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PREVALENCE, INTENSITY AND ABUNDANCE OF HELMINT PARASITES INFECTIONS ON WILD SEA BASS, *Dicentrarchus labrax* (MORONIDAE) FROM BEYMELEK LAGOON LAKE, ANTALYA, TURKEY

Nesrin EMRE*[1], Ayşegül KUBİLAY[2], Ali AYDOĞDU[3]

[1] Mediterranean Fisheries Research, Production and Training Institute, Antalya, Turkey
[2] Süleyman Demirel University, Faculty of Fisheries, Isparta, Turkey
[3] Uludağ University, Mustafakemalpaşa Vocational School, Mustafakemalpaşa, Bursa, Turkey

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*Corresponding Author: Tel.: +90 242 2510585; E-mail: nemre70@hotmail.com

ABSTRACT

A total of 87 wild sea bass, *Dicentrarchus labrax*, was collected from the Beymelek Lagoon, Antalya, Turkey between January 2009 and December 2009, and examined for the presence of helminth parasites. The effects of season, host size, sex and age on helminth infection parameters was investigated. Of 87 *D. labrax,* 62 (71.2%) were found to be infected by at least one parasite. In total, three helminth parasite species were identified: the monogenean *Diplectanum aequens*, the digenean *Acanthostomum absconditum* and the larval nematode *Hysterothylacium* sp.. *D. aequens* was the most common species in this study and a total of 2172 parasites were recorded on 62 fishes. Total prevalence and mean intensity of *D. aequens* were 71.2% and 35.03±31.7 parasite/fish, respectively. A total of 69 *A. absconditum* were found in 9 of 87 fish examined (10.3%; 7.6±7.1 parasite/fish). *Hysterothylacium* sp. larvae was observed in only one fish examined (1.1%). All three helminth species showed a definite seasonal cycle in prevalence of infection. The highest prevalence and significantly higher abundance of infection were observed in the winter for *D. aequens*, and in the autumn for *A. absconditum* and *Hysterothylacium* sp. larvae. Differences were observed in infection levels at different age groups. Prevalence and mean intensity of *D. aequens* were the lowest in age group of 3 whereas the highest in age groups of 1 and 5. *A. absconditum* was found in all age groups of fish, except for age group 3. *Hysterothylacium* sp. larvae were observed only at 5th age group. Present study also showed that the sex of the host also influenced the parasite burden. The prevalence of infection was higher in male fishes. Forty two males were found to be infected by one or more parasites species. The overall prevalence of infection was 84% and 54.1% for males and females, respectively. The larger sized fish were also more heavily infected than small fish. *A. absconditum* is reported for the first time from European sea bass in Turkey and this species is also a new record for the helminth fauna of Turkey.

Key words: *Dicentrarchus labrax, Diplectanum aequens, Acanthostomum absconditum, Hysterothylacium* sp. larvae, seasonal effects, host size, sex, age, Beymelek Lagoon, Turkey

BEYMELEK LAGÜN GÖLÜ'NDEN (ANTALYA, TÜRKİYE) YAKALANAN *Dicentrarchus labrax* (MORONIDAE) DOĞAL LEVREK BALIKLARINDA HELMİNT PARAZİT ENFEKSİYONLARININ YAYGINLIĞI, YOĞUNLUĞU VE BOLLUĞU

ÖZET

Beymelek Lagün Gölü'nden (Antalya, Türkiye) toplam 87 doğal levrek balığı *Dicentrarchus labrax* Ocak-Aralık 2009 tarihleri arasında yakalandı ve helmint parazitleri açısından incelendi. Bu çalışmada, helmint

