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Aestivation and its impact on the survival of snail intermediate hosts and trematode transmission in rice paddies

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Research Paper

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Abstract

In this study, we investigated the diversity and survival of aestivating snails in dry-season rice paddies, focusing on their role as intermediate hosts for trematodes. A total of 1,159 snails from various families were collected and analysed, revealing nine species, primarily from the Bithyniidae family, with *Bithynia siamensis siamensis* being the most abundant. Of the nine species collected, the most common species, *B. s. siamensis*, exhibited a relatively high survival rate of 81.5% throughout the three-month dry period post-harvest, whereas the remaining eight species had survival rates below 24.0% (0–23.1%). Four snail species, *B. s. siamensis*, *Filopaludina martensi martensi, F. sumatrensis polygramma*, and *F. doliaris*, were found infected with larval trematodes, with an overall prevalence of 8.46% (57/674). The larval trematodes identified were categorized into four types: echinostome metacercaria, *Thapariella anastomusa* metacercaria, unidentified metacercaria, and unidentified rediae. These findings demonstrate that trematode infections can persist in aestivating snails despite the challenging conditions during the dry season, emphasizing the potential of aestivating snails to act as vectors for trematode transmission in agricultural settings. This underscores the need for effective management strategies to mitigate the risks associated with trematode transmission.

Introduction

Freshwater snails are relevant for public and veterinary health, as several species act as first and second intermediate hosts for helminths and parasites that cause various food-borne trematode diseases in humans and animals (Chai *et al.* 2005, 2009; Madsen and Hung 2014). In Thailand, several freshwater snail species are reported to be infected by larval trematodes, with high prevalence noted in northern and northeastern regions (Burch and Lohachit 1983; Sri-Aroon 2011; Woodruff and Upatham 1993). However, studies specifically addressing the potential for larval trematode infection in northern Thailand, especially in Chiang Rai Province, remain limited. Recent studies have documented larval trematode infections across various snail species in various ecosystems, with particular emphasis on paddy fields in this region (Chantima *et al.* 2018a, 2018b).

In northern Thailand, rice is generally cultivated once or twice per year. Farmers typically grow rice during the rainy season, with some extending cultivation into the post-rainy season in areas with irrigation access. Following the harvest, the dry season brings unfavourable conditions for freshwater snails in rice paddies. During this period, snails face extreme environmental stresses, particularly drought. To survive, they burrow into the mud and enter a state of aestivation, a physiological process like hibernation. With the arrival of spring rains and monsoon flooding, snails recolonize their habitats and reproduce. Aestivation is a crucial survival strategy for snails facing extreme temperatures and desiccation, particularly in tropical environments. This adaptation is essential not only for their survival, but it also influences the dynamics of trematode transmission (Osborne and Wright 2018; Rubaba *et al.* 2016). The timing of aestivation in relation to infection significantly impacts the development of trematodes within their snail hosts. A study by Badger and Oyerinde (1996) demonstrated that *Biomphalaria pfeifferi* snails that aestivate immediately after infection produce cercariae in greater numbers compared to those that delay aestivation. Furthermore, aestivation can prolong cercarial development, which in turn affects the transmission potential of trematodes in rice paddies. Notably, shorter aestivation durations lead to lower mortality rates for the snail intermediate host of *Opisthorchis viverrini*, *Bithynia siamensis goniomphalos* (Chaiyasaeng *et al.* 2019).

Aestivation serves as a crucial adaptive mechanism for snails; however, it also poses challenges for trematode transmission. Understanding these dynamics is essential for effectively managing snail populations and controlling trematode-related diseases in agricultural settings. Therefore, this study aimed to investigate the diversity and survival of aestivating snails in dry-season rice paddies in Chiang Rai Province, Thailand, with a specific focus on their role as intermediate hosts for trematodes. The findings of this study may enhance our understanding of the ecology of aestivating snails and its implications for public health regarding parasitic infections in agricultural landscapes.

