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Volume7, Issue 4: 1-7



Copepod Parasites of the Gills of *Luciobarbus Callensis* (Valencienne, 1842) and *Carassius Carassius* (Linnaeus, 1758) (Cyprinid Fish) Collected from Beni Haroun Dam (Mila, Algeria).

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ABSTRACT

In aquaculture, the crustaceans are classified among the most dreadful parasites due to their high potential contamination hazards. Some parasitic copepods are responsible for a massive mortality of stocks, however, there has been a lack of knowledge on the Algerian freshwater fish. Therefore, the present work aimed to study the infestation of copepod ectoparasites of 499 fish, including 257 individuals of Luciobarbus callensis (Valencienne, 1842) and 242 of Carassius carassius (Linnaeus, 1758) species found in Beni Haroun Dam (Mila, Algeria). The fish were subjected to some morphometric measurements, and thereby, a gill dissection was measured, harvested, preserved and identified, as well the parasitic indices were calculated. The effect of some parameters (size, sex season, and microhabitat) on the parasite infestation was tested by using χ^2 test. The gill examination of the individuals of the two host populations led to an inventory of eight parasitic species in Luciobarbus callensis callensis (P= 12,84%; I=2.33; A=0.29) (Tab.1). Nevertheless, the seasonal evaluation of the parasite indices of copepods obtained from the same species revealed that the most important values were noted in summer (P=23,63%; A=0,61; I=2,61). Moreover, Carassius carassius provided high infestation rates and parasite infestation charges, respectively during spring (I=1.77; A=0.21) and summer (P=12.76%). The χ^2 values showed the effect of season (χ^2 obs = 18.833 > χ^2 $_{0.05}$ = 6.251 ddl=3) and the size (χ^2 obs = 14,639 > χ^2 0,05 = 4.605; ddl=2) on the parasite infestation of the two host species, since there was no effect due to the microhabitat and sex.

Keywords: Beni Haroun Dam, Luciobarbus Callensis, Carassius Carassius, Parasitic Copepods, Parasitic Indices.

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1. INTRODUCTION

Fish parasites have been important determinants of general health of hosts, and thus, have been the best indicators of hosts and the structure of food chains (Sindermann, 1987). The class of crustaceans contains very diversified parasites, along with highly variable morphology, and also they belong to several large groups in which the most abundant parasitic crustaceans have been associated to the super-order of copepods and the isopoda order (Cressey, 1983). Interestingly, parasitic copepods can cause pathogenic effects on cultured

freshwater fish (Kabata, 1985; Piasecki *et al.*, 2004; Boucenna 2017), and mostly the youngest fish become infested (Piasecki *et al.*, 2004). As a result, various fish can be highly affected by these parasites, i.e some parasitic copepods cause the proliferation of gill tissues, since others are responsible for massive stock mortalities (Fryer, 1968; Johnson *et al.*, 2004, Rückert *et al.*, 2009; Meddour, 2009). The prevention of the infections and epidemics caused by these parasites is generally a prerequisite for the aquaculture success (Euzet *et al.*, 1996). The pathogenic character of parasitic crustaceans effectively requires a better understanding of their taxonomy and the dynamic of the parasite populations. In Algeria, very limited works have been conducted on the parasitic crustaceans of freshwater fish. The parasites of Cyprinidae family have been previously investigated (Meddour, 2009; Meddour et al. 1989)

in Bounamoussa river, (Loucif, 2009) in various water areas of El-Kala National Park, (Meddour et *al.* 1989) in Oubeira lake (Chaibi, 2014) in various continental hydrosystems of the *"Aurés"* and Northern Sahara regions, and (Brahmia, 2017) in Oubeira lake. This work screened the freshwater fish parasites, and subsequently led to an inventory of copepod parasites infesting two populations of Cyprinidae (*Luciobarbus callensis* and *Carassius carassius*) found in Beni Haroun dam (Mila, Algeria). Thus, the present study aimed to evaluate the epidemiological indices of the identified parasites as a function of some biotic parameters of the host (size and sex) and abiotic parameters (season and the microhabitat).

2. MATERIALS AND METHODS

The Beni Haroun dam is one of the huge dams in Algeria, having length of 710m, height of 120m and 710km² of surface, as well as reserving capacity of 960 m³. It is located at 19km away of Mila city, and 15 km north of Constantine city, and it supplies several Eastern Algerian cities (Cities: Jijel, Batna, Oum el Bouaghi, Khenchela, Constantine and Mila), hence, providing a great quantity of irrigation water for hundred hectares of the agricultural holdings (Marmi et *al*, 2008; Mebarki et *al*, 2008).

