COMPARATIVE INVASIVE ASIAN TAPEWORM *Bothriocephalus acheilognathi* INFECTIONS ON THE LOWER KIZILIRMAK DELTA FISHES

Türkay ÖZTÜRK[1]*, Ahmet ÖZER[1]

[1]Sinop University, Faculty of Fisheries and Aquatic Sciences, 57000, Sinop, Turkey

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*Corresponding Author: Tel.: +903682876265; E-mail:turkay.ozturk@gmail.com

ABSTRACT

In the present study, the occurrence of invasive Asian tapeworm *Bothriocephalus acheilognathi* in different fish species from Bafra Fish Lakes, the Lower Kızılırmak Delta, Turkey, was investigated. A total of 1329 fish specimens of *Cyprinus carpio, Vimba vimba, Carassius gibelio, Scardinius erythropthalmus, Neogobius fluviatilis, Proterorhinus marmoratus, Pomatoschistus marmoratus, Mugil cephalus, Liza aurata, Sander lucioperca, Aphanius danfordi, Gambusia affinis, Gasterosteus aculeatus, Atherina boyeri, Cobitis taenia* and *Syngnatus acus* belonging to 10 different families were examined. Routine parasitological investigation procedures were applied to fish specimens. The number of cestode recovered from each fish species was counted and infection prevalence (%) and mean intensity values (number±standard error) were calculated. *B. acheilognathi* was recovered from the intestine of *C. carpio, V. vimba, C. gibelio, N. fluviatilis, P. marmoratus, S. lucioperca, A. danfordii, G. affinis* and *A. boyeri*. *C. carpio* (26.72% and 16.66 ± 2.39) and *V. vimba* (20% and 5.88 ±1.85) had the highest infection prevalence and mean intensity values among all infected fish. This study revealed that *C. gibelio, P. marmoratus, S. lucioperca, A. danfordii* and *G. affinis* are the new host records for *B. acheilognathi* in Turkey.

Key words: *Bothriocephalus acheilognathi,* infection indices, Turkey

AŞAĞI KIZILIRMAK DELTASI BALIKLARINDA KARŞILAŞTIRMALI İSTİLACI ASYA SESTODU *Bothriocephalus acheilognathi* ENFEKSİYONLARI

ÖZET

Bu araştırmada, Asya sestodu istilacı *Bothriocephalus acheilognathi* parazitinin Aşağı Kızılırmak Deltasındaki Bafra Balık Göllerinde yaşayan farklı balık türlerinde bulunuşu araştırıldı. Araştırmada *Cyprinus carpio, Vimba vimba, Carassius gibelio, Scardinius erythropthalmus, Neogobius fluviatilis, Proterorhinus marmoratus, Pomatoschistus marmoratus, Mugil cephalus, Liza aurata, Sander lucioperca, Aphanius danfordii, Gambusia affinis, Gasterosteus aculeatus, Atherina boyeri, Cobitis taenia* ve *Syngnatus acus* olmak üzere 10 farklı familyaya ait toplam 1329 adet balık incelendi. Uzatma ağları ve elektro şok ile yakalanan balıklara rutin parazitolojik metotlar uygulandı. İncelenen balıklarda tespit edilen parazitlerin sayıları kaydedildi, enfeksiyon oranları (%) ve enfekte balık başına ortalama parazit sayıları (adet±standart hata) hesaplandı. *B. acheilognathi* türü bu araştırmada *C. carpio, V. vimba, C. gibelio, N. fluviatilis, P. marmoratus, S. lucioperca, A. danfordii, G. affinis* ve *A. boyeri* türü balıkların bağırsaklarında varlığı belirlendi. En yüksek enfeksiyon *B. acheilognathi* oranı ve yoğunluğu *C. carpio* (%26.72 ve 16.66 ± 2.39) ve *V. vimba* (%20 ve 5.88 ±1.85) bireylerinde belirlendi. Bu araştırma ile *C. gibelio, P. marmoratus, S. lucioperca, A. danfordii* ve *G. affinis* türleri Türkiye'de *B. acheilognathi* sestodunun konaklarına eklendi.

