



Research Article

Histopathological Studies of Pond Reared Indian Major Carp, *Catla catla* Infested with *Argulus japonicus* and Trial for Argulosis Treatment

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Authors' Contributions

MAAM designed the study, performed the investigation, wrote the protocol and prepared the first draft of the manuscript. SN helped in sampling and statistical analysis. SSR helped in writing and executing the reviewers' comments. KSR supervised over all research experiments.

Keywords

Argulus japonicus, *Catla catla*, Histopathology, Hemorrhage, Infestations, Treatments, Mangalore

Abstract | Indian major carp, catla (*Catla catla*) that showed erratic and lethargic movement were collected from College Fish Farm, Mangalore. We examined a total of 20 catla, found all the fish infested with fish louse, firmly attached to the skin, fins, head, and operculum. Based on the morphology of the specimens, the parasites were identified as *Argulus japonicus*. Moribund fishes were examined and up to 350±50 lice were hand-picked from a single fish. Microscopic examinations of *A. japonicus* revealed that most of these were at juvenile stages. Infested fish were transferred to the glass aquaria (60 L) and treated with 4 treatments for 15 days: I) Potassium permanganate II) Aquarium salt III) Formalin and IV) Mechanical. The significant lower ($P<0.05$) parasitic burden were found in mechanical treatment followed by salt, formalin and potassium permanganate. Clinical signs like excessive mucus production, fin erosion, pale gills, and slight hemorrhage were observed in the affected area of skin and operculum. Among the internal organs, liver lost its original reddish-brown and turned to yellowish-brown, intestinal duct remained empty and swelled. Severe infestation in catla caused eye opacity and tail rot indicated the devastating effects of *A. japonicus*. Infestation by this parasite resulted in histopathological changes in the skin and muscle, gill, liver, kidney, heart and different parts of gut tissues. According to the results obtained in the present study, it can be suggested that acute infestation of *A. japonicus* elicited direct effects such as eye opacity, fin rots, scale loss and severe histopathological alterations in catla.

Novelty Statement | Mass infestation and histopathological changes due to heavy infestation of *A. japonicus* in pond reared Indian major carp, *Catla catla* were validated in the present study for the first time in Karnataka, India.

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Introduction

One of the most prevalent parasite of fish is *Argulus*, commonly known as fish lice. It belongs to the large branchiurans (Subclass: Branchiura) ectoparasite group

under the family Argulidae, where one genera, *Argulus* carry at least 129 distinguished species reported from freshwater and coastal water environments globally with exception of Antarctica (William, 2008). Few species, *A. foliaceus*, *A. japonicus*, *A. coregoni*, *A. siamensis*, are involved in the heavy infestation of freshwater fish and often caused mass mortality in cultured and wild habitats (Pekmezci et al., 2011; Steckler and Yanong, 2012). Although the

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highest heterogeneity of *Argulus* were reported in the Afrotropical and Neotropical zone (William, 2008), the rich diversity of this genus was also reported from India. In India, the fish lice were first noticed at 1951 and till date 14 *Argulus* species were validated from different water bodies with the most widespread *A. siamensis* (Kumar *et al.*, 2017). Moreover, the phylogenetic analysis of *Argulus* spp. collected from 13 major aquaculture regions of India revealed 11 population were *A. siamensis* (Sahoo *et al.*, 2013b). Although *A. japonicus* were reported from few population (2 sites) but it can be much more pathogenic and detrimental than *A. siamensis* (Mohanty *et al.*, 2012). The fecundity, prevalence, intensity, and pathogenicity is higher in *A. japonicus*, with faster maturity and size compared to other Argulids. Among *Argulus* spp. in India, including *A. bengalensis* and *A. siamensis* had preferred host of infestation however, *A. japonicus* can infest a wide range of host including common carp (*Cyprinus carpio*) rohu (*Labeo rohita*) catla (*Catla catla*) and mrigal (*Cirrhinus mrigala*) (Mohanty *et al.*, 2012; Banerjee and Saha, 2013). Traditional and intensive aquaculture farms are frequently encountered with Argulosis caused by *A. bengalensis* (Banerjee and Saha, 2013; Patra *et al.*, 2016), *A. siamensis* (Saha *et al.*, 2011; Sahoo *et al.*, 2013b; Patra *et al.*, 2016) and *A. japonicus* (Prabhavathy and Sreenivasan, 1976).

The alien species, *A. japonicus* probably entered India through Southeast Asian countries from its original habitat, Japan. The transboundary fish trading has been spread the *A. japonicus* to the whole world and emerged as epidemic (Avenant-Oldewage, 2001). Generally, *Argulus* per-se do not cause fish mortality in aquatic habitats, however, it acts as a vector for bacterial (Steckler and Yanong, 2012), viral (Ahne, 1985), fungal (Singhal *et al.*, 1990) and parasitic diseases (Noga, 2014) that could lead to mass mortality of fish resulting economic losses. However, heavy infestation with hundreds of *Argulus* in a single fish can cause direct mortality, as reported from different parts of the world (Menezes *et al.*, 1990; Pekmezci *et al.*, 2011). The economic loss in India due to Argulosis was estimated to be US\$ 615 ha⁻¹year⁻¹ (Sahoo *et al.*, 2013a). Moreover, in case of mixed infection, the magnitude of losses increased to US\$ 1428 ha⁻¹year⁻¹ (Mohanty *et al.*, 2012).