Sample collection, and fish dissection

The seasonal sampling was randomly performed by using a gillnet on 499 fish, including 257 of *Luciobarbus callensis* and 242 of *Carassius carassius* individual species. The obtained fishes were indentified exactly according to the nomenclature and criteria given by Fischer et al (1987). The samples were immediately carried to the laboratory, afterwards for being weighted, measured and dissected. Fish sex was determined after dissection of the abdominal cavity. Since the gill arches were delicately detached by two incisions (dorsal and ventral), the gill arches were quickly conserved into pill boxes containing ethanol 70% for 6 to 8 hours after the host death. Also, the gills covering the mucus led to difficult detection of the place in which, the parasite was dead, detached from the gill arch and stuck to the mucus.

Harvesting and identification of parasites

The collected copepod parasites from the two gills were kept in small pill containers containing 5% formalin, each in one pill, and were labeled by the name of fish species, the sampling date and the number of sample. The identification of parasite species was based on the morpho-anatomical characteristic examinations as described by Yamaguti (1963).

Data processing

This step was performed by using the parasitic indices proposed by Margolis *et al* (1982) and Bush *et al* (1997).

Statistical analysis

The independence between the parasitic indices and various factors, including season, size and micro-habitat were statistically analyzed by χ^2 test, using Statistica software for Microsoft Windows (StatSoft, version 8.0). Statistical significance of the means was evaluated at 5% (*P*<0.05).

3. RESULTS AND DISCUSSION

The examination of the gills of 499 fish of the two host species; *Luciobarbus callensis* and *Carassius carassius* caught in the Beni Haroun dam led to collect 114 parasitic copepods. The observation of the morpho-anatomical criteria of the collected parasites promoted identifying eight species, namely *Ergasilus briani, Ergasilus negaceros, Ergasilus sieboldi, Paraergasilus brevigiditus, Neoergasilus japonicus, Lernea cyprinacea, Ergasilus peregrinus and Neoergasilus longispinosis* on the gills of *Luciobarbus callensis and* six species including: *Ergasilus briani, Ergasilus negaceros, Ergasilus sieboldi, Paraergasilus briani, Ergasilus negaceros, Ergasilus sieboldi, Paraergasilus brevigiditus, Neoergasilus japonicus, Lernea cyprinacea* on the gills of *Carassius carassius.*

Variation of the epidemiological indices as a function of season

The highest values of parasitic indices of Luciobarbus *callensis* were observed during the summer (P=23,63%; A=0,61; I=0,61) and autumn (P=15%; A=0,2; I=1,33), but no parasitic infestation was noticed during the winter. On the other hand, *Carassius carassius* showed that the highest values of the parasite indices of six identified parasite species were recorded in the summer (P = 12.76%, A = 0.17, I = 1.33) and autumn (P = 11, 66%, A = 0.15, I = 1.28). The χ^2 test showed the effect of the season on the parasitism in both host species (χ^2 obs = 18.833 χ^2 = 6.251, *ddl* = 3) (**Tab.1**).

Variation of parasitism as a function of sex of fish hosts

As shown in table 2, the prevalence was higher in males of *Luciobarbus callensis* (P = 14.19%), since the intensity and abundance were higher in females of the same species (I = 3, A = 0.36). Also, *Carassius carassius* revealed the highest parasite infestation values in females (P = 11.76%, A = 0.17, I = 1.5), meanwhile both host fish species showed no influence of sex on the copepod infestation (χ^2 obs =0,408 < χ^2 0,05=3,841; *ddl=1*)(**Table.2**)

Variation of parasitism as a function of host fish sizes

Luciobarbus callensis revealed that the counting of parasitic copepods following size classes of host fishes showed a high rate of parasitic infestation in small specimens (P = 18, 18%). The highest parasitic charges were recorded in large specimens (I = 2, 3, A = 0.30) (Tab.3). *Carassius carassius* showed the values of infestation rates and the highest abundances in specimens of size between 25 and 35 cm (P = 15%, A = 0.2), since the highest parasite intensities were noticed in individuals of size ranged between 15 and 25 cm (I = 1.45) (**Table.3**).

 χ^2 test showed the effect of size on the parasitism of the two host species (χ^2 obs =14,639 > χ^2 0,05 =4.605; *ddl=2*).

