



## A Study of Trematoda and Nematoda Prevalence on The Reproductive Organs of Apple Snail (*Pomacea* sp.)

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### Abstract



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Apple snails (*Pomacea* sp.) are Gastropods in rice field ecosystems that consumed by humans and also play an important role as intermediate hosts for various parasitic worms. One part of the apple snail that is consumed by humans is the reproductive organs, which is an ideal place for parasites to develop because it has high metabolic activity. This study aims to identify the types of parasites and analyze the prevalence of infection in the reproductive organs of apple snails. Sampling and laboratory analysis were conducted in September 2025. The research method used was descriptive with simple random sampling techniques at three stations (inlet, center, outlet) in the rice field area Jabungan Village, Semarang. A total of 30 snails were examined through dissection and microscopic observation. The results showed that there are six genera of parasites infecting the reproductive organs (gonads) of apple snails, consisting of two classes, namely the Trematode class (*Opisthorchis*, *Echinostoma*, *Fasciola*, and *Schistosoma*) and the Nematode class (*Gnathostoma* and *Trichuris*). Prevalence analysis showed that the *Opisthorchis* and *Echinostoma* genera dominated with a prevalence value of 100% at all observation stations. The *Gnathostoma* and *Schistosoma* genera had fluctuating prevalence rates between 60% and 80%, while *Fasciola* was found to be the least prevalent (10%-20%). The *Trichuris* genus was only found at the outlet station with a prevalence of 30%. This research is expected to support public health education programs on the risk of parasites in apple snails.

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### 1. Introduction

Apple snail is a species of freshwater Gastropod that often consumed by humans and livestock due to their high mineral protein and lowfat content. Furthermore, apple snails also serve an ecological role as intermediate host, requires y various types of parasitic worms to complete their life cycles before infecting/ definitive hosts (Ghosh *et al.*, 2017; Tiuria & Noviyara, 2020).

The parasitic worms that are often found in apple snails are very diverse, including *Angiostrongylus cantonensis*, *Schistosoma manoni*, *Echinostoma lindoense*, *Paragonimus westermani*, *Clonorchis sinensis*, *Echinococcus granulosus*, *Stroglyoides stercoralis*, *Gnathostoma spinigerum*, dan *Schistosoma japonicum* (El-Zeiny *et al.*, 2025; Martinez *et al.*, 2025; Song *et al.*, 2016). In apple snails, the focus of infection frequently occurs within the reproductive organs, which exhibit distinct physiological characteristics compared to the digestive system. These

organs possess high metabolic activity, high nutrient density and are supported by an extensive blood supply for gametogenesis. Consequently, they provide a highly ideal environment for parasitic development (Martinez *et al.*, 2024).

Each species of parasite that infects the reproductive organs of apple snails has different characteristics and levels of pathogenicity to humans. The risk of transmission of these diseases is exacerbated by the environmental conditions of the Jabungan rice fields, which are prone to domestic waste pollution. This densely populated urban area has a high potential of water pollution in rice fields due to poorly managed domestic waste (D'Souza *et al.*, 2022; Friesen *et al.*, 2022). Consequently, identify the prevalence and type of parasite infecting apple snails in the Jabungan area is a critical step in addressing the local environmental health risks.

## 2. Material and methods

### 2.1 Study Area

The study was conducted in September 2025 on rice field area of Jabungan Village, Semarang which is presented in Figure 1. A total 30 apple snails with a minimum shell

