First Record of *Argulus foliaceus* (Linnaeus, 1758) in Common Carp (*Cyprinus carpio* Linnaeus,1758) in Macedonian Waters with Scanning Electron Microscopy

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Abstract - This study was carried out between January 2009 and December 2013. A total of 958 specimens of common carp (*Cyprinus carpio*) from the most significant and larger cyprinid aquaculture facilities in Republic of Macedonia were examined for parasitological investigations. Fish louse, *Argulus foliaceus* was found on fins and skin in 57 specimens of common carp from four different cyprinid aquaculture facilities. Total, the prevalence with *Argulus foliaceus* in *Cyprinus carpio* was 5,950%, while the mean intensity was 7,807. By seasons, prevalence with *Argulus foliaceus* was as following: spring - 1,587%; summer - 5,026% and autumn - 2,028%, while mean intensity: spring - 5,722; summer - 16,474 and autumn - 5,391. This parasite was not found in winter season. Our findings of fish louse *Argulus foliaceus* in common carp (*Cyprinus carpio*) are first recorded for Republic of Macedonia. At the same time, common carp represent new host for *Argulus foliaceus* in Macedonian waters.

Key words: *Argulus foliaceus*, aquaculture facility, common carp (*Cyprinus carpio*), scanning electron microscopy

1 INTRODUCTION

The Branchiura is a group of crustaceans parasitizing primarily freshwater fishes (Piasecki et al. [1], Møller et al. [2]).

According Noaman et al. [3], *Argulus foliaceus* (Crustacea: Branchiura) or the fish louse, is an ectoparasite of the skin or gill of the fresh water fish species. Clinical signs in infected fish include scratching on aquarium walls, erratic swimming, and poor growth. It causes pathological changes due to direct tissue damage and secondary infections.

It is a cosmopolitan species and its geographical range includes Europe, Central Asia and North America (Rushton-Mellor and Boxshall [4]).

This parasite has a direct life cycle (Mikheev et al. [5]). The mating takes place during the free swimming stage and mature females leave the host and lay several hundred eggs on vegetation and various objects in the water. Eggs are ovoid in shape and are covered by a gelatinous capsule. Depending on the temperature, 40-100 days are required for completion of the life cycle. After being hatched the parasite must find a suitable host within four days, otherwise it will die. Adults may live free from the host for up to 15 days (Bykhovskaya-Pavlovskaya et al. [6]).

According Mirzaei and Khovand [7], *Argulus foliaceus* can act as a potential risk factor for natural ecosystems and native fish population.

The aim of this study was to determine the parasite fauna of common carp (*Cyprinus carpio*) from the most significant and larger cyprinid aquaculture facilities in Republic of Macedonia, especially finding the new parasite species in Macedonian water, using scanning electron microscopy.

2 MATERIALS AND METHODS

This study was carried out between January 2009 and December 2013. A total of 958 specimens of common carp (*Cyprinus carpio*) from eight the most significant and larger cyprinid aquaculture facilities in Republic of Macedonia (fishponds Zhabeni, Bukri, Dolneni, Zhelezara and cage fish farms on reservoirs Tikvesh, Mladost, Globochica and Gradche) were examined for parasitological investigations. The fish were caught using net or hook by local fishermen. The specimens were placed in plastic tanks with fishpond water and immedi-

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ately transferred to the research laboratory. Fish were killed by vertebral dislocation. During the dissection, the gill filaments, the eyes, the fins, the intestines and the skin were examined under the stereomicroscope. All parasites found in each individual fish were identified and enumerated. The parasite specimens were fixed in 70% alcohol to be observed under light microscope and scanning electron microscopy (SEM). During the study period, data on parasite species were categorized according to season. The environmental factors were not measured in this study.

Total numbers of parasites were determined directly by numerical count. The number of fish examined, fish infected, prevalence and mean intensity (total and by seasons) are given in tables.

Classical epidemiological variables (prevalence and mean intensity) were calculated according to Bush et al. [8].

The parasite specimens were identified using reference keys of Bykhovskaya-Pavlovskaya et al. [6].

During the examinations at Laboratory for fish diseases in Hydro - biological Institute in Ohrid (Republic of Macedonia), stereomicroscopes „Zeiss"- Stemi DV4 and „MBS 10", as well as light microscope „Reichart" were used.

The observation under scanning electron microscopy (SEM) was made at Laboratory of electron and confocal microscopy, Faculty of biology, University of Warsaw, Poland.

3 RESULTS

A total of 21 parasite species were found in 958 examined specimens of common carp (*Cyprinus carpio*) from eight the most significant and larger cyprinid aquaculture facilities in Republic of Macedonia.