Argulus spp. is known as an obligate parasite throughout its life. During its attachment in hosts' skin usually feed on the mucus and blood. Infested fish exhibit atypical abnormalities including slow movement, irritation, keep away from the shoal and gathering near the pond wall. In the mass occurrence fish produce excess mucus, pale body, scale loss, fin rots and hemorrhagic spots (Pekmezci *et al.*, 2011). Internal organs such as liver, kidney and intestinal duct often lost its original color and texture. However, limited reports are available on the histological changes of internal organ of the host due to Argulosis. Indian major carps contribute approximate 90% of carp

aquaculture production in India and catla is one of most economic important fish after rohu (Sahoo *et al.*, 2013a). Available reports about Argulosis and its treatment trial on this economically important fish were very sparse and scattered. Therefore, the present study was aimed to report the heavy infestation of *A. japonicus* with its prophylactic measures and to evaluate the histopathological effects of gills, liver, heart, kidney, and gut of catla upon Argulosis.

Materials and Methods

Erratic movement and lethargic fish were observed in a monoculture pond of catla (*C. catla*) that belongs to Aquaculture Department, College of Fisheries, Mangalore, Karnataka Veterinary, Animal and Fisheries Sciences University, Karnataka, India. From that pond, a total of 20 affected live fish were randomly selected and brought to the Aquatic Animal Health Laboratory, Department of Aquaculture. The fishes were kept in 150 L Fibre-reinforced plastic tank with vigorous aeration. Few fishes (n=4) were taken for counting the *Argulus* and histopathological studies, whereas remaining fish (n=16) were transferred to the equal glass aquaria (80 L) for the treatment purpose. Infested fish were treated with four treatments viz., I) Potassium permanganate @ 1ppm II) Aquarium salt @ 2% III) Formalin @ 25 ppm and IV) Mechanical (manual) @ 2 times rubbing with soft nylon net for 15 days period (Table 1). The treatment doses were applied according to Mamun *et al.* (2019a) with slight modifications. Manual treatment and chemicals were applied on 0 day and 7 days to evaluate the best treatment methods. Fishes were fed with commercial pelleted diet (30% crude protein) once in a day and reared in optimal condition. Water was siphoned out in every alternate day and at the same time 10% water was replaced. At the end of the treatment trial (on 15th day) fishes were harvested and parasites on each fish per treatment were counted and documented. SPSS version 20 was used for one-way analysis of variance (ANOVA) at a 5% significant level and Duncan Multiple Range test for measuring significant difference.

Table 1: Parasite (*A. japonicus*) burden on the catla with different treatments observed at the end of 15 days.

Treatments (on 0 and 7 th day)	No. of <i>A. japonicus</i> on each fish
Potassium permanganate @ 1 ppm*	92.67±7.88 ^a
Aquarium salt @ 2%	67±4.16 ^b
Formalin @ 25 ppm	70.67±8.84 ^{ab}
Mechanical (rubbing with soft nylon net) @ 2 times	25±5.03 ^c

Note: *total number of fish (n=4) per treatment. Different superscripts (a, b, c) in the column vary significantly (P < 0.05). Data expressed as Mean ± SE.

The individual fish were weighed and measured carefully avoiding detachment of parasites (Figure 1). Whole

fish body, including head, fins, and scale surface were examined for parasitic infection. Individual *Argulus* with different stages were picked by forceps, hand counted, and microphotographed. The *Argulus* spp. was identified following the protocol of Mohanty *et al.* (2012). No endoparasitic, bacteriology or virology work was performed in this study. In order to find out the histopathological effects at tissue level due to Argulosis, moribund fish were euthanized and different organs including gill, liver, heart, kidney and intestine (foregut, midgut and hindgut) were fixed in 10% neutral buffered formalin for 72 h. The procedures were performed as per the guidelines of the Animal Ethics Committee, College of Fisheries, Mangalore. After fixation, dehydration with graded alcohol were done followed by embedding, sectioning (5 µm) and finally tissues were stained (haematoxylin and eosin). All the protocols were followed according to Bullock (1989). The histological alterations of tissue were analyzed by light microscopy (Olympus, BX3-25ND25, Japan).

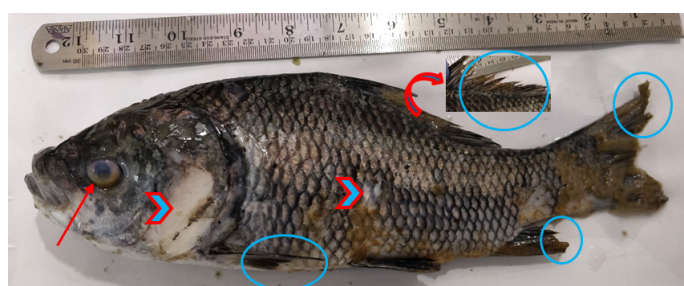


Figure 1: Argulosis in catla showing numerous gross lesions including fin rots (circles), skin and scal loss (arrow heads), corneal opacity (arrow), excessive mucus on the surface of the infested fish.

Results and Discussion

Clinical symptoms of infested Catla

Infested catla exhibited several clinical signs including frayed and eroded fins, scale loss, excessive production of mucus and corneal opacity (Figure 1). During handling fish usually wriggle and show escape movements, but in this case, the fish were calm and lethargic indicating the severity of the stress due to Argulosis. Internal organs showed pale and swollen structure particularly kidney, where liver lost its reddish-brown colour and turned to yellowish-brown, intestinal duct remained empty and swollen (Figure 2b). During necropsy fish body where found in anaemic state. However, no visible lesions of the inner organs were detected in the systematic autopsy. The gill filaments and rakers were found in extreme pale condition accompanied with clogged gill arch (Figure 2b).

Trial for argulosis treatment

Among all the controlling measures mechanical treatment by rubbing the skin with soft nylon net as most effective means of detaching the parasite from fish. At the trial end, we found significant lower level ($P < 0.05$) of attached

parasites on mechanical treatment (Table 1). However, treatments with potassium permanganate, aquarium salt and formalin were not effective as compared to mechanical treatment in detaching the parasites.