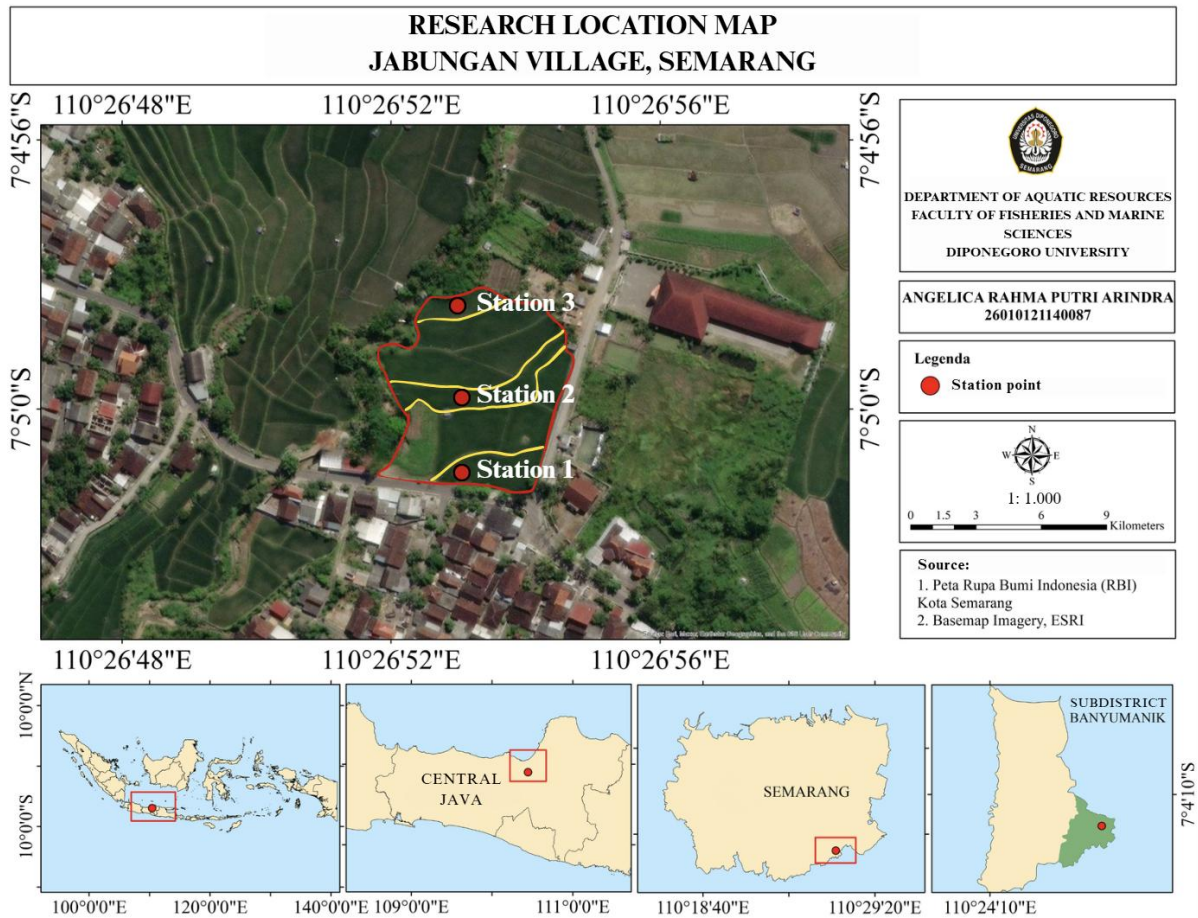


Fig 1. Map of sampling location

### 2.2 Research Equipment and Materials

The equipment utilized in this study comprised sampling tools, laboratory analysis instruments, and data analysis. Apple snail (*Pomacea* sp.) samples were collected and contained in 450 mL plastic jars. For laboratory analysis, a digital caliper with an accuracy of 0.01 cm and a 15 cm ruler with accuracy of 0.1 cm were employed to measure the snail shells. Parasitological analysis was conducted using an Olympus CX-23 binocular microscope equipped with ToupView software, alongside glass slides and coverslips. Additionally, Pasteur pipettes were used for liquid transfer, while a mortar and pestle were utilized to crush the snail shells. Other laboratory apparatus included Petri dishes for sample placement, a dissection kit or personal protection during laboratory procedures. The materials utilized in this research include apple snails as samples and distilled water (aquades). Additionally, a 70% antiseptic alcohol solution was utilized to sterilize and clean the laboratory equipment from residual parasites, while paper towels were used for drying and cleaning the apparatus.

### 2.3 Apple snail sampling

Apple snail samples were collected using simple random sampling, a method where sample members are selected randomly without regard to strata (Sugiyono, 2019). This method was selected because every snail within the rice fields of Jabungan Village had an equal probability of being included in the study. The technique assumes that the snail population across the rice field area is uniformly distributed across the first, second and third stations. The snails were

collected manually by randomly scouring the edges of the rice fields within the designated plots. The selected snails were then placed into biological sample bottles containing 225 ml. of water to maintain their physiological condition and ensure the integrity of their organs remains consistent with their natural habitat.

