



## Helminth communities of herons (Aves: Ardeidae) in southern Italy



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### ABSTRACT

The helminth communities of nine species of herons from southern Italy were studied and compared. Of 24 taxa found including seven digeneans, seven nematodes, six cestodes and four acanthocephalans, only five taxa were found in more than one heron species, and five of the 21 taxa that could be identified to species level were classified as 'heron specialists'. The total number of helminth species per heron species ranged from 1 in *Botaurus stellaris* to 9 in *Ixobrychus minutus* with infection levels generally low. A statistical comparison was carried out for herons with a sample size >5. At the infracommunity level, only *I. minutus* clearly differed from other heron species. Diversity parameters of helminth infracommunities did not significantly differ among heron species. Species richness ranged from just 0.3 to 2.3 helminth taxa per individual host, and the Brillouin index, from 0 to 0.3. Total helminth abundance did not exceed 40 worms per host except in a single case of *Ardeola ralloides*. Infracommunities clearly were dominated by single helminth species. The present study confirms a depauperate helminth community in herons from southern Italy. Comparison with data from Spain and the Czech Republic showed strong quantitative similarities with values obtained in the present study. Results also suggest that the composition of local helminth communities are strongly variable depending on geographical location as is demonstrated by comparison with data from other European areas. However, whether herons in Europe naturally host depauperate helminth communities or these communities are depauperate because of other factors is unknown.

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### 1. Introduction

Herons (Ardeidae) are freshwater and coastal birds that inhabit diverse aquatic habitats where they feed on a wide variety of prey items such as fish, tadpoles, frogs, snakes, snails, crustaceans, aquatic insects and small mammals. The range of prey items that they may ingest depends on the habitat, local availability and host size [1]. Of the 10 heron species occurring in the Palearctic region, at least 9 inhabit temporarily or as resident in wetlands of southern Italy including little bittern *Ixobrychus minutus*, Eurasian bittern *Botaurus stellaris*, black-crowned night heron *Nycticorax nycticorax*, squacco heron *Ardeola ralloides*, cattle egret *Bubulcus ibis*, little egret *Egretta garzetta*, great white heron *Ardea alba*, grey heron *Ardea cinerea*, and purple heron *Ardea purpurea* [1,2].

Reports on helminths in herons are numerous [3–10], but rather surprisingly, surveys on the complete helminth communities of herons from the Palearctic region include only two studies. In the Czech Republic, Sitko and Heneberg [11] suggested that helminth

communities of at least five heron species exhibit strong geographic variation that largely resulted from differences of local availability of intermediate invertebrate hosts. Diverse and abundant helminth communities should be reflective of diverse and rich communities of free-living species in the environment, whereas changing environmental features and the impact of anthropogenic pressures among localities could cause a profound impact in the structure of helminth communities. In fact, Navarro et al. [12] reported depauperate helminth faunas in 6 heron species from western Spain and suggested that these species-poor communities could have partly resulted from habitat degradation and high pollution levels.

Host specificity could also be an important factor influencing structure of helminth communities of herons [6,8,11,12]. Specialist helminths (i.e., those typically restricted to one or a few host species) usually occur at high prevalence and intensity in their preferred hosts over most localities of the hosts' range, whereas infection levels of generalist helminths depend more on the composition of the local host community [13–15]. However, differences observed in several migrating bird groups suggest that host specificity may play a contrasting role in structuring parasite communities depending on geographical region [11,14,16–20].

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**Table 1**

Infection parameters of helminth taxa collected from 9 species of Ardeidae in southern Italy. Upper rows represent prevalence, with 95% CI in parentheses. Lower rows indicate mean intensities, with 95% CI between brackets; when only one or two hosts are infected, raw values of intensity are given.