Anahtar kelimeler: *Bothriocephalus acheilognathi*, enfeksiyon yoğunluğu, Türkiye

INTRODUCTION

Bothriocephalus acheilognathi is an invasive parasite, commonly known as the Asian tapeworm. It was first described in Japan from the intestine of *Acheilognathus rhombea* (Yamaguti 1934). The native hosts of this tapeworm are grass carp *Ctenopharyngodon idella* and silver carp *Hypothalmichthys molitrix* (Choundry et al. 2006). Moreover, it has adapted successfully itself to the common carp, *Cyprinus carpio* (Boomker et al. 1980). It has been widely spread by two ways; i) import and export of exotic cyprinids for culture, ii) grass carp that were used for controlling macrophytes in different regions throughout the world (Boomker et al. 1980, Hoffman 1980, Andrews et al. 1981). The Asian tapeworm is known to infect over 100 species of fish in almost every continent, except Antarctica, and is considered a threat to populations of endemic, commercial and hatchery fishes (Körting 1975, Hoffman 1980, Hoffman and Schubert 1984, Salgado-Maldonado and Pineda-Lopez 2003). High invasiveness and pathogenicity placed it in the list of species selected for the recent Handbook of Global Freshwater Invasive Species (Francis 2011). The overall success of Asian tapeworm can be attributed to its ability to infect a wide variety of fish and copepod hosts and short time requirement to complete its development in the intermediate host (Körting 1975, Dove and Fletcher 2000). Each segment of an adult worm is capable of producing eggs, allowing individual parasites to be able to produce up to 20,000 eggs per day. Eggs are passed with fish faeces, and mobile coracidia emerge from the eggs after embryonation. The coracidia are consumed by the intermediate host, cosmopolitan cyclopoid copepods (*e.g., Acantocyclops, Macrocyclops, Mesocyclops, Tropocyclops* and *Diacyclops*) (Körting 1975, Marcogliese and Esch 1989, Diaz-Castaneda et al. 1995). Fish are the final host of the parasite and the life cycle is completed when fish ingest infected copepods. This comprehensive research study intended to make more contributions to our current knowledge about this parasite and host fish relationship by presenting results on 16 fish species belonging to 10 different families from a previously unstudied area, the Lower Kızılırmak Delta, Samsun, Turkey.

MATERIALS AND METHODS

Fish specimens were collected from Bafra Fish Lakes in the Lower Kızılırmak Delta located along the Black Sea cost, Samsun, Turkey (41° 38' N; 36° 04' E) (Figure 1). The delta covers an area of 50,000 ha, which includes freshwater marshes, swamps and 7 lakes and lagoons (Ulu, Uzun, Cernek, Liman, Karaboğaz, Tatlı and Gıcı). Fish samples were collected with the aid of an electro-schock device and fishing net from December 2010 to November 2011. Totally, 16 fish species belonging to 10 families were investigated (Table 1). After capture, fish were

Figure 1. Map of the Lower Kızılırmak Delta and sampling locations

delivered alive to the laboratory and examined within 48 h using the standard procedures. Briefly, the entire gut of each fish host was examined under a stereomicroscope, and cestodes encountered in each fish were counted. Individual worms then fixed and preserved in 70% alcohol, mounted in glycerine jelly or in ammonium picrate-glycerine under sufficient coverslip pressure to flatten the parasite specimens. Photomicrographs were taken using Olympus BX53 microscope attached with an Olympus DP25 digital camera. For Scanning Electron Microscopy (SEM), some samples of several monogenean species were hydrated, placed in 1% osmium tetroxide overnight, dehydrated in ethanol, air dried and mounted on stubs with double-sided adhesive tape and sputter coated with gold-palladium and examined in Jeol JSM-6510LV at an accelerating voltage of 10kV. Infection prevalence and mean intensity were calculated in accordance with Bush et al. (1997). Water temperature (°C), salinity (ppt), oxygen (mg/L) and nitrate (mg/L) levels were measured using a YSI-Proplus digital water analyser at the sampling sites. Kruskal-Wallis test (Non-parametric ANOVA) was performed to compare differences in the mean intensity values recorded in different seasons. The analyses were carried out using a computer programme (GraphPad Instat 3.0) and P-values less than 0.05 was considered to be significant.