Materials and methods

Study area and sampling localities

The study area in Chiang Rai Province, northern Thailand, within the North Mekong River basin of Thailand, has a tropical monsoon climate with distinctive seasons. The mean annual rainfall of this area varies from 1,200 mm to 2,000 mm (Eastham *et al.* 2008). Eight sampling sites were established across four districts: Mae Lao, Mueang Chiang Rai, Phan, and Wiang Chai, based on the availability of potential intermediate snail hosts, site accessibility, and the tendency of snails to aestivate after the rice harvest during the dry season (February to May) in endemic areas (Chantima *et al.* 2018a, 2018b; Chantima and Rika 2020). Geographic coordinates of sampling sites were recorded using a GPS device or determined as accurately as possible from a map. Sampling sites were then digitally mapped on a dot-by-dot basis on a public domain map (GISTDA 2011; Github 2024). Final maps were compiled using Photoshop CS6 (Adobe Systems Inc., San José, CA, USA). The coordinates of these localities are presented in Table 1 and Figure 1.

Snail and soil sampling

Snail sampling was conducted once a month during three sampling periods from March to May 2020, coinciding with the dry season. The selected sampling sites were located at the dried end of a paddy field. Snails buried in the soil were collected at two depth intervals: from the ground surface to 5 cm and from 5 to 10 cm. A quadrat measuring 0.36 m^2 (0.6 m square) was used to define the sampling area. All snails found within the quadrat were brought to the laboratory for further analysis. Species identification was carried out using conchological methods based on the taxonomic keys of Brandt (1974) and Upatham *et al.* (1983). The snails were subsequently examined for survival and trematode infections.

Table 1. List of localities where snails were collected

Location	Coordinates (UTM)
Ban Pa Ruak, Dong Mada Subdistrict, Mae Lao District (ML1)	47N 574259 2180001
Ban Sri Wang Mun, Buasalee Subdistrict, Mae Lao District (ML2)	47N 579716 2190213
Ban Pong Nam Tok, Ban Du Subdistrict, Mueang Chiang Rai District (MC1)	47N 586290 2210770
Ban Nang Lae Nai, Nang Lae Subdistrict, Mueang Chiang Rai District (MC2)	47N 588413 2214537
Ban San Ko Hiang, Santisuk Subdistrict, Phan District (PH1)	47N 580864 2163834
Ban Dong Lan, Sai Khao Subdistrict, Phan District (PH2)	47N 573946 2177276
Ban Sri Wiang, Wiang Chai Subdistrict, Wiang Chai District (WC1)	47N 595176 2196478
Ban Pong, Wiang Chai Subdistrict, Wiang Chai District (WC2)	47N 597990 2196273

Soil samples were collected in the same quadrat as the snail sampling. At each site, three replicate samples were randomly collected from a 0.36 m^2 area using a metal shovel. Approximately 200 g of sediment per replicate was gathered from each site. Following collection, the samples were sealed in labelled bags and transported to the laboratory for analysis. The soil properties, focusing on pH, electrical conductivity, soluble salts concentration, and cation or anion concentration in soil extract, were determined at each sampling site using the methods by the U.S. Salinity Laboratory Staff (1954) and Sparks *et al.* (1996).

Assessment of aestivating snail species abundance and diversity

To evaluate the abundance and diversity of aestivating snail species, several indices were employed. First, the richness index, specifically Margalef's richness index (D), was utilized and calculated using the following formula:

$$D = \frac{S-1}{\ln n}$$

where S was the total number of species and n was the total number of individuals (Clarke and Warwick 1994; Washington 1984).

Second, Shannon's diversity index (H) was calculated using the following formula:

$$H = -\sum_{i=1}^{n} p_i \ln p_i$$

where p_i was the proportion of the total count arising from the i-th species. A higher value indicated a greater number of species with similar abundances, whereas a lower value indicated low diversity that was dominated by one or a few species (Clarke and Warwick 1994; Hill *et al.* 2005).

Third, species evenness (J'), quantifies the distribution of individuals across different species within an ecosystem. This metric is commonly represented by Pielou's evenness index, which is calculated using the following formula:

$$J' = \frac{H}{\ln S}$$

where H referred to Shannon's diversity index. It values between 0 and 1; values closer to zero represented uneven populations that were dominated by one species, while values closer to 1 represented even populations that were comprised of several species with similar abundances (Brewer 1994; Clarke and Warwick 1994; Hillebrand 2008).

Examination for survival of aestivating snail

The survival of aestivating snails was evaluated by reactivating them in water for a duration of 24 hours. The shells of snails were categorized into three distinct groups: 1) live snails that resumed activity upon immersion in water, 2) dead snails, which either lacked soft tissues or had an operculum, and 3) old shells, indicative of snails that died during the prior dry season, distinguished by their white coloration or discoloration and erosion (Figure 2). The counts of dead and surviving snails, excluding old shells, were recorded to calculate survival rates.

