Study of the helminth fauna in eagle owl (Bubo bubo) in the south of Spain

R. Zafra, F.J. Martínez-Moreno *, P.J. Rufino-Moya, P.N. Gutiérrez, S. Martínez-Cruz, L. Buffoni, A. Martínez-Moreno, I. Acosta

Animal Health Department (Parasitology and Parasitic Diseases), Faculty of Veterinary Medicine, University of Córdoba, Spain

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ABSTRACT

In this study we show the results of the eagle owls’ (Bubo bubo) helminthfauna found in Andalusia. A total number of 50 specimens have been analysed in a period of 10 years (from 2011 to 2020). Prevalence (P%), mean intensity (Ia) and mean abundance (Aa) of parasitation have been obtained. The percentage of parasitation in the total sample was 80% (40 out of 50 eagle owls): 78% nematodes, 8% trematodes, 6% cestodes and 4% acantocephalans. 7 species of helminths were identified: 6 nematodes, and 1 trematode. In the case of cestodes and acantocephalans it was not possible to determine species and only the genus was identified.

The intestinal nematode Capillaria tenuissima (P% = 58% (44–71.2); Ia = 11.52 (5.83–28.9)) was the core species whereas Synhimantus laticeps (P% = 16 (7.5–28.8); IaM = 4 (1.75–7.25)) and Harteria hispanica (P% = 16 (7.5–28.8); IaM = 1.5 (1–2)) were the secondary species. The remainder species were considered satellite species, with low prevalence and average abundance. Likewise, descriptive parameters of the helminth community were determined: species richness, 1.56 (1.29–1.94), total abundance, 12 (7.24–26.40), Brillouin’s diversity index, 0.18 (0.10–0.29) and Berger-Parker dominance index, 0.88 (0.81–0.93).

The data from this study show a non-diverse helminthic community, without species dominance with C. tenuissima as the central species, followed by S. laticeps and H. hispanica as secondary species. Worth mentioning is the presence of H. hispanica, which is considered an endemic species in Spain and specifically in Andalusia.

To the authors’ knowledge, this is the largest population sample taken in parasitological studies about helminths of this raptor in Europe and the first one carried out in the south of Spain (Andalusia).

1. Introduction

The eagle owl (B. bubo) is the largest nocturnal raptor in Europe. As a predatory species, it is at the top of the food chain and is susceptible to acquire parasitic infections mainly by ingesting prey species that act as intermediate hosts for parasites. Factors such as diet, accessibility, and variety of prey species determine the parasite fauna in a specie in each territory (Penteriani and Delgado, 2016). The comparative analysis of the parasites found in specific species in different geographical areas can provide interesting information on aspects related to the ecology of the populations.

The eagle owl (B. bubo) population extends over most of Eurasia and North Africa, with an estimated 12,000–42,000 pairs in Europe (Penteriani and Delgado, 2016). In Spain, there are estimated 2345 pairs, and they can be found almost everywhere on the Iberian Peninsula, with a smaller presence in the northwest (Martínez-Climent and Zuberoigotia, 2003). Its distribution is wide, as it adapts well to diverse habitats, from semi-desert areas without trees to forests and cities, with a preference for areas without too much lush vegetation to be able to fly comfortably (Penteriani and Delgado, 2016).

However, a series of factors have a negative effect on their presence, such as direct persecution, electrocution, and impacts with fences and wire fences (Martí and Del Moral, 2003). Another important aspect that has a major influence on eagle owl populations is the scarcity of the common rabbit (Oryctolagus cuniculus). Inadequate hunting management, the degradation of scrubland areas as well as the impact of infectious diseases, such as haemorrhagic fever and myxomatosis, have led to a significant decline of this mammal, which is the most important prey of the eagle owl (Martí and Del Moral, 2003). Despite this trophic preference, the eagle owl is highly adaptable to food availability and, thus, it is known that its diet can be made up of different species of mammals, birds, reptiles, amphibians, fish, and even invertebrates.

* Corresponding author.
E-mail address: fjmartinez@uco.es (F.J. Martínez-Moreno).

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Specifically in Spain, its main prey are common rabbits, rats, and partridges. In this sense, it has a special predilection for preys with a low physical condition, which in the specific case of rabbits can discriminate through the lower reflectance of their tails (Penteriani and Delgado 2016).