RESULTS

A total of 1328 fish specimens were investigated for parasites and Asian tapeworm was found in the intestine of 116 fish. All developmental stages of adult, juvenile, plerocercoid and egg of *B. acheilognathi* were found. Microphotographs representing those stages under the light microscopy and SEM are shown in Figure 2. Nine of 16 fish species examined in the present study was infected by *B. acheilognathi* and the data on host-list of *B. acheilognathi* with infection prevalence and mean intensities values are presented in Table 1. Common carp, *C. carpio,* had the highest infection rate among all infected fish species (Table 1).

DISCUSSION

Bothriocephalus acheilognathi is a notable species among fish cestodes due to its broad host dispersion. It has been recovered from more than 100 species of freshwater fish around the world (Salgado-Maldonado and Pineda-Lopez 2003). Some characteristics of the parasite such as lack of specificity at both definitive and intermediate hosts and ability to colonize rapidly in new habitats plus an uncontrolled and indiscriminate translocation of cultured fishes led to a successful spread within the aquatic system (Salgado-Maldonado and Pineda-Lopez 2003). This species is of a global

Table 1. Prevalence and mean intensity values of the Asian tapeworm on examined fish species from the Lower Kızılırmak Delta (n=1328)

Examined Fish species	Examined Fish number	Infected Fish Number	Prevalence (%)	Mean Intensity±SE
Cyprinus carpio	232	62	26.72	16.66 ± 2.39
Vimba vimba	40	8	20.00	5.88 ±1.85
Carassius gibelio	32	3	9.37	5.33 ± 2.33
Scardinius erythrophthalmus	28	0		
Neogobius fluviatilis	161	8	4.97	1.37 ± 0.26
Proterorhinus marmoratus	45	8	17.78	1.25 ± 0.16
Pomatoschistus marmoratus	16	0		
Mugil cephalus	254	0		
Liza aurata	46	0		
Sander lucioperca	73	1	1.37	1.00 ± 0.00
Aphanius danfordii	125	1	0.80	1.00 ± 0.00
Gambusia affinis	227	24	10.57	2.04 ± 0.33
Gasterosteus aculeatus	29	0		
Atherina boyeri	10	1	12.5	2.00 ± 0.00
Cobitis taenia	3	0		
Syngnatus acus	8	0		