Examination for trematode infection

The examination of snail infections was conducted using shedding and crushing methods. For the shedding method, individual snails were placed in 15 ml bottles filled halfway with dechlorinated tap

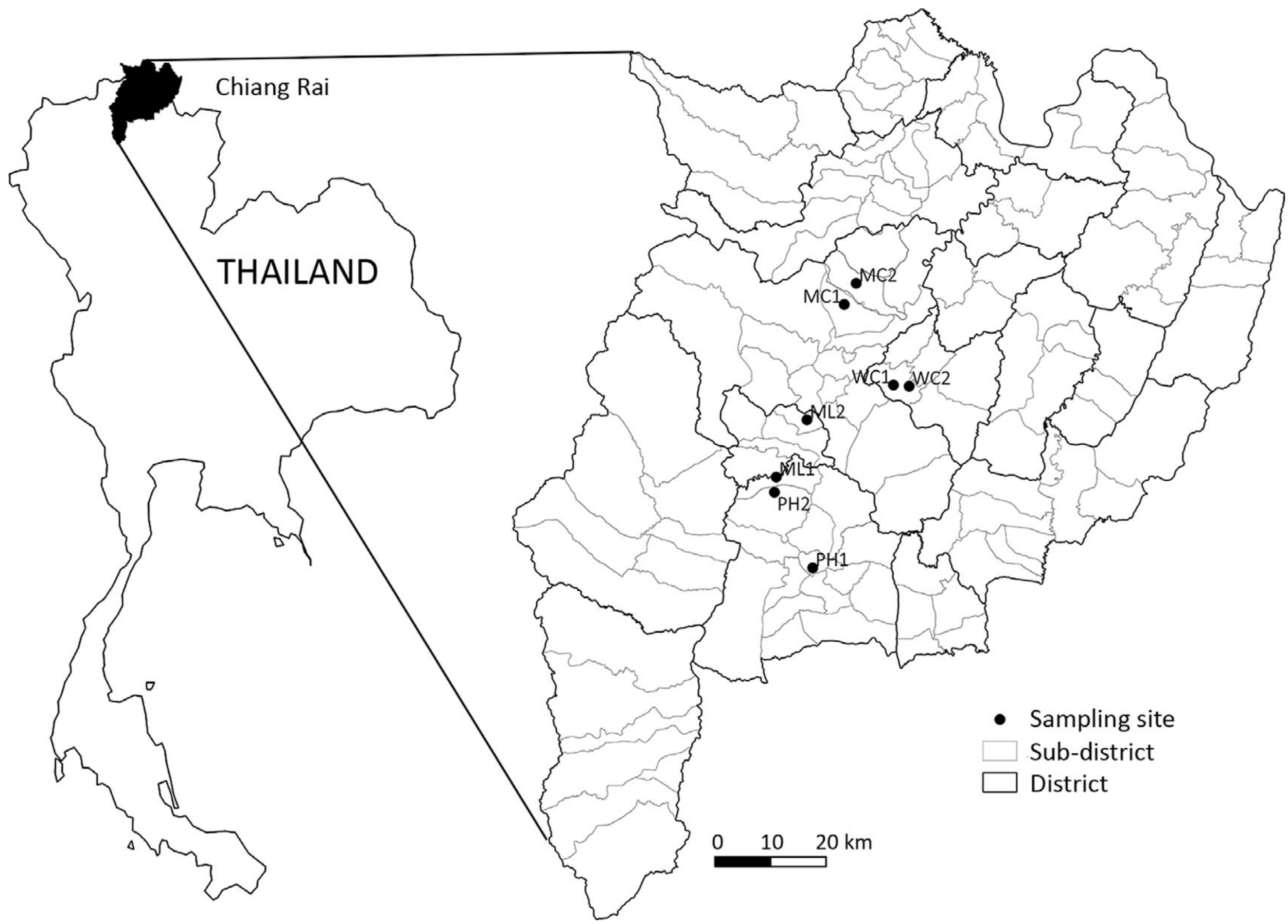

Figure 1. Map of study area, showing the sampling localities and the districts where snails were collected as indicated in Table 1.

