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## Parasites of wild glass eels and cultured elvers of the giant mottled eel (*Anguilla marmorata* Quoy and Gaimard, 1824) in Vietnam

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**Abstract:** This article reports a survey of parasites in wild glass eels following them through culture to elvers in over-flow and re-circulating water systems in Vietnam. The results show that wild glass eels are infected with three parasitic species (*Trichodina* sp., *Ichthyophthirius multifiliis* and *Pseudodactylogyrus anguillae*). Eels in re-circulating systems are infected with three parasites (*Trichodina acuta*, *Trichodina* sp., and *Pseudodactylogyrus anguillae*). Seven parasitic species were found on/in eels in the over-flow system (*T. acuta*, *Trichodina* sp., *I. multifiliis*, *P. anguillae*, *Pseudodactylogyrus bini*, *Centrocestus formosanus* (metacercaria), and, *Haplorchis taichui* (metacercaria). Two parasites, *Trichodina* sp., and *P. anguillae*, were found on wild glass eels and elvers in both rearing systems. *T. acuta* had the highest prevalence (57.3%) in the recirculating system. *P. bini* had the highest prevalence (56.1%) in the over-flow system. The prevalence of *Trichodina* sp. infected fish in the recirculating system was 39.6%. That of *C. formosanus* (metacercaria) was 10.6% in the over-flow system. The prevalence of other parasites in all habitats was less than 10.0%. Two out of 3 parasites found in the re-circulating system had higher prevalence than those in the over-flow system. Results show that elvers reared in over-flow water systems had more parasites than wild glass eels or those reared in recirculating water system.

**Key words:** eel culture, *Anguilla marmorata*, parasites, Vietnam

### Introduction

The glass eel is an early developmental stage in the eels. The giant mottled eel, *Anguilla marmorata* Quoy & Gaimard, 1824, has the widest distribution among anguillid eels, occurring throughout almost the entire Indo-Pacific area (Tesch, 2003). Like other anguillids, adults live in freshwater or estuarine habitats,

are nocturnal, and feed on a wide range of prey, especially crabs, fish and frogs (Skelton, 1993). Adult giant mottled eels spawn offshore and when the eggs hatch a larva called a leptocephalus emerges. The leptocephali spend approximately 114-132 days drifting in the plankton before migrating to river mouths as

glass eels (Arai *et al.*, 2002), where they undergo metamorphosis into elvers in the freshwater.

The giant mottled eel has been widely cultured ('grown-out') in Vietnam for many years. However, there is no artificial breeding technology available at present for the production of glass eels. Therefore, Vietnamese eel culture has to rely on wild-caught glass eels from the Ba River in Phu Yen Province in the South Centre of Vietnam (Cong *et al.*, 2012). In the Ba River, glass-eels swim upstream during the night towards a dam as part of their migration, the water temperature in the river varies from 18-24°C. Close to the dam, local people use scoop nets made of mosquito netting to catch wild glass eels and keep them in clean, fresh water at a temperature of 18–20°C before selling them to fish farmers (Cong *et al.*, 2012).

At the farms, the glass eels are grown in either cement or composite tanks for about 3–6 months before being sold for grow-out as elvers. In Khanh Hoa Province, the rearing systems consist of an over-flow system using water from a reservoir. The water is filtered through sand and flows into a cement tank system, where the eels are reared. Each tank has a volume of 2 m<sup>3</sup>. Water continuously passes through the rearing systems with a constant flow rate which is sufficient to change 100% of the water within 4 hours. The temperature varies between 19–27°C. In Lam

Dong Province, the rearing systems use fiber glass tanks with re-circulated water. Original water is filtered through sand, then through a bio-filter before re-use. The system can replace 100% water in rearing tanks within 2 hours. Temperature in the water varies between 13–18°C.

Recently, eel production has declined for unknown reasons. In an attempt to determine if parasites could be a contributing factor, this study was designed to compare parasite prevalence in glass eels collected from the wild in Phu Yen Province and elvers from the over-flow and re-circulating systems in Khanh Hoa and Lam Dong Provinces, respectively.

## **Materials and Methods**

### **Sources of glass eels and elvers**

Wild glass eels were caught in the mouth of Ba River in Phu Yen Province. Then move to Khanh Hoa and Lam Dong Provinces for rearing. The majority of the glass eels were bathed with formalin (30 ppm) to treat for ectoparasites before being introduced into the rearing systems. They were in rearing systems for about 1 to 2 months before sampling for parasite examination. Subsamples of the glass eels packed in nylon bags inflated with oxygen and filled with clean freshwater and transported to the Research Institute for Aquaculture No.3 for parasite examination. The water occupied 1/3 of the bag volume and temperature in the bags varied between 18–22°C.

