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BODY SIZE AND ECTOPARASITIC INFESTATIONS IN THE MEDITERRANEAN POND TURTLE, *MAUREMYS LEPROSA* (TESTUDINES, GEOEMYDIDAE), IN MAJEN BELAHRITI POND (NORTH-EASTERN ALGERIA)

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Body Size and Ectoparasitic Infestations in the Mediterranean Pond Turtle, *Mauremys leprosa* (Testudines, Geoemydidae), in Majen Belahriti Pond (North-Eastern Algeria). Ramdani, K., Ziane, N., Younsi, M., Ben Krinah, C., Bouchahdane, I. & Rouag, R. — Morphological parameters were measured on a population of the Mediterranean pond turtle, *Mauremys leprosa,* (Schweigger, 1812) in Majen Belahriti pond in the region of Guelma (North-east of Algeria). Females exhibited a longer carapace length (CL) at 177 ± 23.85 mm, surpassing that of males (151.45 ± 31.12 mm), and they also displayed a higher body weight (BW) at 756.52 ± 243.91 g, which was notably greater than that of males (453.09 ± 224.42 g). *Placobdella nabeulensis* Ben Ahmed, Gajda, Utevsky, Kwist & Świątek, 2023 (Hirudinida, Glossiphoniidae) was identified as the only ectoparasite affecting *M. leprosa*, with an average body length and width of 11 ± 5.83 mm and 5.1 ± 2.89 mm respectively. The intensity, represented by the average number of leeches per turtle, was calculated to be 2.21 ± 1.31 for this specific population and a prevalence rate of 21.17 %. The tail region showed the highest parasite load, indicating its increased susceptibility compared to other body parts.
Key words: *mediterranean pond turtle,* morphological parameters, leaches, prevalence, infestation, Algeria.

Introduction

Mauremys leprosa (Schweigger, 1812), also known as the Mediterranean pond turtle, is an endemic species found in the Mediterranean basin (Palacios et al., 2015; Laghzaoui et al., 2020). Two subspecies inhabit the African continent: *M. leprosa leprosa* and *M. leprosa saharica* (Fritz et al., 2006, Veríssimo et al., 2016). In Algeria, the subspecies *M. l. saharica* (Fritz et al., 2006) is widely distributed from the coastal zones (Bakhouche et al., 2019) to the Sahara, although with a relict character (Schleich et al., 1996), and its taxonomic status is still uncertain in the western and central parts of the country (Fritz et al., 2006). Its distribution extends from North Africa to the Southern region of France (Bertolero and Busack, 2017; Laghzaoui et al., 2020). Nevertheless, due to agricultural intensification, water management practices and habitat loss, this species is under serious threat (Mateo et al., 2003; Custodio et al., 2009; Díaz-Paniagua et al., 2015). Several studies, however, have been aware of the important population declines over the past four decades (Campo & Ruiz, 1992; Da Silva, 2002; Araújo & Segurado, 2008; Gálvez & Albero, 2017). The presence of alien species, like the red-eared turtle (*Trachemys scripta elegans*), is also considered an important threat due to interspecific competition (Araújo et al., 1997; Da Silva, 2002; Polo-Cavia et al., 2010; Campos-Such et al., 2016; Domènech et al., 2016) and also for being a potential source of infection of disease and parasites (Martínez-Silvestre et al., 2011; Romero et al, 2014; Meyer et al., 2015). Fortunately, to date this species is still not reported in Algeria as an invasive species. Consequently, the IUCN has designated the Spanish pond turtle as "Vulnerable." Despite extensive research on various aspects of its ecology, demography, diet, and habitats in North Africa, our understanding of leech prevalence in this species remains limited, as noted by Bakhouche et al. (2019). The potential role of leeches as vectors of parasites that could affect turtles is critical to study in evolutionary and ecological contexts, as it can significantly alter host population demography (Iglesias et al., 2015). Therefore, this study focuses on the characterization of morphological parameters of a population of *Mauremys leprosa* situated in Majen Belhriti and also analyzing the prevalence of leeches within this population.