Among them, fish louse *Argulus foliaceus* was found in 57 specimens of common carp from 4 cyprinid aquaculture facilities:

1. On fins and skin in common carp from fishpond Zhabeni, in summer;
2. On skin in common carp from fishpond Bukri, in spring;
3. On fins and skin in common carp from cage fish farm on reservoir Mladost, in autumn;
4. On fins in common carp from fishpond Dolneni, in spring.

Our findings of *Argulus foliaceus* in common carp (*Cyprinus carpio*) are first recorded in Republic of Macedonia. At the same time, common carp represent new host for *Argulus foliaceus* in Macedonian waters.

Data on fish examined, fish infected, as well as the prevalence and mean intensity with *Argulus foliaceus* (total and by seasons) are given in Table 1 and Table 2.

Total, the prevalence with *Argulus foliaceus* in common carp was 5,950%, while the mean intensity was 7,807 (Table 1).

Table 1. Prevalence and mean intensity with *Argulus foliaceus* in common carp (*Cyprinus carpio*) from cyprinid aquaculture facilities in Macedonia

Fish species	Number of examined fish	Number of infected fish	Mean intensity	Prevalence (%)
common carp *(Cyprinus carpio)*	958	57	7,807	5,950

During this study, infection with *Argulus foliaceus* was recorded on fish in all seasons with exception of winter (Table 2). Mean intensity of *Argulus foliaceus* was not varied significantly among the seasons, especially between spring (5.722) and autumn (5.391). The greatest value of mean intensity was recorded in summer (16.474).

Prevalence (%) of *Argulus foliaceus* was the highest in summer (5.026 %) and lowest in spring (1.587 %) (Table 2).

This parasite species was found on all size of host fish.

Table 2. Prevalence (E) and mean intensity (I) with *Argulus foliaceus* in common carp (*Cyprinus carpio*) from cyprinid aquaculture facilities in Macedonia, by seasons

Parasite species	Spring		Summer		Autumn		Winter	
	I	E (%)	I	E (%)	I	E (%)	I	E (%)
Argulus foliaceus	5,722	1,587	16,474	5,026	5,391	2,028	/	/

Kingdom: ANIMALIA Linnaeus, 1758
Phylum: ARTHROPODA Latreille, 1829
Subphylum: CRUSTACEA Lamarck, 1801
Class: MAXILLOPODA Dahl, 1956
Subclass: BRANCHIURA Thorell, 1864
Order: ARGULOIDA Yamaguti, 1963
Family: ARGULIDAE Leach, 1819
Genus: ARGULUS Müller, 1785
Species: *ARGULUS FOLIACEUS* Linnaeus, 1758
Hosts: *CYPRINUS CARPIO*
Localization: fins, skin
Place: cyprinid aquaculture facilities (Zhabeni, Bukri, Dolneni, Mladost)
Season: spring, summer and autumn

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4 DISCUSSION

Argulus foliaceus is an ectoparasite of the skin or fins of the freshwater fish species. Clinical signs in infected fish include erratic swimming and poor growth. Fish are anaemic and anxious. It causes pathological changes due to direct tissue damage and secondary infections. The skin of the fish shows a strong hyper secretion of mucus. The carp offspring with weight up to 2 grams suffer in invasion with 1-2 parasites. Annual fish die if they are infested with 20 or more parasites and older fish if attacked with 100 or more parasites. Infested carps manifested acute haemorrhagic inflamed skin wounds, an increased production of mucosal material, scales spilling and fins corrosion.

Argulus foliaceus is consisting of head, thorax and abdomen (Fig. 1). It is characterized by transparent, oval and flattened dorso-ventral body (Fig. 2 and Fig. 3) and two complex faceted eyes. First maxillae are usually modified as powerful sectorial organs, while the second maxilla, posterior to the sucker, with five segments (Fig. 7).

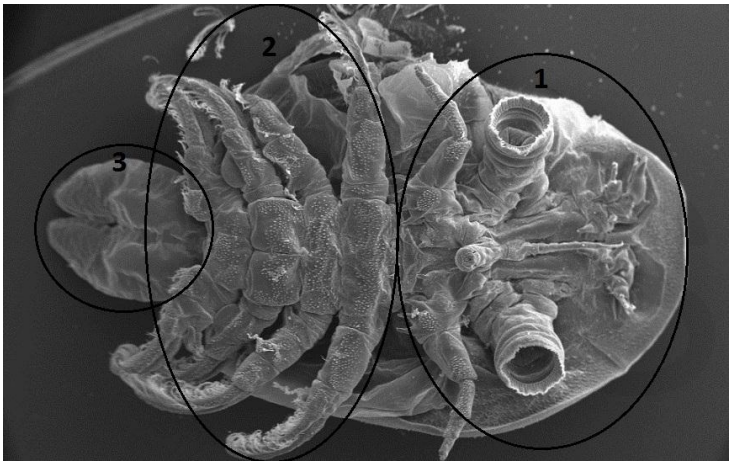

Figure 1. Body of *Argulus foliaceus* (Head – 1; Thorax – 2; Abdomen – 3) (SEM) (original)