Birds of prey, both diurnal and nocturnal, are usually protected species and this fact means that there are few studies on them. Parasitological studies in particular are quite scarce due to this fact and because samples often come from animals collected dead by wildlife rescue centres. As a consequence, the conditions of analysis are not suitable and these parasitological studies cannot reach the level of species identification often.

To the authors’ knowledge, there are only 5 papers providing prevalence data by helminth species specifically on the eagle owl. Only 2 of them quantify parasitism phenomena (Komorova et al., 2017; Sitko and Heneberg 2019). Many of them have been carried out on strigiformes, including some eagle owls. Sometimes, the sample size is limited to 2–5 specimens (Papazahariadou et al., 2008; Komorova et al., 2017). The most extensive studies on strigiformes in Europe cites the analysis of 32 eagle owl specimens (Sitko and Heneberg, 2019) and 34 specimens in Austria (Kutzer et al., 1982). In Spain, similar studies have been carried out, but the number of animals analysed has been much smaller (7 specimens) with only generic identification (Ferrer et al., 2004). Therefore, very limited information on the helminth fauna of this raptor is known to date.

The aim of this study was to deepen the knowledge of the helminth fauna of the eagle owl (B. bubo) by analysing 50 samples in our laboratory over a period of 10 years. This is the first study on the helminth fauna of eagle owls carried out in southern Spain (Andalusia) with the largest number of samples in Europe.

2. Material and Methods

2.1. Samples

A total of 50 eagle owls have been analysed in the period from 2011 to 2020. The raptors came from wildlife rescue centres in the Andalusian provinces of Cordova, Jaen, Seville and Malaga. The causes of death in most cases were related to collisions with fences, wire fences, and in some cases electrocution. Once received in the laboratory, they were identified and frozen at −24 °C in plastic bags for subsequent necropsy.

The processing of the samples started with preliminary thawing. Subsequently, the digestive tract (oesophagus, proventriculus, gizzard, and intestine) was dissected, and the content was washed with saline and placed in settling cups. After that, both the digestive contents and the mucosal surface of the organs were examined under a stereoscopic microscope for helminth identification. The same process was also followed with the liver, the gall bladder, the heart, and lungs. When helminths were detected, they were washed in saline and preserved in 70% ethanol. Finally, for complete species identification, they were mounted in Aman’s lactophenol, and conventional identification keys were followed.

2.2. Parasitological study

2.2.1. Ecological parameters and biological index

For the study of the helminth community, ecological terms were expressed following Bush et al. (1997) for the eagle owls collected in the 10-year period (2011–2020). Core, secondary, and satellite species were determined based on prevalence and abundance (Bush and Holmes, 1986a,b). Thus, core species are those common (present on most hosts) and abundant; satellite species are found on few hosts and low in abundance; and secondary species are those with intermediate characteristics in the community.

The following parameters were used to analyse the characteristics of the helminth community in the owls studied: Species Richness (S); Total Abundance; Brillouin diversity index; and Berger-Parker dominance index.

2.3. Statistical study

Descriptive parameters (prevalence, mean intensity, and mean abundance) were obtained using QPweb version 1.0.14 (Quantitative Parasitology on the web) (Reiczigel et al., 2019). Prevalence was calculated with 95% confidence interval (95% CI) using Sterne’s exact method. For mean intensity and abundance, confidence limits (95% CI) were obtained by the accelerated bias-corrected bootstrap method using 2000 replicates.

Past 4.10 (Paleontological Statistics) was used to calculate the biodiversity indices (Species richness, Total Abundance, Brillouin diversity, and Berger-Parker dominance).

3. Results

3.1. General results

The most prevalent helminths found were nematodes (78%), followed by trematodes (8%), cestodes (6%) and acanthocephalans (4%). The total prevalence of parasitism classified by helminth groups is shown in Table 1.

In the present study, 12 different genera of helminths belonging to 4 taxonomic groups were found: Nematodes, Cestodes, Trematodes, and Acanthocephalans (Table 2). It was not possible to determine the species in some of the helminths studied, namely in the nematode group for Microtetrameres spp. and in some specimens of Synhimantus sp. Thus, it was not possible to identify the species in 17 eagle owls (15 of them parasitised by females of Synhimantus sp. in which the species could not be identified and the remaining 2 parasitised by Microtetrameres sp). On the other hand, with respect to trematodes, there were 3 eagle owls in which the genus could not be identified, reaching only the taxonomic group due to the high degree of decomposition of the parasites. Likewise, for cestodes and acanthocephalans, we were only able to identify the genus (Paruterina sp. in 3 eagle owls and Centrorhyncus sp. in 2 respectively).