Identification of the Argulus species

Based on the morphological differentiation between *A. japonicus* and *A. siamensis*, collected specimens were identified as *A. japonicus*. The dorsal ridges of *A. japonicus* was branched which is one of the most distinguishing feature that separates it from those of *A. siamensis* (Figure 3a-b). Another distinguishing morphological character of *A. japonicus* is bilobed second swimming leg (Figure 3a). *A. japonicus* were firmly attached on whole body of catla especially on dorsal and caudal fins, operculum and ventral side (Figure 2a). Different stages (juvenile and adult) of *A. japonicus* were collected and microphotographed (Figure 4).

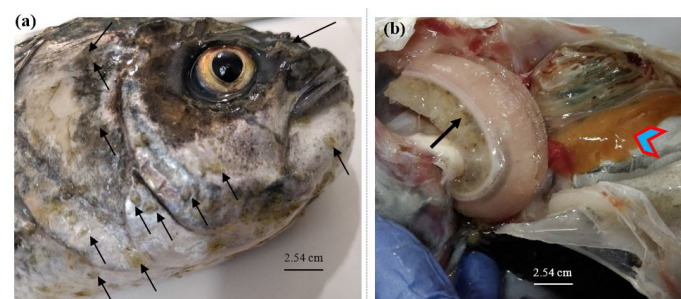


Figure 2: Heavy infestation of *Argulus japonicus* (a) *A. japonicus* firmly attached on the operculum and head surface of catla (arrows); (b) Infested catla showed pale gills and clogged gill arch (arrow) with englarge liver that lost its original reddish-brown colour and turned to yellowish-brown colour (arrow heads).

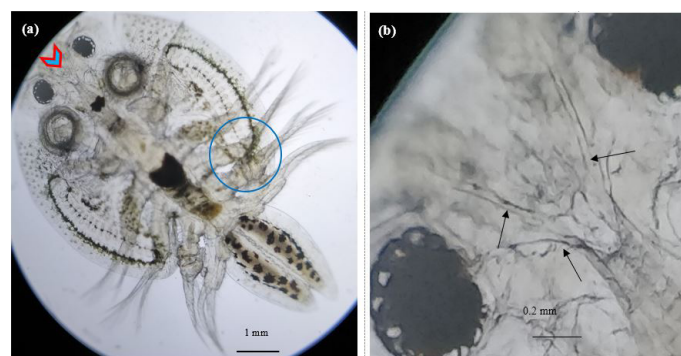


Figure 3: (a) *Argulus japonicus* (Thele 1900) with distinct morphological features including branched dorsal ridge (arrow head) and bilobed second segment (circle); (b) Computer blown up image showing clear image of branched dorsal ridge (arrows).

Histopathological effect in Catla due to Argulosis

Argulosis resulted in severe histological alterations in

different organs of catla. The muscle myofibrils were found broken along with vacuolization in myotomes (Figure 5a). The muscle fibres were splitted and disintegrated myotomes were noticed in the affected muscles of catla (Figure 5b).



Figure 4: Juvenile (arrow head) and adult (arrow) *Argulus japonicus* found in catla.

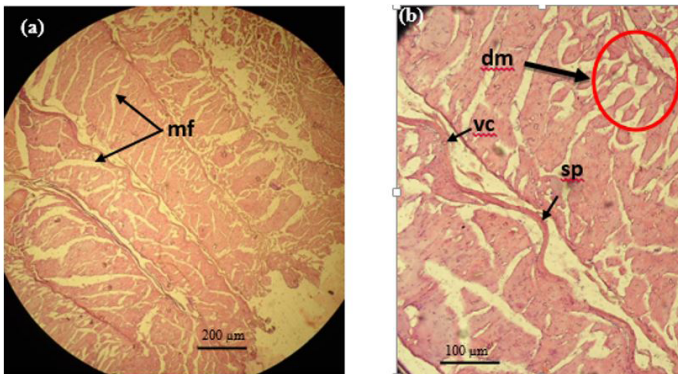


Figure 5: Photomicrographs of the muscle of catla infested with *Argulus japonicus*. a) The broken of myofibrils (mf) (10×); b) Splitting of muscle fibres (sp) disintegrated myotomes (dm) and vacuolation (vc) within the myotomes (40×).

Heavy infestation of *A. japonicus* brought dire consequences in gill anatomy causing complete loss of secondary gill lamellae (Figure 6a-b). The liver histopathology displayed vacuolation, pyknotic nuclei, degenerated hepatocytes cells accompanied with necrosis and atrophy (Figure 7a-b). Vacuolar degeneration, necrotizing endocardium, infiltrated lymphocytes, damage myocardium were noticed in heart ventricular histopathology (Figure 8a-b). Kidney tissues were found in degenerative state in histopathological

studies, displayed disintegration of tubules, glomerular distension, tubular necrosis, and aggregated leukocyte cells (Figure 9a-b). Fore gut were revealed normal architecture (Figure 10a-b) in infested catla, however, short and stout villi, large lumen space, degenerated enterocytes with microvilli, disrupted vacuolization in the mucosal tissues were detected in both mid and hind gut (Figures 11a-b, 12a-b).