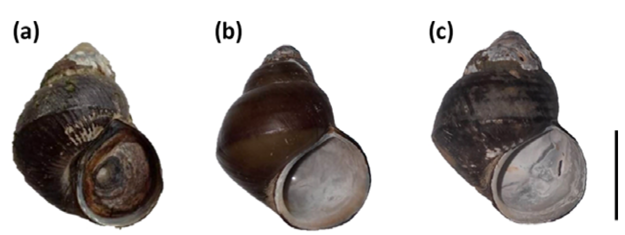

Figure 2. Photographs of aestivating snail, *Filopaludina martensi martensi*: (a) live snail; (b) dead snail; (c) old shell with erosion.

water and left for 24 hours to facilitate the release of cercariae. After this period, the snails were exposed to artificial light for approximately 3 hours to check for the shedding of cercariae. Each snail was individually screened over a period of 48 hours. Prior to the crushing method, the snails were anesthetized by freezing. The shells of the snails were then broken, and the entire body was crushed in a Petri dish. Following this, larval trematode infections were detected by microscopically examining the crushed tissue. The identification of larval trematodes was based on their morphological characteristics, as described by Schell (1962) and Yamaguti (1975), and counts were performed according to the species of snail. Additionally, the prevalence and intensity of infections were assessed, with infection intensity specifically determined by counting only the metacercariae.

Statistical analysis

Statistical analyses were conducted using SPSS v. 29.0.2. Two sets of analyses were performed. The first set employed basic descriptive statistics to summarize soil properties, with values presented as mean±S.D. Descriptive statistics were also used to summarize the prevalence and intensity of trematode infections by snail species. The second set involved correlation tests, where Pearson's correlation was applied to examine the relationship between soil properties and the survival of aestivating snails. Statistical significance was defined as $p < 0.05$.

Results

Aestivating snail abundance and diversity

We collected and examined a total of 1,159 aestivating snails from various species belonging to the families Bithyniidae, Viviparidae, Ampullariidae, Thiaridae, Planorbidae, and Lymnaeidae. Based on conchological assessments, nine species were identified (Figure 3): *Bithynia funiculata* (1.6%), *B. siamensis siamensis* (72.4%), *Filopaludina martensi martensi* (8.4%), *F. sumatrensis polygramma* (6.9%), *F. doliaris* (7.0%), *Pomacea canaliculata* (2.8%), *Melanoides tuberculata* (0.3%), *Indoplanorbis* sp. (0.6%), and *Lymnaea* sp. (<0.1%) (Table 2). Most of the aestivating snails collected belonged to the Bithyniidae, with *B. s. siamensis* being the most abundant and widespread species, present at all sampling sites. Quantification of