enfeksiyon parametreleri üzerine mevsimin, konak büyüklüğünün, cinsiyetinin ve yaşın etkisi araştırıldı. Toplam 87 *D. labrax*'ın 62 (%71.2)'sinin en az bir parazit ile enfekte olduğu tespit edildi. Monogenean *Diplectanum aequens*, digenean *Acanthostomum absconditum* ve Nematode *Hysterothylacium* sp. larvası olmak üzere üç helmint parazit türü tanımlandı. Bu çalışmada, *D. aequens* en yaygın tür olup, incelenen 62 balıkta toplam 2172 parazit kayıt edildi. *D. aequens*'nin enfeksiyon yaygınlığı %71.2 ve yoğunluğu 35.03 ± 31.7 parazit/balık'dır. Seksen yedi balığın dokuzunda toplam altmış dokuz *A. absconditum* incelendi (%10.3; 7.6 ± 7.1 parazit/balık). *Hysterothylacium* sp. larvası sadece bir balık örneğinde gözlemlendi (%1.1). Üç helmint türünün enfeksiyon yaygınlığı belirgin bir mevsimsel döngü gösterdi. *D. aequens* için en yüksek enfeksiyon yaygınlığı ve önemli derecede yüksek enfeksiyon bolluğu kış aylarında, *A. absconditum* and *Hysterothylacium* sp. larvası için sonbahar aylarında gözlemlendi. Farklı yaş gruplarındaki enfeksiyon seviyelerinde farklılılar gözlemlendi. *D. aequens*'in ortalama yoğunluk ve yaygınlık seviyesi üç yaş grubunda en düşük, 1 ve 5 yaş grubunda en yüksektir. *A. absconditum* 3 yaş grubu hariç diğer yaş gruplarında bulundu. *Hysterothylacium* sp. larvası sadece 5 yaş grubu konak bireylerinde gözlemlendi. Bu çalışma, aynı zamanda konak cinsiyetinin de parazit kümelenmesinde etkili olduğunu gösterdi. Enfeksiyon yaygınlığı erkek balıklarda daha yüksek tespit edildi. Kırk iki erkek balığın bir ya da daha fazla parazit türü ile enfekte olduğu, enfeksiyon yaygınlığı erkeklerde %84, kadınlarda %54.1 tespit edildi. Büyük boyutlu balıklar küçük boyutlu balıklara nazaran daha ağır enfektedir. *A. absconditum* Türkiye'de levrek balığından ilk defa rapor edildi ve bu tür aynı zamanda Türkiye helmint faunası için yeni kayıttır.

Anahtar kelimeler: *Dicentrarchus labrax, Diplectanum aequens, Acanthostomum absconditum, Hysterothylacium* sp. larvası, mevsimsel etkiler, konak büyüklüğü, cinsiyet, yaş, Beymelek Lagün Gölü, Türkiye

INTRODUCTION

The parasites of the Sea bass, *Dicentrarchus labrax* in Turkey have been investigated by Kırkım (1998), Tokşen (1999, 2007), Horton and Okumura (2001), Öktener and Trilles (2004), Öktener et al. (2009) and Akmırza (2010). Among the ectoparasites of the sea bass, monogenean parasites, *Diplectanum aequens* were recorded from the Aegean Sea (Tokşen 1999, 2007), Bodrum region, Muğla (Akmırza 2010) and the Black Sea (Öktener et al. 2009). The Crustacean fauna was found to be more diverse than the monogenean fauna. Previously, six species of Crustacea, *Caligus minimus, Lernanthropus kroyeri, Ceratothoa oestroides, C. italica, Anilocra physodes* and *Nerocila orbignyi* have been recorded (Kırkım 1998, Horton and Okumura 2001, Öktener and Trilles 2004, Akmırza 2010). However, when all published information is considered, the data on parasitic helminths of European sea bass in Turkey are still insufficient. Moreover, none of the previous studies have focused on the frequency of parasitic infection and its variation due to season and host size, age and sex. Therefore, the aim of the present study was mainly to fill these gaps in the knowledge on the parasitic helminths of sea bass and to determine how the prevalence and mean intensity of helminth infections varied according to season, the size, age, and sex of the European sea bass.

MATERIALS AND METHODS

The Beymelek Lagoon with a length of 2.5 km, width of 2.1 km and surface area of 225 hectares is located in the western Mediterranean region of Turkey, 36° 00´16´´ N, 30° 00´03´´ E. Fish specimens were collected by fishing net from the Beymelek Lagoon from January 2009 to December 2009. Random samples were taken representing the different fish lengths in every three months. A total of 87 fish specimens were dissected. The fish were placed in plastic containers filled with lagoon water and immediately transferred to the research laboratory, some 2 km away. At necropsy, the total length (as an indicator of host size) sex and age (calculated by analysis of fish scales using the methods of Lagler (1966) and Begenal (1978)) of each fish were determined. Fishes were divided into five age groups. During the dissection, the viscera, gills, gastrointestinal tract liver, kidney, heart, swim bladder, gall bladder, eyes, fins and body surfaces were examined separately for helminths with a dissecting microscope. All helminths found in each individual fish were identified and counted. Monogeneans were permanently mounted