Material and methods

Study area. This study was carried out in Majen Belahriti pond, situated in 36°26'09.9 N and 7°05'11.6 E longitude, in North-eastern Algeria (fig. 1). The climate here is Mediterranean with moderate rainfall concentrated during winter months; the rainiest month is January with an average of 90.79 mm. The pond covers approximately 0.6 hectares and is a shallow water body with minimal vegetation.

Field work. The study was conducted from March to August 2021. Due to the lack of fyke nets ideal for capturing turtles, captures were made by hand and with a dip net. A survey session of the pond was conducted each week by one person. The session consisted of a survey around the pond shore every two hours from 10:00 to 16:00 (4 surveys per day) and each time a turtle was observed, we entered the pond and attempted to capture it. It often happens that no turtle is caught during a session. During this period, the study area was surveyed once every 7 days to facilitate the redistribution of individuals (Gibbons, 1990). From March to August 2021, 24 sessions were conducted and a total of 48 turtles were captured. Surveys were conducted only on sunny days. Individual variables were measured for every specimen such as body size and weight. Carapace length was measured using a digital caliper (precision ± 0.1 mm). The body weight of the turtles was measured using a digital balance (precision ± 1 g). We used the carapace length to estimate the growth rates, and placed this measure against the age of the various individuals (Barbault, 1973).

Turtles were marked by notching its marginal scutes (Cagle, 1939). Measures were taken during the tagging process, in particular the disinfection of the lime and the scutes before and after tagging. Seven morphometric variables were measured on each specimen captured (fig. 2) prior to release at the capture site. Sex was determined by secondary sexual characteristics (e. g., plastral concavity, tail length) (Servan et al., 1989). Turtles less than 100 mm in length were considered too small for sexing and were classified as juveniles.

After measurements, the presence/absence, as well as the total number of leeches and their attachment site, was recorded for each turtle. Leeches were collected using tweezers and stored in vials containing 70 % ethanol (labeled with date and turtle code) until further examination was possible. Leeches were counted and identified to species level in the laboratory using the taxonomic keys for Hirudinea in Tunisia (Ben Ahmed et al., 2015).

All turtle specimens were released alive at their site of origin, and the collected leeches were deposited in the scientific collections of the Biology, Water and Environment Laboratory. Parasitism was expressed by calculating two indicators: (a) infestation prevalence (%) = 100 × number of infested turtles/total number of turtles and (b) infestation intensity = number of leeches/number of infested turtles.