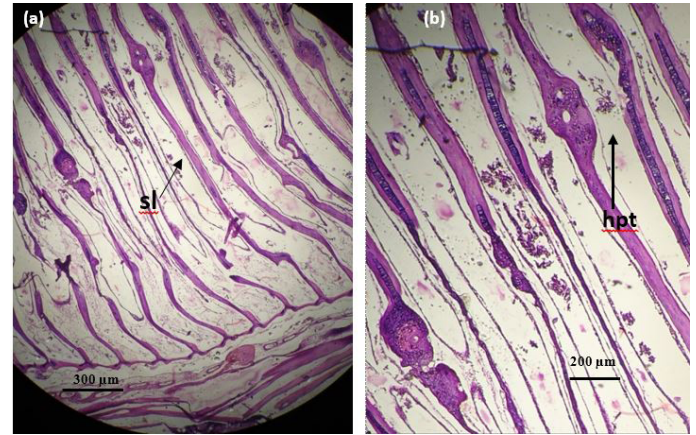


Figure 6: Gill histopathology of catla infected by *Argulus japonicus* a) Displaying complete loss of secondary gill lamellae (sl) and irregular shaped primary gill lamellae (10×); b) Part of the primary gill lamellae become hypertrophied (hpt) (40×).

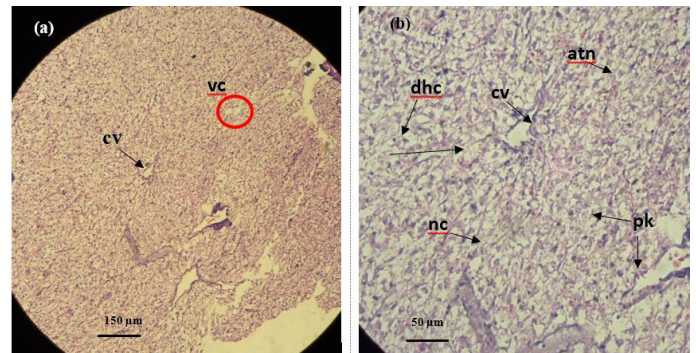


Figure 7: Liver histological section of infested catla by *Argulus japonicus*, a) Central vein (cv) vacuolization (vc) (10×); b) Numerous pyknotic nuclei (pk), disintegrated hepatocyte cells (dhc), atrophied nucleus (atn), necrotic hepatocytes (nc) (40×).

Heavy infestation of *Argulus japonicus* in Indian major carp, Catla and its histopathological analysis were investigated in the present study. Argulosis caused detrimental effect on the catla marked with sever clinical disorders, including structural deformity in the skin and muscles tissues, eroded fins, corneal opacity, and hemorrhagic spots at the site of infestation. In the pond, group of infested fishes were observed with violent erratic movement, while few fishes showed lethargic movement, resting on the pond wall and showed lethargic. The quantum of the clinical signs, and

pathology depends on the population density of *Argulus* in fish (Kumar *et al.*, 2017). Under the chronic state of Argulosis, fishes displayed abrupt movement (Noga, 2014) while under acute infestation fishes with excessive mucus production and sporadic/low intensity haemorrhages on the site of infestation was reported earlier (Roberts, 2012), which was also evident in this study.

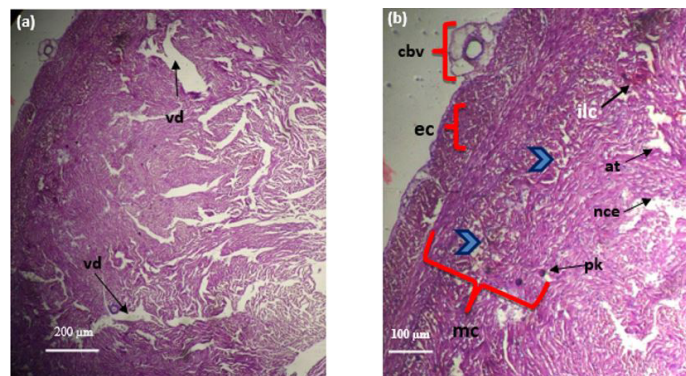


Figure 8: Photomicrograph of heart ventricle of catla infested with *Argulus japonicus*. a) Vacuolar degeneration (vd) accompanied with disrupted myocardium tissues (10×); b) Coronary blood vessel (cbv), epicardium (ec) myocardium (mc), cardiac muscle showed necrotizing endocardium (nce), infiltrated lymphocytes (ilc), degenerated myocardium (arrow heads) accompanied with atrophy (at) and hypertrophied cells (pk) (40×).

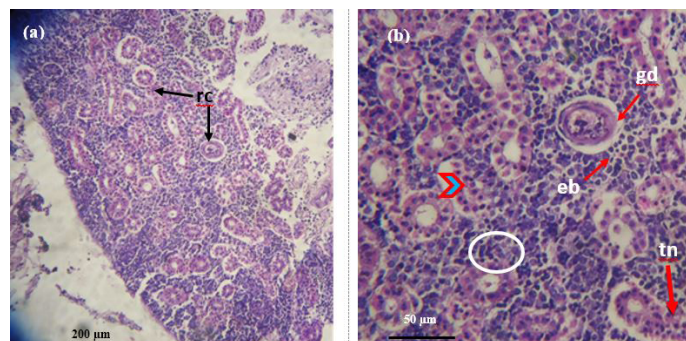


Figure 9: Photomicrographs of the infested kidney of catla by *Argulus japonicus*. a) Showing renal corpuscles (rc) (10×); b) Glomerular distension (gd) accompanied with extended bowmans capsule (eb), vacuolation (arrow head) tubular necrosis (tn), increase in the presence of leukocyte cells (circle) (40×).

