

Research Article

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# Diversity and community structure of ectoparasites infecting some elasmobranch species off the Tunisian coast

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**Abstract:** With more than 62 reported species, Tunisia has a rich diversity of elasmobranchs. However, investigations of their parasites in Tunisian waters remain rare and fragmented. With the global biodiversity crisis that most living species are facing, the study of parasite diversity is crucial for assessing ecosystem health and host-parasite interactions. In this study, 2,092 specimens of cartilaginous fishes (Chondrichthyes: Elasmobranchii) belonging to eight species, six genera and five families were sampled along the Tunisian coast and examined for their ectoparasites. The different host species were each infected by at least one ectoparasite species. A total of 24 ectoparasite species, among which three new species, were collected and identified. Copepods exhibited the highest taxonomic diversity (11 species), followed by ‘monogeneans’ (6 species), isopods (5 species), and leeches (2 species). To understand the diversification and specialisation of the collected ectoparasite species and the factors that may influence them, parasitological indices, parasitic communities’ composition, parasitic richness and seasonal variation are presented in this work. Parasite community structure varied among host species and families. *Mustelus mustelus* (Linnaeus) showed the greatest diversity (Shannon–Wiener  $H' = 1.91$ ; Species richness (SR) = 10), whereas *Torpedo torpedo* (Linnaeus) hosted only a single leech species. The present study demonstrates that parasite community composition and structure of the studied hosts seem to be influenced primarily by the geographic distribution, the sampling effort and the population density, along with other factors such as the phylogeny of the host species.

**Keywords:** Cartilaginous fishes, metazoan parasites, prevalence, diversity indices, host-parasite interaction, seasonality

Parasites are extraordinarily diverse and they represent more than half of all living species (DeMeeus and Renaud 2002, Dobson et al. 2008). Therefore, they are an important component of any ecosystem (Lafferty et al. 2008). Metazoan parasites can induce pathology in their hosts (Benz and Bullard 2004) and thus decrease host fitness (e.g., lower energy reserves) and increase host mortality risk (almost three times higher for infected hosts compared to uninfected) (Robar et al. 2010, McElroy and de Buron 2014, Timi and Poulin 2020, Gérard et al. 2024). Therefore, parasites play a major role in the ecosystem not only by their important diversity but also by their impact on other species.

Healthy ecosystems are known to be rich in parasite species (Marcogliese 2005). Practically all free-living metazoans harbour at least one parasite species (Poulin and Morand 2000). Elasmobranchs are parasitised by several groups of protozoan and metazoan organisms that live either permanently or temporarily on and within their hosts (Schaeffner and Smit 2019). These species (sharks, skates and rays) are hosts to a great variety of parasites in nature

(Merlo-Serna and García-Prieto 2016), and are likely to be much less numerous than the parasite species that infect them (Benz 1994). Studies of several species of elasmobranchs suggest that many, if not most, individuals are infected with at least one species of parasite (Hewitt 1979).

According to Takemoto et al. (2004), it is estimated that each fish species hosts on average ten different parasite taxa. However, estimates for the loss of biodiversity suggest that we are entering a period of mass extinction that is directly comparable to the mass extinctions recorded in the fossil record (Dobson et al. 2008). According to the IUCN Red List of Threatened Species (2025), most elasmobranch species are vulnerable. They are the vertebrate group with the highest extinction risk in the marine realm (Dulvy et al. 2014, Stein et al. 2018). This is mainly due to overfishing, which interacts with other environmental stressors (e.g., habitat loss and degradation, climate change, and pollution), and also the elasmobranch K-selected life history (Martin 2005, Barausse et al. 2014, Sguotti et al. 2016, Dulvy et al. 2021, Gérard et

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**Fig. 1.** Map of the Tunisian coastline showing the sampling sites (indicated by black stars).

al. 2024). The loss of these hosts means not only the loss of top marine predators, but also the loss of the diverse parasite fauna they harbour.

Additionally, studies on parasite diversity of elasmobranchs are quite rare (Henderson et al. 2002, Dallarés et al. 2017, Gérard et al. 2024), and information on most parasitic groups is fragmentary (Schaeffner and Smit 2019). They generally focus on only one group of parasites, notably copepods (Essafi 1975, Benz 1994, Deets 1994, Dippenaar 2016, Youssef et al. 2019) and the artificial taxon of ‘monogeneans’ (Chisholm 1995, Neifar 2001) without taking into consideration the other groups. In the context of global change and the worldwide biodiversity crisis in marine ecosystems, a more comprehensive understanding of the complex interactions between metazoan parasites, elasmobranch hosts, and their environments appears crucial (Gérard et al. 2024).