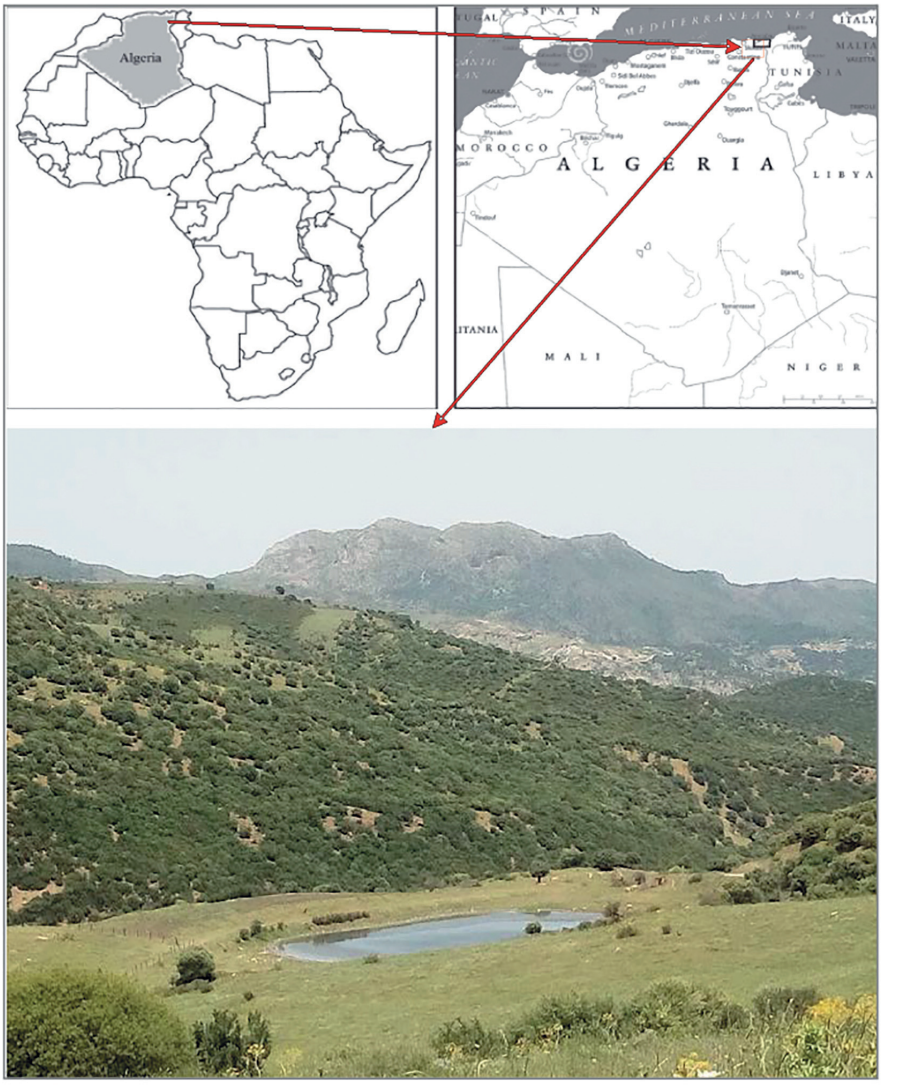

Fig. 1. Location of the study site.

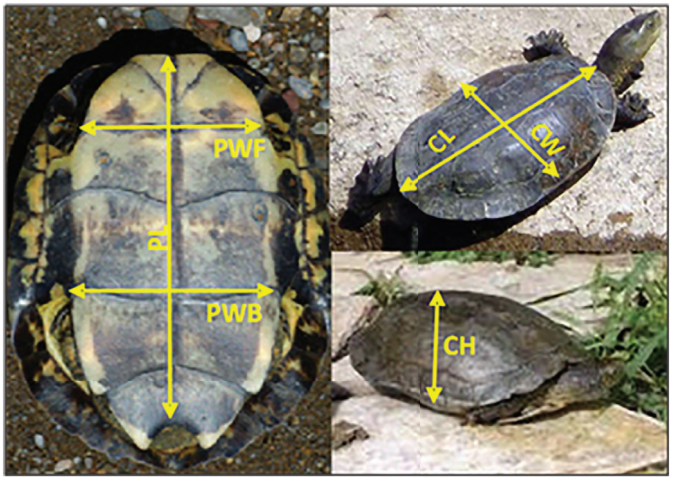

Fig. 2. Morphometrical variables of *Mauremys leprosa* (for abbreviations see table 3).

Statistical analyses. Leech prevalence (%) and intensity, descriptive statistics (Mean ± Sd (Rang) and Pearson's correlation coefficient were also calculated. A t-test was used to analyze differences between genders; differences were considered statistically significant when $P \leq 0.05$. Statistical analysis of the morphological parameters was carried out using the Excel software and Statistix® 8.0.