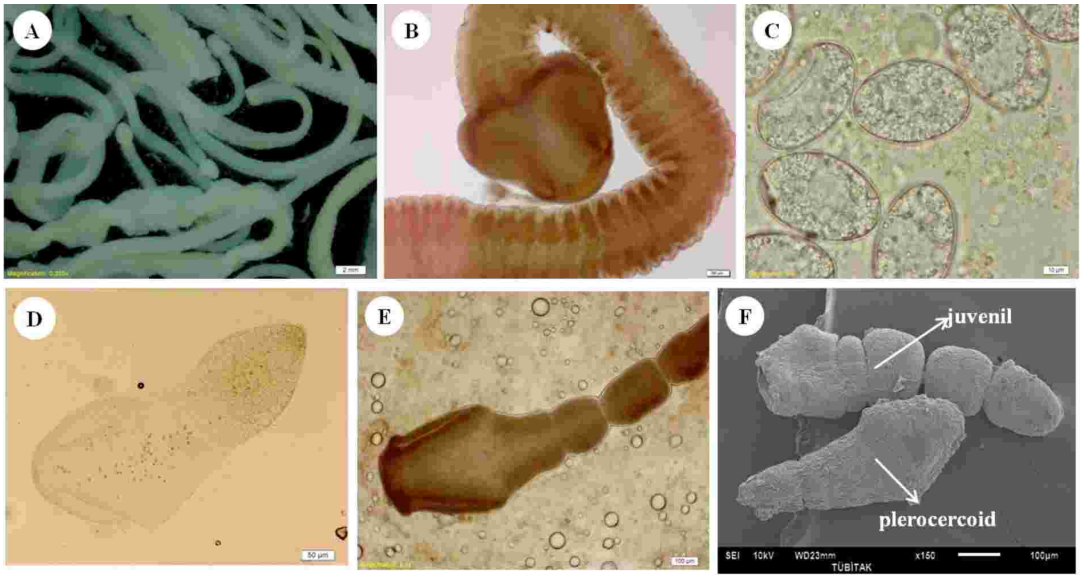

Figure 2. *Bothriocephalus acheilognathi* (original),
A. Adults specimens with eggs, **B.** Anterior portion of an adult specimen, **C.** Embryonated eggs, **D.** Plerocercoid, **E.** Juvenile, **F.** Plerocercoid and juvenile in SEM

interest due to its high pathogenicity for a wide range of fish species. Luo et al. (2002) have reported that some non-cyprinid fishes also harbour this cestode and the geographical distribution was increasing, implying that the parasite might have a high potential to colonise both new definitive hosts and new localities. So far this tapeworm has been reported from a number of species including cyprinids, poecilids, silurids, percides, centrarchids, gobiids, atherinid and cyprinodont fishes (Mitchell 1994, Luo et al. 2002, Öztürk et al. 2002, Velazquez-Velazquez et al. 2011, Çolak 2013a,b). Young of the year common carp, *C. carpio,* and adult western mosquito fish, *G. affinis,* have also been reported to be host for *B. acheilognathi* in the USA (Granath and Esch 1983). *B. acheilognathi* is one of the most extensively studied parasites of many freshwater fish species in different regions of Turkey (Table 2). As can be seen in Table 2, *B. acheilognathi* is widely dispersed among the freshwater fish of Turkey, being recorded in 15 fish species to date. The new host species recorded in this study bring the total number of known host species to 20 by addition of *Aphanius danfordii, Gambusia affinis, Carassius gibelio, Proterorhinus marmoratus* and *Sander lucioperca*. It must be noted that *C. carpio* had adult *B. acheilognathi* specimens while the rest of infected fish had developmental stages of juvenile and/or plerocercoid. In the present study, *C. carpio* had the highest infection prevalence and mean intensity values (26.72% and 16.66 ± 2.39) among all infected fish (Table 1). However, other infected fish species had very low infection indices. Our results are largely in agreement with other reported infection rates in different part of Turkey, except *Atherina boyeri,* which had higher values both infection prevalence and mean intensity (40.6%, 31.83 ± 57.74) (Çolak 2013b). Öztürk (2011) also reported a higher infection prevalence but a similar mean intensity value in *Alburnus escherichii* to those determined in the current study. The higher infection rates determined from *C. carpio* in the present study is not surprising considering that it is the most common host for this parasite (Table 2). Other fish species in the present study, however, had lower levels of infection rates, being in agreement with those reported from other fish species in other parts of Turkey. Brouder and Hoffnagle (1997) reported that four components for a successful invasion by *B. acheilognathi* are presence of intermediate and definitive hosts, suitable locality and water temperature. They also emphasised that the temperature is the most limiting factor for life cycle completion due to requirement for high temperature values above 25 °C. Thus we can speculate that developmental stages observed in all fish species with low level infection rates in the present study could be resulted from low water temperature values preventing further maturation and limiting its dispersal.