### 2.4 Data analysis

Data analysis in this study employed point prevalence calculations to describe the proportion of individuals within a population infected by parasites at a specific point in time, without comparison to previous periods. The prevalence result was used to determine the distribution of parasites infecting apple snails relative to the total number of snails examined in each plot. Prevalence data were analyzed descriptively following the methodology of Dogiel *et al.* (1970), as cited in (Rosnizar *et al.*, 2018), using equation as follow:

$$Prevalence = \frac{\Sigma \text{ of infected snails at each station}}{\Sigma \text{ of snails examined at each station}} \times 100\%$$

## 3. Results

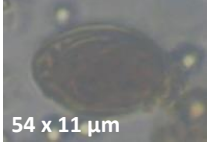

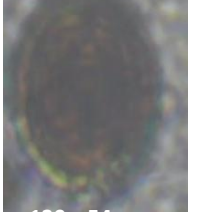
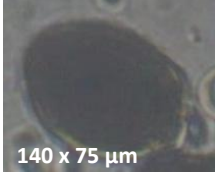

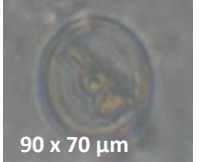
### 3.1 Parasite Identification

The apple snails used as samples in this study exhibited shell sizes ranging from 3 cm to 5 cm. This size variation was selected based on the minimum shell size for adult snails, which typically ranges from 2.5 to 4 cm. The selection of adult snails aimed to ensure that the reproductive organs were sufficiently developed for identification. This maturity facilitated gonadal dissection and parasite

examination and parasitic examination (Dewi *et al.*, 2022; Martinez *et al.*, 2024). Parasitological analysis of the apple

Arindra *et al.*, 2026. *A Study of Trematoda and Nematoda*..... snail's reproductive system showed infection within the gonads. This data is summarized in Table 1.

Table 1. Parasitic identified in 30 apple snails

Genus	Phase	Group	Microscopic characteristics
 54 x 11 µm <i>Gnathostoma</i>	Egg	Nematoda	Cylindrical in shape, the specimen possesses a hooked head and two pairs of cervical sacs (Liu <i>et al.</i> , 2020).
 50 x 22 µm <i>Trichuris</i>	Egg	Nematoda	The eggs exhibit a characteristic barrel-shaped appearance with bipolar opercula; the shell is thick and smooth, displaying a yellowish-brown coloration (García-Sánchez <i>et al.</i> , 2020).
 <i>Echinostoma</i>	Egg	Trematoda	Elliptical in shape, yellowish-brown in color, with an inconspicuous operculum; the shell is relatively thick, exhibiting slight wrinkling and localized thickening (Sah <i>et al.</i> , 2018)
 140 x 75 µm <i>Fasciola</i>	Egg	Trematoda	The eggs are oval or elliptical in shape with a distinct operculum, displaying a pale yellow to yellowish-brown color and a smooth, thick, transparent wall. (Prasetya <i>et al.</i> , 2019)
 30 x 12 µm <i>Opisthorchis</i>	Egg	Trematoda	Oval to elongated in shape, the eggs possess a thin, smooth wall and a small apical operculum. Characteristically, they exhibit shoulders or a prominent <i>abopercular knob</i> . (Liau <i>et al.</i> , 2023)
 90 x 70 µm <i>Schistosoma</i>	Egg	Trematoda	The specimen is spherical or sub-oval in shape, featuring a small and inconspicuous lateral spine, and contains a miracidium. (De Marco Verissimo <i>et al.</i> , 2019)

Based on Table 1, six parasite species were found to infect apple snails (*Pomacea* sp.) on the rice fields of Jabungan Village. These consist of two genera from the Nematoda and four from the Trematoda. Trematodes are flatworms belonging to the phylum Platyhelminthes. They are generally characterized by a dorso-ventrally flattened, leaf-like body covered by a tegument, which is often equipped with scale-like spines (Toledo & Fried, 2017; Waikagul *et al.*, 2015). Conversely, Nematodes are roundworms defined as transparent, bilaterally symmetrical, unsegmented, pseudocoelomate, and triploblastic organisms with a thread-like appearance (Gowda *et al.*, 2022; Li *et al.*, 2017). The parasites identified in the apple snails include the Trematode genera *Echinostoma*, *Fasciola*, *Opisthorchis*, and *Schistosoma*, as well as the Nematode genera *Gnathostoma* and *Trichuris*. All six parasites were localized within the

reproductive organs, with varying distributions across the different sampling sites.

