Aphanius sophiae (Actinopterygii, Cyprinodontidae), a new host for *Ichthyophthirius multifiliis* (Ciliophora) reported from Iran

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Abstract *Aphanius sophiae* (Heckel, 1849) or soffiia tooth-carp is endemic species to the endorheic Kor River basin, Fars Province, Southwest of Iran. In spite of importance of this species as a biologic control of anophelines larvae, little attention has been paid to its diseases particularly parasitic infections. A total of 24 *A. Sophiae* (12 males and 12 females) in Safashahr spring, Kor River basin, were collected by using dip net in February 2012. Surface of all fish were gross examined and screened for any swelling and blisters. All of the examined specimens (100 %) were infected with *Ichthyophthirius multifiliis*. Infected fish typically developed small blister-like raised lesions on the body surface. The current study presents infection of *A. sophiae* with *I. multifiliis* for the first time from Iran. Hence, owing to considerable unexplored area

of Iran further studies are required to increase our knowledge about parasitic infections of this fish.

Keywords Fish · Aquatic · *Aphanius sophiae* · *Ichthyophthirius multifiliis* · Protozoan parasite · Endemic · Iran

Introduction

Species of the killifish genus *Aphanius* Nardo, 1827 (Teleostei, Cyprinodontiformes) are widely distributed throughout the Mediterranean region and in the Middle and Near East in brackish coastal habitats and freshwater to euryhaline inland water bodies (Ferrito et al. 2013). Iran is considered as a center for *Aphanius* diversity, with 14 species recognized and their distribution area include both freshwater and brackish water bodies (Teimori et al. 2012; Gholami et al. 2014a; Esmaeili et al. 2014). Among 14 described *Aphanius* species in Iran, *A. sophiae* (Heckel, 1849) or soffiia tooth-carp is endemic species restricted to the endorheic Kor River basin (Teimori et al. 2012; Gholami et al. 2014b).

The importance of *A. sophiae* is not only owing to being an indigenous fish species but also this species plays a significant role in biologic control of *anopheles* mosquito larvae from streams (Steven 1913).

Ichthyophthirius multifiliis is a ciliate Protozoa which is considered as a significant freshwater teleost pathogen that often causes major economic losses and damages to the aquaculture industry. The disease ichthyophthiriasis, commonly known as white spot disease is the major parasitic hazard to freshwater fish throughout the world (Buchmann et al. 2001). Severe infection of fish by ciliate *Ichthyophthirius* has considerable impact on growth, behavior and

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fish resistance to other stressing factors, susceptibility to predation, high mortality due to epithelial hyperplasia and leukocyte mobilization, predominantly neutrophils and inflammation, as well reduced market ability of trade fish and concomitant economic loss (Swennes et al. 2007).

Despite of all above mentioned facts regarding the importance of this species insufficient data are available on parasitic disease of this valuable fish in Iran. Therefore, the objective of this study was to determine the prevalence of *I. multifiliis* in *A. sophiae* in Kor River basin, Safashahr town, Fars Province, Southwest of Iran.

Materials and methods

This study was done in Safashahr Spring (30°34′50.78″N, 53°31′51.73″E, 2,337 m Altitude) from Kor River basin, Safashahr town, Fars Province, Southwestern Iran in February 2012. The bottom of this shallow spring is generally muddy with rubble and gravel and *A. sophiae* is the only fish fauna inhabiting in the spring. A total of 24 (12 males and 12 females) fish specimens were collected using dip net and preserved in 96 % ethanol. Standard length of the specimens was measured to the nearest 0.05 mm using vernier caliper and their sex was determined considering sex dimorphism characteristics. Entire body of fish was screened both macroscopically and microscopically for the presence of any swelling and blister. In order to detect *Ichthyophthirius* Protozoa, the body mucus (blisters) was scraped gently from skin transferring onto a micro slide and afterwards spread the mucus carefully with a cover slide. *Ichthyophthirius* exposed in Azo-carmin for three days and then washed for three minutes in ethanol containing a drop of added Iodine solution. Next, the smears were mounted between a glass slide and a cover slip in Canada balsam after dehydration (Fernando et al. 1972). Eventually, the parasites were studied using light microscope, and identification of parasites was performed according to all crucial morphological characters of the collected parasites.

