

**Snail composition and its cercariae in rice field of Hoc Mon district,
Ho Chi Minh City, Vietnam**

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ABSTRACT

The research on snail composition and their cercariae by morphological analysis method in Xuan Thoi Thuong rice field of Hoc Mon district, Ho Chi Minh City, Vietnam was carried out in the wet season of August 2022 and the dry season of February 2023. A total of 993 snails were collected and 11 snail species belonging to 9 genera and 5 families were classified. There were 9 snail species collected in the wet season and 7 snail species found in the dry season. *Lymnaea viridis* and *Bithynia siamensis* were infected with trematode (cercariae stage) with the combined prevalence of 14.3% and 4.1%, respectively. The other nine snail species had cercariae free. Two cercariae morphotypes were discovered from snails including *Xiphidio cercariae* and *Echinostome cercariae*. More research on snails and their cercariae in other waterbodies should be done in Hoc Mon district and other places to identify the snail diversity and sources of trematodes affecting fish culture and human health.

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1. Introduction

Hoc Mon is a suburb district of Ho Chi Minh City where rice cultivation still developed well with the total rice field area of 1,583.80 ha (PCHCMC, 2020). Pham et al. (2019) found that silver barb from fishing in Thay Cai - An Ha canal in Hoc Mon district was infected with metacercariae of *Haplorchis pumilio* with the prevalence of 80.0%. The interview's information from the farmers was that all silver barb were mainly in rice fields where snails were available and just followed the water stream to get into the canal. The source of metacercariae in silver barb was questioned whether cercariae existed in snails in rice field and infected or not.

The study on trematode is necessary because foodborne trematodiasis is an emerging public health problem (Keiser & Utzinger, 2005). Little is known about the clinical importance of infections with minute intestinal flukes, but heavy infections can cause serious gastrointestinal symptoms (Nawa et al., 2005). Infections have significant human-health impact and cause substantial clinical or subclinical disease for small liver flukes (Dorny et al., 2009). The life cycles of intestinal flukes and liver flukes are similar (Yu & Xu, 2005). The heterophyid digeneans have a three-host life cycle involving snails as the first intermediate host, fish as the second intermediate host, and fish-eating animals and humans as the definitive host (Elsheikha & Elshazly, 2008).

In the natural condition, cercariae are released from snails then swim freely in the water and usually attract to the second intermediate hosts like fish, crabs, aquatic plants, etc (Thai, 2016). In a shedding experiment, cercariae emerge from infected snails during day and night, but more commonly in the morning and before noon. They are usually found in the lower half of a container with water (Saad, 1994). Cercariae remain active for 12 - 24 h and most die after 72 h. For cercariae

of small liver flukes and intestinal flukes, they are free in the water, attract to fish and penetrate the fish's tissue within 5 - 10 min (Long-Qi et al., 2005).

The snail intermediate hosts for the heterophyid trematode species are primarily species of the Thiaridae and Bithyniidae (Madsen & Hung, 2014). *Melanoides tuberculata*, *Thiara* and *Terabia granifera* are the first intermediate hosts of heterophyids (Waikagul & Radomyos, 2005). *Melanoides tuberculata* is the host of *Haplorchis pumilio* (Khalifa et al. 1977; Wang et al., 2002; Dechruksa et al., 2007) and *Thiara granifera* was found commonly infected with *H. pumilio* in Taiwan (Wang et al., 2002).

Bui et al. (2010) stated that there were 10 snail species in rice fields in Nam Dinh province, and most of snails in the families of Bithyniidae, Stenothyridae and Planorbidae dominated in rice fields. Nguyen et al. (2014) carried out research in An Hoa rice field of Tuy An district, Phu Yen province and found 9 snail species and *Melanoides tuberculata*, *Sermyla tornatella* and *Bithynia* sp. were found infected. In rice fields of Binh Khanh and Ly Nhon communes, Can Gio district of Ho Chi Minh city, Nguyen & Pham (2022) collected 8 snail species and discovered cercariae from *Bithynia* sp. and *Melanoides tuberculata* including xiphidio cercariae, furcocercous cercariae and pleurolophocercous cercariae. If snails in rice field of Hoc Mon district have pleurolophocercous cercariae, one of the questions for metacercariae in silver barb in this district can be answered. Therefore, the research on cercariae from snails in rice fields in Hoc Mon district needed to be implemented.

2. Material and Methods

2.1. Study areas

Xuan Thoi Thuong rice field was chosen to carry out the research because it had the largest

area of rice field in Hoc Mon district of Ho Chi Minh City (PCHCMC, 2020).