Tunisia has a rich diversity of elasmobranchs, with more than 62 reported species (Bradaï et al. 2012). However, investigations of their ectoparasites in Tunisian waters are rare (Essafi 1975, Neifar 2001, Youssef et al. 2019, 2022), and our understanding of species diversity and distribution and parasite communities’ composition is far from being complete. Thus, this study aims to focus on understanding host–parasite associations by exploring parasite species diversity, richness and seasonal variation of this richness, as well as to study the composition of parasitic communities that infect elasmobranchs off the Tunisian coast and the factors that may influence it.

## MATERIALS AND METHODS

### Sampling area and specimen collection

Between 2015 and 2021, 2,092 elasmobranch of eight species of chondrichthyan fishes were examined for ectoparasite species. Samples were collected monthly along the Tunisian coast, focusing especially on the Bay of Bizerte, the Gulf of Tunis, the Gulf of Hammamet, and the Gulf of Gabes (Fig. 1).

### Parasitological examination and parasite sampling

The collected elasmobranch were immediately transported to the laboratory for analysis. The host species were identified using Fischer et al. (1987) and Sêret (2006). Host nomenclature is according to Froese and Pauly (2025).

All body parts (skin, fins, gills, mouth, cloaca) were carefully examined. Gills were removed and placed in petri dishes containing seawater. The date, sampling area, name and the size of the host and the microhabitat of the parasite were noted.

Copepods and isopods were removed from the hosts and preserved in 70% ethanol. Subsequently, specimens were cleared in lactic acid for 2 h before examination by stereo and light microscopy. Specimens were dissected on glass slides and mounted as temporary preparations in lactophenol. Monopisthocotylea and polyopisthocotylean species belonging to the paraphyletic ‘Monogenea’ were stained with iron acetocarmine and examined as permanent mounts in Canada balsam. The leech species were observed alive under a stereo microscope. Then, the specimens were kept in 70% alcohol without relaxation. Subsequently, the fixed parasites were examined again under a stereo microscope.

Specimens of copepods were sent to Geoff A. Boxshall (Natural History Museum, London) for accurate identification. Flatworms and isopods were identified, with the help of Lasaad Neifar (Faculté des Sciences de Sfax, Tunisia) and Zouhir Ramdane (Faculté des Sciences de Bejaia, Algeria).

All parasites were identified to the lowest possible taxonomic level. Copepod species identification was based on morphological features following Wilson (1932) for species of *Eudactylina* Wilson, 1932, Kabata (1964) for species of *Pseudocharopinus* Kabata, 1964, Kabata (1979) for *Lernaopoda* Blainville, 1822 and *Nemesis* Risso, 1826 species, Cressey (1967) for species of Pandaridae Milne Edwards, 1840 and Deets et al. (2025) for species *Kroyeria* Van Beneden, 1853. The identification of the collected isopod species was performed according to Trilles (1979) and Trilles and Raibaut (1971) for species of *Anilocra* Leach, 1818, *Ceratothoa* Dana, 1852, *Emetha* Schioedte et Meinert, 1883, and *Nerocila* Leach, 1818 and Horton (2000) for species of *Ceratothoa*. Identification of ‘monogenean’ species was carried out according to Maillard and Paperna (1978) for species of *Erpocotyle* Van Beneden et Hesse, 1863, Sproston (1946) for *Hexabothrium* von Nordmann, 1840, Neifar et al. (1998) for *Heterocotyle* Scott, 1904, Tazerouti et al. (2011) for *Monocotyle* Taschenberg, 1878 and Chero et al. (2018) for *Hypanocotyle* Chero, Cruces, Sáez, Camargo, Santos et Luque, 2018. The morphometry and identification of leeches (Hirudinea) follow Richardson (1949) (voucher number: LeeTun221018, the parasitological collection of the Zoology Department Museum, College of Science, King Saud University, Riyadh, Saudi Arabia) and Llewellyn (1966).