using the ammonium picrate glycerine procedure according to Gussev (1968) and Fernando et al. (1972). The other helminths were killed in hot saline solution; digeneans were fixed in 70% ethanol stained with iron aceto carmine (Georgiev et al. 1986), cleared and mounted in Entellan. Nematodes were fixed in 70% ethanol, cleared in drop of glycerol and then mounted on glass slides. Data on each species was categorized according to the season of collection and host size, sex and age. All parasites were identified using selected identification keys of Yamaguti (1985a,b), Radujkovic and Euzet (1989), Bray (2001). The prevalence, mean intensity and mean abundance were calculated according to Bush et al. (1997). Standard statistical computation (standard deviation) was carried out using Microsoft Excel (Office 2000). Analysis of regression (Minitab 14.00) was applied to the data to determine the existence of any meaningful association between abundance of helminth species and fish size.

RESULTS

Of 87 *D. labrax* examined, 62 were found to be infected by at least one parasite. Overall 71.2% of the fish were infected. In total, three helminth parasite species were identified: the monogenean specialist *Diplectanum aequens* on the gills; the digenean trematode *Acanthostomum absconditum* in the intestine and the larval nematode *Hysterothylacium* sp. in the abdominal cavity. *A. absconditum* is reported for the first time in Turkey. Data on infection parameters are presented in Table 1-5. A total of 2172 *D. aequens* were recovered from 62 of 87 individuals of *D. labrax* examined (prevalence 71.2% mean abundance 24.9±31.1) (Table 1). Seasonal prevalence varied between 35 and 100%, being the highest in the winter and the lowest in the summer. Mean intensities ranged between 8.9 and 51.4 with the highest value in the winter. Mean abundance varied from 3.2 to 51.4 with the highest in the winter (Table 2). A total of 69 specimens of *A. absconditum* were found in 9 of 87 fish examined (prevalence 9%, mean abundance 0.7±3.1) (Table 1). Seasonal prevalence of the infection was the highest in the autumn with 43.7%. The highest abundance and intensity values were also found in the autumn (Table 2). This species was not detected in the winter, spring and summer samples. Only a single species of nematode, *Hysterothylacium* sp. larvae was observed in one fish examined in the autumn with prevalence of 6.2% and mean intensity of 2 parasites per fish. The prevalence and intensity levels of infections of three parasite species varied with host size (Table 3). Prevalence of *D. aequens* was found between 56.4 and 88.8%. The maximum prevalence and mean intensity levels (37.3 parasites per fish) were in size group III. However, there was no a statistically significant relationship between the size of *D. labrax* and the abundance of *D. aequens* ($r = 0{,}214$; $p > 0.046$). Prevalence of *A. absconditum* was the lowest (5.1%) in size group II while the highest (16.6%) in size group III. The maximum mean intensity (11) was also found in the later group. Prevalence and intensity levels of the parasite species according to age are presented in Table 4. *D. aequens* was found at all age groups of fish with a varying prevalence between 50% and 100%. The highest levels of infection occurred at 1 year-old fish. For *A. absconditum*, higher prevalence was also observed in fish at this age (100%) as compared to the other age groups. *Hysterothylacium* sp. larvae occurred only in host fish at age groups of 5, with a prevalence of 12.5% and mean intensity of 2 parasite per fish (Table 4). Prevalence of *D. aequens* was higher in males (84%) when compared to females (54.1%). Similarly, mean intensity of *D. aequens* was higher in males (41.8 parasite per fish) than females (20 parasite per fish) (Table 5). Although no difference in prevalence of *A. absconditum* could be found

Table 1. Overall prevalence (P%), mean abundance (MA) and mean intensity (MI) of helminths in *Dicentrarchus labrax* caught from the Beymelek Lagoon between January 2009 and December 2009

Helminth species	n	P (%)	MA ± SD	MI ± SD	Intensity (range)
D. aequens	62	71.2	24.90 ± 31.1	35.03 ± 31.7	3-141
A. absconditum	9	10.3	0.70 ± 3.1	7.60 ± 7.1	1-26
Hysterothylacium sp. larvae	1	1.1	0.02 ± 0.2	2	2

n: number of infected fish; SD: standard deviation; prevalence (P%); mean abundance (MA) and mean intensity (MI)