Variation of parasitism as a function of microhabitat

The distribution of parasite indices of copepods collected by microhabitat in *Luciobarbus callensis* showed that the highest prevalence and abundance values were observed in arch 1 of the right gill (P = 5.44%; = 0.07), since the highest values of the mean intensity were observed at the arch 4 of the right gill (I = 2.4). Although, *Carassius carassius* revealed that the highest infestation rates were observed on the right gill and arch I (P = 3.30%), and increased parasite charges were found in arch 4 of the right gill (I = 1.5, A = 0.01) (Tab.4). The prevalence of parasites in the two host species was the same between the right and left gills (χ^2 obs = 0.392 < χ^2 20.05 = 3.841, *ddl* = 1), showing thus no effect of microhabitat on the infestation of the collected parasitic copepod (**Table.4**).



Figure 1: Map of sampling site (Dam Beni Harroun)

 Table 1: Seasonal variation in prevalence, (P%) of mean intensity (I) and average abundance (A) of parasitic copepods in Luciobarbus callensis and Carassius carassius

		Lucio	barbus ca	illensis		Carassius carassius				
Season	NHE	NHI	P(%)	Ι	Α	NHE	NHI	P (%)	Ι	Α
Winter	30	00	00	00	00	60	04	6,66	1	0,07
Springs	77	1	1,29	1	0,01	75	09	12	1,77	0,21
Summer	110	26	23,63	2,61	0,61	47	06	12,76	1,33	0,17
Autumn	40	06	15	1,33	0,2	60	07	11,66	1,28	0,15

NHE: Number of examined hosts; NHI: Number of infested hosts

 Table 2: Prevalence distribution P (%) of mean intensity (I) and mean abundance (A) of parasitic copepods collected as a function of sex in both host species

in both host species									
Li	uciobarbus calle	Carassius carassius							
Sex	ď	ę	ď	ç					
NHE	92	165	106	136					
NHI	13	20	10	16					
P (%)	14,13	12,12	9,43	11,76					
Ι	1,3	03	1,3	1,5					
Α	0,18	0,36	0,12	0,17					

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Luciobarbus callensis							Carassius carassius				
	NHE	NHI	P (%)	Ι	Α	NHE	NHI	Р%	I	Α	
[15-25]	11	2	18.18	1.5	0.27	142	11	7.47	1.45	0.11	
[25-35]	124	15	12.09	2.33	0.28	100	15	15	1.33	0.2	
[35 - 45]	122	16	13.11	2.31	0.30	00	00	00	00	00	

Table 3: Prevalence distribution P (%), Mean intensity (I) and average abundance (A) of copepods collected as a function of size classesin both host species.

Table 4: Prevalence distribution (P%), of mean intensity (I) and mean abundance (A) of copepods by arch gills in both host species.

Luciobarbus callensis	Right gill					Left gill			
Arcs	NHE	NHI	P(%)	Ι	Α	NHI	P(%)	Ι	Α
1	257	14	5,44	1,28	0,07	8	3,11	1,37	0,04
2	257	10	3,89	1,1	0,04	2	0,77	1	0,007
3	257	03	1,16	1	0,016	4	1,55	1	0,01
4	257	05	1,94	2,4	0,04	11	4,28	1,45	0,06
Carassius carassius	NHE	NHI	P(%)	Ι	Α	NHI	P(%)	I	Α
1	242	8	3,30	1,25	0,04	6	2,47	1,16	0,028
2	242	1	0,41	1	0,004	4	1,65	1,25	0,02
3	242	3	1,23	1	0,012	6	2,47	1,16	0,028
4	242	2	0,82	1,5	0,01	1	0,41	1	0,004

4. DISCUSSION

The examination of 257 individuals of the population of Luciobarbus callensis and 242 of Carassius carassius obtained from Beni-Haroun dam promoted to collect 77 and 37 of the individual parasites, respectively in barbel and crucian carp. Moreover, the morpho-anatomical characteristics of the parasites collected from the two species gills led to identify eight species of parasites belonged to two families and four genera (Ergasilus sieboldi, Ergasilus briani, Ergasilus megaceros, Ergasilus peregrinus Paraergasilus bervigiditus, Neoergasilus longispinonis, Neoergasilus japonicus, Lernea cyprinacea), hence, the investigation showed that Luciobarbus callensis exhibited higher parasitic diversity (8 species) than those seen in Carassius carasius (6 species). Brahmia (2016) found in Oubeira Lake that the barbel species hosted a number of parasites, eleven times more than those found in Cyprinus carpio species. Combe (1990) confirmed the existence of variability in the distribution of parasites passing from one species to the other. In addition, Ternengo (2005) proved that each fish species exhibited parasito-fauna characteristics and typical infestation levels, and this was supported by the work of Benmansour (2001) which was carried out in Tunisian coasts, and those done by Ramadhan (2009) and Boualleg (2010) in the Algerian coasts, showing a diversity of fish parasitic copepods from different marine waters. Additionally, Piasecki et al (2004) identified the Ergasilus Sp infestation as a worldwide distributed parasitic disease for agriculture, as well the authors have precisely showed that Ergasilus sieboldi had no specificity for the host, and could infest the majority of freshwater fishes. Whilst, Mokhayer (1981) and Jalali (1998) reported that Lernea cyprinacea had a broad host spectrum,