**Table 1.** Communities of ectoparasites of different elasmobranchs off Tunisia.

Host family	Host species	Copepoda	Isopoda	‘Monogenea’ (Monopisthocotyla and Polyopisthocotyla)	Hirudinea
Dasyatidae	<i>Bathytoshia centroura</i> (Mitchill)	<i>Eudactylinella alba</i> Wilson, 1932 <i>Nemesis</i> sp. <i>Pseudocharopinus bicaudatus</i> (Krøyer, 1837) <i>Pseudocharopinus concaves</i> (Wilson, 1913)		<i>Hypanocotyle</i> sp. <i>Monocotyle myliobatis</i> Taschenberg, 1878	
	<i>Dasyatis pastinaca</i> (Linnaeus)	<i>Eudactylinella alba</i> <i>Pseudocharopinus malleus</i> (Rudolphi in von Nordmann, 1832)		<i>Heterocotyle pastinacae</i> Scott, 1904	
Triakidae	<i>Mustelus mustelus</i> (Linnaeus)	<i>Eudactylinella alba</i> <i>Kroyeria lineata</i> Van Beneden, 1853 <i>Kroyeria</i> sp. <i>Lernaeopoda galei</i> Krøyer, 1837 <i>Nesippus orientalis</i> Heller, 1865 <i>Perissopus dentatus</i> Steenstrup et Lütken, 1861	<i>Anilocra physodes</i> (Linnaeus, 1758) <i>Emetha audouini</i> (Milne Edwards, 1840) <i>Ceratothoa parallela</i> (Otto, 1828)	<i>Erpocotyle</i> sp. 1	
	<i>Mustelus punctulatus</i> Risso	<i>Lernaeopoda galei</i>	<i>Ceratothoa oestroides</i> (Risso, 1826) <i>Ceratothoa parallela</i>	<i>Erpocotyle</i> sp. 2	
Rajidae	<i>Raja clavata</i> Linnaeus	<i>Caligus</i> sp. <i>Lernaeopoda galei</i>	<i>Ceratothoa oestroides</i> <i>Ceratothoa parallela</i> <i>Nerocila orbignyi</i> (Guérin-Méneville, 1832)		
Scyliorhinidae	<i>Scyliorhinus canicula</i> (Linnaeus)	<i>Eudactylinella alba</i>		<i>Hexabothrium appendiculatum</i> (Kuhn, 1829)	
Torpedinidae	<i>Torpedo marmorata</i> Risso	<i>Pseudocharopinus malleus</i>			<i>Pontobdella muricata</i> (Linnaeus, 1758)
	<i>Torpedo torpedo</i> (Linnaeus)				<i>Branchellion tunisensis</i> Youssef, Benmansour, Yurakhno et Mansour, 2024

**Data analyses**

Rates of infection were evaluated using prevalence (P [%]) and mean intensity of infection (MI) as defined by Margolis et al. (1982).

Three indices were calculated to explore parasites’ diversity on the different hosts:

Species richness [SR]: The number of different parasite species infecting a host (Magurran 2004).

Shannon-Wiener index [H’]: To measure the diversity of species (parasites) in a community. It quantifies both the species richness (the number of different species) and the species equitability (how evenly individuals are distributed among those species) in a community. A higher value of H’ indicates greater biodiversity, with a value of 0 representing a community with only one species (Magurran 2004).

Simpson’s index of diversity (1-D): To assess the diversity of a community by taking into account the number of species present as well as the relative abundance of each species. As species richness and evenness increase, so does diversity. The value of this index ranges between 0 (presenting no diversity) and 1 (presenting infinite diversity) (Magurran 2004).

Pielou evenness index [J’] was calculated to measure the distribution patterns of parasite species on all their hosts. A Pielou index value close to 1 indicates high evenness, where most species have a more uniform distribution on the different specimens belonging to a host species, while a value close to 0 signifies low evenness, with a few species dominating the community (Magurran 2004).

Berger-Parker Dominance Index [d] was calculated to quantify the dominance of the most abundant species in a community

of each host species, where higher values indicate greater dominance by a single species (Magurran 2004).

Analyses were performed using Microsoft Excel 2021.

**RESULTS**

**Composition of communities of ectoparasites and dominant species**

The examination of the different host species resulted in finding 24 ectoparasite species belonging to four taxonomic groups (Table 1). Among the studied eight host species, seven species were infected with parasite species belonging to at least two taxonomic groups. The parasite communities of *Mustelus mustelus* (Linnaeus) and *Mustelus punctulatus* Risso are composed of isopods, copepods and ‘monogeneans’. The parasitic communities of *Bathytoshia centroura* (Mitchill), *Dasyatis pastinaca* (Linnaeus) and *Scyliorhinus canicula* (Linnaeus) consist mainly of copepods and ‘monogeneans’, and that of *Raja clavata* Linnaeus is composed of copepods and isopods. The parasitic community of *Torpedo marmorata* Risso is composed of copepods and leeches. Only *Torpedo torpedo* (Linnaeus) was parasitised by a single species of leech (Table 1).

We noticed that *M. mustelus* has the greatest diversity in parasitic copepods (six species), while *R. clavata* has the highest diversity of parasitic isopods (three species), and *B. centroura* has a greater diversity of ‘monogeneans’ (two

**Table 2.** Dominant parasite species on elasmobranchs off Tunisia.

Host species	Dominant species	Berger-Parker dominance index (d)
<i>Bathytoshia centroura</i>	<i>Nemesis</i> sp.	0.53
<i>Dasyatis pastinaca</i>	<i>Heterocotyle pastinacae</i>	0.43
<i>Mustelus mustelus</i>	<i>Erpocotyle</i> sp. 1	0.48
<i>Mustelus punctulatus</i>	<i>Erpocotyle</i> sp. 2	0.54
<i>Raja clavata</i>	<i>Nerocila orbignyi</i>	0.35
<i>Scyliorhinus canicula</i>	<i>Hexabothrium appendiculatum</i>	0.73
<i>Torpedo marmorata</i>	<i>Pontobdella muricata</i>	0.92
<i>Torpedo torpedo</i>	<i>Branchellion tunisensis</i>	1

species) (Table 1). Moreover, leeches (Hirudinea) are only present on Torpedinidae (Table 1).