Taxa	Host species								
	<i>A. cinerea</i> (n = 14)	<i>A. purpurea</i> (n = 5)	<i>N. nycticorax</i> (n = 5)	<i>A. alba</i> (n = 4)	<i>E. garzetta</i> (n = 6)	<i>B. ibis</i> (n = 2)	<i>Ar. ralloides</i> (n = 2)	<i>B. stellaris</i> (n = 3)	<i>I. minutus</i> (n = 52)
<b>Trematoda</b>									
<i>Euclinostomum heterostomum</i> (HG)	–	–	–	–	16.7 (0.9–58.7)	–	–	–	–
					1				
<i>Ophiosoma patagiatum</i> (HS)	–	–	–	–	–	–	–	33.3 (1.7–86.5)	–
								4	
<i>Apharyngostrigea cornu</i> (HG)	35.7 (15.3–62.9)	–	–	25.0 (1.3–75.1)	33.3 (6.3–72.9)	–	–	–	–
	23.4 [14.2–29.4]			3	1–3				
<i>Apharyngostrigea ramai</i> (HG)	–	–	–	–	33.3 (6.3–72.9)	–	–	–	–
					1–9				
<i>Pegosomum asperum</i> (HG)	–	–	–	100 (47.3–100)	–	–	–	–	–
				5.3 [3.3–7.0]					
<i>Sodalis spathulatus</i> (HG)	–	–	–	–	–	–	–	–	19.2 (10.3–32.5)
									2.2 [1.5–3.0]
<i>Ribeiroia ondatrae</i> (TG)	–	–	–	–	–	–	–	–	1.9 (0.1–10.2)
									42
<b>Cestoda</b>									
<i>Dendrouterina herodiae</i> (HS)	–	–	–	–	33.3 (6.3–72.9)	–	–	–	–
					8–12				
<i>Dendrouterina macrosphincter</i> (HG)	–	20.0 (1.0–65.7)	–	–	–	–	–	–	–
		4							
<i>Dendrouterina ixobrychi</i> (HS)	–	–	–	–	–	–	–	–	9.6 (3.9–20.9)
									3.4 [1.4–7.2]
<i>Valipora atrispinosa</i> (HS)	–	–	–	–	–	–	50.0 (2.5–97.5)	–	–
							814		
<i>Neogryporhynchus cheilancristrotus</i> (HG)	21.4 (6.1–50.0)	–	–	–	–	–	–	–	–
	36.0 [13.0–52.3]								
Unidentified cestode	–	–	20.0 (1.0–65.7)	–	–	–	–	–	–
			9						
<b>Nematoda</b>									
<i>Porrocaecum ardeae</i> (HG)	7.1 (3.7–31.7)	–	–	–	–	–	50.0 (2.5–97.5)	–	1.9 (0.1–10.2)
	1						1		1
<i>Contraecum microcephalum</i> (TG)	14.3 (2.6–42.6)	60.0 (18.9–92.4)	80.0 (34.3–99.0)	–	–	–	50.0 (2.5–97.5)	–	25.0 (14.9–38.4)
	1–7	15.7 [12.0–18.7]	17.5 [1.3–33.5]				1		1.2 [1.0–1.3]
<i>Desmidocercella numidica</i> (HG)	14.3 (2.6–46.6)	20.0 (1.0–65.7)	20.0 (1.0–65.7)	50.0 (9.8–90.2)	–	–	–	–	–
	5–7	9	9	2–4					
<i>Avioserpens galliardi</i> (HG)	–	–	–	–	–	–	–	–	1.9 (0.1–10.2)
									1
<i>Desportesius invaginatus</i> (HG)	–	–	–	–	16.7 (0.9–58.7)	50.0 (2.5–97.5)	–	–	–
					16	6			
<i>Desportesius brevicaudatus</i> (HG)	–	–	–	–	–	–	–	–	19.2 (10.3–32.5)
									5.0 [2.2–12.4]
<i>Microtetrameres spiralis</i> (HS)	–	–	–	–	–	50.0 (2.5–97.5)	–	–	–
						36			
<b>Acanthocephala</b>									
<i>Southwellina hispida</i> (TG)	–	–	–	50.0 (9.8–90.2)	–	–	–	–	–
				6–107					
<i>Ardeirhynchus spiralis</i> (HG)	–	–	–	–	–	–	–	–	1.9 (0.1–10.2)
									1
<i>Polymorphus</i> sp.	7.1 (3.7–31.7)	–	–	–	–	–	–	–	–
	1								
<i>Plagiorhynchus</i> sp.	–	–	–	–	–	–	–	–	1.9 (0.1–10.2)
									1

Abbreviations: HG, herons generalist; HS, herons specialist; TG, True generalist.

