SHORT COMMUNICATION

Lernaea cyprinacea (Copepoda, Lernaeidae) Infection on *Glossogobius aureus* (Gobiiformes, Gobiidae) from Naujan Lake under Captive Conditions

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ABSTRACT

Lernaea cyprinacea, a parasitic cyclopoid copepod, is found to parasitize Naujan White Goby, *Glossogobius aureus* (TL = 8–23 cm, BW = 8.6–101 g) maintained in concrete tanks for broodstock development. The infected fish displayed flashing behavior, localized redness, erosion of pectoral, dorsal, and caudal fins, and inflammation of the vent. Red sores were also found on the skin, buccal cavity, and muscle tissue. Sexually mature female *G. aureus* were seen to be more susceptible to *L. cyprinacea* than mature males. *Lernaea* infection was successfully controlled by a salt dip at 10 ppt for 2–3 minutes followed by the salt bath at 2–3 ppt for 21 days. This is the first record of *L. cyprinacea* infection in captive *G. aureus* in the Philippines.

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Lernaeids, often known as "anchor worms," are parasitic copepods with broad distribution due to their lack of host specificity. *Lernaea cyprinacea* is one of the most extensively studied species identified to infect over 100 fish species (Steckler and Yanong 2012; Hossain et al. 2018). This parasite infests both introduced and native species (Waicheim et al. 2018; Innal et al. 2021). In the Philippines, *L. cyprinacea* has been reported to infect *Oreochromis niloticus, Anabas testudineus, Therapon plumbeus*, *Chanos chanos* (Lopez 2001), *Carassius auratus, Channa striata, Glosogobius biocellatus* and *G. guiris* (Arthur and Lumanlan-Mayo 1997). Infections in aquaculture have been reported among ornamental fishes (Trujillo-Gonzales 2018; Vagasam et al. 2006) as well as among commercially cultured food fishes (Rojas and Wadsworth 2007; Bozorgnia et al. 2018; Bilal et al. 2014; Hidayatullah et al. 2020; Halwart et al. 2007).

Lernaeid infection has been reported among wild-caught *G. giuris* in the Philippines (Lopez and Palisoc 1992) and in other tropical (Narciso et al. 2019) and sub-tropical countries (Waichaem et al. 2019). In Malaysia, wild-caught broodstock of marble goby (*Oxyelotris mormorata*) had been reported to be infested by *L. cyprinacea* (Idris and Amba 2011). Lopez and Palisoc (1992) documented a prevalence rate of 25% and a mean intensity of 1.3 for *L. cyprinacea* on wild-caught *G. giuris* from Naujan Lake. However, there has been no report of this parasite occurring among *G. aureus* from a captive environment. This paper presents a case of *L. cyprinacea* infection on adult Naujan White Goby, *Glossogobius aureus*, captured from Naujan Lake and maintained for broodstock development under hatchery conditions, and outlines the treatments conducted to manage the parasite.

Live G. aureus were collected from Naujan Lake (13° 10' 20.424" N, 121° 21' 1.584" E), with sizes ranging from 8 cm to 23 cm in total length (TL) and 8.6–101 g body weight from March to August 2022 (Table 1). The fish samples were visually checked externally for parasites on the skin, nostrils, gills, and base of the fins immediately after collection. Only uninfected fish were transported in oxygenated plastic bags to the fish hatchery of the Bureau of Fisheries and Aquatic Resources (BFAR) Regional Field Office 4B located within the Mindoro State University (MinSU) Bongabong Campus (12° 46' 14" N, 121° 28' 34" E) for conditioning. The collected fish were maintained for broodstock development and captive breeding in three concrete freshwater tanks of 4-ton capacity at 25–30 fish per tank. Male and female fish

Table 1. Prevalence rate of *Lernaea cyprinacea* on wild *Glossogobius aureus* broodstock collected from Naujan Lake.