Results

Population structure. The population is composed of adult individuals, with 89.58 % of those captured (comprising 22 males and 21 females), while only 5 juveniles were found. The domination of adults can be explained by the shorter basking time for juveniles and thus their rare catching by nets. Figure 3 illustrates the size distribution of the studied turtle population. Juveniles, defined as having a carapace length less than 100 mm, made up 10.42 % of the total sample. Among the adults, 39.58 % had a carapace length within the range of 170–200 mm (fig. 3).Haut du formulaire

Sexual dimorphism. The sex ratio was 1.04, it is biased for males. Among the total sample size of 43 turtles, males and females showed significant differences in both carapace length (CL) (males: 151.45 ± 31.12 mm, ranging from 101–202 mm; females: 177 ± 23.85 mm, ranging from 130–225 mm; t-value = –3.01; *p*-value = 0.004; df = 41) and body weight (BW) (males: 453.09 ± 224.424 g, ranging from 132–931 g; females: 756.52 ± 243.91 g, ranging from 333–1327 g; t-value = 4.25; *p*-value = 0.000; df = 41). From morphometrics parameters, it is clearly seen that for all parameters, the average values were higher in female individuals. The t-test shows statistically ($p < 0.05$) differences between males and females for all variables (table 1). The mean carapace length (CL) of juvenile turtles was 71.20 ± 27.44 mm, with a body weight (BW) of 73.60 ± 49.84 g.

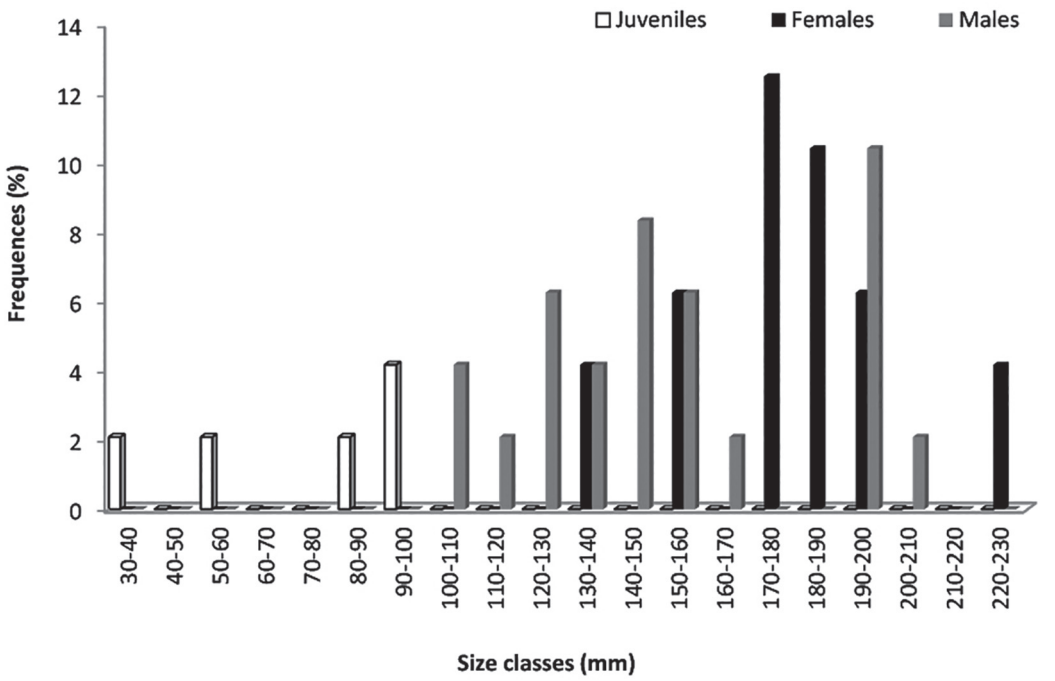

Fig. 3. Size structure of the population of *Mauremys leprosa*.