Here we report on the structure of the helminth communities of nine species of herons from southern Italy and analyze the factors that may influence this structure. In addition, we compared our results with those obtained from previous studies [11,12]. The analyses were driven by the following research questions: (i) Are the helminth communities of herons from southern Italy similar in composition and structure? (ii) To what extent do the helminth communities of herons differ among geographical areas? (iii) What factors might account for the similarities and differences between heron species and geographical areas?

## 2. Materials and methods

### 2.1. Collection data

A total of 93 free-ranging birds of nine heron species (Table 1) that died between January 2005 and December 2014 at two Wildlife Rescue Centers in Calabria region of southern Italy were examined for helminth parasites.

All birds were weighed, sexed, and two age classes were established, i.e. immature and adult individuals, based on phenotypic and biometric

features and gonad development. During necropsy the heart, trachea, lungs, air sacs, kidneys, spleen, pancreas, liver, gallbladder, and the whole digestive tract of birds, including esophagus, stomach, and intestines (duodenum, jejunum, ileum, ceca, and cloaca), were examined. Helminths were collected, identified, and counted following the techniques described by Krone [21]. Worms were washed in saline solution and fixed in 70% ethanol; trematodes and cestodes were stained with carmine, dehydrated using a graded alcohol series, cleared in methyl salicylate and mounted on slides in Canada balsam. Nematodes and acanthocephalans were cleared in lactophenol on a glass slide for identification and then returned to the preservative. Voucher specimens are deposited in the University of Nebraska State Museum, Systematics Research Collections (accession numbers: 91967 to 91977 and 101828 to 101832), Lincoln, Nebraska, USA.

## 2.2. Statistical analyses

For the quantitative comparison of parasite faunas between heron species we used the approach described in Santoro et al. [17,18]. Helminth species were classified by host specificity based on data from the Host–Parasite Database of the Natural History Museum, UK (<http://www.nhm.ac.uk/research-curation/scientific-resources/taxonomy-systematics/host-parasites>), and specific parasitological references on herons [6–8,10–12 and references therein]. We considered ‘heron specialists’ as helminth species that are known to reproduce only in one heron species; ‘heron generalists’ as species that are shared by many heron species, and ‘true generalists’ as those species that are widespread in birds and mammals.

Infracommunities were described using total abundance (i.e., the total number of all helminth species), species richness, and the Berger–Parker dominance index. Helminth infracommunity parameters were calculated for each heron individual, and compared between heron species with  $n \geq 5$  using Kruskal–Wallis tests with post hoc comparisons [22]. The 95% confidence interval (CI) for prevalence was calculated with Sterne’s exact method, and for mean values of intensity, total abundance, species richness, Brillouin’s index, and Berger–Parker index, with the bias-corrected and accelerated bootstrap method using 20,000 replications [23,24]. Differences of community structure between herons with a sample size  $\geq 5$  were investigated with a non-parametric analysis of similarities (ANOSIM) [25]. The number of individuals of each helminth species from each infracommunity was square-root transformed, and the Bray–Curtis similarity coefficient was calculated among individual hosts that harbored at least 1 helminth species [see 17 for details]. The overall comparison was followed by pair-wise comparisons between heron species. Only comparisons for which the total number of possible permutations was  $>400$  were considered.

To investigate geographical predictability in helminth communities of herons, we qualitatively compared diversity parameters, at infracommunity level, of each heron species between southern Italy (this study), eastern Spain [12] and the Czech Republic, in central Europe [11]. At component community level, we compared community composition, and differences of prevalence, for helminths from each heron species among geographical regions. For this comparison, we selected heron species with a sample size  $n \geq 20$ , and helminth species with a prevalence  $\geq 5\%$ , in at least one region [see 11]. Differences of prevalence were assessed with exact Chi-square tests. It should be noted that some comparisons involved small sample sizes and, therefore, results of these tests are conservative due to reduced power. The free software Quantitative Parasitology v. 3 [26] was used to set 95% confidence intervals of parameters, the package Primer v.6 [25] for the ANOSIM, and the cluster analyses, Statxact v. 11 for exact Chi Square tests, and the statistical package SPSS v. 17 for the remaining analyses. Statistical significance was set at  $p < 0.05$ . In pairwise comparisons, probability values were also corrected by the sequential

Bonferroni procedure [27]. Both the nominal and the corrected  $p$ -values are reported.