The Berger-Parker dominance index (d) allowed us to define the dominant parasite species among the parasite community of the different host species (Table 2). Four ‘monogenean’ species (*Heterocotyle pastinacae* Scott, 1904, *Erpocotyle* sp. 1, *Erpocotyle* sp. 2 and *Hexabothrium appendiculatum* [Kuhn, 1829]) were the dominant species within the parasite communities of their respective hosts (Table 2). For *B. centroura*, the copepod, *Nemesis* sp., was the most dominant species (Table 2). The parasite community of *R. clavata* was dominated by the isopod *Nerocila orbignyi* (Guérin-Méneville, 1832) (Table 2). Both leeches were dominant within their hosts’ parasite communities (Table 2).

The dominance index was quite high for most species, with the lowest recorded index *N. orbignyi*. This demonstrates that a single parasitic species strongly dominates each parasite community (Table 2).

**Parasitological indices of the collected parasite species**

Our analysis shows that the prevalence of isopod species is the lowest among the different taxonomic groups. The highest prevalence for an isopod species is recorded for *N. orbignyi* on *R. clavata*, while the lowest values were recorded for *Anilocra physodes* (Linnaeus, 1758) and *Emetha audouini* (Milne Edwards, 1840) on *M. mustelus* (Table 3).

The prevalence of the different copepod species was relatively low as well. The highest prevalence was recorded for *Nemesis* sp. on *B. centroura* (Table 3). The lowest prevalence among the collected copepods, and in this work, was recorded for *Caligus* sp. on *R. clavata* (Table 3).

Several copepod and isopod species were collected on different host species. Among the four host species infected by *Eudactylinella alba* Wilson, 1932, the highest prevalence was recorded on *D. pastinaca* (Table 3). *Lernaeopoda galei* Krøyer, 1837 exhibited the highest prevalence on *M. punctulatus* (Table 3). *Pseudocharopinus malleus* (Rudolphi in von Nordmann, 1832) had a higher prevalence on *D. pastinaca* than on *T. marmorata* (Table 3).

*Ceratothoa oestroides* (Risso, 1826) prevalence on *M. punctulatus* was slightly higher than on *R. clavata* (Table 3). Among the three host species infested by *Ceratothoa parallela* (Otto, 1828), the highest prevalence was recorded on *M. punctulatus* (Table 3).

‘Monogenean’ species exhibited higher prevalence, and *H. pastinacae* on *D. pastinaca* presented the highest preva-

**Table 3.** Survey of hosts, their ectoparasites and infection parameters.

Host	No. host examined	Hosts infected	Parasite species	Prevalence (%)	Mean intensity
<i>Bathytoshia centroura</i>	129	4	<i>Eudactylinella alba</i>	3.1	1.0
		21	<i>Nemesis</i> sp.	16.3	1.6
		3	<i>Pseudocharopinus bicaudatus</i>	2.3	1.0
		6	<i>Pseudocharopinus concavus</i>	4.6	1.0
		4	<i>Hypanocotyle</i> sp.	3.1	1.5
		7	<i>Monocotyle myliobatis</i>	5.4	1.6
		6	<i>Eudactylinella alba</i>	4.5	1.5
<i>Dasyatis pastinaca</i>	132	8	<i>Pseudocharopinus malleus</i>	6.1	1.0
		34	<i>Heterocotyle pastinacae</i>	25.8	1.2
		7	<i>Eudactylinella alba</i>	1.5	1.1
<i>Mustelus mustelus</i>	480	19	<i>Kroyeria lineate</i>	4.0	1.0
		5	<i>Kroyeria</i> sp.	1.0	1.0
		12	<i>Lernaeopoda galei</i>	2.5	1.2
		2	<i>Nesippus orientalis</i>	0.4	1.0
		4	<i>Perissopus dentatus</i>	0.8	1.0
		2	<i>Anilocra physodes</i>	0.4	1.0
		2	<i>Emetha audouini</i>	0.4	1.0
		6	<i>Ceratothoa parallela</i>	1.3	1.2
		29	<i>Erpocotyle</i> sp. 1	6.0	2.0
		7	<i>Lernaeopoda galei</i>	3.2	1.0
<i>Mustelus punctulatus</i>	216	2	<i>Ceratothoa oestroides</i>	0.9	1.0
		4	<i>Ceratothoa parallela</i>	1.9	1.5
		12	<i>Erpocotyle</i> sp. 2	5.6	1.5
		1	<i>Caligus</i> sp.	0.2	6.0
<i>Raja clavata</i>	480	4	<i>Lernaeopoda galei</i>	0.8	1.0
		4	<i>Ceratothoa oestroides</i>	0.8	1.5
		7	<i>Ceratothoa parallela</i>	1.5	1.4
		11	<i>Nerocila orbignyi</i>	2.3	1.3
<i>Scyliorhinus canicula</i>	480	5	<i>Eudactylinella alba</i>	1.0	1.0
		41	<i>Hexabothrium appendiculatum</i>	8.5	2.0
<i>Torpedo marmorata</i>	63	1	<i>Pseudocharopinus malleus</i>	1.6	1.0
		13	<i>Pontobdella muricata</i>	20.6	2.0
<i>Torpedo torpedo</i>	112	24	<i>Branchellion tunisensis</i>	21.4	1.0