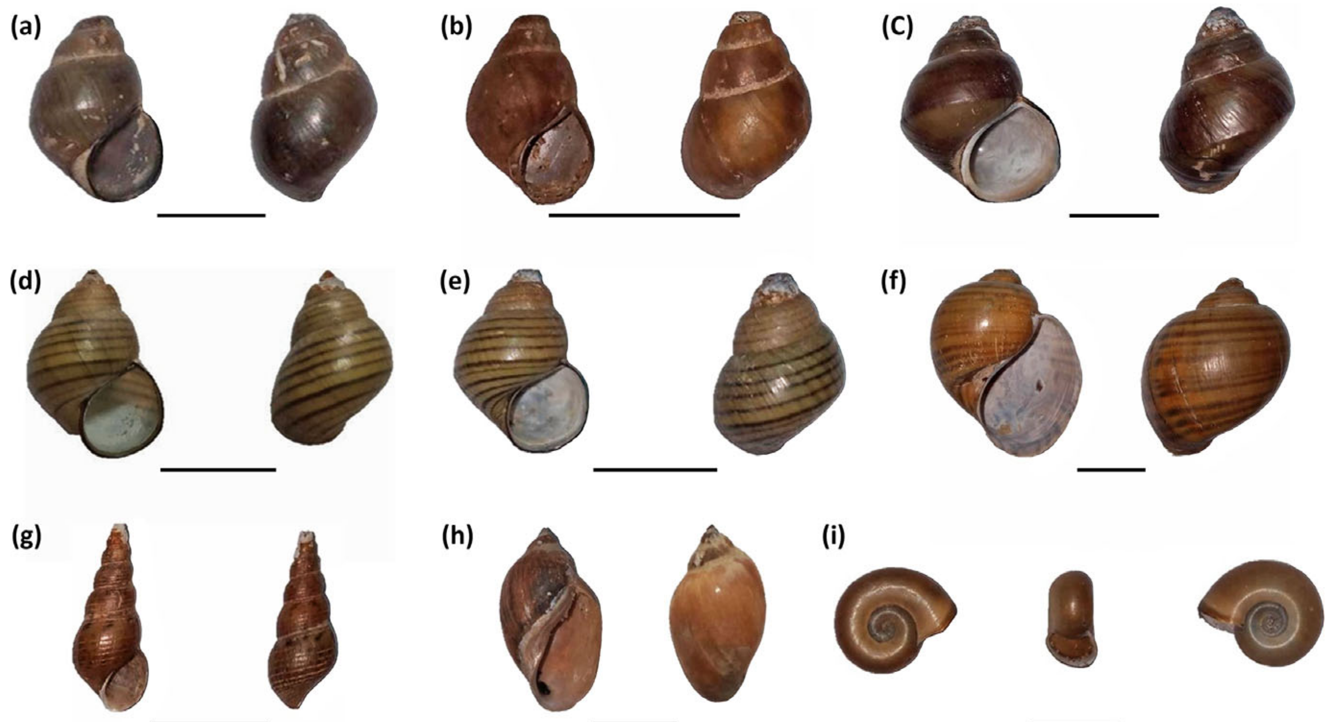

Figure 3. Shell morphology of aestivating snails collected from rice paddies during the dry season: (a) *Bithynia funiculata*; (b) *B. siamensis siamensis*; (c) *Filopaludina martensi martensi*; (d) *F. doliaris*; (e) *F. sumatrensis polygramma*; (f) *Pomacea canaliculata*; (g) *Melanoides tuberculata*; (h) *Lymnaea* sp.; (i) *Indoplanorbis* sp. Scale bar: 1 cm.

Table 2. Snail fauna, survival, and mortality (percent) of aestivating snails*

Family	Species	No. of total snail	%	No. of old shells	No. of live snail (% survival)	No. of dead snail (% mortality)
Bithyniidae	*Bithynia funiculata*	18	1.6	5	3 (23.1)	10 (76.9)
	Bithynia siamensis siamensis	839	72.4	45	647 (81.5)	147 (18.5)
Viviparidae	*Filopaludina martensi martensi*	97	8.4	39	4 (6.9)	54 (93.1)
	Filopaludina sumatrensis polygramma	80	6.9	20	11 (18.3)	49 (81.7)
	Filopaludina doliaris	81	7.0	38	8 (16.7)	35 (83.3)
Ampullariidae	*Pomacea canaliculata*	32	2.8	15	1 (5.9)	16 (94.1)
Thiaridae	*Melanoides tuberculata*	4	0.3	1	0 (0)	3 (100)
Planorbidae	*Indoplanorbis* sp.	7	0.6	0	0 (0)	7 (100)
Lymnaeidae	*Lymnaea* sp.	1	0.1	0	0 (0)	1 (100)
Total		1,159	100.0	163	674 (67.7)	322 (32.3)

*The old shells were excluded from the survival and mortality rate calculations.

species richness and diversity revealed low values of 0.932±0.375 (*D* range: 0.314–1.431) and 0.792±0.572 (*H* range: 0.072–1.741), respectively. The evenness index was also low ($J' = 0.497 \pm 0.306$; J' range: 0.066–0.865). However, some sampling sites exhibited higher values, with J' approaching 1 ($J' > 0.8$).

Survival and mortality rates of aestivating snails

Aestivating snails were found only within a depth of 0 to 5 cm from the ground surface. None were found 5–10 cm from the ground surface. After reactivating the snails in water, they were categorized into three groups: live snails (674), dead snails (322), and old shells (163). The overall survival and mortality rates, excluding the old shells, were 67.7% and 32.3%, respectively (Table 2). The survival rate of aestivating snail species ranged from 5.9% to 81.5%, with *B. s. siamensis* exhibiting the highest survival rate. In contrast, the mortality rate for all species, except *B. s. siamensis*, was greater than 70%. Specifically, *M. tuberculata*, *Indoplanorbis* sp., and *Lymnaea* sp. had a 100% mortality rate, meaning that no individuals of these species survived the aestivation period. This 100% mortality rate reflects the complete loss of individuals within these species, with all individuals perishing during aestivation.