## 3. Results

### 3.1. General data

Twenty-four helminth taxa, including seven digeneans, seven nematodes, six cestodes and four acanthocephalans, and 1603 helminth individuals were collected from the nine heron species (Table 1); however, 814 individuals (50.7% of all individual parasites found) belonged to the cestode *Valipora atrispinosa* and were found in a single *Ar. ralloides*. Samples of all helminth taxa included adult specimens acquired by ingestion. All taxa were found in the gastrointestinal tract except the digenean *Pegosomes asperum* found in the liver of *A. alba* and the nematode *Avioserpens galliardi* in the subcutaneous tissues of the neck of *I. minutus*.

As many as 19 helminth taxa (79.2%) were found in a single heron species. Out of the 5 helminth taxa that were shared among herons, 4 were nematodes, namely *Desportesius invaginatus*, *Porrocaecum ardeae*, *Desmidocercella numidica* and *Contraecum microcephalum*, which were found in 2, 3, 4 and 5 heron species, respectively; the digenean *Apharyngostrigea cornu* was also shared between 2 heron species, and no helminth species was found in all herons (Table 1). Despite the high number of helminth taxa found only in a single heron species, only 5 (23.8%) of the 21 taxa that could be identified at species level were classified as ‘heron specialists’; 13 (61.9%) were considered as ‘heron generalists’, and 3 (14.3%) as ‘true generalists’ (Table 1).

### 3.2. Helminth communities

The total number of helminth species per heron species ranged from 1 in *B. stellaris* to 9 in *I. minutus* (Table 1). Infection levels were generally low. Only two species, i.e., the digenean *P. asperum* in *A. alba*, and the nematode *C. microcephalum* in *A. purpurea* and *N. nycticorax*, had a prevalence  $>50\%$ . Also, mean intensities were  $<40$  individuals per infected host except for the cestode *V. atrispinosa* in *Ar. ralloides* and the acanthocephalan *Southwellina hispida* in *A. alba*. At the helminth infracommunity level, we found significant compositional dissimilarities among the 5 heron species with  $n \geq 5$  (ANOSIM,  $R = 0.346$ ,  $p = 0.005$ ). Pair-wise comparisons were performed between all herons except the pair *A. purpurea* and *N. nycticorax* (possible number of permutations = 126). All comparisons involving *I. minutus* were significant even after the sequential Bonferroni correction (minimum  $p = 0.02$ ). *I. minutus* shared only 2 of 9 helminth taxa with 4 other herons, i.e., the nematodes *P. ardeae* (shared with *A. cinerea*), and *C. microcephalum* (with *A. cinerea*, *A. purpurea* and *N. nycticorax*). The comparison between *A. purpurea* and *N. nycticorax* was nominally significant ( $p = 0.019$ ) and those between *A. purpurea* and *E. garzetta* (nominal  $p = 0.079$ ), and between *A. cinerea* and *N. nycticorax* (nominal  $p = 0.079$ ), were close to significance; none was after the sequential Bonferroni correction.

Diversity parameters of helminth infracommunities for each of the 9 herons are shown in Table 2. These parameters did not significantly differ among host species, i.e., distribution of values in heron individuals were similar among heron species (Kruskal–Wallis tests, minimum nominal  $p > 0.05$ ). Average species richness ranged from just 0.3 to 2.3 helminth taxa per individual host, and the Brillouin index, from 0 to 0.3. Total helminth abundance did not exceed 40 worms per host except in the case of *Ar. ralloides* (Table 2). Also, according to the Berger–Parker index, infracommunities were clearly dominated by single helminth species (Table 2).

Available data on helminth infracommunity parameters of herons from Spain and the Czech Republic indicated strong quantitative similarities with values obtained in the present study, i.e. herons harbor depauperate infracommunities with low total helminth abundance

**Table 2**

Infracommunity parameters of the helminth fauna of herons (Ardeidae) collected in three localities from western Europe, i.e., southern Italy (this study), eastern Spain ([12] Navarro et al. 2005) and Czech Republic (central Europe) [11] (Sitko and Heneberg 2015).

