External Parasitic Worms and Protozoans of the Fish Caught in Shapour River and Seyyed Hossein Spring in Kazerun, Iran

Alamdari, A.[1] ;Golchin Manshadi, A.R.[2*] ;Tarahomi , M. [3]

[1]Gratuated from Faculty of Veterinary Medicine, Kazerun Branch, Islamic Azad university, Kazerun, Iran;[2,3]Department of Aquatic Animal Health, Faculty of Veterinary Medicine, Kazerun Branch, Islamic Azad university, Kazerun, Iran.

Coresponding author: A.R. Golchin Manshdi, Department of Aquatic Animal Health, Faculty of Veterinary Medicine, Kazerun Branch, Islamic Azad University, Kazerun, Iran, *Email:golchinalireza@yahoo.com*

(Received: March 22, 2017; Accepted: June 12, 2017)

Abstract

In this study, 38 fish in four species of Shapour River and Seyyed Hossein Spring were caught in 2016, including: *Albumus mossulensis*, *Cyprinion macrostomum*, *Capoeta barroisi persica* and *Garra rufa* and they were studied for external worm parasites and protozoans. Totally, 10 species of parasites were isolated and recognized, including: five species of monogenes (*Dactylogyrus alatus, Dactylogyrus carassobarbi, Dactylogyrus pulcher, Dactylogyrus holciki* and *Gyrodactylus* sp.),one species of digenes (*Centrocestus* sp.),one species of crustacean (*Lamproglena* sp.),two species of protozoan (*Ichthyophthrius multifilliis*, *Chilodonella sp.*) and one species of Myxozoa (*Myxobolus sp.*). The results showed that the highest percent of parasitic infection was for *Dactylogyrus carassobarbi* and the least amount was reported for *Lamproglena* sp. and *Myxobolus* sp. Moreover, *Capoeta barroisi persica* and *Albumus mossulensis* had the highest and lowest percent of parasitic infection, respectively. There wasn't any meaningful relationship between the infection of different species of the fish and their infected organs (P>0/05).

Keywords: *Protozoa, Metazoa, Seyyed Hossein Spring, Shapour River, Fish*

Introduction

Parasites in fish have been a great concern since they often produce disease conditions in fish which will lead to reduced growth, increase in the fishes' susceptibility to other diseases as well as fish loss. Parasites may be the cause of growth reduction, fatality, delayed puberty or infertility in the fish and they are often the reason for microbial, viral and fungal diseases. In some cases, parasites are the cause of high fatality of the fish (Abbasi, 2003).Studies on parasites of Iranian fish species date back to 1949, when Bychowsky reported three Dactylogyrus species and one Ancyrocephalus on the gills of fishes in Karkheh River. Since then other researchers reported more parasites from Iranian freshwater fish. Various studies have been conducted with respect to the study of parasite infection of freshwater fish in Mesopotamian and the other areas in Iran. The freshwater fish in western and southern areas belongs to Mesopotamian area which is located in Iraq, Turkey and Syria (Jalali, 1998). The most important studies in this area are as: Mortezaei et al, (2000) on parasite infection of freshwater fish in Hoorolazim and

Shadegan Suns, Farahnak et al, (2002) in Khuzestan Basins, Abbasi et al, (2003) and Barzegar et al, (2004) studying parasites of the Behesht Abad River, and Raissy et al, (2009) in identifying the parasites of 3 kinds of *Barbus capito* in Kiar and Behesht Abad Rivers in Chahar Mahal and Bakhtiari Basins. The importance of the identification of the parasites is that, on one hand, a large group of animals are studied and different kinds of parasites as infecting a host could be recognized and on the other hand, they can infect the farmed fish, as well.

Shapour River originates from Ranjan Spring and connects Sasan Spring in Tange Chowgan area and flows toward Bushehr. In Bushehr, this river touches Dalaki River and then finally joins Persian Gulf called Hilla River. Shapour River is very vital in Kazerun, providing this area's water which is consumed for agriculture. Seyyed Hossein Spring is located twenty-three kilometers away in Kazerun Road. The fish species in this river have nutritional value and are caught recreationally or for eating, thus recognizing their parasite gamut is very essential and this study has been carried out accordingly. This study, therefore, aimed at detecting the occurrence of parasites in fish in Shapour River and Seyyed Hossein Spring, Kazerun Province, with regard to the importance of native fish population in the river.