The most widespread *A. siamensis* is often confused with *A. japonicus* because they having similar anatomical structure. Therefore, proper identification of *A. japonicus* from *A. siamensis* is imperative to understand the nature and site of infestation along with its epidemics. The *Argulus* specimens collected in the present study were identified as *A. japonicus* based on the morphological differentiation from *A. siamensis*. The detailed morphological features

of these two *Argulus* spp. were reported by Mohanty *et al.* (2012) with microphotography. Among the many distinct characteristics between these two species, the most distinguishable features were dorsal ridge and second swimming leg of male which was reported unbranched and bilobed respectively (Mohanty *et al.*, 2012), are similar with the present study. *Argulus japonicus* were reported from the capital city of Karnataka, Bangalore which is nearby of Mangalore (Sahoo *et al.*, 2013b). Molecular based species identification revealed that Bangalore, Karnataka and Mandi, Himachal Pradesh clusters were *A. japonicus* (Sahoo *et al.*, 2013b). However, they reported *A. japonicus* from common carp (*Cyprinus carpio*) and gold fish (*Carassius auratus*) respectively. Two sub species of *Argulus* were reported from Chitradurga and Bangalore district of Karnataka (Bai *et al.*, 1988) whereas *A. mangalorensis* were delineated from the planktonic sample of Mangalore's estuarine water (Natarajan, 1982).

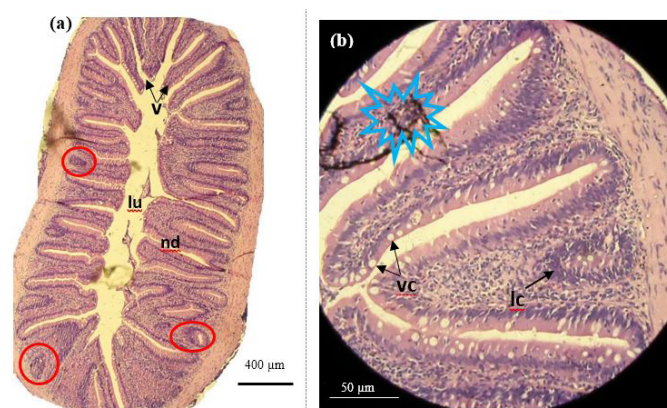


Figure 10: Histology section of fore gut of catla infested with *A. japonicus*. a) Normal architecture of villi (v) with numerous gut associate lymphoid tissues in mucosal tissues (red circle) (10×); b) Emptied mucous cells (mc), aggregated lymphocyte cells (lc) (40×) =artifacts.

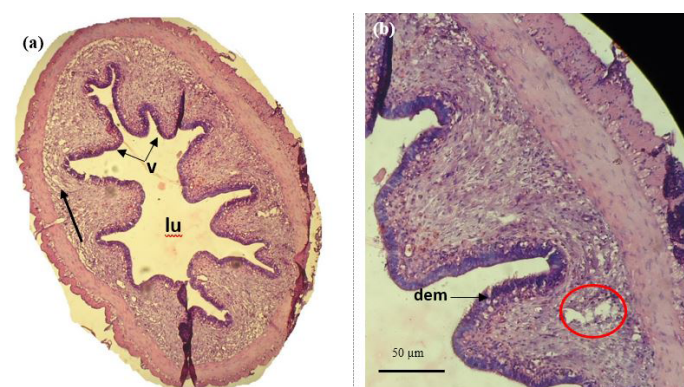


Figure 11: Histological analysis of mid gut of catla infested with *Argulus japonicus*. a) Transvers section showing short and stout villi (v) large lumen space (lu) with lamina propria (lp) disruption of mucosal and sub-mucosal tissues (arrow) (10×) b) Damage of enterocytes and microvilli (dem) with large vacuolar space (circle) (40×).

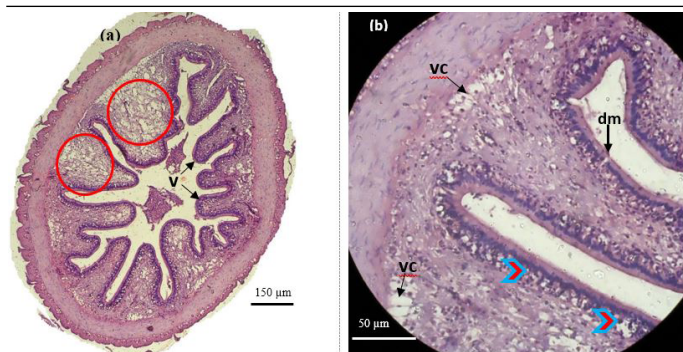


Figure 12: Histology section of hind gut of catla infested with *Argulus japonicus*. a) Short and stout villi (v), disruption and vacuolization (vc) in the mucosal tissues (red circle) (10×); b) disintegration of enterocytes (arrow heads), damage of microvilli (dm) (40×).

In this study, at least 350 ± 50 *A. japonicus* of different stages were hand-picked from a single pond reared catla. The lentic water such as ponds, lakes and reservoir are the preferred habitat for *A. japonicus* (Rushton-Mellor, 1992) where they can able to breed throughout the year by continuous breeding pattern (Shafir and Oldewage, 1992). Severe infestation of *A. foliaceus*, up to 800-1000 lice were reported from pond reared *Cyprinus carpio* in Turkey (Pekmezci *et al.*, 2011). Our study corroborates with Kruger and Van-As (1983) where they described *A. japonicus* with 90% prevalence in different fish species of western Transvaal, South Africa. *A. japonicus* become potential threat to the wild and cultured fishes of UK and authorities were suggested for the careful examination of ornamental fishes before entry to the UK environment (Rushton-Mellor, 1992). *Argulus japonicus* were found highly pathogenic (Gresty *et al.*, 1993) and it was interesting to find that *A. japonicus* dominated over *A. siamensis* when mixed infection occurred in rohu (*Labeo rohita*) and mrigal (*Cirrhinus mrigala*) (Mohanty *et al.*, 2012). The continuous spawning pattern (Shafir and Van-As, 1986), off-host ability, and degree pathogenicity made *A. japonicus* into more virulent (Mohanty *et al.*, 2012) and opportunistic (Shafir and Van, 1985). The therapeutic agents like potassium permanganate, salt and formalin were frequently used chemicals that are used against *Argulus* spp. (Rintamäki *et al.*, 1994). Mechanical treatments (rubbing/shaking) found extremely effective in detachment of *A. coregoni* (Hakalahti-Sirén *et al.*, 2008). Earlier studies demonstrated that removal of *A. japonicus* in mechanical method reduced the burden of parasites from the body of different fish species (Mamun *et al.*, 2019a)