Relationship between soil properties and survival of aestivating snails

There was considerable variation in the recorded soil properties among the sampling sites. Soil pH varied from 4.01 to 6.10

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(mean 4.88±0.75), while electrical conductivity varied between 0.0061 and 0.3248 ds/cm (mean 0.1063±0.1087). Soluble salts concentration in soil extracts also fluctuated, with values from 3.904 to 207.872 mg/l (mean 68.048±69.579), Cation or anion concentration in soil extracts ranged from 0.0061 and 0.3248 mg/l (mean 0.1063 ±0.1087). The analysis of soil properties in relation to the survival of aestivating snails revealed a positive relationship between electrical conductivity, soluble salt concentrations, and cation/anion concentrations in soil extracts with the survival of viviparid snails, including *F. m. martensi*, *F. s. polygramma*, and *F. doliaris* ($p < 0.05$). However, no significant correlations were observed for other snail species (*B. funiculata*, *B. s. siamensis*, and *P. canaliculata*) ($p > 0.1$).

Trematode infections in aestivating snails

The investigation of trematode infections in 674 aestivating snails from dry-season rice paddies revealed that none of the snails shed cercariae after being activated in water for 48 hours. Among the total snails examined, 8.46% (57/674) were infected with larval trematodes. Four types of larval trematodes were identified in this study, including three different kinds of metacercariae: echinostome metacercaria, *Thapariella anastomusa* metacercaria, and an unidentified metacercaria, as well as rediae (Figure 4).

The infection rates and the number of metacercariae recovered from various snail species are presented in Table 3. Echinostome metacercariae, which were predominantly found clumping together in the pericardial sac of the infected snails, were the most commonly identified, with an overall prevalence rate of 6.37% (43/674). Throughout the survey, a total of 224 echinostome metacercariae were recovered from bithyniid and viviparid snails, including *B. s. siamensis*, *F. m. martensi*, *F. s. polygramma*, and *F. doliaris*. The infection rates for individual species ranged from 5.24% to 100%, and the intensity of infection varied from 3.00 to 28.33 metacercariae per infected snail, with an average intensity of 5.20 metacercariae per infected snail. Metacercariae of *T. anastomusa* were found co-infecting with echinostome metacercariae, and were recovered from a single snail species, *F. doliaris*. Out of 674 snails examined, 4 (0.59%) were infected, with a mean intensity of 2.25 metacercariae per infected snail. Additionally, an unidentified metacercaria co-infection with echinostome metacercariae was detected in *B. s. siamensis*, exhibiting a prevalence of 0.3% (2/674). Rediae were also observed in this snail species, with a prevalence of 2.07% (14/674).

Discussion

This study focused on investigating the diversity of aestivating snails and their potential role in the transmission of trematodes in dry-season rice paddies in Chiang Rai Province, Thailand. We identified nine aestivating snail species across eight locations with most belonging to the family Bithyniidae. *B. s. siamensis* was the most frequently encountered species, consistent with the previous findings highlighting its abundance in rice paddies and its broad physiological and ecological tolerance (Pratumchart *et al.* 2019; Suwannatrai *et al.* 2011). In contrast, species such as *M. tuberculata*, *Indoplanorbis* sp., and *Lymnaea* sp. were less common, likely due to habitat preferences favouring stream environments (Chantima *et al.* 2018a, 2020; Krailas *et al.* 2014).

Variability in snail diversity among sites was linked to the different agricultural practices, particularly chemical applications,