Material and Methods

Early morning, the fish were caught using gillnet and Sachuk Net and they were transferred alive to the Laboratory of Parasitology, Faculty of Veterinary Medicine, Islamic Azad University, Kazerun and were kept in the aquarium. The fish were anaesthetized using *Eugenia* sp. solution of 170 ppm (Rastiannasab et al, 2014). Immediately after anesthesia, they were identified based on recognition keys of Berg (1964), and Coad (1992). Their eyes, skin and gills were then examined for parasitological information. Using an optical microscope, the sample of their gills and skin was studied in order to separate metazoan parasites. The parasites were picked by Pastor Pipette and through using Ammonium Picrate, they were fixed on a slide following Fernando et al, (1972) and Gussev (1983) instructions. Identification of the kinds of isolated monogenes was based on recognition key by Gussev (1987). After gills' sampling, using Fernando et al, (1972) instruction in order to identify the protozoans, the sample was fixed and they were recognized using Lom and Dykova (1992) specification clue. Stabilizing and clearing parasite samples were performed using Fernando et al, (1972) instructions. In this study, the data were processed using SPSS 18 software. The frequency and one-way ANOVA were used to compare the infection among different kinds of fish and the organs.

Results

Totally, 10 external parasites were isolated from the skin and gill of 4 fish species caught in the river. They were identified for their sex and species, including: five Monogenes (*Dactylogyrus holciki*, *Dactylogyrus pulcher*, *Dactylogyrus carassobarbi*, *Dactylogyrus alatus*, frequency 65.7 %), (*Gyrodactylus* sp., frequency 31.5 %). one Digene (*Centrocestus* sp., frequency 31.5 %), one crustacean (*Lamproglena* sp., frequency10.5 %), 2 protozoans (*Ichthyophthirius multifilliis* and *chilodonella* sp., frequency 21% and 13.1%, respectively) and one Myxozoa (*Myxobolus* sp., frequency 10.5 %). Therefore as the results showed, *Dactylogyrus carassobarbi* demonstrated the highest and *Lamproglena* sp. and *Myxobolus* sp. presented the lowest frequencies. The highest infection was detected in *Capoeta barroisi persica* with 100 % frequency and the least infection was observed in *Albumus*

[DOR: 20.1001.1.2717378.2017.1.1.3.9]

mossulensis with 60 percent frequency. The average intensity of infection in *Capoeta barroisi persica* with 2.74 (±2.43),*Cyprinion macrostomum* with 2.9 (±4), *Albumus mossulensis* with 0.72 (±1.24), and *Garra rufa* with 0.48 (±0.48) was calculated. Using one-way ANOVA, the infection among different kinds of the fish and the infected organs was evaluated. As the findings showed, no meaningful relationship was detected (P>0/05).

Table 1 represents data related to the isolated parasites, their isolated organs and hosts. Diagram 1 and 2 represent the amount of infection identified in fish under study, and also the frequency percentage of the isolated parasites. Diagram 3 represents the infection average.

Table 1. Isolated external metazoan and protozoans of the fish in Shapour River and Seyyed Hossein Spring based on the infected organ and its host

Parasite	Infected Organ	Fish Species
Ichthyophthirius multifiliis	Gill	*Capoeta barroisi persica*
Dactylogyrus pulcher		
Centrocestus sp.		
Myxobolus sp.		
Ichthyophthirius multifiliis	Skin	
Dactylogyrus carassobarbi		
Gyrodactylus sp.		
Chilodonella sp.		
Ichthyophthirius multifiliis	Gill	*Albumus mossulensis*
Dactylogyrus alatus		
Dactylogyrus holciki		
Chilodonella sp.		
Myxobolus sp.		
Ichthyophthirius multifiliis	Skin	
Dactylogyrus carassobarbi		
Gyrodactylus sp.		
Chilodonella sp.		
Centrocestus sp.	Gill	*Cyprinion macrostomum*
Ichthyophthirius multifiliis		
Chilodonella sp.		
Myxobolus sp.		
Lamproglena sp.		
Ichthyophthirius multifiliis	Skin	
Dactylogyrus carassobarbi		
Gyrodactylus sp.		
Chilodonella sp.		
Dactylogyrus alatus	Gill	*Garra rufa*
Gyrodactylus sp.	Skin	