lence. The lowest prevalence among ‘monogenean’ species is recorded for *Hypanocotyle* sp. on *B. centroura* (Table 3).

Both species of leeches had high prevalence with P = 21.4% for *Branchellion tunisensis* Youssef, Benmansour, Yurakhno et Mansour, 2024 on *T. torpedo*, and P = 20.6% for *Pontobdella muricata* (Linnaeus, 1758) on *T. marmorata* (Table 3).

The mean intensity of the different species was relatively low (1 or 2 parasites per infected host). *Caligus* sp. on *R. clavata* had the highest mean intensity (MI = 6) followed by *P. muricata* on *T. marmorata* (MI = 2), *Erpocotyle* sp. 1 on *M. mustelus* (MI = 2) and *H. appendiculatum* on *S. canicula* (MI = 2) (Table 3).

**Table 4.** Indices of the diversity of ectoparasites of elasmobranchs off Tunisia.

Host	Specific richness (SR)	Simpson's index of diversity (1-D)	Shannon-Wiener index (H')
<i>Bathytoshia centroura</i>	6	0.67	1.40
<i>Dasyatis pastinaca</i>	3	0.46	0.80
<i>Mustelus mustelus</i>	10	0.82	1.91
<i>Mustelus punctulatus</i>	4	0.77	1.36
<i>Raja clavata</i>	5	0.64	1.24
<i>Scyliorhinus canicula</i>	2	0.10	0.22
<i>Torpedo marmorata</i>	2	0.11	0.22
<i>Torpedo torpedo</i>	1	0	0.00

### Parasite richness per host family

Among the five families studied, the Triakidae has the highest parasitic richness, with 12 species of parasites. The families Dasyatidae (RS = 8) and Rajidae (RS = 5) also have a high parasitic richness. On the other hand, families Torpedinidae and Scyliorhinidae have parasitic richness = 2 (Table 1).

### Parasite richness per host species

The different fish species were infected by at least one parasitic species (Tables 1, 3 and 4). The analysis of the specific richness (SR) revealed that *M. mustelus* has the highest diversity in ectoparasites, with 10 different species, followed by *B. centroura* (SR = 6) and *R. clavata* (SR = 5). Four host species (*M. punctulatus*, *D. pastinaca*, *T. marmorata* and *S. canicula*) presented a relatively low diversity (SR ≤ 4). *Torpedo torpedo* displayed the lowest richness with a single parasite species (SR = 1) (Table 4).

The Simpson's index of diversity (1-D) varied between 0 and 0.82. *Mustelus mustelus* exhibited the highest value, followed by *M. punctulatus*, *B. centroura* and *R. clavata* (Table 4). This means that the species richness is quite high and evenly distributed on these hosts. Furthermore, the very low values of this index for *S. canicula* and *T. marmorata* confirm that their parasite communities have extremely low diversity and are strongly dominated by a single parasite species (Table 4).

The highest value of the Shannon-Wiener index (H') was observed for the parasite community of *M. mustelus*, followed by those of *B. centroura* and *M. punctulatus* (Table 4). Thus, these hosts have relatively high species richness, and the different parasitic species of each host have fairly similar abundances. Furthermore, the low Shannon-Wiener index (H') index for *S. canicula* and *T. marmorata* reflect the low diversity of their parasite commu-

nities, which is a result of both low species richness and a strong dominance by a single species (Table 4). The value of the Shannon-Wiener index was 0 for *T. torpedo* indicating infection with a single species of parasite (Table 4).

### Seasonal variation of specific richness

The study of parasite richness in the different host species revealed some variation during different seasons (Table 5). The Shannon-Wiener index was higher during spring in most studied species, namely *D. pastinaca*, *M. mustelus*, *M. punctulatus*, *R. clavata* and *S. canicula*, indicating a high species richness (Table 5). This index was the highest during the summer for *B. centroura* and during the autumn for *T. marmorata* (Table 5). Furthermore, this index decreased to reach the minimum recorded, indicating low species richness, during winter for *B. centroura*, during summer for *D. pastinaca* and *M. mustelus*, and during autumn for *R. clavata* (Table 5). The Shannon-Wiener index was low during autumn and winter for *M. punctulatus* and *S. canicula* revealing a very low species richness (Table 5).