Avian host	Species richness	Total helminth abundance	Brillouin index	Evenness	Berger–Parker index
<i>Italy</i>					
<i>Ardea cinerea</i> (n = 14)	1.0 ± 1.2	17.6 ± 23.4	0.33 ± 0.41	0.57 ± 0.38	0.82 ± 0.26
<i>Ardea purpurea</i> (n = 5)	1.0 ± 0.7	12.0 ± 9.0	0.09 ± 0.19	0.64	0.96 ± 0.08
<i>Ardea alba</i> (n = 4)	2.3 ± 1.0	35.8 ± 51.7	0.46 ± 0.42	0.73 ± 0.37	0.71 ± 0.31
<i>Nycticorax nycticorax</i> (n = 5)	1.2 ± 0.5	16.6 ± 17.5	0.07 ± 0.16	0.47	0.96 ± 0.08
<i>Egretta garzetta</i> (n = 6)	1.3 ± 1.0	8.5 ± 10.5	0.19 ± 0.34	0.61 ± 0.31	0.90 ± 0.18
<i>Bubulcus ibis</i> (n = 2)	1.0 ± 0.0	21.0 ± 21.2	0.0 ± 0.0	–	1 ± 0.0
<i>Ardeola ralloides</i> (n = 2)	1.5 ± 0.7	408.0 ± 575.6	0.004 ± 0.006	0.01	0.99 ± 0.0
<i>Botaurus stellaris</i> (n = 3)	0.3 ± 0.6	1.3 ± 2.3	0.0	–	1
<i>Ixobrychus minutus</i> (n = 52)	0.9 ± 0.9	3.0 ± 7.0	0.13 ± 0.22	0.72 ± 0.30	0.90 ± 0.18
<i>Spain<sup>a</sup></i>					
<i>Ardea cinerea</i> (n = 25)	1.3 ± 1.3	52.9 ± 154.8	NC NA <sup>b</sup>	0.23 ± 0.35	NC NA
<i>Ardea purpurea</i> (n = 4)	1.0 ± 1.2	14.8 ± 24.5	NC NA	0.23 ± 0.27	NC NA
<i>Nycticorax nycticorax</i> (n = 2)	1.5 ± 2.1	7.5 ± 10.6	NC NA	0.27 ± 0.38	NC NA
<i>Bubulcus ibis</i> (n = 9)	1.7 ± 1.3	95.9 ± 112.5	NC NA	0.25 ± 0.33	NC NA
<i>Egretta garzetta</i> (n = 20)	0.5 ± 0.8	9.6 ± 35.9	NC NA	0.06 ± 0.23	NC NA
<i>Ixobrychus minutus</i> (n = 5)	1.2 ± 1.3	12.0 ± 25.2	NC NA	0.31 ± 0.45	NC NA
<i>Czech Republic</i>					
<i>Ardea cinerea</i> (n = 506)	2.6	71.0	NC NA	0.65	0.26
<i>Ardea alba</i> (n = 13)	1.7	40.6	NC NA	0.80	0.26
<i>Nycticorax nycticorax</i> (n = 6)	1.4	52.7	NC NA	0.70	0.44
<i>Botaurus stellaris</i> (n = 7)	1.4	15.1	NC NA	0.91	0.36
<i>Ixobrychus minutus</i> (n = 25)	0.1	3.5	NC NA	0.92	0.67

<sup>a</sup> Evenness for data from Spain was calculated using base-2 logs.

<sup>b</sup> NCNA: not calculated available from the original publication.