A total of 7 helminth species were completely identified: 6 nematodes and 1 trematode (Table 3). Considering the above, the species with the highest prevalence was C. tenuissima (58%), followed by S. laticeps (16%), and H. hispanica (16%). The remaining species (Synhimantus affinis, Cyrnea spinosa, Cyrnea ficheiri and Strigea falconis) showed a prevalence of less than 10%.

3.2. Helminth community structure

As is common in parasitic infections, individuals often harbour more than one helminth species. Thus, of the 40 owls parasitised, 13 were parasitised by more than one species. The frequency distribution according to the number of species found in each animal is shown in Fig. 1.

The total number of helminths collected from the parasitised eagle owls was 521. From these parasites, 409 helminths were completely identified and for the remaining 112 only the genus could be determined (Synhimantus sp. = 98; Paruterina sp. = 3; Unidentified Trematoda = 6; Table 1)

| Helminth Group | N | Prevalence (%) |
|----------------|---|----------------|
| Nematoda       | 39 | 78             |
| Trematoda      | 4  | 8              |
| Cestoda        | 3  | 6              |
| Acanthocephala | 2  | 4              |
| Total Owls Parasitised | 40 | 80            |
Table 2
Helminths found in the eagle owls examined for the period 2011–2020. Numbers in parentheses are the 95% confidence intervals of each parameter.

| Taxonomic Group | Species                  | N  | P%  | \( \text{M} \) | \( \text{M} \) |
|-----------------|--------------------------|----|-----|----------------|-------------|
| Nematoda        | Capillaria tenuissima     | 33 | 58 (44.71.2) | 11.52 (5.83–28.9) | 6.58 (3.4–16.4) |
|                 | Synhimanus affinis        | 2  | 4 (0.7–13.7)  | 6.5 (2–6.5)    | 0.26 (0–1)  |
|                 | Synhimanus laticeps       | 8  | 16 (7.5–28.8) | 4 (1.75–7.25)  | 0.64 (0.22–1.51) |
|                 | Synhimanus sp²            | 15 | 30 (18.6–44)  | 6.53 (2.73–14) | 1.96 (0.78–4.83) |
|                 | Harteria hispanica        | 8  | 16 (7.5–28.8) | 4 (1.75–7.25)  | 0.64 (0.22–1.51) |
|                 | Cyrena spinosa            | 2  | 4 (0.7–13.7)  | 3 (2–3)        | 0.12 (0–0.4) |
|                 | Cyrena ficheuri           | 3  | 6 (1.7–16.7)  | 2 (1–2.67)     | 0.12 (0.02–0.33) |
|                 | Microtetrameres sp²       | 2  | 4 (0.7–13.7)  | 1.5 (1–1.5)    | 0.06 (0–0.18) |
| Trematoda       | Unidentified¹             | 3  | 2 (0.1–10.6)  | 6               | 0.12 (0.02–0.34) |
|                 | Strigea falconis          | 1  | 2 (0.1–10.6)  | 1               | 0.06 (0–0.12) |
| Cestoda         | Paruterina²              | 3  | 6 (1.7–16.7)  | 1               | 0.04 (0–0.1)  |
| Acanthocephala  | Centrorhynchus sp.³       | 2  | 4 (0.7–13.7)  | 1               | 0.04 (0–0.1)  |

N*: number of eagle owls parasitised; P%: prevalence; \( \text{M} \): Mean intensity; \( \text{M} \): Mean abundance.

1, 2, 3 & 4: Not able to reach species level.

*: High decomposition.