Results

Overall, all the examined specimens of 24 (100 %) fishes (12 males and 12 females) had blister-like lesions namely white spot (Fig. 1a–b). The standard length of fishes ranged from 21.2–29.6 mm and 22.2–33.7 mm for males and females, respectively.

Plenty of blisters and lesions were observed on skin and fins of examined fishes. Gill examination revealed numbers of white spots and gills were pale and very swollen. Also fish eyes were seen cloudy. The causative agent of blisters

Fig. 1 Photographs of *Aphanius sophiae* collected from Safashahr Spring, Kor River basin, Fars Province, Southwestern Iran, **a** infected male, **b** infected female having blisters containing *Ichthyophthirius multifiliis*, known as white spot disease or ichthyophthiriasis

was recognized as *I. multifiliis*. Additionally, some dead fish were observed on the surface of spring water due to high rate of infection (field observation).

Discussion

According to the obtained results, the prevalence rate of *I. multifiliis* in *A. sophiae* was high (100 %). Different prevalence rates in various fish species has been reported (Kuperman 2002; Masoumian et al. 2005; Jalali et al. 2008; Raissy et al. 2010). Some acceptable justifications for this phenomenon might be related to the innate fish resistance, different environmental and nutritional factors. High infection rate in our specimens may be due to that there is a positive phototaxis in *Ichthyophthirius*, indicating they prefer to infect pelagic hosts such as *A. sophiae* (Wahli et al. 1991).

Additionally, nutritional conditions can be associated to both host and parasite. Because when there are not enough foods in the habitats, fish will become weak with low immunity system and these stressful conditions force fish prepared to accept parasites easily. On the other hand, Nigrelli et al. (1976) displayed that the young free-living ciliates must find a proper host during their infective period, otherwise they starve to death (Nigrelli et al. 1976).

Moreover, the study area (Kor River Basin) is small and *A. sophiae* species is the only fish fauna dwelling in this spring. Therefore, the opportunity of parasite for attack will be more favorable, and many fish individuals will be infected by this parasite. Besides, high stock densities are considered as a common cause of white spot problems in fisheries. Indeed this makes easier for parasites to find a fish to infect and can help them to spread rapidly.

Likewise, favorable conditions for parasite development include high stocking density of fish, unhealthy conditions of fish, host nutritional deficiencies, low values of water conductivity and organic dry matter (Aihua and Buchmann 2001; Garcia et al. 2009).

As previously described, *I. multifiliis* from *Aphanius dispar* were also observed in Mehran River, Hormozghan Province, Iran (Gholami et al. 2014a). Furthermore, the infection of *A. sophiae* with *Tetracotyle* sp. has been reported (Gholami et al. 2011). Malek and Mobedi (2001) indicated infection of *A. sophiae* with *Clinostomum complanatum* (Malek and Mobedi 2001).

In summary, the results indicated exposure of all examined specimens to *I. multifiliis* and very high prevalence of this devastating protozoal infection. Hence, owing to considerable unexplored area of Iran further studies are required to increase our knowledge about this parasitic infection. Considering that *A. sophiae* is indigenous species and are used as a biologic control of *anopheles* mosquito larvae and Iran is an endemic area for malaria, protection and conservation of this species against this ectoparasite is highly recommended. Also preventive measures including regular monitoring of water quality and managing stock density should be taken into consideration owing to the significance of the disease. Present study reported *A. sophiae* as a new host for *I. multifiliis* in Iran.

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Conflict of interest We declare that we have no conflict of interest.