Elvers were collected from the respective rearing systems in Khanh Hoa and Lam Dong Provinces. They were transported from the collection sites to the Research Institute for Aquaculture No. 3 (RIA3) in Nha Trang City, Khanh Hoa Province, in the same manner as the glass eels above.

At RIA3, both glass eels and elvers were

kept in 3 m<sup>3</sup> cement tanks before being examined for parasites within 24 hours of arrival. The tanks were constantly supplied with clean, fresh water and 15–20 fish collected at a time to avoid crowding in the tanks. Table 1 shows the number, length, and weight of wild glass eels and reared elvers collected from different sites.

**Tab. 1: Number, length and weight of wild glass eels and elvers collected from different sites.**

Sampling sites	Fish number (specimens)	Mean length ± SD (Range)	Mean weight ± SD (Range)
Phu Yen (Wild glass eel)	57	48.4 ± 3.5 (44.0-58.0)	0.13 ± 0.05 (0.10-0.20)
Khanh Hoa (Elvers)	66	57.4 ± 4.9 (45.0-65.0)	0.25 ± 0.07 (0.10-0.32)
Lam Dong (Elvers)	96	55.2 ± 6.3 (45.0-63.0)	0.24 ± 0.08 (0.10-0.30)

### Parasite examination and identification

Each eel was scraped to collect external mucus. In addition, they were dissected, the gills, liver, gall bladder and viscera and examined for the presence of parasites. All parasites found were fixed following the methodologies of Lom and Dykova (1991) and Berland (2005). They were then photographed, drawn and measured for aid in identification.

### Data analysis

Standard parasitological parameters follow-

ed the recommendations in Bush *et al.* (1997). The prevalence of each parasitic species in each host group was compared between different sites using a Chi-square test with Yates correction being employed if 5 or more fish were infected. When less than 5 fish were infected, the Fisher-exact test (two tails) was used, significance level of  $\alpha = 0.05$ .

### Results

Prevalence of parasites from wild glass eels, and elvers from over-flow and re-circulating

systems is presented in Table 2. Three parasitic species infected wild glass eels, 7 parasitic species were found from elvers in over-flow systems (Khanh Hoa Province) and 3 parasitic species found from elvers in re-circulating systems (Lam Dong Province). The prevalence of *T. acuta* was 57.3% in re-circulating systems, and 33.3% in over-flow systems. The prevalence of *Trichodina* sp. was 39.6% of elvers in re-circulating systems, 1.5% of elvers in over-flow systems and 1.7% in wild glass eels. The prevalence of *P. bini* was 56.1% in

over-flow systems, while this parasite was not found in either elvers from re-circulating systems or wild glass eels. *P. anguillae* was found in both over-flow and re-circulating systems, with a prevalence of 7.6% and 6.3 %, respectively. *Ichthyophthirius multifiliis* and metacercariae of *C. formosanus* were found only in over-flow systems, at prevalence of 3.0% and 10.6%, respectively. Metacercariae of *H. taichui* were found in elvers from over-flow systems with a prevalence of 9.0%.

**Tab. 2: Prevalence of parasites from wild glass eels, and elvers from over-flow and re-circulating systems.**

Parasites	Number fish infected/number fish examined (%)		
	Wild	Over-flow system	Re-circulating system
<i>Trichodina acuta</i>	0/57 (0.0)	22/66 (33.3)	55/96 (57.3)
<i>Trichodina</i> sp.	1/57 (1.7)	1/66 (1.5)	38/96 (39.6)
<i>Ichthyophthirius multifiliis</i>	2/57 (3.4)	2/66 (3.0)	0/96 (0.0)
<i>Pseudodactylogyrus anguillae</i>	1/57 (1.7)	5/66 (7.6)	6/96 (6.3)
<i>Pseudodactylogyrus bini</i>	0/57 (0.0)	37/66 (56.1)	0/96 (0.0)
<i>Centrocestus formosanus</i> (Metacercariae)	0/57 (0.0)	7/66 (10.6)	0/96 (0.0)
<i>Haplorchis taichui</i> (Metacercariae)	0/57 (0.0)	6/66 (9.0)	0/96 (0.0)

Table 3 shows statistical comparison of the prevalence of the parasites found among glass eels and elvers. Results of comparison between wild glass eels (Phu Yen Province) and elvers from over-flow systems (Khanh Hoa Province) showed that the prevalences of *T. acuta*, *P.*

*bini*, *C. formosanus*, and metacercariae of *H. taichui* were statistically significantly higher on elvers ( $P < 0.05$ ), while the prevalence of *Trichodina* sp., *I. multifiliis* and *P. anguillae* were not statistically significantly different between eels from the two sites.