2.2. Sampling of snails

Two cross-sectional studies on snails were carried out in August 2022 (the wet season) and in February 2023 (the dry season). Snail sampling was done by using hand nets and hands with gloves to collect snails in the standard cell of 0.4 m wide x 10 m long x 0.1 m deep along the bank of rice field. Rice fields were in the rice-seedling stage in August 2022 and already harvested in February 2023. Fifteen samples per season were collected in each rice field with 500 m between the two sampling sites. The collected snails were washed and transferred to cloth bags and transported to the laboratory to analyze. Snails were classified into species following the keys of Dang et al. (1980).

2.3. Examination of snails for cercariae

The shedding method (Frandsen & Christensen, 1984; Bui et al., 2010) was used to examine for trematode infection (cercariae stage). Each snail was kept separately in 200 mL beakers and left for 24 h for shedding. Cercariae were checked twice per day at 8:00 AM and 14:00 PM in two days. Cercariae were recognized following the systematic key references (Frandsen & Christensen, 1984; Schell, 1985).

2.4. Data analysis

Microsoft Excel 2010 and SPSS (Statistical Package for Social Sciences version 20; SPSS Inc., Chicago, Illinois) was used for data entry and data analysis. The Chi-squared test was used to compare the difference of prevalence between the two seasons. A value of $P < 0.05$ was considered significant.

3. Results and Discussion

3.1. Snail composition and distribution in rice fields

Eleven snail species belonging to 9 genera and 5 families were collected and classified (Table 1). Nine snail species were found in the wet season and 7 snail species were caught in the dry season. This agreed with Brockelman et al. (1986) that snail populations are typically more abundant in the rainy season which provides good conditions for the development of snails. In addition, increasing rain in the wet season leads to an increase of snail populations in the area (Khamboonruang et al., 1997).

Total snail species in this study were much higher than what Bui et al. (2010) collected in rice field in Nam Dinh province with 10 snail species, or Nguyen et al. (2014) found in An Hoa rice field of Tuy An district, Phu Yen province with 9 snail species. Moreover, it was much higher than the finding from Nguyen & Pham (2022) with 8 snail species in two rice fields in Can Gio district, also belonging to Ho Chi Minh City, but Can Gio was located in brackish water site and Hoc Mon was in freshwater area; therefore, snails in Hoc Mon must be more abundant. Generally, it can be commented that total snail species in rice fields were not the same, but not very different among the North (Nam Dinh province), the Central (Phu Yen province) and the South of Vietnam (Ho Chi Minh City). Therefore, further research on snails in rice fields should continue doing to confirm the similarity of number of snail species in rice fields in Vietnam.

Table 1. Snail composition in rice field of Hoc Mon district, Ho Chi Minh City, Vietnam

| Family | Genus | Snail species | Number of samples in the wet season | Number of samples in the dry season |
|---------------|------------------------|--|-------------------------------------|-------------------------------------|
| Ampullariidae | <i>Pomacea</i> | <i>Pomacea canaliculata</i> (Lamarck, 1828) | 240 | 197 |
| Bithyniidae | <i>Bithynia</i> | <i>Bithynia siamensis</i> (Lea, 1856) | 111 | 10 |
| Viviparidae | <i>Angulyagra</i> | <i>Angulyagra polyzonata</i> (Frauenfeld, 1862) | 1 | 0 |
| | <i>Cipangopaludina</i> | <i>Cipangopaludina chinensis</i> (Gray, 1834) | 9 | 0 |
| | | <i>Cipangopaludina japonica</i> (Martens, 1861) | 154 | 0 |
| | <i>Filopaludina</i> | <i>Filopaludina sumatrensis</i> (Dunker, 1852) | 11 | 101 |
| | | <i>Filopaludina martensi martensi</i> (Frauenfeld, 1865) | 21 | 4 |
| | <i>Sinotoia</i> | <i>Sinotoia aeruginosa</i> (Reeve, 1863) | 3 | 0 |
| | <i>Hippeutis</i> | <i>Hippeutis umbilicalis</i> (Benson, 1836) | 0 | 1 |
| Physidae | <i>Physa</i> | <i>Physa acuta</i> (Draparnaud, 1805) | 36 | 59 |
| Lymnaeidae | <i>Lymnaea</i> | <i>Lymnaea viridis</i> (Quoy & Gaimard, 1832) | 0 | 35 |

A total of 993 samples of snails were collected in August 2022 (the wet season) with 586 snails and in February 2023 (the dry season) with 407 snails. The snail species had high numbers including *Pomacea canaliculata* (44.0%), *Cipangopaludina japonica* (15.5%), *Bithynia siamensis* (12.2%), *Filopaludina sumatrensis* (11.3%), *Physa acuta* (9.6%), *Lymnaea viridis* (3.5%) and *Filopaludina martensi martensi* (2.5%). The other four snail species had low percentage including *Cipangopaludina chinensis* (0.9%), *Sinotoia aeruginosa* (0.3%), *Angulyagra polyzonata* (0.1%)

and *Hippeutis umbilicalis* (0.1%) (Table 2).