[DOR: 20.1001.1.2717378.2017.1.1.3.9]

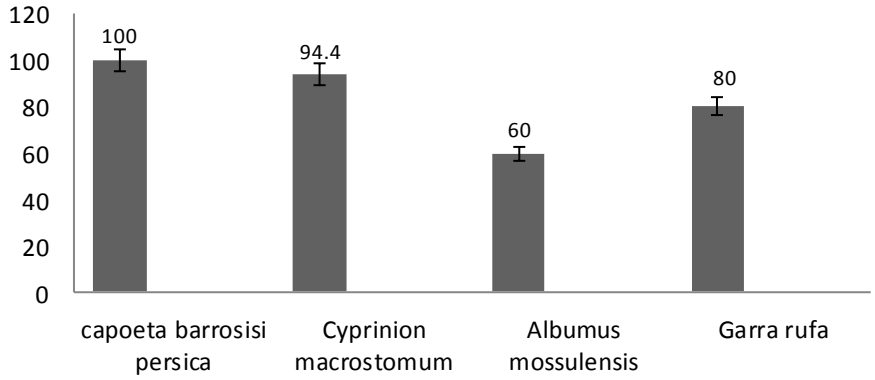


Diagram 1. Infection Percent of the Fish Caught in Shapour River and Seyyed Hossein Spring

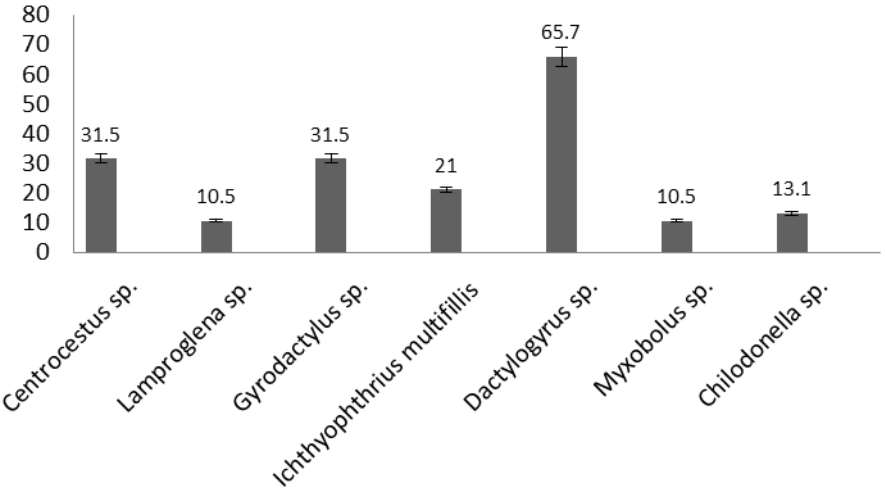


Diagram 2. Infection Frequency Percent of the isolated Parasites of the Fish in Shapour River and Seyyed Hossein Spring

[DOR: 20.1001.1.2717378.2017.1.1.3.9]