**Tab. 3: Statistical comparison of the prevalence of the parasites found among glass eels and elvers.**

Parasites	Wild (Phu Yen) vs.	Wild (Phu Yen) vs.	Over-flow (Khanh Hoa) vs.
	over-flow	re-circulating	re-circulating
	(Khanh Hoa)	(Lam Dong)	(Lam Dong)
	P value	P value	P value
<i>Trichodina acuta</i>	<0.01	<0.01	0.09
<i>Trichodina</i> sp.	1.00	<0.01	<0.01
<i>Ichthyophthirius multifiliis</i>	1.00	<0.01	0.17
<i>Pseudodactylogyrus anguillae</i>	0.21	0.25	0.76
<i>Pseudodactylogyrus bini</i>	<0.01		<0.01
<i>Centrocestus formosanus</i> (Metacercariae)	0.01		<0.01
<i>Haplorchis taichui</i> (Metacercariae)	0.03		<0.01

Results of comparison between wild glass eels (Phu Yen Province) and elvers from re-circulating systems (Lam Dong Province) showed that the prevalence of three parasitise (*T. acuta*, *Trichodina* sp., and *Ichthyophthirius multifiliis*) were statistically significantly higher in elvers. The prevalence of *P. anguillae* was not statistically different between wild glass eels and elvers from re-circulating systems.

The results show that there are statistically significant differences between prevalence of 4 parasitise from over-flow systems and re-circulating systems. The prevalence of *Trichodina* sp. was higher in elvers from re-circulating systems, while that of *C. formosanus*, *P. bini* and *H. taichui* were higher in elvers from over-flow systems. Prevalences of other parasites were not statistically significantly different.

## Discussion

## The parasite composition

The parasites of eels have been studied comprehensively in Europe, while in Vietnam, only two references mention parasites of eels (Arthur and Bui, 2006; Ha and Bui, 2007). These documents report 4 parasitic species: 1 nematode, 1 monogenean, and 2 protozoa. This paper finds three parasites on wild glass eels, and, 7 and 3 parasitise from elvers in over-flow systems and re-circulating systems, respectively. Sasal *et al.* (2008) reported 8 taxa of parasites found from giant mottled eel on the Island of Reunion, including *Pseudodactylogyrus anguillae*, *P. bini*, *Anguillicoloides crassus*, *Paraquimperia africana*, *Acanthocephalus reunionensis*, *Bothriocephalus claviceps*, and, cestode and nematode larvae. Only two of these parasites (*P. anguillae* and *P. bini*) were found in this study.

*Trichodina acuta* is a non-host specific ciliate found worldwide on various fish species such as

*Cyprinus carpio*, *Carassius auratus*, *Oncorhynchus mykiss*, *Salmo trutta*, *Phoxinus phoxinus* (Gaze and Wootten, 1998); *Anguilla anguilla* (Imai et al., 1991). In Vietnam, this species has been reported from *Pangasius micronemus*, *P. hypophthalmus*, *Cirrhina mrigala*, *Labeo rohita*, *Cyprinus carpio*, *Oreochromis niloticus niloticus*, *Ctenopharyngodon idellus* (Arthur and Bui, 2006; Ha and Bui, 2007), this is the first record on *A. marmorata*.

*Ichthyophthirius multifiliis* is a common disease causing ciliate of many freshwater fish species of various areas in the world (Lom and Dyková, 1992). Ha and Bui (2007) reported it from *A. marmorata* in Vietnam.

*Pseudodactylogyrus anguillae* was originally described from the Japanese eel *Anguillae japonica* as *Dactylogyrus bini* Kikuchi (1929), after several systematic changes, it finally became *Pseudodactylogyrus anguillae*. Today, this monogenea is reported from various species of eel. Marcogliese and Cone (1993) reported it from the American eel *Anguilla rostrata*. Sasal et al. (2008) reported it from *Anguilla mossambica* and *A. marmorata* from Reunion Island. Ha and Bui (2007) reported it from *Oxyeleotris marmoratus*, *Anguilla japonica* and *A. marmorata* in Viet Nam.

*Pseudodactylogyrus bini* was originally described from the Japanese eel *Anguillae japonica* as *Dactylogyrus bini* Kikuchi (1929), Gusev (1965) re-described it as

*Pseudodactylogyrus bini*. This species was later found from different eels in various areas, Hayward et al. (2001) reported it from the American eel *Anguilla rostrata* in North America. Ogawa and Egusa (1976) reported it from the European eel *A. anguillae*. This is the first record of this species in Viet Nam, and the first record on *A. marmorata*.