Histopathological alterations derived from the argulosis has not gained much attention. Therefore, this study was aimed to report the histopathological findings of catla heavily infested with *A. japonicus*, a ubiquitous *Argulus* species with cosmopolitan distribution. The muscle

histopathology revealed broken myofibrils and numerous vacuoles accompanied with disintegrated myotomes. Continuous feeding of mucus and blood through proboscis by inserting stylet from the skin musculature could alter the normal anatomy of the skin and muscle. The hooks, appendages of *Argulus*, and repeated penetration of stylet were involved in mechanic damage of skin and muscle (Noga, 2014). Several histopathological changes including aggregated melanocytes accompanied with edema, hyperplasia and necrosis were found in the skin and muscle of gold fish (*Carassius auratus*) (Al-Darwesh *et al.*, 2014). Similar pathological changes were observed in the skin and muscle of Indian major carps (*Catla catla*, *Labeo rohita*) (Das *et al.*, 2009), common carp (*Cyprinus carpio*) (Al-Hamdanne and Al-Tae, 1995), mrigal (*Cirrhinus mrigala*) and rohu (*Labeo rohita*) (Ahmad *et al.*, 2016). Gills, in the present study were the most affected organ due to heavy infestation of *A. japonicus*, virtually lost its original anatomical structure. The histopathological study revealed complete loss of secondary gill lamellae with hypertrophied primary gill lamellae indicated the severity of the *Argulus* attack. Prolonged infestation with huge number of *A. japonicus* may alter the gill structure in catla. The gill rakers were found clogged with numerous debris particle may led to the termination of secondary gill lamellae. These results are in agreement with the findings of Dash *et al.* (2009). Ahmad *et al.* (2016) reported fusion of gill lamellae with hypertrophy and hyperplasia and these alterations due to the hypoxia. The pathology derived from Argulosis related to the continuous irritating behaviour of fish and abrupt swimming with feeding regime and attachment of *Argulus* (Roberts, 2012).

The liver histopathology in the present study revealed vacuolization, necrosis, pyknosis and disintegration of hepatic cells. Approximately 40 percent hepatic cells lost its polygonal shape and turned into irregular shape. Most importantly the nucleus of the individual hepatic cell disappeared and/or atrophied. Although central vein remained normal but disintegrated hepatic cells dominated over normal hepatocytes. Similar liver pathologies including vacuolation, congestion were reported from tissue section of mrigal (*Cirrhinus mrigala*) (Ahmad *et al.*, 2016). Heart ventricular tissue sectioned displayed vacuolar degeneration accompanied with necrotizing endocardium. Moreover, disintegrated myocardium, pyknotic cells, along with atrophy were also detected in the present study. The presence of renal corpuscle, hematopoietic tissues in the kidney histology indicated there were no bacterial infection in the infested fish. Bacterial infection often caused severe pathological changes where fish lost its original anatomy of kidney (Mamun *et al.*, 2020). However, in kidney histology we found few alterations related to heavy infestation of *A. japonicus* in catla which were glomerular distension, vacuolization, tubular necrosis and aggregated lymphocytes cells. The results of the present study are corroborated with

Ahamad *et al.* (2016) reported mild tubular damage and necrotizing kidney tissues. In the present study, Indian major carp, catla infested with *A. japonicus* in long back that could lead to the anaemic and hypoproteinemia due to the overfeeding of the fish's mucus and blood rendered tissue alteration in liver, heart and kidney.

The Histopathological studies related to heavy infestation of *Argulus* is very scarce (Sahoo *et al.*, 2013a; 2013b, Kumar *et al.*, 2017). Short and stout villi were noticed in the intestinal histopathology of catla. Though fore gut were found in normal architecture however, both mid and hind gut showed severe histopathological changes, including vacuolization, disintegration of mucosal and sub-mucosal tissues and damage of enterocytes and microvilli. Moreover, the absence of goblet cells and wider lumen in the present study indicated the weak immune system of catla (Mamun *et al.*, 2019b). The surface of the villi are very important for the efficient absorption of digested nutrients. In addition, the enterocytes along with goblet cell produce necessary enzymes not only for proper digestion but also enhanced the surface microbial trapping. The short and stout villi related to the disruptive digestive system of the infested catla. Malabsorption of nutrients may lead to various physiological disorders including poor growth, skeletal deformity, and reproductive imbalance. Argulosis responsible for the reduction in aesthetic beauty to the recreational fishery (Taylor *et al.*, 2016). Heavy infestation of *Argulus* not only cause fin rots, dermal haemorrhages and ulceration but also responsible for huge economical loss due to growth reduction, higher feed conversion efficiency. The monetary loss due to Argulosis in aquaculture farm of India were estimated US\$615 h⁻¹y⁻¹ where US\$504 (82%) was due to the growth retardation (Sahoo *et al.*, 2013a). The growth impairment in fish might be due to direct cause of blood feeding by the *Argulus* and also other indirect effects including gut intestinal alterations that virtually ceased the nutrient absorption through lumen. The gut histopathology in the present study validated the tissue alteration due to Argulosis could primarily be attributed for the growth impairment. Several studies noted decreased growth rate during infestation of *Argulus* from different fish species (Singhal *et al.*, 1990; Mohanty *et al.*, 2012; Kumar *et al.*, 2017).