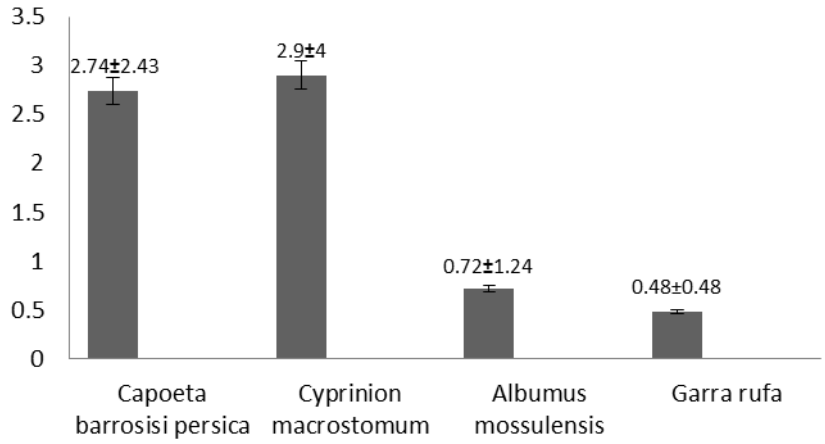


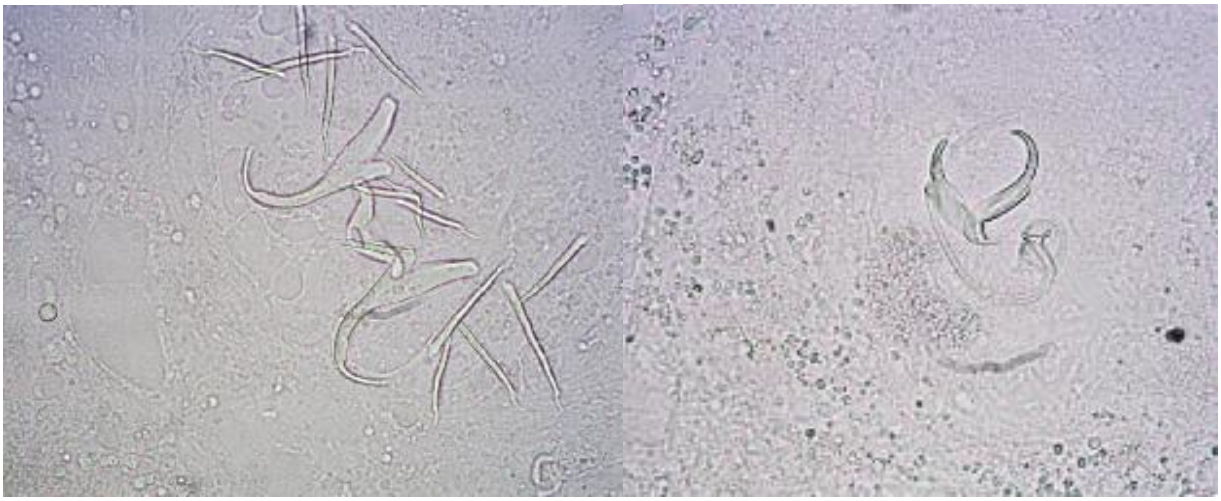
Diagram 3. The Average Intensity of the Infected Fish in Four Studied Groups

Figure 1. *Gyrodactylus* sp. (×3000)

[DOR: 20.1001.1.2717378.2017.1.1.3.9]

A:Main Hook and Mini Hooks B: Sexual Organ

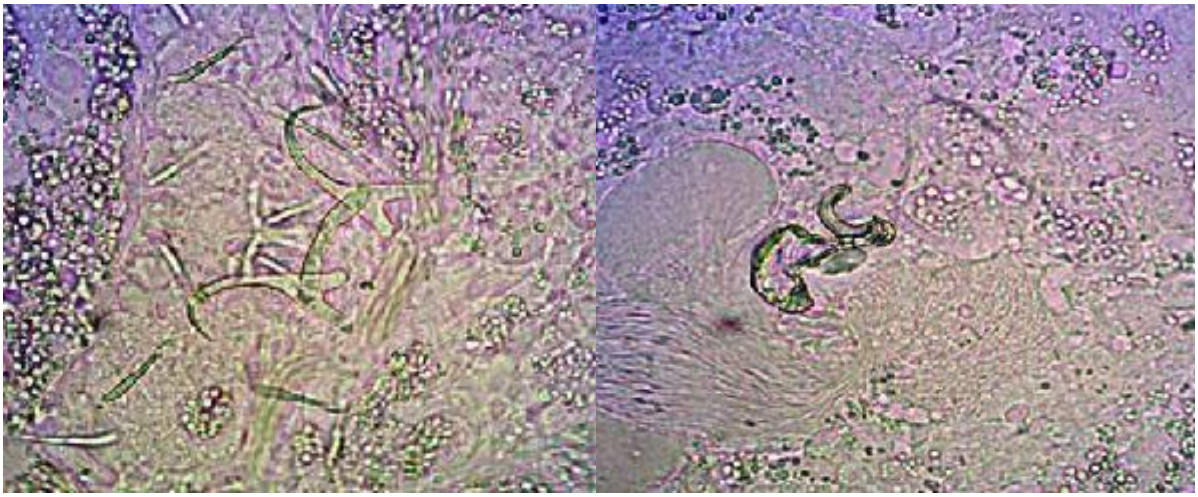
Figure 2. *Dactylogyrus* sp. (×7500)