Table 2: Seasonal changes in infection parameters of the helminths in *D. labrax* caught from the Beymelek Lagoon between January 2009 and December 2009

Season	Autumn (n=16)				Winter (n=30)			
Helminth species	P	MA±SD	MI±SD	Range	P	MA±SD	MI±SD	Range
D. aequens	93.7	29.6±20.8	31.6±19.9	3-75	100	51.4±34.9	51.4±34.5	7-141
A. absconditum	43.7	4.1± 6.5	9.4± 7.1	3-20	6.6	0.1± 0.4	1.5± 0.7	1-2
Hysterothylacium sp. larvae	6.2	0.1± 0.5	2	2	-	-	-	-
Season	Spring (n=21)				Summer (n=20)			
Helminth species	P	MA±SD	MI±SD	Range	P	MA±SD	MI±SD	Range
D. aequens	47.6	4.2±6.4	8.9±6.7	1-26	35	3.2±5.7	9.1±5.7	2-20
A. absconditum	-	-	-	-	-	-	-	-
Hysterothylacium sp. larvae	-	-	-	-	-	-	-	-

P: prevalence (%); MA: mean abundance; MI: mean intensity: SD: standard deviation.

Table 3: Infection parameter variations the helminthes due to host size groups in *D. labrax* caught from the Beymelek Lagoon between January 2009 and December 2009

Size groups	I (n= 30)	II (n=39)	III (n=18)
Length (cm)	25.1-30	30.1-35	35.1<
Diplectanum aequens			
Prevalence (%)	83.3	56.4	88.8
Mean intensity ±SD	29.4±28.2	19.5±31,3	37.3±37.2
Total parasite number	737	763	672
Acanthostomum absconditum			
Prevalence (%)	13.3	5.1	16.6
Mean intensity ±SD	7.5±8.5	3.0±1.4	11.0±7.2
Total parasite number	30	6	33
Hysterothylacium sp. larvae			
Prevalence (%)	0	0	5.5
Mean intensity ±SD	0	0	2
Total parasite number	0	0	2

Table 4. Infection parameter variations of the helminthes due to host ages in *D. labrax* caught from the Beymelek Lagoon between January 2009 and December 2009

Age groups	1	2	3	4	5
Sample size	1	45	25	8	11
D. aequans					
Prevalence (%)	100	71.9	50	85.7	87.5
Mean intensity	44	34.6±30.2	16.5±13.3	24.6±24.6	63.5±44.5
Total parasite no	44	1419	116	148	445
A. absconditum					
Prevalence (%)	100	8.7	0	28.5	12.5
Mean intensity	4	6.4±7.7	0	10±9.9	13
Total parasite no	4	32	0	20	13
Hysterothylacium sp. larvae					
Prevalence (%)	0	0	0	0	12.5
Mean intensity	0	0	0	0	2
Total parasite no	0	0	0	0	2

Table 5. Infection parameter variations of the helminthes due to host sex in *D. labrax* caught from the Beymelek Lagoon between January 2009 and December 2009

Helminth species	FEMALE (n:37)			MALE (n:50)		
	Prevalence (%)	Mean intensity	Total parasite no	Prevalence (%)	Mean intensity	Total parasite no
Diplectanum aequens	54.1	20.6±16.2	413	84	41.8±35.1	1759
Acanthostomum absconditum	10.8	9.7± 6.2	39	10	6±7.9	30
Hysterothylacium sp. larvae	2.7	2	2	0	0	0

between males (10.8%) and females (10%), its mean intensity was higher in female (9.7 parasite per fish) than males (6 parasite per fish) (Table 5). Only one *Hysterothylacium* sp. larvae was found in a female fish (Table 5).