### 3.2 Parasitic Infection

Samples of apple snail were collected from three stations, consisting of 10 individuals each, as illustrated in the following Figure 2. Based on the analysis, parasitic infections in apple snails (*Pomacea* sp.) within the rice fields of Jabungan Village exhibited varying prevalence rates across different irrigation flow points. The parasites *Opisthorchis* and *Echinostoma* showed the highest prevalences at 100% across all stations. In contrast, the prevalence of *Gnathostoma* and *Schistosoma* fluctuated between 60% and 80%, while *Fasciola* was the least common, with a low prevalence ranging from 10% to 20%. Notably, station 3 (outlet) demonstrated the highest parasitic diversity, marked by presence of *Trichuris* at a 30% prevalence rate a species not recorded at any other station.

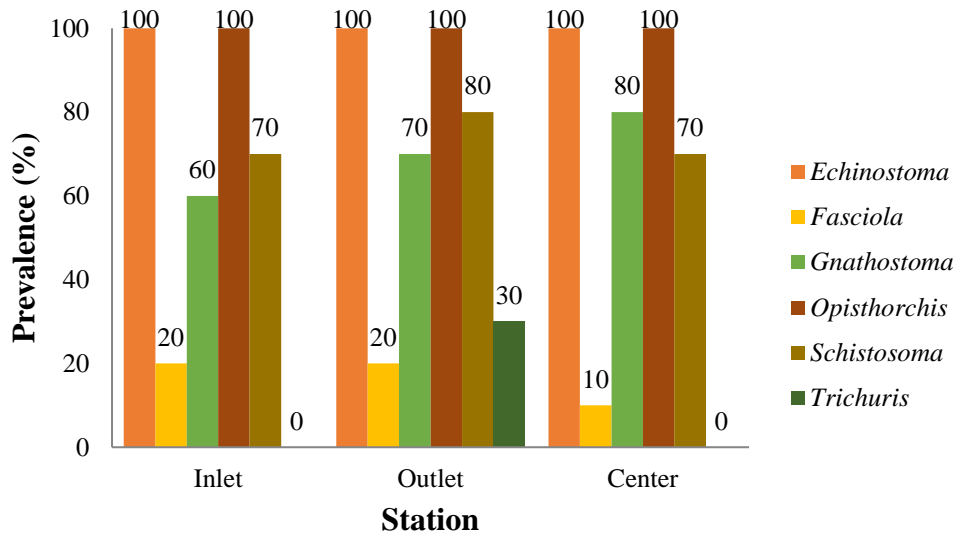


Fig 2. Prevalence of parasites in the reproductive organs of apple snails (n=10 per station)

The publication screening process is shown in the PRISMA flow diagram (Fig 1). Initially, 540 papers were retrieved using the systematic literature search phrases listed in Table 2. Subsequently, after literature screening that included title, abstract, and full-text screening, 89 final papers were selected for further data extraction. Overall, 16% of the initially recognized publications in publication databases (Scopus, ResearchGate, and Google Scholar) were included in the extraction and analyses process. Quite a few publications were registered in more than one database, hence counted as duplicates (Fig 1). The cutoff date for the literature search was June 27, 2025.

## 4. Discussion

### 4.1 Parasite Diversity in Gonadal Tissue

In this study, six parasites were identified with distinct morphological characteristics consistent with literature on Trematoda and Nematoda, specifically from the genera *Echinostoma*, *Fasciola*, *Gnathostoma*, *Opisthorchis*, *Schistosoma*, and *Trichuris*. These six genera comprise two species from the class Nematoda and four from the class Trematoda. Nematodes are microscopic, unsegmented, elongated, roundworms without appendages, occurring in freshwater, marine, and terrestrial environments (Gowda *et al.*, 2022; Li *et al.*, 2017). Meanwhile, Trematodes are a class of flatworms characterized by complex life cycles involving at least two hosts and alternating sexual and asexual reproductive stages, characterized by a dorsoventrally flattened and unsegmented leaf-like shape, trematodes possess specialized suckers for attachment to the host tissue (Toledo & Fried, 2017; Waikagul *et al.*, 2015). Notably, in this study, only eggs of each species were recovered. Morphological observation under a microscope reveal distinct differences in egg characteristics between Trematodes and Nematodes. The majority of Trematode eggs possess an operculum and exhibit a symmetrical oval or barrel-shaped morphology, whereas Nematode eggs lack an operculum and display asymmetrical or highly diverse shapes (Greenwood, 2012; Mkandawire *et al.*, 2022; Pyarogo *et al.*, 2025).