Table 1. Morphometrics variables

Parameters	Males (N = 22)		Females (N = 21)		*t*-Test	
	X ±SD	Min-Max	X ±SD	Min-Max	t	p
BW, g	453.1 ± 224.4	(132–931)	756.5 ± 243.9	(333–1327)	–4.25	0.000
CL, mm	151.81 ± 31.10	(101.64–202.45)	177.22 ± 23.91	(130.59–225.48)	–2.99	0.005
CW, mm	104.83 ± 14.62	(73.97–127.94)	122.62 ± 18.73	(95.60–176.38)	–3.48	0.001
CH, mm	49.51 ± 8.03	(36.72– 67.50)	60.08 ± 9.67	(40.55–76.03)	–3.90	0.000
PL, mm	122.72 ± 2.17	(73.02–157.18)	50.43 ± 16.76	(114.61–188.22)	–4.48	0.000
PWF, mm	42.31 ± 16.18	(19.27–68)	55.50 ± 18.83	(27.04–82)	–2.47	0.018
PWB, mm	46.18 ± 15.71	(20.08–66)	59.19 ± 19.26	(28.19–85)	–2.43	0.020

Note. X — mean; SD — standard deviation; Min-Max — minimum and maximum measurements; BW — body weight; CL — carapace length; CW — carapace width; carapace height — CH; PL — plastron length; PWF — plastron width front; PWB — plastron width back. t-Test (t) and level of significance (*p*) between males and females ($\alpha = 0.05$).

Growth. A relative constancy of the size of the individuals based on their weight is observed both in males ($r = 0.89$, $p < 0.0001$) and in females ($r = 0.88$, $p < 0.0001$) with a net increase of size and body weight in females (fig. 4).

Prevalence and intensity of leeches infection. A total of 31 leeches were collected and carefully examined and the identification showed the presence of a single species of the leech, *Placobdella nabeulensis* Ben Ahmed, Gajda, Utevsky, Kwist & Świątek, 2023 (Hirudinida, Glossiphoniidae) (fig. 5) a new species described from Tunisia and Algeria (Ben Ahmed et al., 2023) and most closely related to the European species *Placobdella costata* (Fr. Müller, 1846), usually considered to be a Mediterranean species that has spread over large parts of Central and Eastern Europe (Bielecki et al., 2012).

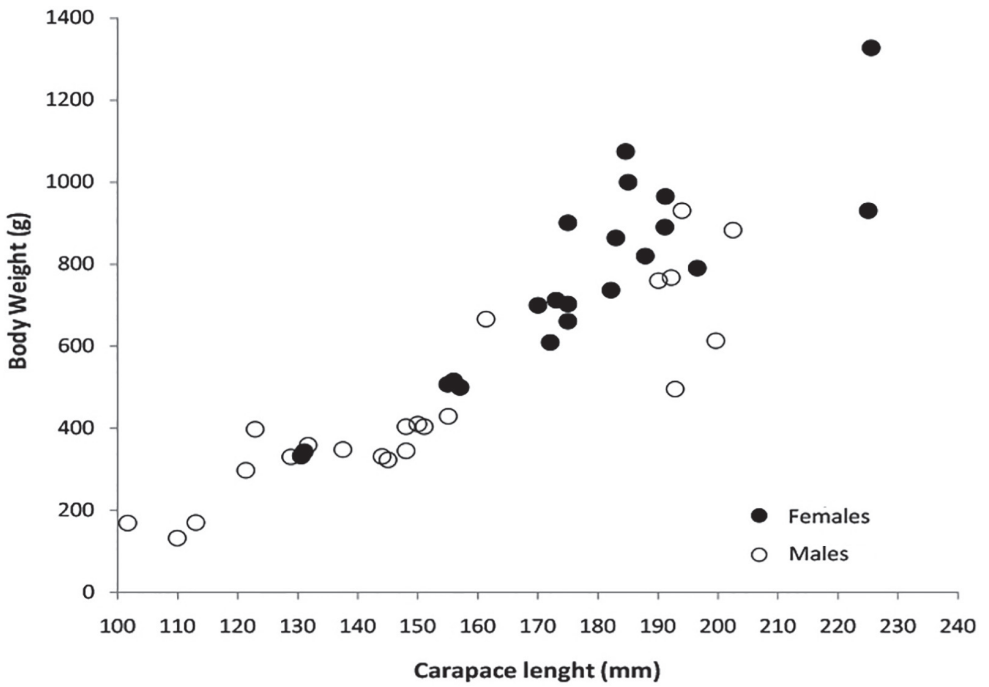

Fig. 4. Linear regression of body weight (BW) on carapace length (CL) for *Mauremys leprosa* (N = 43).

