae *et al*., 2014). Males, however, leave the burrow when they reach adulthood and can be found wandering in search of a female during the mating period (Fig. 1E-F). *Amblyocarenum walckenaeri* (Lucas, 1846) is the only known representative of the genus in the Iberian Peninsula (World Spider Catalog, 2021), mostly distributed in the southern and eastern of Spain, with some records from central Spain (Decae *et al*., 2014; Branco *et al*., 2019).

In this pilot study, we measured the viability of *A. walckenaeri* populations in areas of different anthropic impact of La Safor region (Valencia, Spain) by analyzing the composition of year classes through the study of trapdoor sizes. This method might expose the disturbing effect of the rapid urban expansion of the area on trapdoor spider populations.

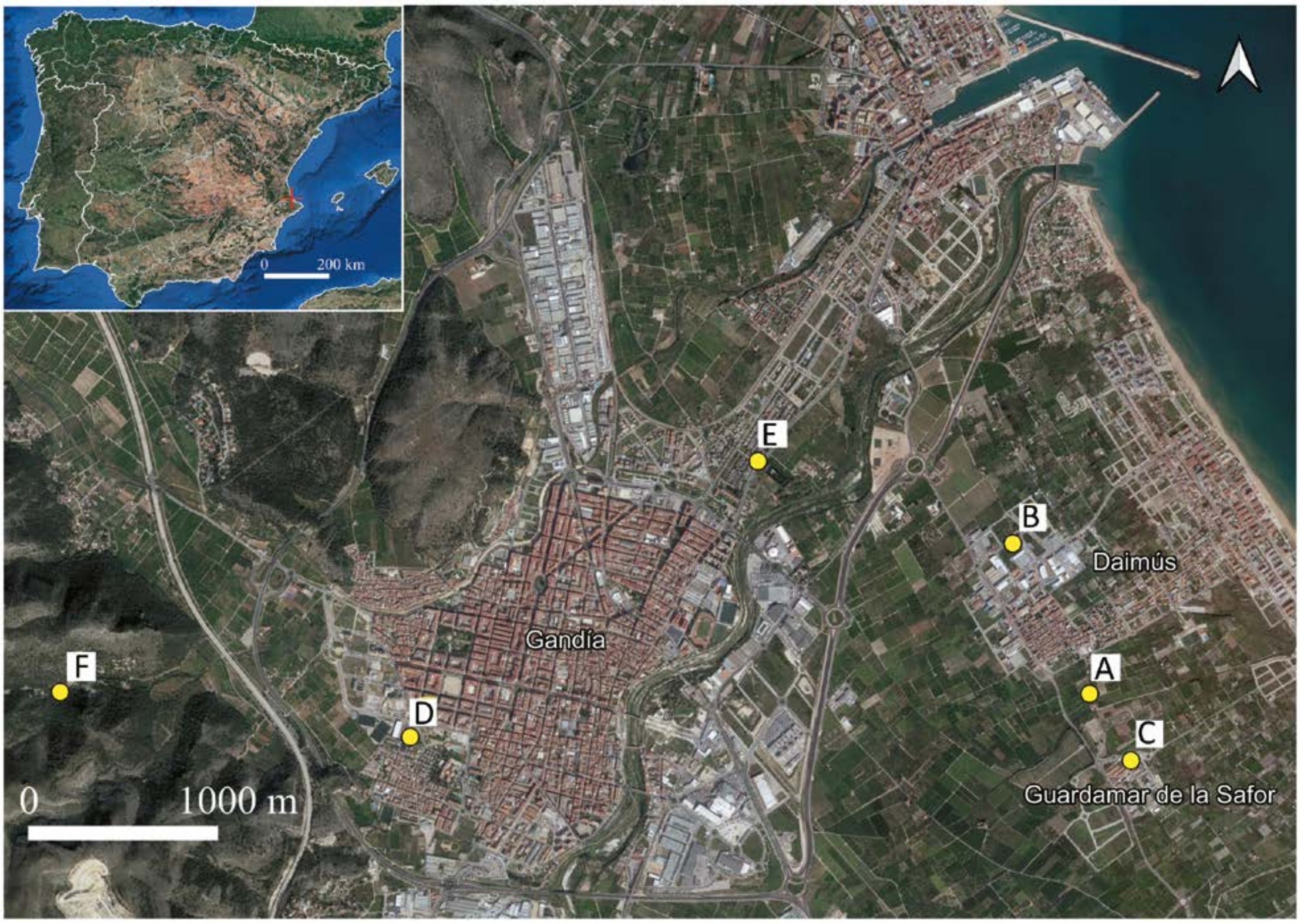

Fig. 2.– Map of the sampled localities. A) citric orchards in urban peripheries; B) abandoned land within an industrial area; C) abandoned land in a dogs park; D) garden hedges of an urban park; E) garden hedges of sidewalks; F) mountain slope of Serra de la Falconera.

Fig. 2.– Mapa de las localidades muestreadas. A) Cultivos de naranjos en la periferia urbana; B) terreno abandonado junto a área industrial; C) terreno abandonado en un parque para perros; D) setos de un parque urbano; E) setos de una acera; F) pendiente de montaña en la Serra de la Falconera.

Material and methods

Field sampling took place on 13th and 14th November 2021, in the coastal localities of Gandía, Daimús and Guardamar de la Safor (Valencia, Spain) (Fig. 2).

The anthropic environments where the species is present range from urban parks to abandoned lands and orchards. The most important orchard in the coastal localities of Valencia is citric trees, usually located at urban peripheries (Fig. 3A). The trees grow in little ground mounts to avoid floods. These ground mounts harbour the typical burrows of the species. Abandoned lands in the sampled localities consist of isolated patches covered by native vegetation like the wild blackberry (*Rubus ulmifolius*) or invasive vegetation, such as the sugarcane (*Saccharum officinarum*) or the bermuda buttercup (*Oxalis pes-caprae*). The burrows of *A. walckenaeri* were found in the ground edges of