A: Main Hook and Mini Hooks B: Sexual Organ

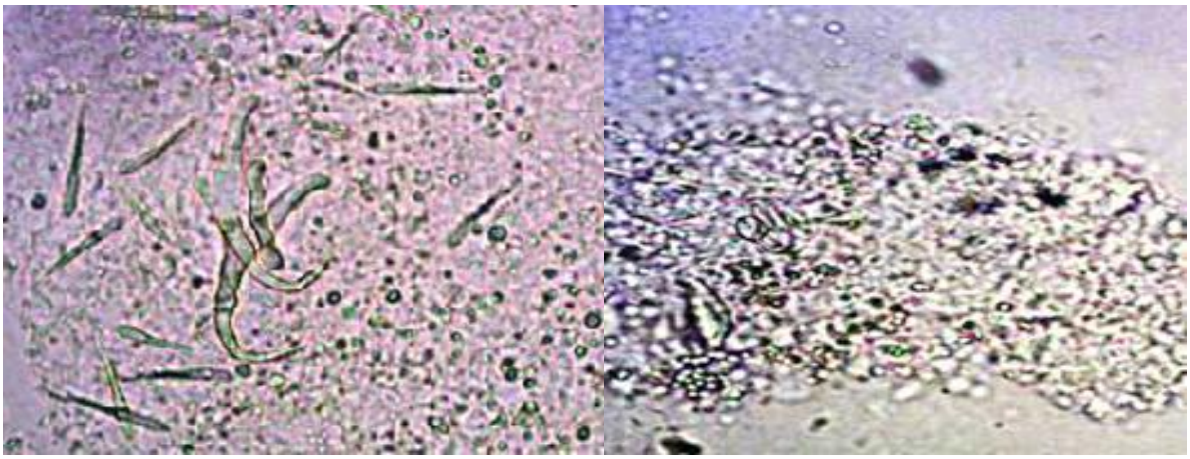
Figure 3. *Dactylogyrus carassobarbi*(×3000)

[DOR: 20.1001.1.2717378.2017.1.1.3.9]



A: Main Hook and Mini Hooks B: Sexual Organ

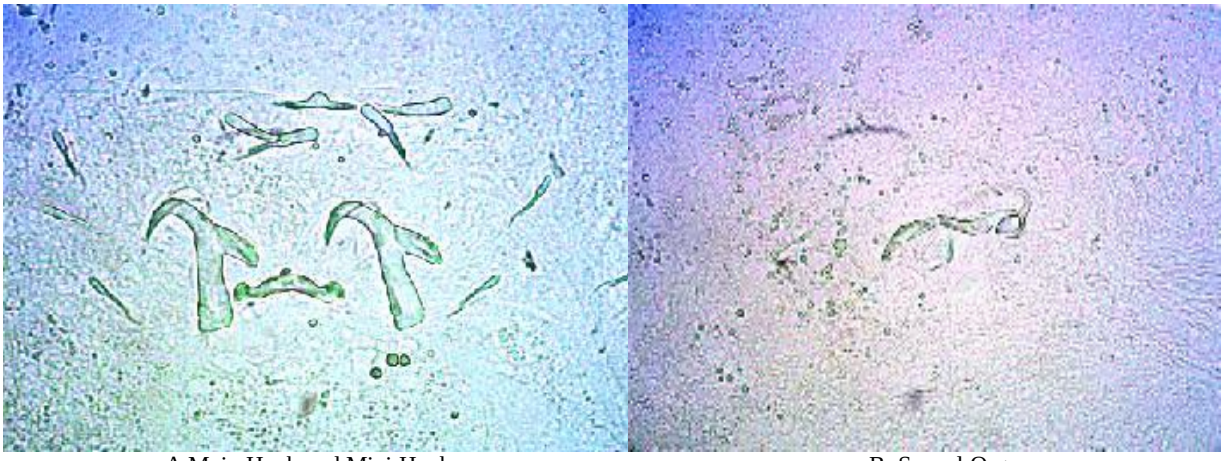
Figure 4. *Dactylogyrus holciki*(×3000)