The localization of helminth parasites within the gonads of the apple snail (*Pomacea* sp.) represents a specialized ecological and physiological adaptation. Firstly, the snail gonad is a highly vascularized organ characterized by an abundant supply of essential nutrients, particularly

lipids, glycogen, and proteins, which are dynamically synthesized for gametosis (Martinez *et al.*, 2024). Parasitic helminths, especially Trematodes and Nematodes, have high metabolic demands to support their intensive egg production and larval development. Consequently, the gonad serves as an ideal metabolic sink, providing the rich energetic resources necessary for the parasites' reproductive success (Gilardoni *et al.*, 2018; Jithila & Prasadana, 2018). Secondly, the migration pathway of these parasites within the gastropod host naturally leads to the digestive gland (hepatopancreas) and the adjacent gonadal complex. In *Pomacea* sp. the gonad and the digestive gland are anatomically intertwined, sharing common hemolymph space (sinuses). Trematode and Nematoda may penetrate the snail integument migrate via hemolymph vessels and utilize open circulatory space or actively bore through tissue to reach the visceral mass, resulting in co-infecting and egg deposition within the gonadal lumen (Ghosh *et al.*, 2017).

### 4.2 Prevalence Patterns Among Sampling Stations

In this study, the genera *Echinostoma* and *Opisthorchis* were identified as the two parasitic taxa infecting the reproductive organs of the apple snails (*Pomacea* sp.) examined at each sampling station. These findings align with existing literature, which states that both genera tend to develop within the reproductive tissues of Gastropods as a site for larval development and life cycle completion. Furthermore, variations in the community composition of freshwater snails, such as the apple snail, significantly influence the diversity of Trematode species (Aryaeipour *et al.*, 2023; Duan *et al.*, 2021). Microhabitats within the snail's reproductive organs provide an optimal environment for the development of sporocysts and rediae. A study by Martinez *et al.* (2024) revealed that snail gonad tissues are primary targets for larval invasion; beyond providing the necessary energy for proliferation, infecting the host's gonads allows parasites to enhance their survival conditions and successfully complete their life cycles. The occurrence of *Echinostoma* and *Opisthorchis* in this study area suggests that distribution patterns and diversity are frequently influenced by host dispersal. Furthermore, rice fields rich in organic matter serve as potential hotspots for the transmission of early Trematode stages, including the miracidium phase and the subsequent development of sporocysts and rediae within the snail host (Wang *et al.*, 2022; Zemmer *et al.*, 2020).

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organs that provide ideal physiological conditions for the development of Trematode and Nematode larvae. The highest parasitic loads are typically found in the hepatopancreas, mantle cavity, renal cavity, and digestive tract, whereas the shell and umbilicus often serve as attachment sites for epiphytic organisms such as leeches and copepods. Furthermore, Zhang *et al.* (2024) demonstrated that nearly all cercariae-stage parasites survive exposure to 10 mg/L of powder detergent and 100 mg/L of bar soap for 60 minutes, though powder detergent proved to be more lethal than bar soap.

Prior to consumption, local communities typically boil apple snails in salted water or soak them in vinegar and lime juice for 15–30 minutes to eliminate the fishy odor. However, it is crucial to ensure that the snails are cooked thoroughly to eradicate any infecting parasites through heat exposure. According to the FAO (2021), most thermal processing techniques, including boiling, are effective in neutralizing these parasites. Temperatures ranging from 55°C to 70°C for 30 minutes are generally sufficient; however, if parasites are in the egg stage, a minimum temperature of 100°C for 15 to 40 minutes is required. Furthermore, the FAO (2021) suggests that freezing infected food materials at -20°C for 48 hours is also capable of killing parasitic eggs. These guidelines serve as a vital reference for safe processing to prevent parasitic transmission in humans. Alternatively, risk can be mitigated by consuming only non-infected tissues, such as the foot muscle.