Although Hoc Mon and Can Gio were districts of Ho Chi Minh City, snail species and snail composition were almost different. In rice fields of Can Gio district, the common snail species were *Sermyla tornatella* (47.6%), *Pomacea canaliculata* (27.2%) and *Melanoides tuberculata* (14.0%) (Nguyen & Pham, 2022) while the high percentage of three snail species in Hoc Mon district were *Pomacea canaliculata* (44.0%), *Cipangopaludina japonica* (15.5%) and *Bithynia siamensis* (12.2%). Nguyen et al. (2014) also found the three highest snail species

in rice fields of Phu Yen province were *Bithynia* sp., *Pomacea* sp. and *Tarebia grannifera*. It can be concluded that snail species and composition in

rice fields in different areas were different from each other.

Table 2. Percentage contribution of each snail species in rice field of Hoc Mon district, Ho Chi Minh City, Vietnam

| Snail species | Wet season (August 2022) | | Dry season (February 2023) | | Total | |
|--|-----------------------------|------|-------------------------------|------|-------|------|
| | N | (%) | N | (%) | N | % |
| <i>Angulyagra polyzonata</i> (Frauenfeld, 1862) | 1 | 0.2 | 0 | 0 | 1 | 0.1 |
| <i>Bithynia siamensis</i> (Lea, 1856) | 111 | 18.9 | 10 | 2.5 | 121 | 12.2 |
| <i>Cipangopaludina chinensis</i> (Gray, 1834) | 9 | 1.5 | 0 | 0 | 9 | 0.9 |
| <i>Cipangopaludina japonica</i> (Martens, 1861) | 154 | 26.3 | 0 | 0 | 154 | 15.5 |
| <i>Filopaludina sumatrensis</i> (Dunker, 1852) | 11 | 1.9 | 101 | 24.8 | 112 | 11.3 |
| <i>Filopaludina martensi martensi</i> (Frauenfeld, 1865) | 21 | 3.6 | 4 | 1.0 | 25 | 2.5 |
| <i>Hippeutis umbilicalis</i> (Benson, 1836) | 0 | 0 | 1 | 0.2 | 1 | 0.1 |
| <i>Lymnaea viridis</i> (Quoy & Gaimard, 1832) | 0 | 0 | 35 | 8.6 | 35 | 3.5 |
| <i>Sinotoia aeruginosa</i> (Reeve, 1863) | 3 | 0.5 | 0 | 0 | 3 | 0.3 |
| <i>Pomacea canaliculata</i> (Lamarck, 1828) | 240 | 41.0 | 197 | 48.4 | 437 | 44.0 |
| <i>Physa acuta</i> (Draparnaud, 1805) | 36 | 6.1 | 59 | 14.5 | 95 | 9.6 |
| Total | 586 | 100 | 407 | 100 | 993 | 100 |

3.2. Cercariae morphotypes infected in snails

The prevalence of cercariae infection in the dry season (14.3%) was significantly higher than in the wet season (4.5%) ($P < 0.05$). For the overall prevalence in two seasons for each snail species, *Lymnaea viridis*, the host of *Fasciolidae* (Thai, 2016), had the highest prevalence of 14.3% ($N = 5/35$) and *Bithynia siamensis*, a potential host for both Heterophyidae and Opisthorchiidae (Madsen et al., 2015), had the prevalence of 4.1% ($N = 5/121$). The other nine snail species had cercariae free (Table 3).