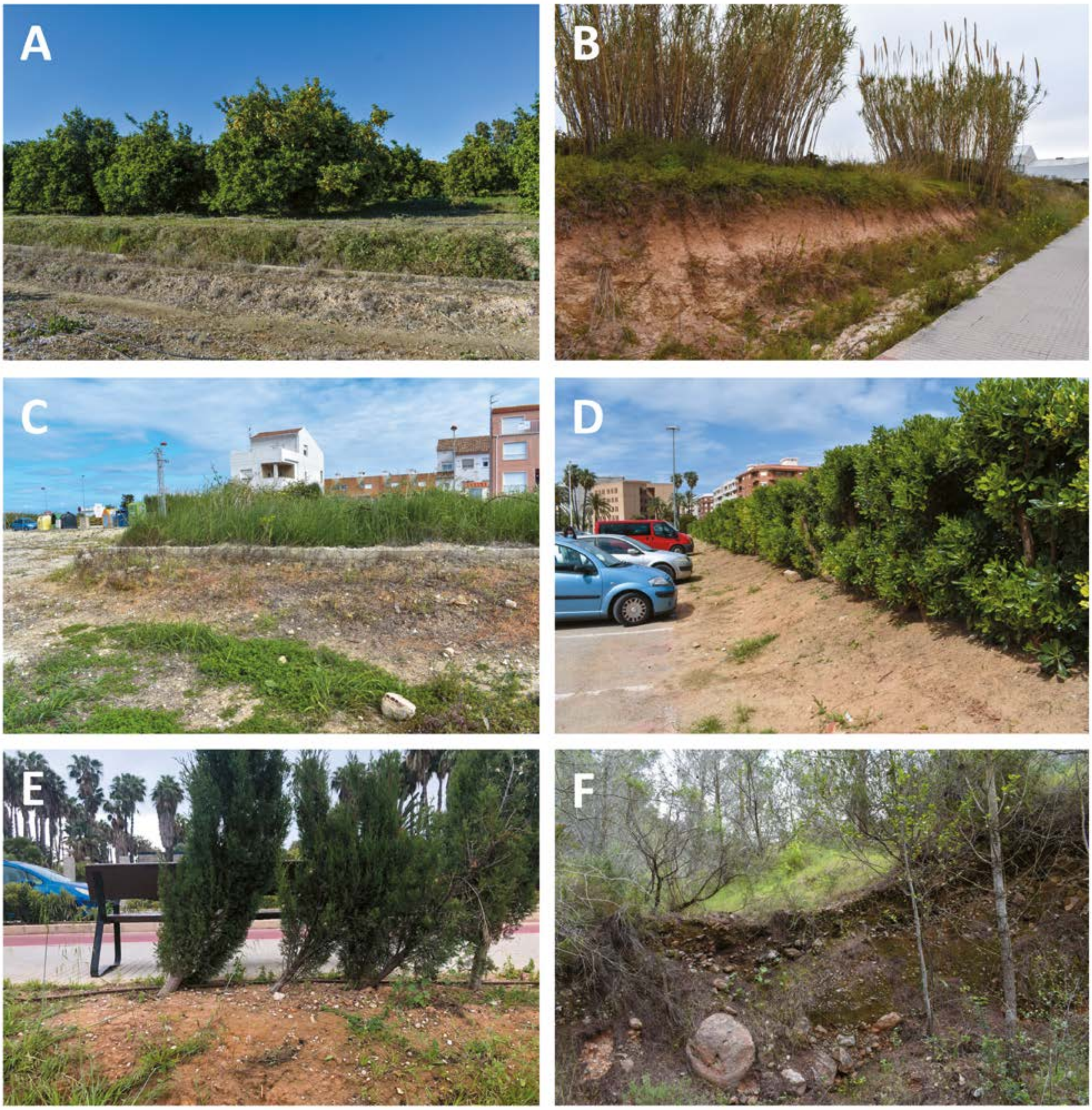

Fig. 3.– Urban and natural *Amblyocarenum walckenaeri* habitat photographs. A) citric orchards in urban peripheries; B) abandoned land within an industrial area; C) abandoned land within a dogs park; D) garden hedges of an urban park; E) garden hedges of a sidewalk; F) mountain slope of Serra de la Falconera.

Fig. 3.– Fotografías de los hábitats urbanos y naturales de *Amblyocarenum walckenaeri*. A) Cultivos de naranjos en la periferia urbana; B) terreno abandonado junto a área industrial; C) terreno abandonado en un parque para perros; D) setos de un parque urbano; E) setos de una acera; F) pendiente de montaña en la Serra de la Falconera.

the patches. These ground edges can be almost vertical and higher, like in an abandoned land at the industrial area of Daimús (Fig. 3B) or subtle and lower, like in the dogs park of Guardamar de la Safor (Fig. 3C). In urban parks, burrows were found at the borders, in ground ramps close to the garden hedges (Fig. 3D). In addition, the species is also present in garden hedges of sidewalks (Fig. 3E). The closest natural habitat in the sampled localities is Serra de la Falconera, a small group of low mountains placed 6 km from the coast. In this habitat, burrows of *A. walckenaeri* were frequently seen in small mountain slopes (Fig. 3F).

Samplings consisted of measuring all burrows found in a 50 m^2 transect (1 m long × 50 m wide; except for Serra de la Falconera habitat, which was 2 m long × 25 m wide, due to longer slopes) in each environment where burrows were detected.

In order to establish the viability of populations, we compared the size of trapdoors between the natural habitat (considered as the ideal population) and each of the anthropic populations using the Mann-Whitney-Wilcoxon test (Gehan, 1965). Normality was previously rejected with the Kolmogorov-Smirnov test (Lopes *et al.*, 2007). Analyses and graphics were performed with RStudio software (RStudio Team, 2020). Maps were performed with the geographic information system QGIS 3.10.

Results

A total of 250 *A. walckenaeri* trapdoors were measured in the anthropic areas and the natural habitat. Only the citric orchards (A) and the natural population (F) contain spiderlings (small trapdoors), whereas adult and subadult spiders were present in all populations (medium to big trapdoors). Trapdoor size range and number of burrows of each environment is summarized in Table 1. Boxplots of each environment including the burrow diameters are showed in Fig. 4.