Month	No. of broodstock	No. of infected	Prevalence %	Ambient temp °C *
March	59	2	3	27.99
April	56	20	36	29.12
May	50	38	76	30.68
June	22	2	9	30.6
July	10	0	0	29.91
August	10	3	30	30.22

*www.pagasa.org.ph/weather/heat-index. Accessed on April 4, 2022.

were kept in separate tanks and fed a mixed diet of commercial feeds, trash fish, and live earthworms. Fish identification was based on the arrangement of pit organs and sensory papillae, as described by Akihito and Meguro (1975), Abdulmalik-Labe et al. (2022), and Basa et al. (2024).

The presence of adult lernaid parasites was noted on some individuals two weeks after stocking. Initially, the parasites were removed from the fish body using forceps and preserved in 70% ethyl alcohol for morphological identification by light microscopy following Kabata (1970), Kabata (1985), and Demaree (1967). Skin scrapings from dead and moribund fish were also taken to enable microscopic observations of other parasitic stages of *L. cyprinacea*. Currently, the cephalic anchors, known as "holdfast" or "horns" of metamorphosed females, have been used to identify *Lernaea* species. The prevalence rate (Pr, %) of this lernaeid parasite on *G. aureus* was determined as the percentage of fish infected with a parasite species related to the total number of fish examined (Bush et al. 1997).

Infected fish were used to test the effectivity of two therapeutic controls to manage *L. cyprinacea* infection in *G. aureus*: Treatment 1- bathing infected fish in 2.5 ppt *Methylene blue* overnight as per manufacturer's instruction; and Treatment 2 - dipping of infected fish in 10 ppt water for 2–3 minutes followed by bathing in 2–3 ppt water for 21 days (modified from Kabata, 1985). Treatment 3 served as a control without any therapeutant. All treatments involved three infected fish with 7–15 adult *L. cyprinacea* attached to their skin. Test animals were randomly stocked in glass aquaria (70 cm x 40 cm x 40 cm) and replicated thrice. Fish were fed with earthworm (African Night Crawler) at 3% body weight.

The lernaeid parasites from the infected *G. aureus* are characterized by well-developed dorsal and ventral pairs of branches. The anchors are cylindrical, with Y-shaped or T-shaped processes on the dorsal pair and a smaller, simpler ventral pair typical of *L. cyprinacea* (Figure 1). Adult *L. cyprinacea* was observed visually from individuals being conditioned 11 days after collection (Figure 2A). It was hypothesized

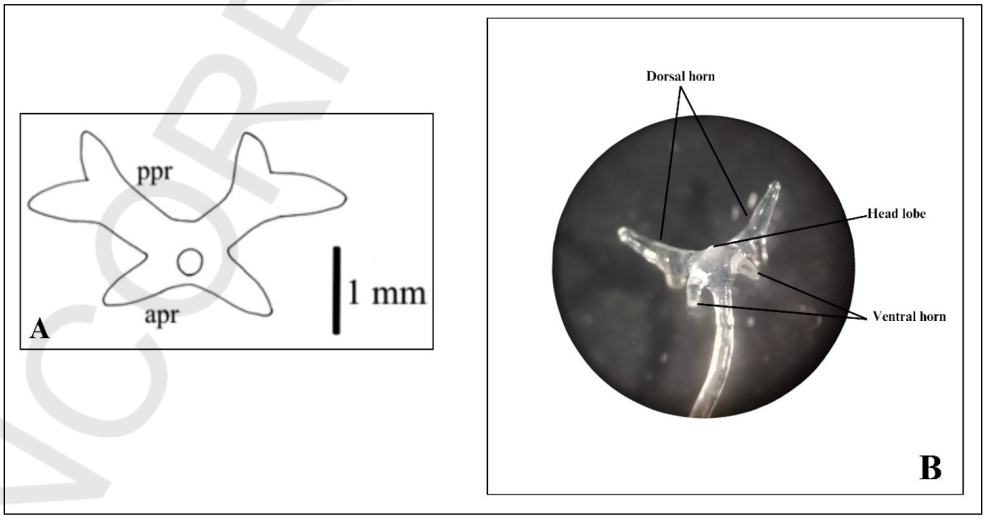

Figure 1. (A) Cephalic structure of *Lernaea cyprinacea* modified from Demaree (1967): ppr = posterior protuberance; apr = anterior protuberance; (B) Ventral view of the anchor shape of *L. cyprinacea* from the skin surface of *Glossogobius aureus* from Naujan Lake (4x magnification).