DISCUSSION

The purpose of this study was to determine how the prevalence and mean intensity of helminth infections varied according to season, the size, ages and sex of the European sea bass, *D. labrax*. Throughout the study period, *D. aequens* was the dominant species with a prevalence of 71.2% and mean intensity of 35.03 parasite/fish. Previously, few studies have reported parasites of *D. labrax* in Turkey (Tokşen 2007, Öktener et al. 2009, Akmırza 2010). In Bodrum, Muğla, Akmırza (2010) reported a prevalence of 23.88% and 21.54% for this parasite in cultured and wild sea bass, respectively. In an investigation conducted in Ordu, Öktener et al. (2009) determined an overall infestation prevalence of 100% and mean intensity of 3.45 parasite/fish for *D. aequens* on cultured sea bass. There are also other studies on cultured and wild sea bass population in different regions (Gonalez-Lanza et al. 1991, Sterud 2002, Mladineo 2005, Dezfuli et al. 2007, Fioravanti et al. 2007). Ganzala-Lanza et al. (1991) reported *D. aequens* and *D. laubieri* on the sea bass from the Spanish Mediterranean area. Further, Sterud (2002) reported 100% prevalence of *D. aequens* on the sea bass from Norway. They reported that *D. aequens* are pathogenic for the gills. Mladineo (2005) also determined a prevalence of 62.03% for *D. aequens* on farmed *D. labrax* in the Eastern Adriatic Sea. Similarly, Dezfuli et al. (2007) reported prevalence of 73.6% and mean intensity of 34.6 parasite/fish for this parasite in cultured *D. labrax* populations. Fioravanti et al. (2007) reported a prevalence of 39.6% for this parasite on sea bass held in extensive and intensive farming systems in Italy. Our findings about the prevalence and mean intensity of this parasite are similar to those reported by Dezfuli et al. (2007). We found a digenetic trematodes *A. absconditum* in the intestine of wild *D. labrax*. *A. absconditum* is reported for the first time from the European sea bass in Turkey and also this species is a new record for the helminth fauna of Turkey. Sterud (2002) noted the trematoda was the dominant parasite group in his study and recorded eight of the digenean species, *Brachyphallus crenatus, Derogenes varicus, Timoniellaim butiforme, T. praeterita, Labratrema minimus, Cainocreadium labracis* and *Bucephalus* spp. on *D. labrax*. On the other hand, Mladineo (2005) and Fioravanti et al. (2007) did not sample any digenean species in farmed *D. labrax* in the Adriatic Sea and Italy, respectively. To our knowledge, *A. absconditum* has been reported as a species infecting Bagrid fish (*Bagrus bayad* and *B. docmac*) from Egypt (Fishcthal and Kuntz 1963, Ibraheem 2006), Sudan (Khalil 1963, 1969, 1971) and Ghana (Thomas 1958). In addition, Moravec (1976) reported this species from African Bagrid fish. The inconsistent reports about the digenetic trematodes of sea bass may be attributed to the differences in the ecological and hydrobiological conditions of the localities. Furthermore, Sterud (2002) assumed that "On the basis of their indirect life cycle, which includes two intermediate host- a mollusk and a benthic fish - digeneans are not likely to be encountered as adults in farmed sea bass'', being in line with findings of Mladineo (2005) and Fioravanti et al. (2007), who did not reported digenetic trematodes in farmed sea bass. Other parasitic helminth species reported in this study was *Hysterothylacium* sp. larvae from the abdominal cavity of one host fish. Sterud (2002) reported presence of *H. aduncum* larvae on the same host species and indicated that larval stages of *H.*