We found significant differences regarding the mean size of the trapdoors between each anthropic population and the natural one, with p-values as follows: 0.0043 in the citric orchards (A); $5.604e^{-8}$ in the abandoned land within an industrial area (B); $1.996e^{-10}$ in the abandoned land within a dogs park (C); $1.631e^{-10}$ in the garden hedges of an urban park (D) and $2.199e^{-9}$ in the garden hedges of a sidewalk (E).

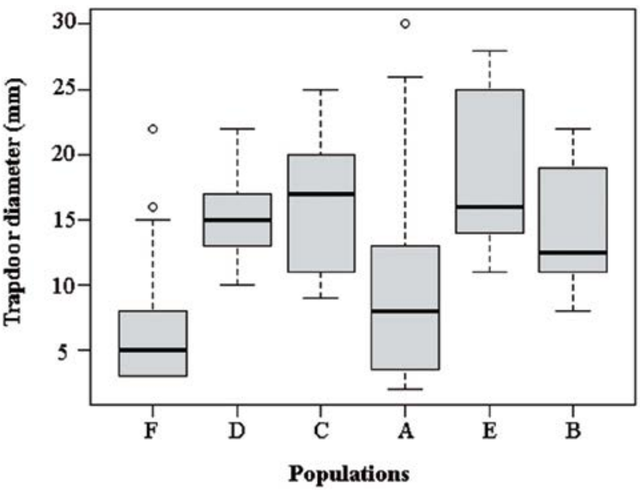

Fig. 4.– Box plot showing the trapdoor diameters of each population sampled. The letters from the horizontal axis correspond to the letters in figures 2 and 3.

Fig. 4.– Diagrama de cajas mostrando los diámetros de las trampillas de cada población muestreada. Las letras del eje horizontal corresponden a las letras de las figuras 2 y 3.

Discussion

Trapdoor spider populations constitute a perfect model for studying the consequences of a rapid urban expansion. Except the citric orchards, none of the measured urban populations contain spiderlings, the burrows range from medium to big (Fig. 5A). In contrast, burrows from the natural populations of Serra de la Falconera and specially from the citric orchards, present a wide range of sizes, showing the presence of spiderlings and different age groups (Fig. 5B) (Table 1) (Fig. 4). Recruitment is apparent when spiderlings and different age groups are well represented in a population. The presence of different age groups is determined by the size of burrows (Mason *et al.*, 2016). The absence of small burrows in most anthropic populations indicates the lack of spiderlings, having severe difficulties to recruit new generations of specimens and being inviable in the long-term. By contrast, the natural and the citric orchard populations present recruitment and seem to be viable. Up to approximately 100 spiderlings were observed emerging from an *Amblyocarenum* nest (Marta Calvo pers. obs.). However, the biology of the early stages of *Amblyocarenum*, including how long spiderlings remain in the maternal burrow before dispersal, and the mortality rates of juveniles before they successfully make their burrow, is still unknown (Decae *et al.*, 2014). In any case, a large percentage of small burrows would be expected in a viable population. Considering this fact, the natural population of Serra de la Falconera would be an ideal population due to the large number of spiderlings found; 75% of burrows (n= 92) were smaller than 5 mm. The other viable population was found in a citric orchard in the urban peripheries of Daimús. Nevertheless, the results show that this habitat seems to be suboptimal, due to a significantly smaller number of spiderlings; 40 % of burrows (n= 26) were smaller than 5 mm. Several studies have reported relatively high diversity and abundance of arthropods in citric orchards from all over the world (Deshmukh & Chaudhari, 2016; Gama, 2017; Michael *et al.*, 2021), and especially