Fig. 5. Adult *Placobdella costata* attached to the carapace of *Mauremys leprosa* (Photos by K. Ramdani).

Table 2. Measurements of leeches

Parametrs	N; Mean ± Sd ; (Min-Max)
Lenght (mm)	31 ; 11±5.83 (4-22)
Width (mm)	31 ; 5.1±2.89 (2-13)

Leeches differed in age and ranged in size from about 4 mm length for the young to 22 mm length for adults. The measurements of the specimens reveal an average length of the body of 11 ± 5.83 mm, which varies between 4 and 22 cm and an average width of the body of 5.1 ± 2.89 mm, which varies between 2 and 13 mm (table 2).

Of the 48 turtles examined, leech infestations were found in 14 individuals, representing approximately 21.17 % of the total sample. More specifically, approximately 31.82 % of male turtles and 33.33 % of female turtles were affected, while no juveiles were parasitised (table 3). On average, each turtle had an intensity of 2.21 ± 1.31 leeches, ranging from 1 to 4 leeches per turtle. It is important to note that there was no statistically significant difference between the mean parasite intensities observed in males and females (t-value = -1.49; p-value = 0.161; df = 12), with respective values of 2.71 ± 1.25 and 1.71 ± 1.25 leeches per turtle.

The correlations between the intensity of the infestation and the individual morphometric parameters of the turtles revealed a single positive relationship with the average front plastron width (PWF; r = 0.544; p = 0.044). The other parameters are negative (PWB r = 0.519, p = 0.057; CH r = -0.102, p = 0.730; CW r = -0.338, p = 0.237; CL r = -0.159, p = 0.587; BW r = -0.360, p = 0.205)

Table 3. Prevalence and intensity of infestation

Turtle data	Leech Prevalence (%)	Intensity		
		N	Mean±Sd	Range
Males (n = 22)	31.82	7	2.71 ± 1.25	(1-4)
Females (n = 21)	33.33	7	1.71 ± 1.25	(1-4)
Juveniles (n = 05)	0	0	–	-
Total (n = 48)	21.17	14	2.21 ± 1.31	(1-4)

Table 4. Correlations between mean parasite intensity and turtle morphometric parameters

(N = 14)	(N = 48)	*r*	*p*
Mean parasite intensity *vs*	Body weight (BW)	-0.360	0.205
	Carapace length (CL)	-0.159	0.587
	Carapace width (CW)	-0.338	0.237
	Carapace height CH)	-0.102	0.730
	Plastron length (PL)	-0.180	0.538
	Plastron Width Front (PWF)	0.544	0.044
	Plastron Width Back (PWB)	0.519	0.057

Table 5. Prevalence of attachment sites on turtles

Attachement sites	Turtles (n = 14)	Leeches		
		N	Mean ± Sd	Range
Tail	5	9	0.64 ± 1.08	(0–3)
Carapace	6	6	0.43 ± 0.51	(0–1)
Plastron	6	6	0.43 ± 0.51	(0–1)
Hind limbs	2	4	0.29 ± 0.83	(0–3)
Fore limbs	2	4	0.29 ± 0.83	(0–3)
Head	1	2	0.14 ± 0.53	(0–2)

Attachment sites. The carapace and plastron exhibited the highest prevalence of infection, each accounting for 42.86 % (6/14). The tail follows closely with a prevalence of 35.71 % (5/15). In contrast, both the hind limbs and fore limbs have a lower prevalence of 14.29 % (2/14) each. The head shows the lowest infection rate at 07.14 % (1/14). Regarding leeches load, the tail, carapace, and plastron of the turtles carry the highest number, with 0.64 ± 1.08 specimens for the tail and 0.43 ± 0.51 ones for both the carapace and plastron, respectively, making them the most susceptible sites. The hind limbs and fore limbs have a comparatively lighter load, with 0.29 ± 0.83 leeches. The head remains the least affected site, with only 0.14 ± 0.52 leeches (table 5).