Table 3
Helminths species found in the eagle owls examined for the period 2011–2020. Numbers in parentheses are the 95% confidence intervals of each parameter.

| Taxonomic Group | Species                  | N  | P%  | \( \text{M} \) | \( \text{M} \) |
|-----------------|--------------------------|----|-----|----------------|-------------|
| Nematoda        | Capillaria tenuissima     | 29 | 58 (44.71.2) | 11.52 (5.83–28.9) | 6.58 (3.4–16.4) |
|                 | Capillaria tenuissima*    | 4  | 6.5 (2–6.5)   | 0.26 (0–1)    | 0.44 (0–0.88) |
|                 | Synhimanus affinis        | 2  | 4 (0.7–13.7)  | 6.5 (2–6.5)   | 0.26 (0–1)  |
|                 | Synhimanus laticeps       | 8  | 16 (7.5–28.8) | 4 (1.75–7.25) | 0.64 (0.22–1.51) |
|                 | Synhimanus sp²            | 15 | 30 (18.6–44)  | 6.53 (2.73–14) | 1.96 (0.78–4.83) |
|                 | Harteria hispanica        | 8  | 16 (7.5–28.8) | 4 (1.75–7.25) | 0.64 (0.22–1.51) |
|                 | Cyrena spinosa            | 4  | 1.5 (1–2)     | 0.06 (0–0.2)  | 0.26 (0–0.52) |
|                 | Cyrena ficheuri           | 2  | 2.6 (1–3)     | 0.12 (0–0.4)  | 0.28 (0–0.84) |
|                 | Microtetrameres sp²       | 4  | 6 (1.7–16.7)  | 1.5 (1–1.5)   | 0.06 (0–0.18) |
| Trematoda       | Unidentified¹             | 3  | 2 (0.1–10.6)  | 6               | 0.12 (0.02–0.34) |
|                 | Strigea falconis          | 1  | 2 (0.1–10.6)  | 1               | 0.06 (0–0.12) |
| Cestoda         | Paruterina²              | 3  | 6 (1.7–16.7)  | 1               | 0.04 (0–0.1)  |
| Acanthocephala  | Centrorhynchus sp.³       | 2  | 4 (0.7–13.7)  | 1               | 0.04 (0–0.1)  |

N*: number of eagle owls parasitised; P%: prevalence; \( \text{M} \): Mean intensity; \( \text{M} \): Mean abundance.

*: Core species; #: Secondary species.

In addition, to determine the importance of the different species in the eagle owl helminth community, we applied Bush and Holmes (1986a,b) criteria that distinguish between core, secondary, and satellite species based on the prevalence and abundance with which they occur in a host species. Using these criteria, C. tenuissima was clearly identified as a core species with a prevalence of 58% and a mean abundance of 6.58. S. laticeps was assigned the category of secondary species with a prevalence of 16% and a mean abundance of 0.64. A similar status was observed for H. hispanica, with a prevalence of 16% and a mean abundance of 0.12. The remaining helminths were considered as satellite species with a mean prevalence of lower than 10% (Table 3).

As community descriptors, the parameters of Species Richness (S), Total Abundance, and the Brillouin biodiversity and Barger-Parker dominance indices were obtained (Table 4).

No previous data on these parameters in the species in question exist to date, so we cannot establish comparisons with the helminthic communities of eagle owls in other regions. The only available data are those obtained on other Strigiformes species in Calabria (Southern Italy) (Santoro et al., 2012) and will be commented on in the discussion section.

4. Discussion

Considering taxonomic groups the most prevalent helminths in our study were nematodes (78%), followed by trematodes (8%), cestodes (6%), and acanthocephalans (4%). From the total of helminths recovered, a total of 7 helminth species were identified completely: 6 nematodes and 1 trematode (Table 3). In Strigiformes, this predominance of nematodes over other helminths has been previously reported in Spain, specifically in Andalusia (Illescas et al., 1993) and Catalonia (Ferrer et al., 2004). The nematodes prevalence in our results is higher than described both in Andalusia in 1993 (64.3%) and Catalonia in 2004 (51%). On the other hand, in comparison with studies carried out in Europe, our results are clearly higher than those described in Slovakia too, where nematodes prevalence reached a percentage of 49% (Komorová et al., 2017).

Parasitisation by cestodes was less frequent than by nematodes in our study, with a prevalence of 6% (Table 2) in the genus Paruterina (Fuhrmann, 1906). Nevertheless, specific identification and quantification was not possible due to the fragmented state of the strobili. Although without statistical differences, this prevalence is higher than the prevalence described in B. bubo by Sitko and Heneberg (2019) in the Czech Republic (3%). This non-significant prevalence increase could be related to their size sample of 32 eagle owls.