## 5. Conclusions

This study demonstrates a remarkably high prevalence and diversity of parasitic helminths infecting the reproductive organs (gonads) of the golden apple snail (*Pomacea* sp.) in the agricultural ecosystem of Jabungan Village, Semarang. The identification of six distinct parasitic genera, consisting of four trematodes (*Echinostoma*, *Fasciola*, *Opisthorchis*, and *Schistosoma*) and two nematodes (*Gnathostoma* and *Trichuris*). From a public health perspective, these findings present a profound epidemiological warning regarding foodborne zoonotic risks. Given that golden apple snails are widely utilized as a low-cost protein source by local communities, the presence of highly pathogenic parasites capable of causing severe human illnesses (such as opisthorchiasis, echinostomiasis, and gnathostomiasis) necessitates strict safety guidelines.

## Ethics approval

No permits were required.

## Data availability statement

The database consisting of the results of this review can be accessed through the supplementary materials.

## Author contributions

ARPA was responsible for conceptualization, data curation, methodology, resources, software visualization, as well as writing and editing. WTT and DET contributed to conceptualization, supervision, validation, formal analysis, as well as review & editing.

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This high prevalence driven by environmental influences, agricultural runoff and the distribution of definitive hosts. The sampling locations, primarily characterized by slow flowing irrigation channels and stagnant rice fields, provide an ideal microhabitat for trematode transmission. Furthermore, agricultural runoff rich in organic nutrients can stimulate algal blooms, which increases the food availability for the snail, leading to higher snail population densities and consequently facilitating dense, continuous parasite transmission cycles (Cable *et al.*, 2017; Widiastuti *et al.*, 2015). The high prevalence of *Opisthorchis* and *Echinostoma* is heavily influenced by the constant biotic input from definitive hosts within the surrounding environment. *Echinostoma* species typically utilize semi-aquatic birds (such as ducks and herons) and small mammals as final hosts, which frequently forage in rice fields and deposit feces laden with parasite eggs directly into the water (Campbell *et al.*, 2016). Similarly, *Opisthorchis* species rely on mammalian hosts (including humans, domestic cats, and dogs) near agricultural settlements. The integration of livestock farming and poor sanitation barriers in rural irrigation systems ensure an uninterrupted, high-density influx of miracidia into the snail habitat (Campbell *et al.*, 2016; Tiuria & Noviyara, 2020; Widiastuti *et al.*, 2015)

## 4.3 Ecological and Public Health Implications

Apple snails are known to serve as intermediate hosts for several types of parasites, particularly those from the Nematoda and Trematoda group that can infect humans and animals (Widiastuti *et al.*, 2015). According to Martinez *et al.* (2025), the identification of intermediate stage of Trematoda and Nematoda in apple snail that this species plays an epidemiological role in maintaining the life cycle of parasitic worms in freshwater ecosystems. The risk of infection is influenced by contaminated water, snail population density, and interactions with definitive host such as rats or waterfowl, which serve as sources of contamination for the parasite's larvae (Testai *et al.*, 2023). The life cycles of these two groups of parasites are fundamentally different. The life cycle of Trematodes begins when eggs are released into environment, usually through the feces of a definitive host such as a mammal, bird, or fish. These eggs then flow through the water in rice paddies and hatch into ciliated larvae called mirasidia, which must immediately find their first host, such as a apple snail (Billet *et al.*, 2023).

Beyond environmental influences, the presence of these parasites in apple snails warrants concern due to their zoonotic potential. If these snails are consumed by humans without adequate thermal processing, the parasites may undergo host transition and pose severe health risks. The identified taxa are known to cause diseases such as echinostomiasis, fascioliasis, gnathostomiasis, opisthorchiasis, schistosomiasis, and trichuriasis. Each condition presents distinct clinical manifestations, ranging from diarrhea, abdominal pain, dyspepsia, fever, and anemia to more severe complications, including hepatic cellular damage, meningitis, and malignancy (Manz *et al.*, 2017; Nogrado *et al.*, 2023; Qi *et al.*, 2024; Rodari *et al.*, 2025; Sripta *et al.*, 2025; Toledo & Esteban, 2016).

Given its abundance and high protein content, the apple snail is frequently consumed by local communities near rice fields (Prabewi *et al.*, 2019). However, without proper processing, the presence of anti-nutritional factors within the snail meat may limit its utility. While some consumers utilize the entire organism, others only consume specific organs. Damborenea *et al.* (2017) noted that parasite distribution within the snail is not uniform; rather, it is concentrated in

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#### Declaration of competing Interest

None

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