(Table 2). Prevalences were also low regardless of geographical region, with values  $\geq 50\%$  in only 6 out of 54 helminth taxa recorded in 5 heron species (Table 3). However, faunal composition was highly variable depending on the region. In the comparisons of *I. minutus* and *A. cinerea*, which involved all regions (Table 3), as many as 17 helminth were found in single regions (i.e., 77% of total taxa with prevalence  $\geq 5\%$ ); only 2 taxa of *I. minutus* were shared between two regions, and 3 taxa from *A. cinerea* among the three regions. This pattern was supported by the observation that most comparisons of prevalence were significant (Table 3). The comparisons of *E. garzetta*, *A. alba*, and *N. nycticorax* involved samples of southern Italy, and central Europe or eastern Spain. Host sample sizes in Italy were very low and, not surprisingly, most prevalence comparisons were not significant (Table 3). However, it is very interesting to note that, again, as many as 16 helminth were found in single regions (i.e., 80% of total), and only 4 were shared between two regions (Table 3). Moreover, the helminth species shared, namely, *A. cornu*, *D. numidica* and *C. microcephalum*, are the same detected in the geographical comparisons of *I. minutus* and *A. cinerea* (Table 3). This would suggest that these parasites are ecologically ubiquitous and geographically widespread.

#### 4. Discussion

In southern Italy, herons serve as definitive hosts for 24 helminth taxa. Species with known life cycles all require freshwater intermediate hosts (Table 1). Considering the total number of taxa here found in the whole sample of herons no helminth group appeared to be dominant; we found seven digeneans, seven nematodes, six cestodes, and four acanthocephalans. A similar lack of dominance was reported for herons in Spain (six nematodes, five digeneans and two cestodes) [12], but contrasted with data from the Czech Republic where digeneans were a clearly dominant group (26 taxa) over cestodes (5 species), nematodes (3 species) and acanthocephalans (1 species) [11]. The observation that digeneans are dominant is evident also from other studies on herons of central and eastern Europe [8,28]. Sousa and Grosholz [29] described a variety of habitat-related biotic and abiotic factors influencing the rate of transmission of parasites. Of these, the most probable factor that could account for the geographic difference of digenean diversity in herons is the higher abundance of wetland areas in central and eastern Europe compared to southern Italy and eastern Spain. Wetlands are key habitats for intermediate and paratenic hosts for digeneans, such as



**Table 3**  
Comparison of prevalence (%) of helminth taxa in 5 heron species in three European regions. Data from Central Europe and eastern Spain were obtained from [11,12], respectively. Only helminths with a prevalence  $\geq 5\%$  in at least one region are shown. Taxa shared between two or more regions are in bold. (–) species not detected.

Host species	Parasite taxon	Region		
		Central Europe	Eastern Spain	Southern Italy
<i>Ixobrychus minutus</i>		(n = 25)	(n = 5)	(n = 52)
	<i>Sodalis spathulatus</i> * <sup>a</sup>	–	–	19.2
	<i>Ribeiroia ondatrae</i> **	–	20.0	–
	<i>Apharyngostrigea cornu</i> **	–	20.0	–
	<i>Dendroustera ixobrychi</i>	–	–	9.6
	Cyclophyllidae gen. sp.**	–	20.0	–
	<b>Contracaecum microcephalum</b>	8.0	–	25.0
	<b>Avioserpens galliardi</b>	–	20.0	1.9
	<i>Desportesius brevicaudatus</i> * <sup>a</sup>	–	–	19.2
	<i>D. spinulatus</i> **	–	20.0	–
	Capillariidae gen. sp.**	–	20.0	–
<i>Ardea cinerea</i>		(n = 506)	(n = 25)	(n = 14)
	<i>Echinocasmus beleocephalus</i> **	38.5	–	–
	<i>Uroprostepisthimium bursicola</i> **	35.8	–	–
	<i>Posthodiplostomum cuticola</i> **	40.9	–	–
	<b>Apharyngostrigea cornu</b> **	67.4	48.0	35.7
	<i>Bilharziella polonica</i>	5.3	–	–
	<b>Neogryporhynchus cheilancristrotus</b> **	30.8	8.0	21.4
	<b>Desmidocercella numidica</b>	28.7	36.0	14.3
	<i>Contracaecum yamaguti</i> **	–	16.0	–
	<i>Desportesius spinulatus</i> **	–	16.0	–
	<i>Porrocaecum ardeae</i> **	–	–	7.1
	<i>Contracaecum microcephalum</i> **	–	–	14.3
	<i>Polymorphus</i> sp.**	–	–	7.1
<i>Egretta garzetta</i>			(n = 20)	(n = 6)
	<i>Euclinostomum heterostomum</i>	–	–	16.7
	<b>Apharyngostrigea cornu</b>	–	10.0	33.3
	<i>Apharyngostrigea ramai</i> * <sup>a</sup>	–	–	33.3
	<i>Desmidocercella numidica</i>	–	30.0	–
	<i>Avioserpens galliardi</i>	–	5.0	–
<i>Desportesius invaginatus</i>	–	–	16.7	
<i>Ardea alba</i>		(n = 38)		(n = 4)
	<i>Pegosomum</i> sp.	39.0	–	–
	<i>Pegosomum asperum</i> **	–	–	100.0
	<i>Uroprostepisthimium bursicola</i>	29.0	–	–
	<i>Posthodiplostomum cuticola</i>	21.0	–	–
	<b>Apharyngostrigea cornu</b>	45.0	–	25.0
	<b>Desmidocercella numidica</b> * <sup>a</sup>	5.0	–	50.0
	<i>Contracaecum microcephalum</i>	5.0	–	–
<i>Porrocaecum ardeae</i>	11.0	–	–	
<i>Nycticorax nycticorax</i>		(n = 29)		(n = 5)
	<i>Opistorchis longissimus</i>	10.0	–	–
	<i>Posthodiplostomum cuticola</i>	7.0	–	–
	<b>Contracaecum microcephalum</b> * <sup>a</sup>	55.0	–	80.0
	<i>Neogryporhynchus cheilancristrotus</i>	62.0	–	–
	<i>Desmidocercella numidica</i>	–	–	20.0
	Cestoda fam. gen. sp.	–	–	20.0