Discussion

Furthermore, the population shows sexual dimorphism in all morphometric parameters. Sexual dimorphism is related to differences in history strategies of both sexes (Olivier, 2002). When smaller males could be advantaged in their mobility and search for sexual partners; females tend to a larger size which can be correlated with the clutch size (Olivier, 2002).

The studied population is composed by a high proportion of adult individuals (89.58 %). Also in other populations of *Mauremys leprosa* in the Mediterranean basin, Keller (1997) reported a proportion of adults/sub-adults of 94.3 %, in the National Park of Doñana (South-western Spain). Araújo et al. (1997) noted that 67 % of a population in Portugal was adult and Guerdoud and Benyahia, (2021) noted in the "Lac Noir" pond in Northeastern Algeria a high proportion of adult individuals (88.78 %) The sexual dimorphism is similar to that reported in several European populations: in France (Fraysse, 2002), in South-western Spain (Muñoz and Nicolau, 2006). A very similar demographic structure was observed in *Mauremys leprosa* population of Lac Reghaia, in Northern Algeria (Bakhouche et al., 2019). Sexual dimorphism is a variable trait in Chelonians (Agha et al., 2018), and in *Mauremys leprosa,* it is variable over its distribution. The sex ratio plays a crucial role in the ecology of animal species, significantly impacting population dynamics. Ideally, the primary sex ratio should be 1 : 1, but numerous studies, such as Gibbons (1990), have revealed the presence of imbalanced sex ratios. In this particular case, the number of females is nearly equal to that of males, with a sex ratio of 1.04.

Only one species of leech parasitizes the studied population, which is *Placobdella nabeulensis* sp. n**.** (Hirudinida, Glossiphoniidae). This species most closely related to the European species *Placobdella costata* (Fr. Müller, 1846) known for its haematophagous ectoparasitic feeding on freshwater turtles, in Europe mainly on *Emys orbicularis* (Vamberger & Trontelj, 2007), but it has also been reported on *Mauremys leprosa* (Romero et al., 2014) and *Mauremys caspica* (Yadollah & Kami, 2014). Leeches have a smaller size compared to specimens in Northern Iran where the length varies between 7 mm and 26 mm (Bashirichelkasari & Yadollahvandmiandoab, 2017) and in the North-West of

France from 30 mm to 70 mm (Lacaplain & Noel, 2019). In Morocco the average length is 1.9 ± 23.6 mm (min = 13-max = 28) and the width is 1.3 ± 7.9 mm (6.51–14.22) (Laghzaoui et al., 2020).

Our results suggest that males are more infested than females. Various results have been found in Eastern Poland (Bielecki et al., 2012), North-eastern Algeria (Fediras et al., 2017) and Morocco (Laghzaoui et al., 2020). This difference can be explained by two factors: (1) the high level of testosterone in males, which is considered to decrease the immune response (Freedberg et al., 2008; Laghzaoui et al., 2020). Indeed, increased testosterone levels during the mating season in males may reduce the energy reserved for defense against parasites. (2) The movements of males in search of females and competition with other males make them more vulnerable to many parasites (Diaz-Paniagua, et al., 2001). Juvenile turtles are healthy because they occupy marginal habitats (Berry and Shin, 1980; Amo et al., 2004).

Analysis of the attachment sites of leeches on *M. leprosa* revealed that there are preferences for specific sites. The tail, carapace and plastron were the most affected, while other sites showed a less affected parasite. Leeches generally prefer to attach to softer skin areas; hence, the specific attachment sites observed in our study were the tails. Our results are similar to those found in other studies of leech attachment sites in Spain (Domínguez & Villarán, 2017), North-eastern Algeria (Fediras et al., 2017) and Morocco (Laghzaoui et al., 2020) where the preferred attachment part is the posterior.

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