References

- Aihua L, Buchmann K (2001) Temperature and salinity dependent development of a Nordic strain of *Ichthyophthirius multifiliis* from rainbow trout. *J App Ichthyol* 17:273–276
- Buchmann K, Sigh J, Nielsen CV, Dalgaard M (2001) Host responses against the fish parasitizing ciliate *Ichthyophthirius multifiliis*. *Vet Parasitol* 100:105–116
- Esmaili HR, Teimori A, Gholami Z, Reichenbacher B (2014) Two new species of the tooth-carps *Aphanius* (Teleostei: cyprinodontidae) and the evolutionary history of the Iranian inland and inland-related *Aphanius* species. *Zootaxa* 37:246–268
- Fernando CH, Furtado JJ, Gussev AV, Hanek G, Kakong SA (1972) Methods for the study of freshwater fish parasites. 1st Edn. University of Waterloo, Biology Series
- Ferrito V, Pappalardo AM, Canapa A, Barucca M, Doadrio I, Olmo E, Tigano C (2013) Mitochondrial phylogeography of the killifish *Aphanius fasciatus* (Teleostei, Cyprinodontidae) reveals highly divergent mediterranean populations. *Mar Biol* 160:3193–3208
- Garcia F, Fujimoto RY, Martins ML, Mand Moraes FR (2009) Protozoan parasites of *Xiphophorus* spp. (Poeciliidae) and their relation with water characteristics. *Arq Brasil Med Vet Zootec* 61:156–162
- Gholami Z, Akhlaghi M, Mobedi I, Esmaili HR (2011) First report of *Aphanius sophiae* (Heckel, 1849) (Actinopterygii: cyprinodontidae) with *Tetracotyle* sp. in Cheshmeh-Ali Damghan, Semnan, Province North of Iran. *Vet Res Forum* 2:605–608
- Gholami Z, Esmaili HR, Reichenbacher B (2014a) Phylogenetic analysis of *Aphanius* from the endorheic Kor River Basin in the Zagros mountains, South-western Iran (Teleostei: cyprinodontiformes: Cyprinodontidae). *J Zoo Sys Evol Res* 52:130–141
- Gholami Z, Rahimi MT, Zarei R, Kia EB, Mobedi I, Vatandoost S (2014b) First report of *Ichthyophthirius multifiliis* (Ciliophora: oligohymenophorea) from *Aphanius dispar* (Cyprinodontidae) in Iran. *J Coastal Life Med* 2:490–495
- Jalali B, Barzegar M, Nezamabadi H (2008) Parasitic fauna of the spiny eel, *Mastacembelus mastacembelus* banks et Solander (Teleostei: mastacembelidae) in Iran. *Iran J Vet Res* 9(2):23–25
- Kuperman BI (2002) Matey VE, Warburton ML, Fisher RN. Introduced parasites of freshwater fish in southern California. The tenth international congress of parasitology. Vancouver, Canada, August
- Malek M, Mobedi I (2001) Occurrence of clinostomum complanatum (Rudolphi, 1819) (Digenea:clinostomatidae) in capoeta capoeta gracilis (Osteichthys: Cyprinidae) from Shiroud River Iran. *Iranian J Publ Health* 30:95–98
- Masoumian M, Pazooki J, Yahyazadeh M, Teymornezhad A (2005) Protozoan (*sic*) from fresh water fishes of northwest Iran. *Iran J Fish Sci* 4:31–42
- Nigrelli RF, Pokorny KS, Ruggieri GD (1976) Notes on *Ichthyophthirius multifiliis*, a ciliate parasitic on freshwater fishes, with some remarks on possible physiological races and species. *Trans Am Micro Soc* 95:607–613
- Raissy M, Fadaeifard F, Ansari M, Tajizadegan H, Hosseini R (2010) Parasites of fish in Sooleghan Lagoon. *Vet. J. Islam. Azad Uni. Garmsar Branch* 5:143–148
- Steven WSR (1913) Report on an investigation in regard to the prevalence of malaria amongst the troops stationed at Karachi. *J R Arm Med Corps* 24:251–261
- Swennes G, Findly R, Dickerson W (2007) Cross-immunity and antibody responses to different immobilization serotypes of *Ichthyophthirius multifiliis*. *Fish Shellfish Immunol* 22:589–597
- Teimori A, Esmaili HR, Gholami Z, Zarei N, Reichenbacher B (2012) *Aphanius arakensis*, a new species of tooth-carp (Actinopterygii, Cyprinodontidae) from the endorheic Namak Lake basin in Iran. *Zookeys* 215:55–76
- Wahli T, Meier W, Schmitt M (1991) Affinity of *Ichthyophthirius multifiliis* theronts to light and/or fish. *J Appl Ichthyol* 7:244–248