\* Nominal  $p < 0.05$ .

\*\* Nominal  $p < 0.001$ .

<sup>a</sup> Not significant after the sequential Bonferroni correction.

aquatic snails, frogs and fish, which all exhibit low vagility [14,29,30] and likely have more reduced populations in localities from southern Europe.

At a component community level, of the 21 helminth taxa identified to species level in our study most were generalists in herons (12

species), or in birds and mammals (4 species) (Table 1). However, and rather strikingly, of the 24 taxa found, most (19 species or 79.2%) were found in just one heron species. At first glance, one might think that a small sample host size coupled with low helminth recruitment rates (see below) could limit the possibility of finding parasites common to all host groups. However, this pattern is shared with other surveys. In six species of herons, Navarro et al. [12] found a total of 15 helminth taxa, of which all those identified to species level (12 spp.) were generalists, and more than a half (8 species, 61.5% of total) were found in single host species. Likewise, Sitko and Heneberg [11], using much larger host samples sizes, found similar results for at least five heron species: most of helminth species were generalists, but 22 of the 33 species (66.7%) were found in just a single host species [11]. This pattern is corroborated by the geographical comparison shown in this study. To explain why generalist parasite species may infect single, and different, host species depending on the geographical area, we should take into account two key aspects related to the biology and ecology of the host group. First, most herons included in the study are highly vagile, migratory birds, particularly trans-Saharan migrants such as *I. minutus*, *N. nycticorax*, *Ar. ralloides*, and *A. purpurea* [1,2] that exploit different wetlands temporarily. Thus, herons may be exposed to different local pools of parasites depending, not only on the local community of intermediate and paratenic hosts [19], but also on the temporal dynamics of the specific assemblage of birds visiting each wetland [31]. For instance, during their migration, aquatic birds visit different wetlands, and even use different migratory corridors, accumulating different helminth species from each area they use [14,20].

A second key factor is the habitat use, feeding strategies and diversity of prey of each heron species. With regard to the feeding spectrum, *I. minutus* preys mainly on aquatic adult and larval orthopterans, lepidopterans and coleopterans; *Ar. ralloides* on insects, frogs and fish; *B. ibis* mainly on terrestrial insects, especially orthopterans, dipterans and lepidopterans; *N. nycticorax* is an opportunistic feeder taking fish, amphibians, reptiles, and insects; and *Ardea* spp., *B. stellaris* and *E. garzetta* prey mainly on fish and small mammals [32]. All digeneans and cestodes here found use fish as second intermediate hosts, and acanthocephalans use freshwater amphipods and decapod crustaceans [33]; within the nematodes, *Contracaecum* spp., *Desmidocercella* spp., and *Desportesius* spp. use fish as intermediate or paratenic hosts; *Avioserpens* spp. use copepods as intermediate hosts and fish, frogs, and dragon fly larvae as paratenic hosts [34,35]. In contrast, the life cycles of *Porrocaecum* spp. and *Microtetrameres* spp. are terrestrial, including earthworms and orthopterans, respectively, as intermediate hosts [34,35]. Results of component communities here obtained indicate that herons feed intensively on fish. The finding of the nematode *M. spiralis* in just *B. ibis* may be related to the fact that this heron feeds mainly on orthopterans, and the presence of *P. ardeae* in *A. cinerea*, *Ar. ralloides*, and *I. minutus* confirms a wider alimentary spectrum for these heron species. On the other hand feeding habitats may also differ among heron species. For instance, bitterns usually favor reed beds, *A. cinerea* and *E. garzetta* prefer more open waters, whereas *B. ibis* mainly exploits pastures and open land [1,2,32].