Acanthocephalans were found in only 2 of the owls examined, with only 1 specimen in each (Table 2). The 2 acanthocephalans found had an invaginated proboscis, which prevented us from determining the
number and distribution of the hooks and made it impossible to identify the species. However, we were able to identify the genus Centrorhynchus (Amin, 2013). The only data on the prevalence of acanthocephalans in the species. However, we were able to identify the genus Centrorhynchus (Amin, 2013). The only data on the prevalence of acanthocephalans in eagle owls has been reported in the Czech Republic with a prevalence of 6% of the species C. alacoris (Sitko and Heneberg, 2019). Our results are in line with those of these authors, as a prevalence of 4% for these helminths was found in our study.

Regarding nematode species (Table 3), C. tenuissima was the most prevalent species in our study (58%) with an intensity mean value of 11.52. Capillaria spp. had been reported previously in B. bubo in Spain (Ferrer et al., 2004), Austria (Kutzer et al., 1982), and Italy (Santoro et al., 2012). Although Capillariae genus (Komorová et al., 2016) and other Capillaria species have been described in B. bubo, such as Capillaria strigia (Papazahariadou et al., 2008) or Baruscapillaria falconis (syn. Capillaria falconis) (Illescas et al., 1993; Sitko and Heneberg, 2019), the species identified in our study was C. tenuissima (Rudolphi, 1803). This intestinal Capillaria had been reported also in the American Eagle Owl (Bubo virginianus) (Ramalingam and Samuel, 1978) and the Magellanic Owl (Bubo magellanicus) (Grandon-Ojeda et al., 2018). The prevalence observed in our study is similar to that detected in Catalonia (57%) by Ferrer et al. (2004), although in this study the sample was lower (only 7 owls) without identifying the species. In comparison with our study, other studies with a larger number of samples (32 owls) showed an important decrease in prevalence (11%) although the species identified (B. falconis) showed similar intensity mean (11.41) (Sitko and Heneberg, 2019). This higher prevalence found in our study could be related to the number of samples analysed (50) as well as the generalist nature of this genus, which can parasitize both diurnal and nocturnal raptors from different geographical areas (Baras and Sergejeva, 1989). Litte is known about the life cycle of this intestinal nematode frequently observed in raptors. Authors discuss about whether C. tenuissima has an indirect or direct life cycle (Anderson, 2000; Atkinson et al., 2008). In any case, it seems that earthworms are involved and could act as intermediate hosts. As paratenic hosts, earthworms as well as rodents are also mentioned; this would explain the generalist nature of this parasite and its presence in the studied birds.

The second most important species was S. laticeps. This is a frequent species in raptors, found with equal prevalence in Accipitriformes, Falcoformes, and Strigiformes (Sitko and Heneberg, 2019). The highest prevalences of S. laticeps in the Iberian Peninsula have been obtained in short-eared owls (62%) and sparrowhawks (60%) (Sanmartín et al., 2004). In B. bubo, S. laticeps was reported for the first time by Illescas et al., in 1993. Regarding S. laticeps prevalence, the percentage obtained in our study (16%) is lower than that reported by Ferrer et al. (2004) in Catalonia (43%) although on a much smaller sample (7 individuals). On the one hand, this result could be related to the frequency and abundance of dietary insects in different areas of distribution, since they use insects and/or isopod crustaceans as intermediate hosts (Penteriani and Delgado, 2008a). On the other hand, it could be related, to the sample size. In any case, an underestimation of the prevalence of this species should be considered (see comments below).

Hartertia hispanica also showed a prevalence of 16%. So, alongside S. laticeps it is considered the second most important species with respect to the helminth fauna found in B. bubo. However, it should be noted that H. hispanica showed lower intensity and abundance (Table 3) than S. laticeps. It is worth mentioning that this is the third description of H. hispanica in B. bubo, being originally described by López-Neyra in 1947 and later by Illescas et al., in 1993. More recently, it has been found in other Strigiformes (Asio otus) and in two Accipitriformes (Circaetus gallicus and Buteo buteo) also in Andalucía by our group (unpublished data). To our knowledge, no records are known from other regions (Spain or Europe), so it is likely that it is a species endemic to the south of the Iberian Peninsula.