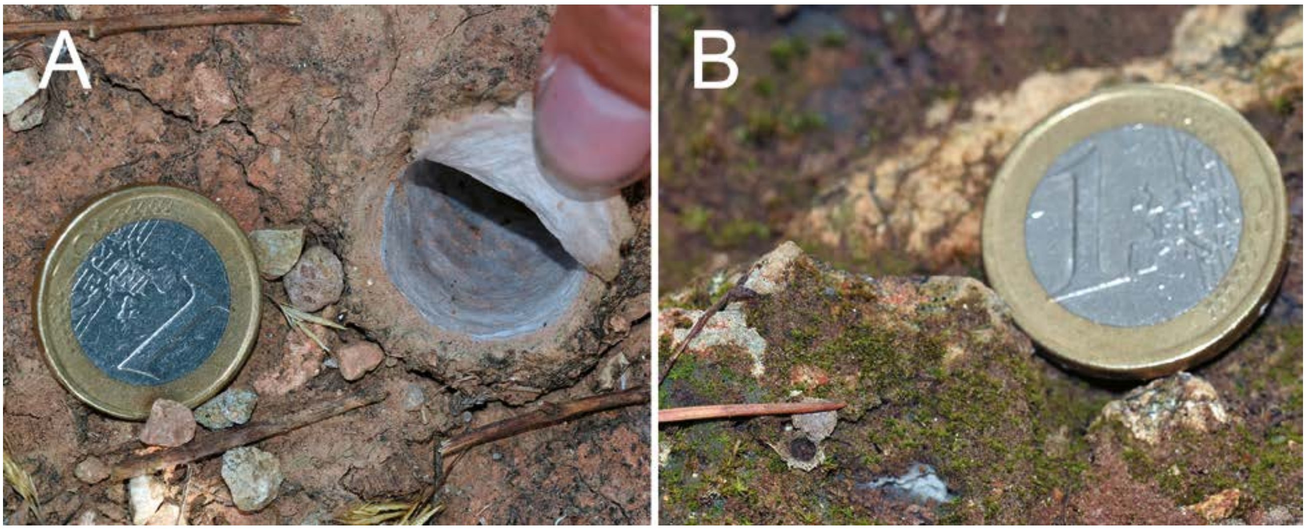

Fig. 5.– Trapdoor burrows, size comparison. A) Big sized burrow from a garden hedge of a sidewalk; A) Small sized burrow from Serra de la Falconera.

Fig. 5.– Comparación de los tamaños de los nidos. A) Nido grande en los setos de una acera; A) Nido pequeño en la Serra de la Falconera.

in the Valencian ones (González *et al*., 2008). Citric orchards of the studied area have formed large areas of potential habitat for *A. walckenaeri* for reasons such as: they are usually connected, placed at the urban peripheries and remain relatively stable through time, thus reducing the risk of mortality related to dispersal; they also provide a suitable habitat for the species with humidity, slopes and abundance of different preys.

In the last two decades, human development has changed dramatically the land uses in the sampled areas; citric orchards have decreased, continuous urban fabric has appeared, and industries have been established (Fig. 6). Since citric orchards seem to be suitable for *A. walckenaeri*, urban fabric and industries could act as a barrier between natural and urban populations, fragmenting the habitats even more. The roaming males leave their burrows after the last moult to mate, traveling large distances until they die (Buchli, 1966; Decae *et al*., 2014). The dispersal period seems to be the most dangerous period for males, increasing the probabilities of a premature death before they mate. In fact, many males got trapped in houses or swimming pools, presumably attempting to traverse between patches (Decae *et al*., 2014; Tamajón Gómez *et al*., 2018). Drownings seem to be extreme during the tempest nights of summer; even three different males were observed in the same pool during the same day (pers. obs.). Also, their matting period is in summer, when the human population of the area is quadruplicated due to massive tourism (Instituto Nacional de Estadística, 2021). If reproductive rates had subsequently been reduced due to higher male mortalities because of the urban expansion, remnant populations would have formed. Considering high mobility of males and the rapid urban expansion, future studies on the species dispersal will shed light into the population dynamics of the species in urban environments.

As a pilot study, this work attempts to establish the firsts steps into the conservation of the Mediterranean endemic *Amblyocarenum walckenaeri*. Additional studies in different populations following this method are expected to show if the remnant populations would be the rule rather than the exception. Furthermore, the methodology of the study could also be applied

Table 1.– Number of burrows and range diameter of trapdoors from the sampled populations. All measurements are in millimeters.

Tabla 1.– Número de nidos y rango del diámetro de las trampillas de las poblaciones muestreadas. Todas las medidas están en milímetros.