*Centrocestus formosanus* is a zoonotic parasite found in many freshwater fish species world-wide. In Vietnam, metacercariae of this species are found in *Cyprinus carpio*, *Clarias fuscus*, *C. macrocephalus*, *C. batrachus*, *Hypophthalmichthis harmandii*, *Barbodes gonionotus*, *Monopterus albus*, *Labeo rohita*, *Cirrhinus mrigala*, *Catla catla*, *Osphronemus gorami*, *Oreochromis niloticus*, *Ctenopharyngodon idellus*, *Colossoma macropomum* (Ha and Bui, 2007); in *Aristichthys nobilis*, *Cyprinus carpio*, *Ctenopharyngodon idellus*, *Cirrhinus mrigala*, *Labeo rohita*, *Hypophthalmichthis molitrix*, *Oreochromis niloticus* (Chi et al., 2008); and, in *Pangasiannodon hypophthalmus*, *Anabass testudineus*, *Ctenopharyngodon idellus*, *Cyprinus carpio* (Thien et al., 2007). This is the first record from *A. marmorata*.

*Haplorchis taichui* is a zoonotic trematode. It has been intensively studied in many countries. Its metacercariae have been confirmed to have a wide range of fish hosts and wide geographical distribution, and have been reported in different fish species in Vietnam. Ha

and Bui (2007) reported them from Silver carp (*Hypophthalmichthys harmandi*), Grass carp (*Ctenopharyngodon idellus*), Common carp (*Ciprinus carpio*), Climbing perch (*Anabas testudineus*). Thien *et al.* (2009) found them in Giant gourami (*Osphronemus gourami*), Kissing gourami (*Helostoma temmincki*). Adults have been found in dog, cat and humans in Vietnam, Taiwan, The Philippines, Bangladesh, Palestine, China, Thailand (Chai *et al.*, 2005). Dung *et al.* (2007) confirmed its presence in human in Vietnam. This is the first record from *A. marmorata*.

#### **The prevalence of parasites of eels from different sites**

Several studies have reported different infection of fish with *Trichodina* spp. due to different habitat conditions. Madsen *et al.* (2000) showed that eels reared in water that had high contents of organic parameters, low concentration of nitrate and high pH suffered more problems with *Trichodina jadranica* than eels in water that had low contents of organic parameters, high concentration of nitrate and low pH. Ogut and Palm (2005) showed that the prevalence of *Trichodina* spp. on the gills of Whiting (*Merlangius merlangus*) varied seasonally; and, that its variation correlated with the concentration of nitrite, nitrate, phosphate, oxygen, and, water temperature.

Results of this study show that in 11 out of 18 cases there are significant differences ( $P <$

0.05) between parasite prevalence from eels of 3 different systems and 7 cases with no significant difference found.

The fish in this study collected from different systems, which could have had different organic and inorganic contents, for ex. some organic contents could be higher in over-flow systems than those in re-circulating systems. This might be one of the reasons for the differences of prevalences. The results show that wild glass eels were infected with 3 species at low prevalence (1.7 – 3.4 %), while elvers in re-circulating systems were also infected with 3 species, but at much higher prevalence (6.3 – 57.3 %). Elvers in overflow systems were infected with 7 species and prevalence varied between 1.5 and 56.1 %. The wild glass eels had recently entered the river mouth, where they encountered a sudden reduction in salinity; this probably reduced any ectoparasites that they brought from the sea. They had not been long enough in the brackish/fresh water to be infected with other parasites. Both factors may help explain why the wild glass eels were infected with only 3 species at low prevalence.

There were no statistically significant differences ( $P > 0.05$ ) between the prevalence of wild glass eels and elvers from over-flow systems infected with the same parasitic species (*Trichodina* sp., *I. multifilis*, and *P. anguillae*). The over-flow systems use a reservoir water source run through a sand filter, thus many environmental parameters might be

similar to those in the habitat where the wild glass eels had been collected.

There was no statistically significant difference in prevalence of *P. anguillae* of wild glass eels and elvers from re-circulating systems. This parasite is not commonly found in Vietnam, and its prevalence was very low on the fish collected from the two systems, this could make statistical detection differences difficult, or, there may truly be no statistically significant differences in this case.

Results also show that *Trichodina* sp. prevalence was not statistically significant different between wild glass eels in Phu Yen and elvers from Khanh Hoa Province; in these two provinces, water temperatures are almost identical. Comparison between elvers from Lam Dong and Khanh Hoa, as well as between elvers from Lam Dong and wild glass eels from Phu Yen, demonstrated statistically significant differences in both cases. Lam Dong has lower water temperatures than the other two provinces. Thus temperature might have important effect on the prevalence of parasites in eels in this case.

-Both wild glass eels and elvers were infected with parasites. Elvers reared in over-flow systems were infected with significantly more parasitic species than wild glass eels or elvers reared from re-circulating systems.

-Elvers reared in over-flow water systems had higher prevalences of parasites than wild glass eels or elvers reared in re-circulating water

systems.

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