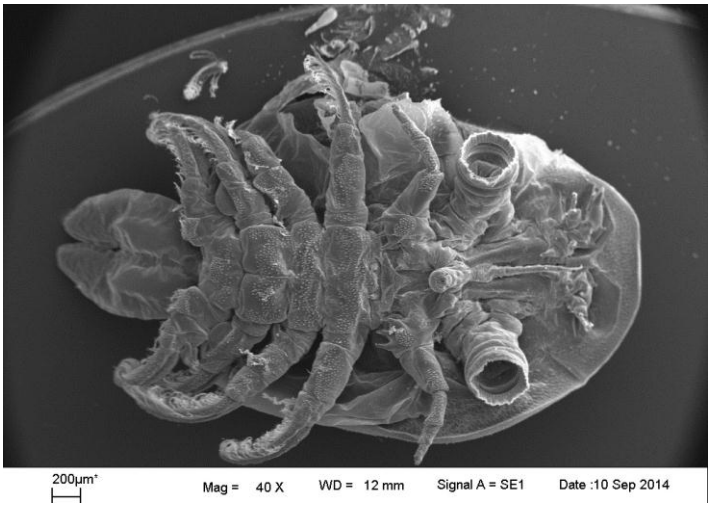

Figure 2. Ventral part of *Argulus foliaceus* (SEM) (original)

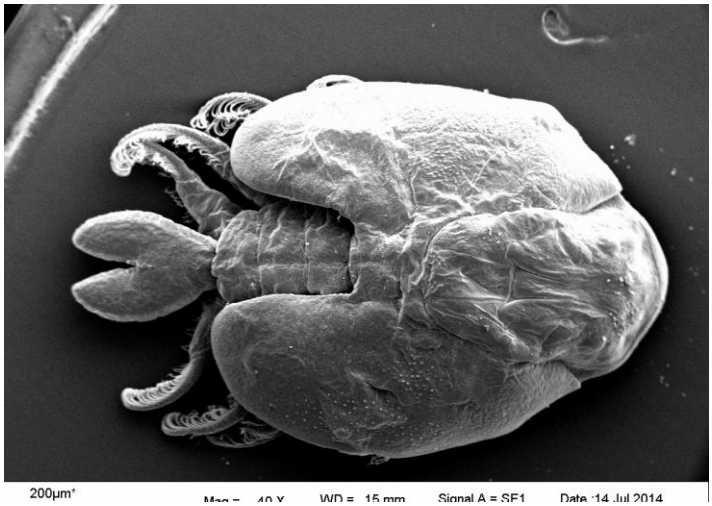

Figure 3. Dorsal part of *Argulus foliaceus* (SEM) (original)

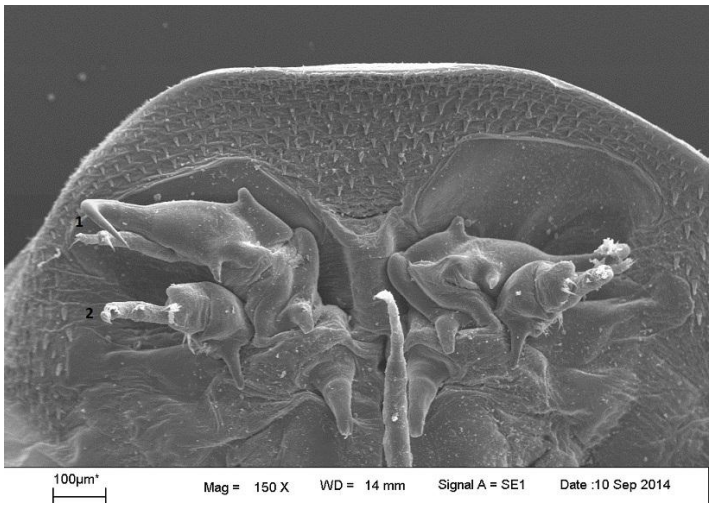

Figure 4. First antenna (1) and second antenna (2) (SEM) (original)

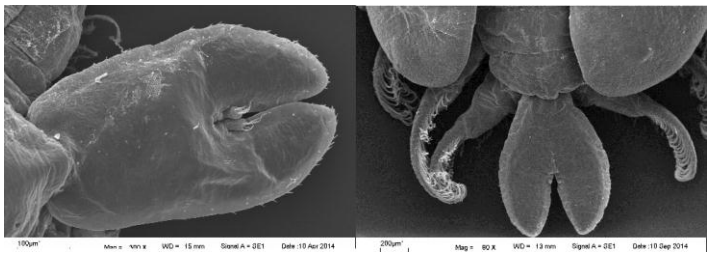

Figure 5. Abdomen (SEM) (original)