Location	Trapdoor diameter (mm)	N° of burrows
A) citric orchards in urban peripheries	3–30	64
B) abandoned land within an industrial area	8–22	14
C) abandoned land within a dogs park	9–25	18
D) garden hedges of an urban park	10–22	17
E) garden hedges of a sidewalk	11–28	14
F) mountain slope of Serra de la Falconera	3–22	123

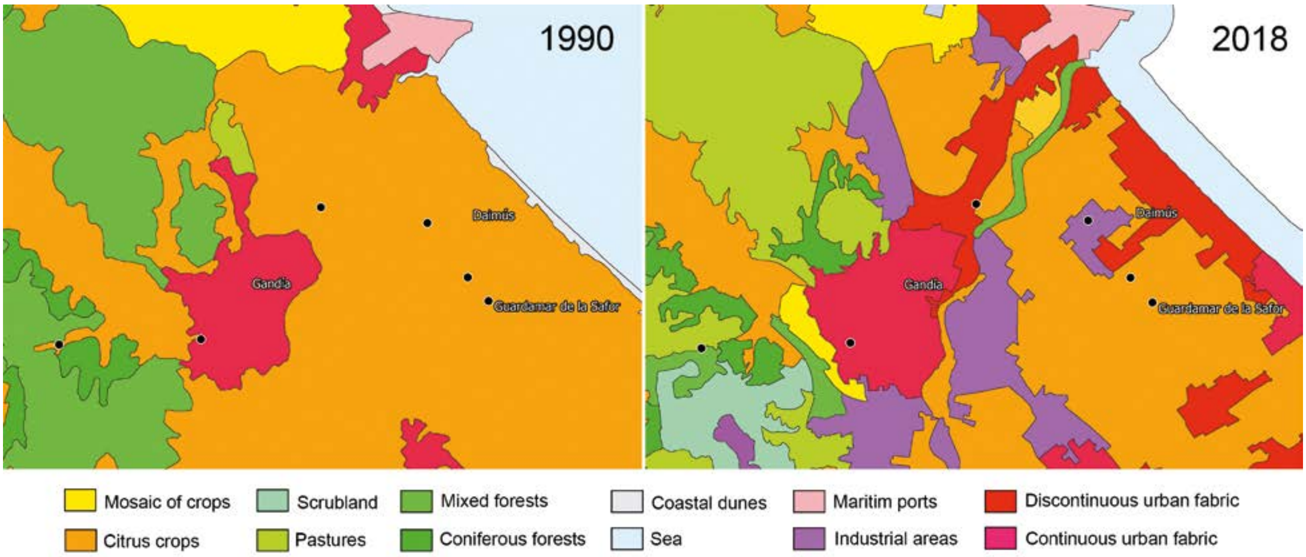

Fig. 6.– "Corine Land Cover" comparison between 1990 and 2018 in the sampled localities of Gandía, Daimús and Guardamar de la Safor (Valencia, Spain). Black dots represent the habitats of figure 2. Maps are available at "Centro de descargas del CNIG".

Fig. 6.– Comparación del "Corine Land Cover" entre 1990 y 2018 en las localidades muestreadas de Gandía, Daimús y Guardamar de la Safor (Valencia, España). Los puntos negros representan los hábitats de la figura 2. Los mapas están disponibles en el "Centro de descargas del CNIG".

to other species in the highly diverse Iberian trapdoor spider fauna and elsewhere in the Mediterranean biodiversity hotspot. Another point to take into consideration is that incorporation of molecular data in challenging groups such as mygalomorphs spiders has disclosed many previously overlooked species (Hamilton *et al*., 2011; Ferretti *et al*., 2019). In this sense, despite the relatively broad range extension of the species, molecular approaches in *A. walckenaeri* could uncover independent lineages, restricting its distribution range and considering them as single conservation units.

Acknowledgements

We are deeply thankful to Alberto Sánchez Vialas, José Antonio Luis de la Iglesia and Arthur E. Decae for their advice and improvement of the manuscript. Thanks also to Susana Ortiz and María Fàbregas for providing the drowned males, to Cristian Ibáñez for his help during fieldwork and to Roser Mascarell for English suggestions.

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