Conclusion

The present experiments showed that *A. japonicus* can be detached effectively by mechanical treatment with rubbing the skin with soft nylon net. Histopathological changes due to heavy infestation of *A. japonicus* in pond reared Indian major carp, *Catla catla* were validated in the present study. Acute infestation caused fin rots, scale loss, anaemic gills and swelled inner organs of catla. The histopathological analysis revealed severe alteration in the gills, liver, heart and gut morphology might be due

to the direct and indirect effect of highly invasive species *A. japonicus*. Further studies need to endorse Argulosis treatment in field level and the cause of histopathological changes in *Argulus* infested fishes.

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Conflict of interest

The authors have declared no conflict of interest.

References

- Ahamad, D.B., Punniamurthy, N., Kumar, V.S., Selvaraj, J. and Gomathinayagam, S., 2016. Pathomorphology of argulosis in fresh water carps in Thanjavur region of Tamil Nadu. *Shanlax Int. J. Vet. Sci.*, **3**: 28-34.
- Ahne, W., 1985. *Argulus foliaceus* L. and *Philometra geometra* L. as mechanical vectors of spring viremia of carp virus (SVCV). *J. Fish Dis.*, **8**: 241-242. <https://doi.org/10.1111/j.1365-2761.1985.tb01220.x>
- Al-Darwesh, A.A., Al-Shabbani, M.A.A., and Faris, B.H., 2014. Diagnostic and pathological study of *Argulus japonicus* in goldfish (*Carassius auratus*). *Glob. J. Biosci. Biotech.*, **3**: 384-387.
- Al-Hamdanne, A.H., and Al-Tae, A.F., 1995. Pathological study of experimental infection of the common carp with fish lice *Argulus foliaceus*. *Iraq. J. Vet. Sci.*, **8**: 109-112.
- Avenant-Oldewage, A., 2001. *Argulus japonicus* in the Olifants River system- possible conservation threat. *Afr. J. Wildl. Res.*, **31**: 59-63.
- Bai, A.S., Seenappa, D., and Deveraj, K.V., 1988. Oviposition and sex ratio of *Argulus siamensis* var. *siamensis* and *Argulus siamensis* var. *Hessarghattaris* (Crustacea: Branchiura) parasitic on freshwater fishes. *Curr. Sci.*, **57**: 685-686.
- Banerjee, A., and Saha, S.K., 2013. Biphase control of (*Argulus bengalensis*) Ramakrishna (1951) (Crustacea: Branchiura) with plant derivatives. *Aquac. Res.*, **44**: 202-209. <https://doi.org/10.1016/j.aquaculture.2013.07.044>
- Bullock, A.M., 1989. Laboratory methods. In: Fish Pathology (ed. R.J. Roberts), London: Bailliere Tindall, UK. pp. 374-406.
- Das, G., Parida, S.K., and Sahoo, S.N., 2009. Histopathological observations on carps infected with *Argulus* in freshwater bheries of West Bengal. *J. Appl. Biosci.*, **35**: 159-162.
- Gresty, K.A., Boxshall, G.A., and Nagasawa, K., 1993.