Table 2. Known distribution of *B. acheilognathi* in Turkey freshwater fishes

Fish Species	Prevalance (%)	Locality	References
Cyprinus carpio	10.0	Van	Topçu and Taşçı 1993
	12.4	İznik Lake, Bursa	Aydoğdu and Altunel 2002
	8.0	Karacaören Dam Lake Isparta-Burdur	Kır et al. 2004
	43.4	Eber Lake, Afyonkarahisar	Öztürk 2005
	-	Keban Dam Lake, Elazığ (*)	Dörücü and İspir 2005
	18.7	Sapanca Dam Lake, Sakarya	Uzunay and Soylu 2006
	13.7	Selevir Dam Lake, Afyonkarahisar	Öztürk and Bulut 2006
	14.1	Karamık Lake, Afyon	Kutlu and Öztürk 2006
	20.6	Kovada Lake, Isparta	Kır and Tekin-Özan 2007
	7.3	Beyşehir Lake, Konya	Tekin-Özan et al. 2008
	25.0	Kunduzlar Dam Lake, Eskişehir	Öztürk 2011
	18.2	Sığırcı Lake, Edirne	Çolak 2013a
Tinca tinca	1.9	Kovada Lake, Isparta	Kır and Tekin-Özan 2005
	3.5	Sapanca Lake, Sakarya	Akbeniz and Soylu 2006
	2.0	Terkos Lake İstanbul	Demirtaş 2011
Leuciscus cephalus	23.3	Örenler Dam Lake, Afyonkarahisar	Kurupınar and Öztürk 2006
	6.2	Kunduzlar Dam Lake, Eskişehir	Öztürk 2011
	2.5	Doğancı Dam Lake, Bursa	Aydoğdu et al. 2001
Atherina boyeri	40.6	İznik Lake, Bursa	Çolak 2013b
Alburnus alburnus	8.3	Mustafakemalpaşa Stream, Bursa	Aydoğdu and Selver 2006
Alburnus escherichii	50.0	Kunduzlar Dam Lake, Eskişehir	Öztürk 2011
Alburnus chalcoides	1.9	Tödürge Lake, Sivas	Yıldırım and Ünver 2012
Rutilus rutilus	0.8	Kocadere Stream, Bursa	Selver et al. 2009
Barbus plebejus escherichi	8.5	Doğancı Dam Lake, Bursa	Aydoğdu et al. 2002
Gobius fluviatilis	34.3	Uluabat Lake	Öztürk et al. 2002
Squalis cephalus	7.0	Serban Dam Lake, Afyonkarahisar	Açıkel and Öztürk 2012
Silurus glanis	-	Hirfanlı Dam Lake	Aydın 2003
Capoeta trutta	-	Keban Dam Lake, Elazığ (*)	Dörücü and İspir 2005
C. regium	-	Keban Dam Lake, Elazığ (*)	Dörücü and İspir 2005
C. Capoeta umbla	-	Keban Dam Lake, Elazığ (*)	Dörücü and İspir 2005

(*) Reported as Bothriocephalus gowkongensis.

CONCLUSION

The data presented new information on host range of *B. acheilognathi* with the addition of one endemic, *Aphanius danfordii,* two introduced fish species *Gambusia affinis* and *Carassius gibelio* and two native fish species *Proterorhinus marmoratus* and *Sander lucioperca* in the Lower Kızılırmak Delta in Turkey. This work clearly indicates a potential threat of this tapeworm for our endemic, native and introduced fish species. *B. acheilognathi* is known to be highly pathogenic to its hosts, causes serious damage and even death to fry and small fishes in case of high infections, suggesting that care should be taken into consideration when introducing a fish species to natural environments. This is particularly important in an introduction of a non-native fish before pre-stocking health control. This will contribute to prevent the introduction of parasites like *B. acheilognathi* as well as other harmful fish pathogens.

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