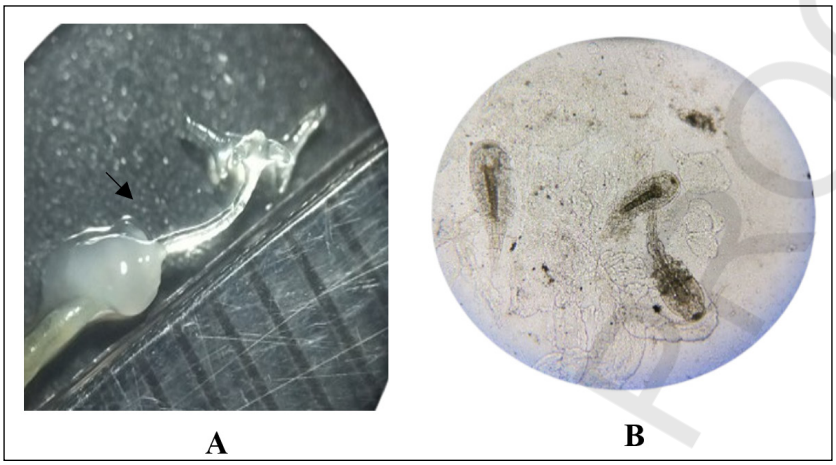

Figure 2. Anchor worm, *Lernaea cyprinacea* collected from *Glossogobius aureus* viewed under Kern dissecting microscope OSE-4. (A) anchor worm of 0.9 mm in length with mucus on the swimming leg (arrow). (B) copepodid individuals (infective stage) identified from scraped samples from the skin of *G. aureus* (4x magnification).

that some of the breeders were already infected in their natural habitat by the microscopic copepodid (infective stage) of *L. cyprinacea* (Shields 1978) based on the copepodids of *L. cyprincea* found in the scraped samples from the skin of the fish (Figure 2B). The larvae attached to the host's skin surface may have metamorphosed into an adult *L. cyprinacea* under favorable conditions. The parasite thrives at elevated temperatures, with optimal development between 23–30°C (Baur 1962). Lernaeid specimens collected were stored in 70% ethyl alcohol for further study.

The parasite's 3% prevalence rate was noted in March 2022 (Table 1). Subsequent collection of broodstocks showed a higher prevalence of infection by L. cyprinacea, which was collected in April, May, and August. The higher parasite infection of the collected broodstocks may have been promoted by low water quality from uneaten commercial feed (weaning diet), making fish more vulnerable to infections. A higher percentage of female G. aureus were infected, with a 65% prevalence compared to 31% in male G. aureus. A significant increase in susceptibility, characterized by high parasite load and mortality, was observed in sexually mature female trout (Currie and Woo 2007). It was hypothesized that 17β-estradiol, the major female reproductive hormone during spawning, may be a contributing factor in increasing or promoting parasite proliferation in females. The findings of this study suggest that lernaeosis can decrease the reproductive ability of the broodstocks if not controlled.