Pielou evenness index (J') was relatively high during spring for *D. pastinaca*, *M. mustelus*, *M. punctulatus*, and *R. clavata*, confirming the significant parasitic diversity in these hosts and that the different parasitic species presented similar abundances (Table 5). In addition, this index was high for *B. centroura* during autumn, spring and summer and decreased during winter, suggesting that the parasite diversity of this host is quite low during the winter season (Table 5). As for *S. canicula*, Pielou evenness index (J') was low during autumn, winter and spring, which confirms that the specific richness in this host is quite low and that the majority of parasite specimens correspond to a single species (Table 5). However, Pielou evenness index (J') increased during summer, which indicates that the parasite species that appeared in spring had increased in abundance to levels comparable with the other parasite species of this host (Table 5).

### DISCUSSION

Studying biodiversity is crucial to fully appreciate important biological issues such as speciation, ecosystem function, species interaction (competition, symbiosis, predation and parasitism), ecological importance (productivity and food networks), and economic importance to humans (Hausdorf 2011). Nonetheless, parasites have been largely ignored in biodiversity surveys and ecological studies (Timi and Poulin 2020).

**Table 5.** Seasonal variation of species richness indices of ectoparasites of elasmobranchs off Tunisia.

Seasons	Autumn			Winter			Spring			Summer		
	SR	H'	J'	SR	H'	J'	SR	H'	J'	SR	H'	J'
<i>Bathytoshia centroura</i>	4	1.00	0.72	2	0.41	0.37	5	1.2	0.80	5	1.37	0.85
<i>Dasyatis pastinaca</i>	3	0.73	0.66	3	0.79	0.72	3	0.97	0.88	3	0.60	0.54
<i>Mustelus mustelus</i>	5	1.31	0.81	5	1.26	0.78	10	2.06	0.89	5	1.08	0.67
<i>Mustelus punctulatus</i>	2	0.56	0.80	2	0.56	0.80	4	1.35	0.97	3	0.86	0.78
<i>Raja clavata</i>	3	1.01	0.91	3	1.03	0.93	5	1.54	0.95	4	1.09	0.78
<i>Scyliorhinus canicula</i>	1	0	0	1	0	0	2	0.33	0.47	2	0.20	0.75
<i>Torpedo marmorata</i>	2	0.36	0.51	0	***	***	1	0	0	1	0	0
<i>Torpedo torpedo</i>	1	0	0	1	0	0	1	0	0	1	0	0

Caption. H' – Shannon-Wiener index; J' – Pielou evenness index; SR – specific richness

During this study, 24 ectoparasite species belonging to four taxonomic groups (Isopoda, Copepoda, 'Monogenea' and Hirudinea) were collected from eight elasmobranch species. Moreover, primary morphological examinations suggest that *Caligus* sp., *Kroyeria* sp. and *Hypanocotyle* sp. could represent undescribed species, and further studies are being conducted to accurately identify them.

Among the collected parasites, copepods exhibited the highest diversity with 11 species, followed by 'monogeneans' (six species) and isopods (five species), while only two species of leeches were collected. This taxonomic distribution aligns with previous studies demonstrating that copepods constitute the most diverse group on elasmobranchs, followed by 'monogeneans' (Benz and Bullard 2004, Carrier et al. 2012).

The results of this study reveal that most communities are host-specific and strongly dominated by a single species, as shown by a high Berger-Parker dominance index. *Mustelus mustelus* had the highest parasite diversity, confirmed by the Shannon-Wiener index ( $H' = 1.91$ ) and a high specific richness ( $SR = 10$ ), while other species (*Scyliorhinus canicula*, *Torpedo marmorata* and *Torpedo torpedo*) exhibited low Simpson's index of diversity and Shannon-Wiener index, indicating low species richness. Seasonal analysis showed parasite diversity and evenness (Pielou evenness index) peaked for most hosts in spring, proving a temporal variation.

According to Poulin (1995) and Dallas et al. (2020), parasite variation across hosts, community composition and parasite richness are the result, among other things, of interactions between the evolutionary history and the ecological characteristics of the hosts. The study of the composition of the parasitic communities of different host species revealed that the taxonomic composition of these communities varies from one host to another. Yet, we noticed some similarities among the communities' composition of host species belonging to the same family. Both *M. mustelus* and *Mustelus punctulatus* (Triakidae) share parasitic communities composed of isopods, copepods and 'monogeneans', while the parasitic communities of *Bathytoshia centroura* and *D. pastinaca* (Dasyatidae) were composed of copepod and 'monogenean' species, and the parasitic communities of Torpedinidae species (*T. torpedo* and *T. marmorata*) were primarily composed of leeches species. These results show that parasite communities varied significantly across hosts but exhibited phylogenetic coherence within host families. According to Poulin (2014) and Wells et al. (2019), close phylogenetic relationships between host species may yield more similar parasite community composition.