making the species as hoghly pathogenic in the family of Cyprinidae. On top of that, Meddour (2009) has reported the presence of Lernea cyprinacea species on the Barbus callensus gills in lake Oubeira and Bounamoussa river (northeastern of Algeria), since Chaibi (2014) has identified Ergasilus sp and Lernea sp from the individual gills of Barbus sp caught respectively in the Timgad dam (Batna) and the El Ghaicha dam (Laghouat). Furthermore, Boucenna (2017) reported the presence of Neoergasilus japonicus and Ergasilus sieboldi of Carrassius carrassius in the Foum El- Khonga and Ain El Dalia dams (Souk-Ahras), including also the species, namely Ergasilus sp and Neoergasilus sp, Gnathia sp, Lernea cyprinacea and Argulus foliaceus of Barbus callensis and Cyprinus carpio. In contrast, the host species of Abramis brama showed no parasitic infestation in the two studied zones. On the other hand, Barbus Sp obtained from the lake Oubeira revealed the predominance of parasitic crustacean (Argulus foliaceus) and the nematode Contracaecum sp in Cyprinus carpio, and so these differences were probably due to the geographical particularity of the study sites (Brahmia, 2017). Sasal (1997) declared that different scales could lead to opposite results or conclusions.

The results of the parasitic indices of copepods collected from the eight parasite species showed variations associated to the four seasons, indicating that infestation rates and parasite charges in *Luciobarbus callensis* were found to be highly elevated during the summer and autumn, somewhat weak in the spring, and then no parasite infestation in the winter. In *Carassius carassius*, the highest infestation rate was recorded in the summer, since the parasitic charges were found as important in the spring, along with no parasitic infestation in the winter. These data were in line with those reported by Brahmia (2017), proving that the infestation rate of *Barbus callensis* and Cyprinus *carpio* by the copepod parasites were

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high in the summer and spring, but weak in the winter. Nonetheless, Boucenna et al (2015) have shown that the infestation of Cyprinus carpio by Ergasilus sieboldi, Ergasilus peregrinus and Lernea cyprinacea was high in the summer and spring, and absent in the winter. The data of this study concurred with those reported by Nematoallah et al (2013) in pisciculture centers of Mashad (northeast of Iran), showing that the infestation of Cyprinus carpio by Lernea cyprinacea was maximal in the spring and absent in the winter. In this context, Benmansour (2001) reported that the seasons played a very important role in the development and abundance of the parasitic copepods, whilst the time acted as one of the essential factors causing seasonal fluctuations in the parasite populations. According to Koskivaara et al 1991, the change in water temperature was generally considered to be one of the most important factors determining the presence and abundance of parasites. As reported, the changes of water temperature have been generally considered as a crucial factor, determining the presence and abundance of parasites (Koskivaara et al., 1991). The statistical analysis confirmed the influence of seasons and size on the infestation of the copepod parasites in the two host fishes. Importantly, the study of parasitism considering the function of sex in the species of Luciobarbus callensis and Carassuis carassuis proved no significant differences in the prevalence of caught copepods found between both sexes (male/female) of the two host fish species, thus indicating that the two fish sexes were infested in the same way. Of note, the absence of the influence of fish sexes on the parasitic infestation has been already investigated by Bilong-Bilong (1995) on monogenic gills of Hemichromis fasciatus, by Kassi et al. (2009) on Cichlido gyrusacerbus and Cichlido halli in Ayame dam (Ivory Coast), and by Boucenna (2017) on Barbus callensis, Carrassius carassius and Cyprinus carpio infested by Ergasilus sp, Neoergasilus sp and Lernea sp in Ain El Dalia dam (Souk-Ahras). Besides, Abdulhusein (2014) has shown that the prevalence of *Ergasilus sp* was influenced by the sex of Cyprinus carpio and Oreochromis miloticus; and likewise Anvarifar et al. (2014) found significant differences in the prevalence of the two sexes of Caporta gracilis infested by copepods, Tracheliates polycolus. The parasitism study as a function of size classes of Luciobarbus callensis and Carassius carassius showed that the highest parasitic charges were noticed in the big-size specimens. The results did not concord with those reported by Perez-Bote (2000) in the river of Guadiana on Leuciscus alburnoides, Chondrostoma willkommii ,Barbus sclateri host species, Talib & Jawad (2011) in Iraq on Cyprinus carpio, Ibrahimi (2012), on Tilapia zillii, Stavrescu-Bedivam et al (2014) in Romani on Lepomis gibbosus and by Boucenna et al (2015) on Cyprinus carpio in Foum El Khanga dam and Carassius carassius in Ain El Dalia dam, showing that big-size fishes are highly infested by copepod parasites. Moreover, Sasal et al (1997) suggested that the big-size hosts are sensitive to provide a great number of parasitic richness. The long-term exposure of a big fish to parasites would likely increase the infestation by diversified parasite communities. On the other hand, Luque and Alves (2001) considered the relationship between size of hosts, and the parasitic prevalence and intensities as a large model observed in marine and freshwater fish. In contrast, Poulin (2000) claimed that this model cannot be generalized due to some quantitative and