aduncum could cause considerable mortality in halibut fry fed with natural zooplankton. In the present study, *D. aequens, A. absconditum* and *Hysterothylacium* sp. larvae showed a definite seasonal cycle in terms of prevalence. The highest prevalence and significantly higher mean abundance of infection were observed in the winter for *D. aequens*, and in the autumn for *A. absconditum* and *Hysterothylacium* sp. larvae than other seasons. However, Mladineo (2005) pointed out that abundance and prevalence of *D. aequens* on sea bass in cages from the Adriatic increased in the summer and fall, and decreased in the winter. Our results coincides with those of Bauer (1957), Lom (1970), Chubb (1977), Hanzelova and Zitnan (1985), Öztürk and Altunel (2006) and Turgut et al. (2011), who found that monogenean parasites showed higher abundance and prevalence in the spring and early summer. This corresponds with the spawning of the host fish. Similarly, Hanzelova and Zitnan (1985) reported a higher monogenean infection during the spawning period. Kennedy (1968) pointed out that the seasonal prevalence of *Caryophyllaeus laticeps* (cestoda) in dace showed a decrease in the winter and an increase in the summer. He explained that this trend was resulted from changes in fish resistance to the infection during spawning as well as variations in water temperature, which was also the case in study of Kennedy and Walker (1969). The latter suggested that seasonal changes in prevalence of *C. laticeps* are related to changes in the immune response of host fish at different temperatures. In our study area, the spawning of *D. labrax* population takes place in the winter months and over winter in lagoons rather than open sea. Our data of seasonal prevalence of *D. aequens* suggest that these changes are generally associated with the spawning and over wintering of the host fish. However, our findings are also in agreement with the results of Öztürk and Altunel (2006) and Hanzelova and Zitnan (1985). Prevalence and mean intensities of *A. absconditum* and *Hysterothylacium* sp larvae were the highest in the autumn (Table 2). This could be related to changes in abundances of plankton and food compositions as well as food preference of *D. labrax* in different seasons. In agreement with our findings, many studies have been published on the seasonal changes of helminth infections in fish (Dogiel 1962, Wooten 1978, Lassiere 1988, Aydoğdu et al. 2003, Tekin-Ozan et al. 2008). The highest prevalence and mean intensity of *D. aequens* were observed in larger fish (>35.1cm) compared with the other groups. The size of fish has been shown to affect their monogenetic parasites simply because of larger fish provide more space for parasite attachment (Aydoğdu et al. 2003, Tekin-Ozan et al. 2008). Accordingly, these authors determined a higher prevalence of infection on the largest size class than smaller classes. Our findings are in agreement with the results of Aydoğdu et al. (2003), Özer and Öztürk (2004), Tekin-Ozan et al. (2008). There are other studies showing a preference of monogenean parasites for small size classes (Pojmanska 1994, Öztürk 2002). Variations for *A. absconditum* and *Hysterothylacium* sp. larvae infections depending on host size and age were also similar to that of *D. aequens* infection, suggesting that larger fishes are more heavily parasitized than smaller ones (Table 3 and 4). There are several studies regarding the relationship between the infection levels of endohelminth parasites and age of host fish (Dogiel 1958, Karanis and Taraschewski 1993, Aydoğdu et al. 2003, Tekin-Ozan et al. 2008). The findings of the present study are consistent with those authors who found that prevalence and mean intensity levels of endo helminth infection increased with the increase of age and length of the host fish. Existing literature observations indicate that the food preference by a host may determine the kind of its parasite fauna. In this study, larger fishes are more heavily parasitized with *A. absconditum* and *Hysterothylacium* sp. larvae than smaller ones, impliying that larger fish (older fish) in the Beymelek Lagoon feed selectively on mollusk and benthic fish which serve as intermediate hosts for digenean species, and zooplankton which are intermediate hosts for nematoda species. In this study, it is clear that there were significant differences between the overall prevalence infections of male and female fishes. Males were more frequently infected with helminth parasites than females (overall prevalence 84% for males and 54.1% for females). However, prevalence and mean intensity of parasitic helminth infections varied in both sexes. The overall prevalence of *D. aequens* on male seems to be higher than female. Özer (2002) have reported that higher *Dactylogyrus extensus* and *D. anchoratus* infection in males than those on females of *Cyprinus carpio*. Pickering and Christie (1980) determined higher prevalence and intensities on male than females. They

also explained that this might be a result of differences in color, hormonal states and mucus between female and male fish. However, Özer and Öztürk (2004) reported that a higher *Gyrodactylus arcuatus* infection in females than in males. Sönmez (1996) also reported that a higher *D. extensus* infection in female than males of *C. carpio*. Similar results were obtained by Kır (1998) for the infestation of *D. extensus* on the same host species. A higher prevalence of *D. aequens* in male observed in this study confirms the findings of Özer (2002) and Pickering and Christie (1980). Prevalence of *A. absconditum* showed similar prevalence in females and males (10.8 vs 10%). This is consistent with the fact that sea bass of both sexes feed on similar the benthic or planktonic invertebrates which serve as intermediate hosts for digenean species. In conclusion, these variations in infections of the three parasitic helminth species according to seasons, host length, size, age and sex might be influenced by various factors such as the hydrobiological conditions of water, changes in host diets and feeding habits and physiological state of fish (Bauer 1959, Kennedy 1969, Collard 1970). However, future studies with a more extensive sampling are needed in order to better understand prevalence and intensity of infections of three parasitic helminthes due to season, host fish size, age and sex.

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