Lymnaea sp. was found infected with cercariae, but the snail species had the highest

prevalence was *Melanoides tuberculata* in Nguyen et al. (2014). *Lymnaea* sp. was not found in Can Gio rice field in the research by Nguyen & Pham (2022) and in Cu Chi rice field in the study by Pham & Duong (2023). Thirty five *Lymnaea viridis* which were collected in Hoc Mon rice field in this research with the trematode prevalence of 14.3% were the new finding in the rice field of Ho Chi Minh City area. *Bithynia siamensis* was the second infected snails with the prevalence of 4.1%. This finding agreed with Nguyen & Pham (2022) that *Bithynia* in rice fields got the top prevalence. It was also like what Bui et al. (2010) and Nguyen et al. (2014) reported that *Bithynia* in canals and ponds had the high prevalence. For

the cercariae infection, the prevalence in the dry season was significantly higher than in the wet season ($P < 0.05$). This finding is similar to what Nguyen et al. (2014) that infection by trematode larvae in snails was high in the dry season and low in the wet season because of the temperature. The finding in this study also agreed with the publish by Nkwengulila & Kigadye (2005) that the prevalence of cercariae fluctuated by seasons, it was high in the dry season and decreased in the wet season. However, the result in this research

was completely different from what Nguyen & Pham (2022) that the prevalence in the wet season was much higher than in the dry season. Although two districts belonged to Ho Chi Minh City but the result in Hoc Mon in this research was different from the finding from Can Gio; therefore, the different natural conditions might affect the distribution of snails and the trematode prevalence in snails when comparing between seasons.

Table 3. Trematode prevalence in snails in rice field of Hoc Mon district, Ho Chi Minh City, Vietnam

| Snail species | Wet season (August 2022) | | Dry season (February 2023) | | Total | |
|---------------------------|-----------------------------------|----------------|-----------------------------------|----------------|-----------------------------------|----------------|
| | Infected snails/ Collected snails | Prevalence (%) | Infected snails/ Collected snails | Prevalence (%) | Infected snails/ Collected snails | Prevalence (%) |
| <i>Bithynia siamensis</i> | 5/111 | 4.5 | 0/10 | 0 | 5/121 | 4.1 |
| <i>Lymnaea viridis</i> | 0 | 0 | 5/35 | 14.3 | 5/35 | 14.3 |

No *Pleurolophocercous cercariae* was found in snails from the research rice field. Xiphidiocercariae was more common and discovered from *Lymnaea viridis* and *Bithynia siamensis*. *Echinostome cercariae* was got in only one *Bithynia siamensis*. It was obvious that *Bithynia siamensis* had 2 different cercariae morphotypes whereas *Lymnaea viridis* had only one cercariae morphotype (Table 4). *Xiphidio cercariae* seemed to have more frequent occurrence than the others as it was recovered from 9 of 10 snails. This is similar to Nkwengulila & Kigadye (2005) that *Xiphidio cercariae* was the most prevalent in ponds and stream. It was also like what Pham & Tran (2021) that *Xiphidio cercariae* appeared the most in ponds of giant gourami culture in ponds.

Silver barb from rice field in Hoc Mon and released into Thay Cai - An Ha canal in Hoc Mon district was infected with metacercariae of *Haplorchis pumilio* (Pham et al., 2019), the species belongs to Heterophyidae (Chai et al., 2005). Pham & Nguyen (2005) stated that *Pleurolophocercous cercariae* was the cercariae of Heterophyidae; however, no *Pleurolophocercous cercariae* was found in two morphotypes of cercariae in this research in August 2022 and February 2023. Therefore, the infected source of metacercariae in silver barb has not been identified yet. More research on cercariae in snails in other rice fields of Hoc Mon in the different months should be done to check whether they exist to find the answer for one of the reasons why silver barb was infected.

Table 4. Cercariae morphotypes in infected snails in rice field of Hoc Mon district, Ho Chi Minh City, Vietnam

| Cercariae | Wet season (August 2022) | | Dry season (February 2023) | |
|------------------------------|---------------------------|------------------------|----------------------------|------------------------|
| | <i>Bithynia siamensis</i> | <i>Lymnaea viridis</i> | <i>Bithynia siamensis</i> | <i>Lymnaea viridis</i> |
| <i>Xiphidio cercariae</i> | 4 | 0 | 0 | 5 |
| <i>Echinostome cercariae</i> | 1 | 0 | 0 | 0 |

4. Conclusions

Eleven snail species belonging to 9 genera and 5 families were identified. *Lymnaea viridis* and *Bithynia siamensis* were infected with trematode (cercariae stage) with the overall prevalence of 14.3% and 4.1%, respectively. The other nine snail species had cercariae free. Two cercariae morphotypes were recovered from snails including *Xiphidio cercaria* and *Echinostome cercariae*. Further research on this subject in different months and in other water bodies should be done in Hoc Mon district and other places to identify the sources of trematodes infected to fish in cultured ponds.

Conflict of interest

The authors declare no conflict of interest.

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