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Figure 6. Central movable parts (1); Mouth cone (2); Pre-oral spine (3) (SEM) (original)

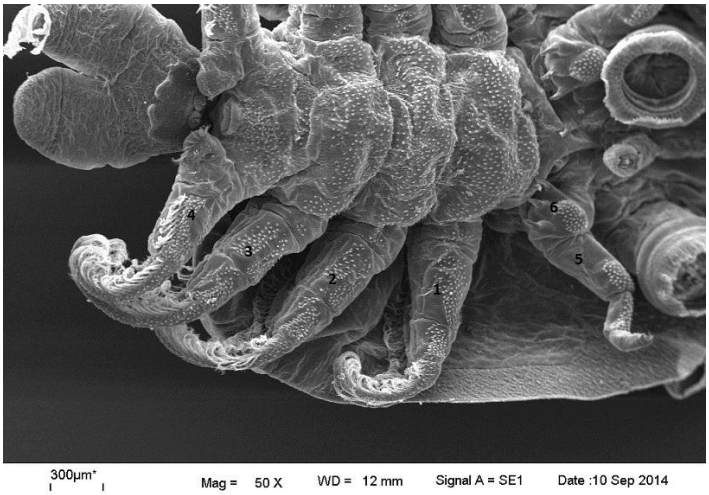

Figure 7. Thoracopods (1-4); Maxilla (5); Maxilla proximal segment (6) (SEM) (original)

5 CONCLUSION

Infested common carps manifested acute hemorrhagic inflamed skin wounds, an increased production of mucosal material, scales spilling and fins corrosion. Heavy infestations can cause serious damage to the skin and subsequent mortality.

Argulus foliaceus has been detected in Macedonian waters for the first time. Common carp (*Cyprinus carpio*) represent new host for *Argulus foliaceus* in Republic of Macedonia and it is first recorded in this paper.

Total, the prevalence with *Argulus foliaceus in Cyprinus carpio* was 5,950%, while the mean intensity 7,807.

Prevalence with *Argulus foliaceus* in common carp (*Cyprinus carpio*) from cyprinid aquaculture facilities in Macedonia, by seasons was as following: spring - 1,587%; summer - 5,026% and autumn - 2,028%, while the mean intensity was: spring - 5,722; summer -16,474 and autumn - 5,391. This parasite was not found in winter season.

It is very important for further researches to examine factors which influence the population dynamics of *Argulus foliaceus* in common carps from cyprinid aquaculture facilities in Republic of Macedonia.

REFERENCES

[1] W. Piasecki, A. Avenant-Oldewage, "Diseases caused by crustacean". In: Eiras, J.C., Segner, H., Wahli, T. and Kapoor, B.G. (eds) Fish Diseases. Science Publishers, New Hampshire, USA, pp. 1115–1200, 2008.

[2] O.S. Møller, J. Olesen, A. Avenant-Oldewage, P.F. Thomsen, H. Glenner, "First maxillae suction discs in Branchiura (Crustacea): development and evolution in light of the first molecular phylogeny of Branchiura, Pentastomida, and other 'Maxillopoda'". Arthropod Structure and Development, 37, pp. 333–346, 2008.

[3] V. Noaman, Y. Chelongar and A. H. Shahmoradi, "The First Record of Argulus foliacesus (Crustacea: Branchiura) Infestation on Lionhead Goldfish (Carassius auratus) in Iran", Iran J. Parasitol. 5(2): pp. 71–76, 2010.

[4] S. K. Rushton-Mellor and G. A. Boxshall, "The developmental sequence of Argulus foliaceus". J. Nat. Hist. 28: pp. 763-785, 1994.

[5] V.N. Mikheev, A.F. Pasternak, E.T. Valtonen, Y.A.N. Lankinen, "Spatial distribution and hatching of overwintered eggs of a fish ectoparasite, Argulus coregoni (Crustacea: Branchiura)". Dis. Aquat. Org. 46: pp.123–128, 2001.

[6] I. E. Bykhovskaya-Pavlovskaya, A. V. Gusev, M. N. Dubinina, N. A. Izyumova, T. S. Smirnova, I. L. Sokolovskaya, G. A. Shtein, S. S. Shulman, V. M. Epstein, "Key to Parasites of Freshwater Fish of the U.S.S.R". Leningrad, 1964.

[7] M. Mirzaei and H. Khovand, "Prevalence of Argulus foliaceus in ornamental fishes [goldfish (Carassius auratus) and Koi (Cyprinus carpio)] in Kerman, southeast of Iran". J. Parasit Dis., 39(4): pp.780-2, 2015.

[8] A.O. Bush, K.D. Lafferty, J. M. Lotz and A.W. Shostak, "Parasitology meets ecology on its own terms: Margolis et al. Revisited". J. Parasitology., 83: pp. 575- 583, 1997.