qualitative differences in the study of fish class-sizes. The distribution of parasitic indices of copepods collected by microhabitat in Carassius carassius and Carassius carassius has shown that the arches of the right gill are highly infested as compared to those of the arches of the left gill, and hence the highest values of the infestation and abundance rates have been recorded at the arch 1, since the intensity increases at the gill arch 4. The work of Boucenna et al. (2015) carried out on Cyprinus carpio in Foum El-khanga dam showed significant differences in the distribution of Ergasilus sieboldi, Ergasilus peregrinus and Lernea cyprinacea between gill arches. Indeed, the levels 1 and 2 of the right gill arches and levels 2 & 3 of the left gill arches were found to have the highest infestation rates. Also, Boucenna et al. (2015) reported that in the species Carasisus carassius found in Foum El-khanga dam, the highest infestation rates were recorded in the arches 3 and 4, whilst the highest parasitic charges were observed at the arches 2 & 3. No significant differences were noticed in the distribution of these parasites between the gill arches of the two sides. As reported by Oliver (1987), the differences in the localization of various parasitic groups in the microhabitat of gill parasite have been explained by the different physiological and ecological needs as revealed by each group. Timi (2003) reported that Lernanthropus cypnoscicola occupies different attachment sites in Cynosciongua tucupa, since the highest prevalence values were recorded in the arches 4 followed by arches 1, 2, and 3. Many advanced hypotheses have elucidated the distribution of the parasites on the biotope of gill teleost fishes. Some authors (Hanek and Fernando, 1978; Adams, 1986; Koskivaara and Valtonen, 1991; Gutierrez and Martorlli, 1994) showed that the median arches 2 and 3 have been highly infested because of the large volume of the blistering currents of water carrying the parasites. Furthermore, Koskivaar et al. (1991) have found that the preference of median arches has been associated to the large attachment surface offered to parasites.

5. CONCLUSION

The examination of 499 fish belonging to two species, namely *Carassius carassius* and *Carassius carassius* obtained from Béni-Haroun (Mila, Algeria) dam led to collecting 114 parasites and identifying eight species of copepod parasites (*Ergasilus sieboldi , Ergasilus briani, Ergasilus megaceros, Paraergasilus bervigiditus ,Neoergasilus japonicus* and *Lernea cyprinacea Ergasilus peregrinus* and *Neoergasilus longispinosus*); and other freshwater surfaces, including dams, rivers and lakes in Algerian territory; increasing the sampling effort for the precise identification of some parasitic communities; knowing the effect of these parasites on growth and development of the host fish;

monitoring the evaluation of *Luciobarbus callensis*, exhibiting higher important diversified parasites (8 species) than that of *Carassius carasius* (6 species).

The study of the epidemiological indices showed that:

- *Luciobarbus callensis* and *Carassius carasius* have not been the same in facing parasitisms;
- The species *Luciobarbus callensis* has been highly infested by the copepod parasites;

- The copepod parasites greatly infested both host species during summer and fall;
- The sex and the microhabitat of the two host species had no effect on the parasitic infestations;
- The season and the size of the two host fish influenced on the infestation rates caused by copepod parasites.
- The work required further studies for being more elucidated (Perspectives of the study):
- The knowledge on the copepod parasites of freshwater fish should be increased, in order to develop similar studies of the other -teleost fish that was not included in this investigation;
- The study should be extended on the abiotic and anthropic factors as temperature, pH, salinity, and its impact on the parasitism.

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