A: Main Hook and Mini Hooks B**:** Sexual Organ

Figure 5.*Dactylogyrus pulcher* (×3000)

[DOR: 20.1001.1.2717378.2017.1.1.3.9]



A:Main Hook and Mini Hooks B: Sexual Organ

Figure 6. *Dactylogyrus alatus* (×3000)

Figure 7. *Myxobolus* sp. (×3000)

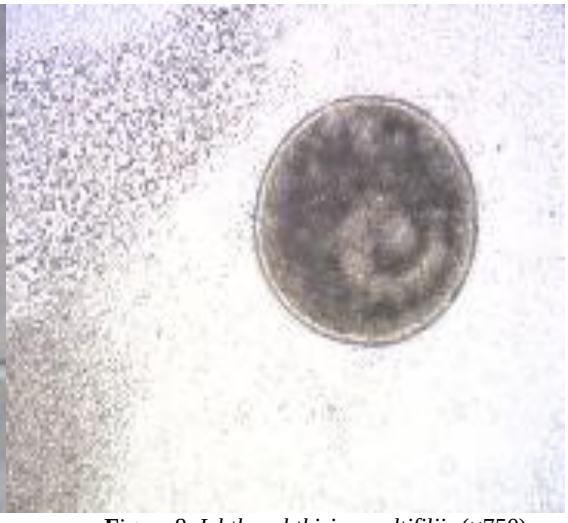


Figure 8**.** *Ichthyophthirius multifiliis* (×750)

[DOR: 20.1001.1.2717378.2017.1.1.3.9]

Figure 9. Metacercaria of the *Centrocestus* sp. (×3000)

Figure 10. Cystic Stage of the *Centrocestus* sp. (×3000)

Figure 11. *Lamproglena* sp. (×40)

Discussion

Throughout the time, a balanced condition would be developed among parasites and the host in natural environments, but in high fish density in cultural and environmental stressful conditions, parasites would make many problems for the reproduction (Bush et al, 2001) and despite parasitic infection of the fish, no clinical signs of the disease were observed which confirm the balance of parasite and the host in natural environments. Among the reported parasites in this study, a Ciliated Protozoan (*Ichthyphthirius multiphilis*) is found to be very important. This parasite lacks the host specificity and infects different species of the fish and kills them or decreases their growth.

[DOR: 20.1001.1.2717378.2017.1.1.3.9]

This parasite spreads globally and all freshwater fish are sensitive to it as existed in many wild and cultured fish all over the country (Jalali, 1998). Mokhayer (2003) reported this parasite in *Barbus capito, Capoeta damascina*, and *Cyprinus carpio* in Sefidrood River. Moreover, Masumian et al, (2003) identified it in *Capoeta damascina* in GhezelDagh. In the present study, this parasite was isolated from the gill and skin of the 3 fish species (*capoeta barroisi persica, Cyprinion macrostomum*, and *Albumus mossulensis*) with 21 percent of infection. As the findings revealed, this parasite lacked host specialization. *Chilodonella* sp. infects approximately all freshwater fish, especially fingerlings. It seems that they have dispersed by the infected fish all over the world. Chilodonellosis is more dangerous than *I. multifiliis* since a serious damage would be developed before the emergence of symptoms. In this study, *Chilodonella* sp. was isolated from *capoeta barroisi persica* and *Cyprinion macrostomum* with a frequency of 13.1 percent. Myxozoa are a large group of parasites and *Myxobolus* sp. is very important among these fish. This parasite produces cysts in different tissues and while considering the tissue and the infected organ, it makes various infectious effects (Woo, 2006). Many studies have shown the existence of this parasite in Iran. Barzegar et al. (2004) reported the existence of this parasite inZayande Rood River. In a study by Masoumian and Pazooki (1998), *M. musayevi* was detected and reported in *Capoeta capoeta* fish in Tajan River. Masumian et al. (2003) also reported *M. kovali* in *Capoetada mascina, M. squamae* in *Barbus capito,* and *M. rutili* in *Rutilus firisii kutum.* In this study, *Myxobolus* sp. was isolated from *Cyprinion macrostomum, capoeta barroisi persica*, and *Albumus mossulensis.*