Severe damage to fish tissue and health was observed to have a fatal effect of *L. cyprincea* on the fish. The attachment sites were distinguished by visible reddening and swollen margins (Figure 3A). Excessive mucus secretion was observed on the

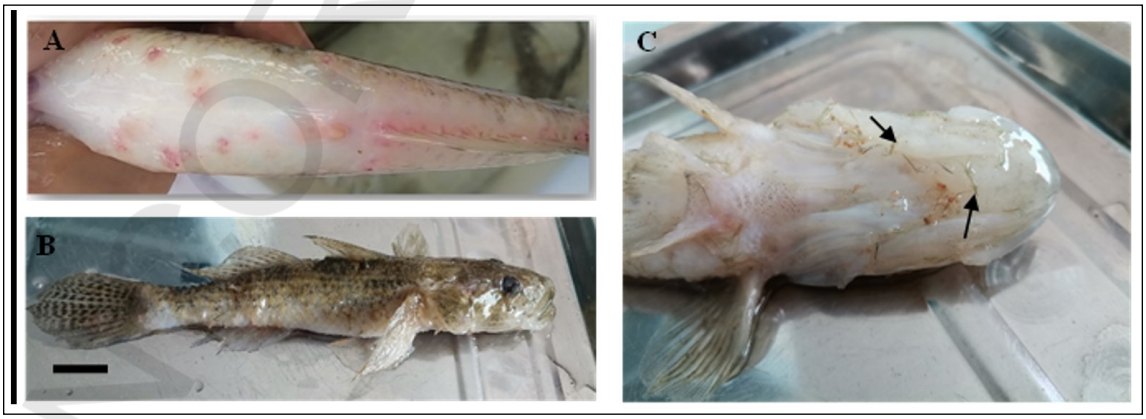

Figure 3. (A) Lesions on the abdominal part of adult *G. aureus* characterized as red spots caused by *L. cyprinacea*. (B) moribund *G. aureus* with excessive mucus secretions as the first line of defense against *L. cyprinacea* infection (scale bar = 1 cm). (C) adult *L. cyprinacea* attached to a dead *G. aureus* (arrow).

body of the infected fish, forming a physical barrier against *L. cyprinacea* infection (Figure 3B). *The lernaeid parasite* was first seen on the dorsal fin, and the infection then spread to the caudal fin, pelvic fin, head, and other muscular body parts (Figure 3C). The infected fish also had pectoral, dorsal, and caudal fin erosion, vent inflammation, and open wounds.

Infested *G. aureus* were subjected to methylene blue and salt bath treatments. Control of the parasite was effective by dipping the infected fish in 10 ppt water for 2–3 minutes and transferring it to 2–3 ppt water for 21 days. After treatment, nine individuals (100%) treated with salt dip and bath, and six individuals (67%) treated with methylene blue survived, with zero adult parasites on the external body surface. However, a 100% mortality rate was recorded in the control group, with one individual having more than 30 adult *L. cyprinacea* anchored on the fish's body. Khalifa and Post (1976) found tissue debris and erythrocytes in the alimentary canal following *L. cyprinacea* infestation, indicating that the parasite feeds on both tissue and blood. This feeding may have caused the weakening and eventual mortality of *G. aureus* observed in this study. It should be noted that the salinity applied in this study is lower than the optimum salinity treatment (10 ppt) for *L. cyprinacea* used by Idris and Amba (2011) over 21 days.

The need to therapeutically treat wild-sourced broodstock of *G. aureus* in 10 ppt for 2–3 minutes and hold them in 2–3 ppt for 21 days is emphasized to control possible infection. The collection of broodstock for aquaculture purposes is suggested to be done after the warm months, particularly in July. Further research is needed to elucidate the interactions between *G. aureus* and *L. cyprinacea*, focusing on tissue, cellular structures, and molecular mechanisms. Understanding these factors is crucial in developing protocols for proper diagnosis, treatment, and prevention against lernaeosis.

This is the first record of *L. cyprinacea* infection on wild-caught *G. aureus* in captivity in the Philippines.

AUTHOR CONTRIBUTIONS

Balagtas AF: Investigation and Data Curation. **Guibone EY:** Supervision, Data Curation, Visual Presentation, and Writing - Original draft. **Sanchez-Escalona KP:** Conceptualization, Data Curation, Writing - Review and editing, Project Administration, and Funding Acquisition.

CONFLICTS OF INTEREST

We declare no conflict of interest among the researchers.

ETHICS STATEMENT

The researchers followed all institutional and national guidelines for the care and use of laboratory animals.

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