The combined effect of hosts' vagile behavior and different diets should result in somewhat idiosyncratic diversity and composition of their helminth communities in each locality. This could explain why the composition of helminth fauna of each heron species was hardly repeatable from one geographic region to another. Effects at infracommunity level appeared to be idiosyncratic as well. Helminth communities from *I. minutus* from the study area were comparatively most distinct with respect to other heron species, but this was not the case neither in central Europe [11] nor in Spain [12]. In addition, mean values of diversity and total helminth abundance per host were not clearly related to host body size in any survey.

Hérons from southern Italy harbored depauperate helminth communities. Furthermore, when we compared parameters (ie. species richness, abundance and diversity) with the other surveys on herons

from Europe [11,12], no noticeable differences were found. The species-poor helminth infracommunities with low abundances that we found in herons were unexpected because aquatic birds, as a group, harbor the richest and most diverse helminth communities of all vertebrates [15]. Among migrating birds, it is believed that two major factors play a role in affecting parasite diversity. The first is a shift in exposure to potential intermediate hosts; the other one, profound changes in host diet during migration cause significant changes in bird physiology, which in turn may influence the parasite diversity [15,36,37]. It is believed that intensity of helminth infection decreases during the migration and reaches its maximum on the breeding grounds [36–39]. During migration, birds aggregate at the same stop-over sites, with high local density and species diversity favoring conditions that increase parasite transmission and switching [14,40–42]. In addition, it is expected that migratory animals using larger geographical ranges are exposed to a greater number of parasites compared to residents [20,40,41]. In fact, Koprivnikar and Leung [20] found that migratory birds host significantly higher richness of nematodes compared to residents, and that migratory and resident species harbored dissimilar nematode faunas. Nevertheless, it has been also suggested that migratory animals may harbor fewer parasites if they spend time in habitats with a low risk of parasite exposure, or if heavily infected individuals are removed from the population by the high demands of migration [42].

To explain the depauperate nature of helminth communities of herons from Spain, Navarro et al. [12] proposed several factors, including (i) the loss of vagility of hosts, i.e., an increasing tendency of many individuals from migratory heron species to become year-long residents; (ii) the strong trophic dependence on a few prey species such as the invasive crayfish *Procambarus clarkii*, and, (iii) stress birds suffered before death that may have caused loss of parasites. Interestingly, Sitko and Heneberg [11] indicated that most of the herons they studied were hunted or wounded, then probably most were immunocompromised individuals in which, the number of parasite species and the intensity of infection should increase [11].

Violante-González et al. [19] collected non-stressed individuals of two species of herons (*A. alba* and *Nyctanassa violacea*) from two coastal lagoons of Guerrero State in Mexico, and found very high values of helminth communities compared with those found in the European surveys. Thus, the question that arises is the extent to which the latter represent reliable examples of the helminth fauna from free-ranging birds. The herons examined in this study came from the same rescue centers used in previous studies on raptors (hawks and owls) [17,18] whose results strongly suggested meaningful contrasting patterns of helminth community structure that were not affected by the fact that animals were wounded. Thus, there is no a priori reason to expect a stronger influence of this bias on the helminth fauna of herons. However, more samples of wild birds from other geographic regions would be necessary to assess the influence of captivity conditions. This type of sampling, however, will be difficult because herons are protected in most European countries and/or are not game species that are easily available.

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