Five Monogenean parasites, as some other dangerous species, were detected in this study. They are as *Dactylogyrus alatus, D.carassobarbi, D.holciki, D.pulcher* and *Gyrodactylus* sp. Different kinds of this species have been reported in the gills of freshwater fish. Jalali and Molnar (1990) identified *D.chramulii, D.graciliis, D.lenkorani*, and *D.pulcher* in *Capoeta damascina* in Sefidrood. Moreover, Mokhayer (2003) reported *D.vastator* in *Capoeta damascina* in Sefidrood. In this study, *Centrocestus* sp. was isolated from *Cyprinion macrostomum* and *capoeta barroisi persica*. The fish can be infected as the second host and this cycle is completed at the time that the fish are eaten by the final host including rats, penguins, birds and mammals and they become mature.

Some human infections have also been reported by this parasite. For the first time, this parasite was detected in Asia (Chen, 1942) and later, it was reported in Mexico, Australia, Croatia and Turkey (Evans and Yildiz, 2005; Scholz and Salgado-Maldonado, 2000; Gjurcevic et al., 2007; Lester, 2001). Mood et al. (2010) identified this parasite in some imported aquarium fish. However, there has not been any report of its existence in inland waters. *Lamprolegna* sp. was another isolated parasites of gills of *Cyprinion macrostomum* in this study. Raissy et al. (2009) reported *Lamproglena chinensis* in the gill of *Capoetada mascina* in Kiar River.

Conclusion

Between fish, all *C. barroisi persica* were infected by the parasites and the genus of Dactylogyrus has the highest percentage of isolation. High prevalence of Dactylogyrus may affect native fish population. *Centrocestus* sp. collected from *C.barroisi persica* and *C. macrostomum* are reported for the first time in Iran. *C. barroisi persica* and *C. macrostomum* are also reported as a new host for *Centrocestus* sp.

[DOR: 20.1001.1.2717378.2017.1.1.3.9]

References

Abbasi K, Moradkhah S and Rahimi R. Identification and study of Black River fish distribution of Siah Darvishan Anzali Wetland, Bony fish research center of Khazar Sea. Research and development Magazine, 2003; 19(1):27-39.

Barzegar M, Asadollah S, HemmatZadeh A, JalaliJafari B and Rahnama R. Fish parasites of Behesht Abad River (Chahar Mahal and Bakhtiari Province). Journal of Comparative Pathology, 2004; 1(1): 67-74.

Berg LS. Freshwater Fishes of USSR and adjacent countries, Vol,3 (English version).Nauka, Mos, 1964; pp: 926-1382.

Bush OB, Ferandez JE, Esch GW and Seed JR. Parasitism, the diversity and ecology of animal parasites.Cambrigde university press, 2001; p: 652.

Bychowsky BE. Monogenetic trematodes of some fish of Iran collected by E.N. Pavlowsky.TrudiZoologicheskovoInstitutaAkademiya, 1949; 8(4): 870-878.

Chen, HT. The metacercaria and adult of Centrocestus formosanus (Nishigori, 1924) with notes on the natural infection of rats and cats with C. armatus (Tanabe, 1922). Journal of Parasitology, 1942; 28(2):285-98.

Coad BW. Freshwater Fishes of Iran.A checklist and bibliography Ichthyology Section. Canadian museum of Nature, Ottawa, Ontario, Canada, 1992; pp: 66-90.

Evans BB and Lester RJG. Parasites of ornamental fish imported into Australia. Bulletin of the European Association of Fish Pathologists, 2001; 21(2):51-55.

Farahnak A,.Mobedi I and Tabibi R. Fish anisakidae helminthes in Khuzestan province, South West of Iran. Iranian Journal of Public Health, 2002; 31(2): 129-132.

Fernando CH, Furtado JI, Gussev AV Kakong SA and Hanek AV. Methods for the study of fresh water fish parasites. University of waterloo, Biology series, 1972; p: 76.