The non-random distribution of parasite species across host species (Vázquez et al. 2005, Poulin 2014), which was observed in the distribution of the collected parasite species among hosts, was evident, particularly among copepods, which infected the largest diversity of host species and were collected from seven out of the eight host species. *Mustelus mustelus* has the highest species richness in copepods among the different host species studied (Table 1). This host shares some ecological traits, such as gregarious behaviour, active swimming (Smale and Compagno 1997),

and demersal habitat (Mytilineou et al. 2005) with other host species studied. It is one of the most common elasmobranch species encountered during this study (with 480 individuals examined – Table 3) confirming its high density along the Tunisian coasts (Bradaï 2000). High species richness in copepods was previously reported for *M. mustelus* in the Mediterranean Sea (Raibaut et al. 1998).

Parasitic isopods were less frequent and found on three host species (*Raja clavata*, *M. mustelus* and *M. punctulatus*), with *R. clavata* exhibiting the highest species richness (Table 1). These parasites seem to be quite rare on the studied elasmobranchs. Of the eight elasmobranch species examined, only *R. clavata* and *T. marmorata* were previously reported to host parasitic isopods (Bariche and Trilles 2008, Öktener et al. 2009). However, the relatively high specific richness of these parasites on *R. clavata* may reflect ecological overlap between host and parasite habitats; the seasonal migration of this ray species to shallow coastal zones (Holden 1975) coincides with the depth distribution of cymothoid isopods (<200 m; Smit et al. 2014), which may enhance the possibility of getting infected by different parasitic isopods.

'Monogenean' species are typically host-specific ectoparasites (Neifar 2001). During this study, these parasites were found on five host species, with *B. centroura* exhibiting the highest species richness ( $SR = 2$ ), while *R. clavata*, *T. torpedo* and *T. marmorata* were not infected by any species.

*Raja clavata* is known to host several species of 'monogeneans', including *Leptocotyle minor* (Monticelli, 1888) (Henderson et al. 2002) and *Rajonchocotyle batis* Cerfontaine, 1899 (Neifar et al. 1998). *Torpedo marmorata* was reported to host *Amphibdelloides kechemirae* Tazerouti, Neifar et Euzet, 2006, *Amphibdelloides vallei* Llewellyn, 1960, *Empruthotrema raiae* (Maccallum, 1916), *Empruthotrema torpedinis* Kearn, 1976 and *Epicotyle torpedinis* (Price, 1942) (Kearn 1976, Chisholm and Whittington 1999, Tazerouti et al. 2006, Derbel et al. 2022). Two species of 'monogeneans' (*Amphibdella paronaperugiae* Llewellyn, 1960 and *Amphibdelloides benhassinae* Tazerouti, Neifar et Euzet, 2006) were identified as parasitic species of *T. torpedo* as well (Derbel et al. 2022).

The observed low diversity of 'monogenean' species may be explained by two hypotheses; first, the low local abundance or loss of rare 'monogenean' species in the study area, or the environmental stressors, particularly warming trends in the Mediterranean Sea (Giorgi 2006), that may affect these parasite populations. Indeed, alterations in environmental conditions have been linked to changes in the structure and diversity of fish parasite communities (Braicovich et al. 2020). However, more in-depth research is needed to explain these results.

Leeches are common on elasmobranchs, and many species have been reported from the skins of sharks and rays, including thornback ray (*R. clavata*), sand-tiger shark (*Carcharias taurus* Rafinesque) and Argentina angel shark (*Squatina argentina* [Marini]), from regions such as the Indian Ocean, the Atlantic Ocean, Japan, and southern Brazil (Oka 1910, Soto 2000, Wunderlich et al. 2011). The gath-



ered Hirudinea seem to prefer Torpedinidae among the different host species examined. *Pontobdella muricata* is an ectoparasite of benthic elasmobranch species rarely found on teleost fish (Minelli 1979). Its presence on *T. marmorata* can be related to the fact that this host is the only species that lives in a benthic habitat. However, we cannot dismiss environmental factors (salinity, temperature, etc.) since these species were only found in the Gulf of Tunis.

Both leech species exhibited a fairly high prevalence, despite they are being well known for their pathogenic impact on their host. Most leeches are well known to have high prevalence on their host (Bolognini et al. 2017). The results indicated that isopod species and most copepod species displayed relatively low prevalence values. This is possibly due to interspecific competition for space and feeding resources (Karvonen et al. 2011). The notably high prevalence of the copepod *Nemesis* sp. (prevalence 16%) on *B. centroura* suggests species-specific adaptation or favourable microhabitat conditions on gill lamellae. However, further morphological and molecular studies are needed to identify this species.