- The fine structure and function of the cephalic appendages of the branchiuran parasite, *Argulus japonicus* Thiele. *Philos. Trans. R. Soc. Lond., B, Biol. Sci.*, **339**: 119-135. <https://doi.org/10.1098/rstb.1993.0009>
- Hakalahti-Sirén, T., Mikheev, V.N., and Valtonen, E.T., 2008. Control of freshwater fish louse *Argulus coregoni*: a step towards an integrated management strategy. *Dis. Aquat. Org.*, **82**: 67-77. <https://doi.org/10.3354/dao01971>
- Kruger, I., and Van AS, J.G., 1983. Observations on the occurrence of the fish louse *Argulus japonicus* Thiele, 1900 in the western Transvaal. *Afr. Zool.*, **18**: 408-410. <https://doi.org/10.1080/02541858.1983.1447848>
- Kumar, S., Kumar, T.S., Vidya, R., and Pandey, P.K., 2017. A prospective of epidemiological intervention in investigation and management of argulosis in aquaculture. *Aquac. Int.*, **25**: 303-325. <https://doi.org/10.1007/s10499-016-0030-0>
- Mamun, M.A.A., Nasren, S., Rathore, S.S., Abhiman, P.B. and Ramesh K.S., 2019a. Studies on infection variation of fish to *Argulus japonicus* and trial for argulosis treatment. Asian-Pacific Aquaculture June 19-21, 2019, organized by World Aquaculture Society, Chennai, Tamil Nadu, India.
- Mamun, M.A.A., Nasren, S., Rathore, S.S., and Alam, M.M.M., 2020. Histopathological analysis of striped catfish, *Pangasianodon hypophthalmus* (Sauvage 1878) spontaneously infected with *Aeromonas hydrophila*. *Jordan J. Biol. Sci.*, **15**: 2022.
- Mamun, M.A.A., Nasren, S., Rathore, S.S., Sidiq, M.J., Dharmakar, P., and Anjusha, K.V., 2019b. Assessment of probiotic in aquaculture: functional changes and impact on fish gut. *Microbiol. Res. J. Int.*, **29**: 1-10. <https://doi.org/10.9734/mrji/2019/v29i130156>
- Menezes, J., Ramos, M.A., Pereira, T.G., and Da Silva, A.M., 1990. Rainbow trout culture failure in a small lake as a result of massive parasitosis related to careless fish introductions. *Aquac. Res.*, **89**: 123-126. [https://doi.org/10.1016/0044-8486\(90\)90304-6](https://doi.org/10.1016/0044-8486(90)90304-6)
- Mohanty, J., Sahoo, P.K., Garnayak, S.K., and Kar, B., 2012. Mixed infection of *Argulus japonicus* and *Argulus siamensis* (Branchiura, Argulidae) in carps (Pisces, Cyprinidae): Loss estimation and a comparative invasive pattern study. *Crustaceana*, **85**: 1449-1462. <https://doi.org/10.1163/156854012X651501>
- Natarajan, P., 1982. A new species of *Argulus* Muller (Crustacea: Branchiura), with a note on the distribution of different species of *Argulus* in India. *Anim. Sci. J.*, **91**: 375-380. <https://doi.org/10.1007/BF03186132>
- Noga, E.J., 2014. *Fish disease: Diagnosis and treatment*, 2nd ed. Wiley India Pvt. Ltd., Ansari Road, Daryaganj, New Delhi, India. pp. 497.
- Patra, A., Mondal, A., Banerjee, S., Adikesavalu, H., Joardar, S.N., and Abraham, T.J., 2016. Molecular characterization of *Argulus bengalensis* and *Argulus siamensis* (Crustacea: Argulidae) infecting the cultured carps in West Bengal, India using 18S rRNA gene sequences. *Mol. Biol. Res. Commun.* **5**: 156-166
- Pekmezci, G.Z., Yardimci, B., Bolukbas, C.S., Beyhan, Y.E., and Umur, S., 2011. Mortality due to heavy infestation of *Argulus foliaceus* (Linnaeus, 1758) (Branchiura) in pond-reared carp, *Cyprinus carpio* L., 1758 (Pisces). *Crustaceana*, **84**: 553-557. <https://doi.org/10.1163/001121611X574317>
- Prabhavathy, G., and Sreenivasan, A., 1976. Occurrence of *Argulus japonicus* in brood fish ponds in Tamil Nadu. *J. Inland Fish. Soc. India*, **8**: 131-133.
- Rintamäki, P., Torpström, H. and Bloigu, A., 1994. *Chilodonella* spp. at four fish farms in northern Finland. *J. Eukaryot Microbiol.*, **41**: 602-607. <https://doi.org/10.1111/j.1550-7408.1994.tb01522.x>
- Roberts, R.J., 2012. *The parasitology of teleost*. In: *Fish pathology* (eds. R.J. Roberts). Blackwell Publishing Ltd. West Sussex, UK. pp. 339-381. <https://doi.org/10.1002/9781118222942.ch8>
- Rushton-Mellor, S.K., 1992. Discovery of the fish louse, *Argulus japonicus* Thiele (Crustacea: Branchiura), in Britain. *Rev. Fish. Sci. Aquac.* **23**: 269-271. <https://doi.org/10.1111/j.1365-2109.1992.tb00618.x>
- Saha, S.K., Guha, A., and Banerjee, A., 2011. Feeding apparatus and associated glands in the freshwater fish ectoparasite *Argulus siamensis* Wilson, 1926 (Branchiura). *Crustaceana*, **84**: 1153. <https://doi.org/10.1163/156854011X587469>
- Sahoo, P.K., Mohanty, J., Garnayak, S.K., Mohanty, B.R., Kar, B., Prasanth, H., and Jena, J.K., 2013a. Estimation of loss due to argulosis in carp culture ponds in India. *Indian J. Fish.*, **60**: 99-102.
- Sahoo, P.K., Mohanty, J., Garnayak, S.K., Mohanty, B.R., Kar, B., Jena, J.K., and Prasanth, H., 2013b. Genetic diversity and species identification of *Argulus* parasites collected from major aquaculture regions of India using RAPD-PCR. *Aquac. Res.*, **44**: 220-230. <https://doi.org/10.1111/j.1365-2109.2011.03025.x>
- Shafir, A. and Van-As, J.G., 1985. The opportunistic nature of *Argulus japonicus* in maintaining host-parasite relationships. *S. Afr. J. Sci.* **81**: 638-638.
- Shafir, A. and Van As, J.G., 1986. Laying, development

- and hatching of eggs of the fish ectoparasite *Argulus japonicus* (Crustacea: Branchiura). *J. Zool.*, **210**: 401-413. <https://doi.org/10.1111/j.1469-7998.1986.tb03645.x>
- Shafir, A., and Oldewage, W.H., 1992. Dynamics of a fish ectoparasite population: opportunistic parasitism in *Argulus japonicus* (Branchiura). *Crustaceana*, **62**: 50-64. <https://doi.org/10.1163/156854092X00046>
- Singhal, R.N., Jeet, S., and Davies, R.W., 1990. The effects of argulosis-saprolegniasis on the growth and production of *Cyprinus carpio*. *Hydrobiologia*, **202**: 27-31. <https://doi.org/10.1007/BF00027090>
- Steckler, N., and Yanong, R.P., 2012. *Argulus* (fish louse) infections in fish. Fisheries and Aquatic Sciences Publications. Florida: University of Florida, pp. 1-4. <https://edis.ifas.ufl.edu/fa184>. Cited Sep 2020.
- Taylor, N.G.H., Sommerville, C., and Wootten, R., 2006. The epidemiology of *Argulus* spp. (Crustacea: Branchiura) infections in still water trout fisheries. *J. Fish Dis.*, **29**: 193-200. <https://doi.org/10.1111/j.1365-2761.2006.00704.x>
- William, J.P., 2008. Global diversity of fish lice (Crustacea: Branchiura: Argulidae) in freshwater. *Hydrobiology*, **595**: 209-212. <https://doi.org/10.1007/s10750-007-9015-3>