Gjurcevic E, Petrinec Z, Kozaric Z, Kuzir S, GjurcevicKantura V, Vucemilo M and Dzaja P. Metacercariae of Centrocestus formosanus in goldfish (Carassius auratus L.) imported into Croatia. Helminthologia, 2007; 44(4): 214-216.

Golchin Monshadi A, Jalali B, Barzagar M, Mostafavi E and Raissy M. Studying Monogenetic native fish and introduced from Parishan Lake, Iranian Journal of Pathbioology, 2010; 7(1): 189-194.

Golchin Manshadi AR, Masoumian M, Jalali B and Barzegar M. Protozoan and Myxozoan infections in ome Fishes of Parishan Lake, Asian Journal of Animal and Veterinary Advances, 2012; 10(4):1-9.

[DOR: 20.1001.1.2717378.2017.1.1.3.9]

Gussev AV. Key to parasites of freshwater fishes of the soviet USSR.Izd.Nauka, Leningrad, USSR, 1987; pp: 379-523.

Gussev AV. The methods of collection and processing of fish parasitic Monogenean materials (In Russian). Nauka, Leningrad, 1983; pp: 5-48.

Jalali B and Monlar K. Occurance of Monogeneans of freshwater fishes of Iran, Dactylogyrus spp.On cultured Iranian fishes.Acta Veterinary Hungaria, 1990; 38(5):339-342.

Jalali B. Freshwater fish parasites and parasitic diseases in Iran, 1st Edition, Deputy Proliferation and Aquaculture Fisheries Organization, Tehran, Iran, 1998; pp: 229-301.

Lom J and Dykova I. Protozoan parasites of fishes (developments in aquaculture and fisheries science), Elsevier Science, Amesterdam, 1992; P: 316 .

Masoumian M and Pazooki J. Infection of some fish speies to Myxospora in Gilan and MazandaranProvines. Iranian Journal of fisheries science, 1998; 7(3): 56-67.

Masumian M, Pazuki J and Ghasemi R. The infection of 3 species of Barbus fish in the Caspian Sea with Myxobolus parasites. Journal of Veterinary Medicine, Tehran University, 2003; 5(4): 329-334.

Mokhayer B. Studying fish parasites in Sefidrood River.a letter from Faculty of Veterinary Medicine, 2003; 36(4): 61-75.

Molnar K. Report of the fish pathology course held in Iran. From 24 Nov to 21 Dec. Fisheries Company of Iran., 1990; P:11.

Mood SM, Ebrahimzadeh Mousavi HA, Mokhayer B, Ahmadi M, Soltani M and Sharifpour I. Centrocestus formosanus metacercarial infection of four ornamental fish species imported into Iran. Bulletin of the European Association of Fish Pathologists, 2010; 30(4):146-149.

Raissy M, Ansari M, Jalali B. Identification of parasites of black fish in Kiar and Behesht Abad Rivers, Chahar Mahal Bakhtiari province, and the 1st report of Lamproglena chinensis in Iran. Veterinary Pathobiology, 2009; 1(1): 18-22.

Rastiannasab A, Hosseini SA, Sharif Rohani M, Gandomkar H, Nekoyiefard A and Gorjipour E. The control of recovery process with clove tree (Eugenia caryophyllata) in rainbow trout (Oncorhynchusmykiss). Iranian Journal of Fisheries Sciences, 2014; 23(1): 117-123.

Scholz T and Salgado-Maldonado G. The introduction and dispersal of Centrocestus formosanus (Nishigori, 1924) (Digenea: Heterophyidae) in Mexico: A review. American Midland Naturalist, 2000; 143: 185-200.

[DOR: 20.1001.1.2717378.2017.1.1.3.9]

Woo PTK. Fish diseases and disorders, protozoan and metazoan parasites. CAB international, UK,2006; pp: 32-245.

Yildiz HA. Infection with metacercariae of Centrocestus formosanus (Trematoda:Heterophyidae) in ornamental fish imported into Turkey. Bulletin of the European Association of Fish Pathologists, 2005; 25(5):244-246.

[DOR: 20.1001.1.2717378.2017.1.1.3.9]