Among the nematode species with the lowest prevalence values, we found S. affinis, C. spinosa, and C. ficheuri. Regarding S. affinis, this is a specific parasite of Strigiformes (Illescas et al., 1993; Etchegoin et al., 2000; Sanmartín et al., 2004; Papazahariadou et al., 2008; Santoro et al., 2012; André Tomás et al., 2017; Komorová et al., 2017; Sitko and Heneberg, 2019). Interestingly, despite being specific to Strigiformes, its prevalence is very low (only 4%) in our study, where it is considered a satellite species in the eagle owl helminth fauna. This fact could be related to a geographical influence on the biological cycle of this species. However, as commented above, there were 15 Synhimantus spp. specimens whose species could not be identified (Table 2). Therefore, it is possible that the prevalence of both S. laticeps and S. affinis was underestimated.

### Table 4

| Biological index found in the eagle owls parasitised for the period 2011–2020. Numbers in parentheses are the 95% confidence intervals of each parameter. |
|---------------------------------------------------------------|
| Species richness                                             | 1.56 (1.29–1.94) |
| Total abundance                                              | 12 (7.24–26.40)  |
| Brillouin index                                              | 0.18 (0.10–0.29) |
| Berger-Parker index                                          | 0.88 (0.81–0.95) |

![Fig. 2. Prevalence of identified species.](image)
In relation to the genus *Cyrena* (Seurat, 1914), the species found were *C. spinosa* (Gendre, 1922) and *C. ficheuri* (Seurat, 1916) with prevalences of 4% and 6% and mean intensities of 3 and 2 respectively. Of the 3 owls parasitised with this genus, 2 of them had mixed infections and the third had a single infection with *C. ficheuri*.

*C. spinosa* is a frequent species in the common kestrel (*Falco tinnunculus*), the raptor in which it was originally described (Gendre, 1922; Barus, 1966; Sitko and Heneberg, 2019). In addition, it has been found in *F. naumanni* by our group (unpublished data), *F. subbuteo* (Komorova et al., 2017) and *Circus cyaneus* (Lacina and Bird, 2000). In Andalusia it was found in *B. bubo* by López-Neyra in 1947 and by Illescas et al., in 1994.

*C. ficheuri* (Seurat, 1916) was described for the first time in the cattle egret (*Bubulcus lucids*) in Africa. Since then, it has been found several times in both ardeids and striigiformes (Gendre, 1922; Orlette, 1938; Chabaud and Brygoo, 1950). Even though it is a species whose origin and location are in Africa and no observation in Europe, it has been described in Andalusia by our group in *F. naumanni* and *B. bubo* (unpublished data), representing the only record in this host, as well as the first made in Europe to date.

The prevalence of *Cyrena* spp in nocturnal raptors is low (Sanmartin et al., 2004; Ferrer et al., 2004b; Santoro et al., 2012). In this sense, the results obtained in our study agree with those described by the authors mentioned above.

From 4 owls parasitised by trematodes, it was possible to identify the species *Strigea* only in 1 (Table 3). This trematode has been specific of nocturnal raptors (Lacina and Bird, 2000; Heneberg et al., 2018). In the Iberian Peninsula it has been recorded in *T. Alba, Buteo buteo*, and *Milvus milvus* (Simón-Vicente et al., 2004). In our study we found the lowest prevalence for this parasite (2%). The trematodes life cycle is linked to the presence of Planorbidiae snails, amphibia and reptiles as intermediate hosts whose presence is conditioned by environmental characteristics (Odening, 1967; Sitko et al., 2006). Thus, the environmental conditions in which the life cycle of these parasites develops cause great differences in presentation according to geographical regions, being in our case (southern Spain) a parasitosis with little relevance both in prevalence and abundance.

Our results contrast with other studies conducted in northern Spain (Ferrer et al., 2004) and the Czech Republic (Sitko and Heneberg, 2019) where prevalence of 14% and 21% respectively were reported. Considering the geographical conditioning factor, related to the life cycle, these differences may be related also to the sample size in the case of *T. Alba*.

In conclusion, this is the first study on the helminthfauna in eagle owls carried out in southern Spain (Andalusia) with the largest number of samples in Europe. The data from this study show a non-diverse helminthic community, without species dominance and where the core species is *T. tenuissima*, followed by *S. laticeps* and *H. hispanica* as secondary species. It is worth mentioning the presence of *H. hispanica*, which is considered an endemic species in Spain and specifically in Andalusia.

**Declaration of competing interest**

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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