In contrast, most ‘monogenean’ species (classes Monopisthocotyla and Polyopisthocotyla) exhibited higher parasitological indices than isopods and copepods encountered during this work. Parasites belonging to these classes are reported to have a high parasitological load on their hosts (Neifar 2001). This may be due to their reproduction strategy (a direct life cycle and high reproduction rate) (Ferreira-Sobrinho and Tavares-Dias 2016). Their relatively small size and reduced pathological impact in comparison with the other collected ectoparasite species may explain their persistence at higher density.

The analysis of the species richness revealed that *S. canicula*, *T. marmorata* and *T. torpedo* were characterised by a low species richness ( $RS \geq 2$ ), which is strongly dominated by a single species (*Hexabothrium appendiculatum* on *S. canicula*, *P. muricata* on *T. marmorata* and *Branchellion tunisensis* in *T. torpedo*). Our results are consistent with previous studies of Dallarés et al. (2017) and Santoro et al. (2022), where the authors highlighted the low species richness in *S. canicula* in the Mediterranean. On the other hand, *M. mustelus* presented a relatively rich and diversified parasitic fauna ( $RS = 10$ ) dominated by *Erpocotyle* sp.1.

Furthermore, the species richness varies from one host family to another. The Triakidae exhibited the highest parasitic richness with 12 different species, while the parasitic richness of the Scyliorhinidae and the Torpedinidae is equal to two. This difference may be primarily due to the sampling effort (696 fish from the Triakidae, 480 specimens from Scyliorhinidae and 335 specimens from the Torpedinidae), as sampling effort is of fundamental importance in determining parasite richness (Feliu et al. 1997). Furthermore, it is important to take into consideration the extent of the sampling areas of the different hosts, as representatives of Triakidae and Scyliorhinidae were sampled in the different gulfs of Tunisia, while Torpedinidae were only harvested in the Gulf of Tunis. According to Kamiya et al. (2014), the geographical range of a host correlates positively with the number of parasitic taxa it harbours.

In addition, the species richness of the host should be considered at a geographical scale (Poulin et al. 2011). Mediterranean small to medium-sized sharks generally have less numerous parasite communities compared to their conspecifics from European Atlantic waters (Isbert et al. 2015). According to Raibaut et al. (1998), the Carcharhinidae and the Triakidae exhibit the highest parasitic richness among the Mediterranean Elasmobranch species. However, we noticed that this richness varies among host species even within a single family sharing the same ecological traits (migration, gregarious behaviour or habitat preferences).

In some host taxa, interspecific variation in parasite richness correlates with host species characteristics such as body size, diet, or geographic distribution (Lindenfors et al. 2007). During the present survey, *M. mustelus* presented a higher parasite richness than *M. punctulatus*. These two host species have the same etho-ecological characteristics (Bradaï et al. 2012). This discrepancy may be due to the smaller sample size of *M. punctulatus* and low population density of this host along the Tunisian coasts compared to *M. mustelus*. Kamiya et al. (2014) identified host population density as one of the main universal determinants of interspecific variation in parasite richness. Although *M. punctulatus* is common along the Tunisian coasts, it is considered less abundant than *M. mustelus* (Bradaï 2000).

The composition and richness of parasitic communities exhibited clear seasonal variation. Some parasitic species were detected exclusively during specific seasons, such as *Caligus* sp. which appeared only in summer, while *Emetha audouini* was observed solely in spring. This seasonal fluctuation seems to be related to the life cycles of some parasite species. During warm seasons, environmental conditions become favourable for egg hatching and reduce the time to maturity for ‘monogenans’ (Brazenor et al. 2015), which may explain their proliferation observed during the summer in this study. The life cycle of the host species seems to influence the occurrence of leech species. According to Bolognini et al. (2017), these parasites proliferate during the reproduction period of their hosts.

The search for the determinants of local biodiversity and its spatial and temporal variation remains a central objective of contemporary ecology (Kamiya et al. 2014). In this context, marine parasite communities can be considered effective bio indicators of environmental conditions and trophic network status (Sures et al. 2017). The present study highlights rich and diverse parasitic communities infecting some elasmobranch species off the Tunisian coast. Parasite variation across hosts may be attributed to a complex interplay of ecological and evolutionary factors (Dallas et al. 2020). This study suggests that parasite community composition and structure of the studied hosts are primarily influenced by the geographic distribution, the population density along the Tunisian coasts and the sampling numbers.

The phylogeny of the host species appears to be an important factor as well. According to Poulin (1995), through the phylogenetic history of host species within a given clade, parasite species are acquired or lost like other traits, and can be mapped onto a host phylogeny. Nonetheless,

this study was limited to eight hosts. Therefore, it is imperative to study a larger number of host species and explore other possible intrinsic factors (morphology, diet, age, etc.) and extrinsic factors (environmental variables, geo-morphological characteristics, etc.) to learn more about the different factors that can influence parasite biodiversity, which will provide essential information on the identification of the underlying mechanisms of diversity, as well as on the conservation of biodiversity.

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