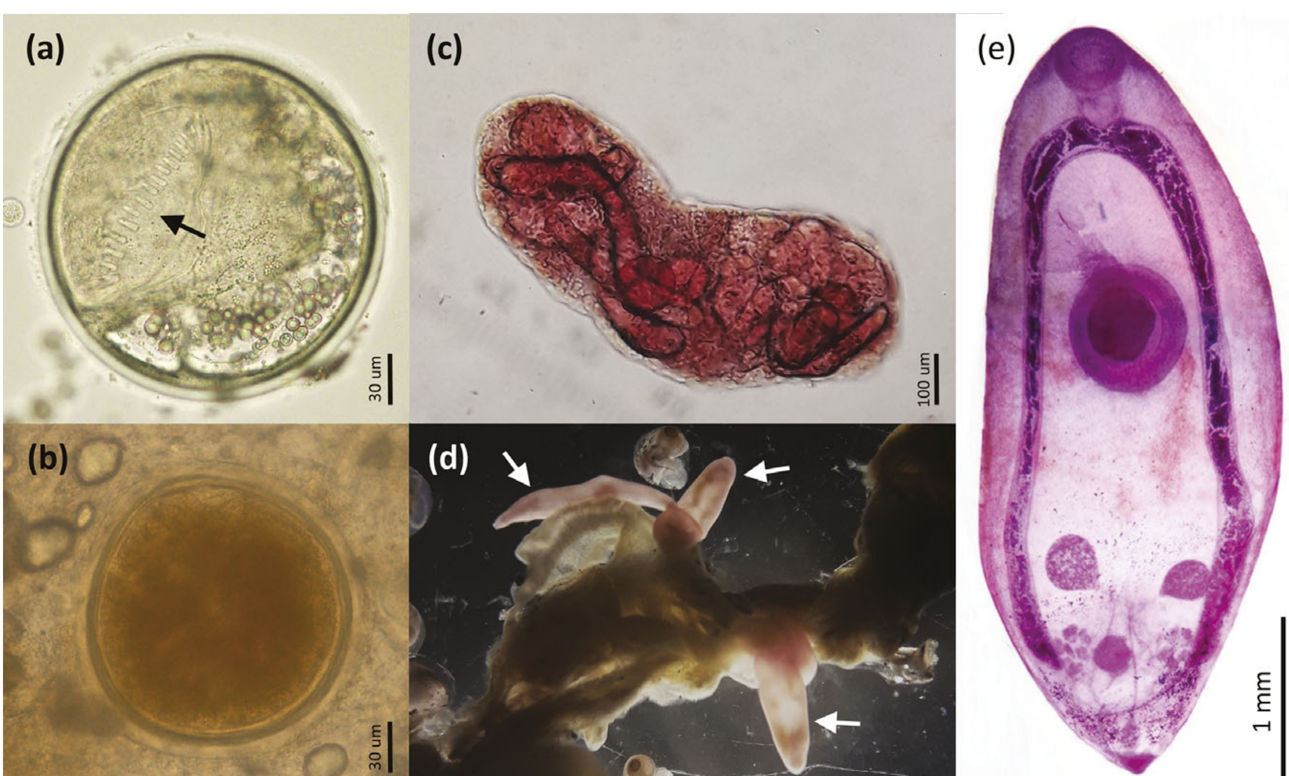

Figure 4. Morphotypes of metacercariae and redia recovered from aestivating snail: (a) An isolated echinostome metacercaria showing head collar with collar spines (arrowhead); (b) An isolated unidentified metacercaria; (c) Redia stained with 0.5% neutral red; (d) *Thapariella anastomusa* metacerariae were free, not encysted in the tissue of snails (arrowhead); (e) An isolated *T. anastomusa* metacercaria stained with hematoxylin.

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Table 3. Prevalence and intensity of trematode infections in aestivating snails

Snail species	No. of snail examined	Echinostome metacercaria		*Thapariella anastomusa*		Unidentified metacercaria		Redia
		No. of snail infected (% prevalence)	No. of metacercaria (intensity)	No. of snail infected (% prevalence)	No. of metacercaria (intensity)	No. of snail infected (% prevalence)	No. of metacercaria (intensity)	No. of snail infected (% prevalence)
B. funiculata	3	0	0	0	0	0	0	0
B. s. siamensis	647	34 (5.24)	133 (3.91)	0	0	2 (0.31)	3 (1.5)	14 (2.16)
F. m. martensi	8	2 (28.57)	6 (3.00)	0	0	0	0	0
F. s. polygramma	11	3 (27.27)	85 (28.33)	0	0	0	0	0
F. doliaris	4	4 (100)	20 (5.00)	4 (100)	9 (2.25)	0	0	0
P. canaliculata	1	0	0	0	0	0	0	0
Total	674	43 (6.37)	224 (5.20)	4 (0.59)	9 (2.25)	2 (0.30)	3 (1.5)	14 (2.07)

which may have molluscicidal effects (Min *et al.* 2020; Qiu *et al.* 2020). Seasonal changes in rice paddies, including paddy preparation and harvesting, also influence snail populations. The golden apple snail, *P. canaliculata*, was notably scarce, contrasting with its widespread abundance in other rice-growing regions of Thailand (Chaichana and Sumpan 2015; Ng *et al.* 2020). Since its introduction in the 1980s, this species has significantly impacted rice cultivation (Chanyapeth and Achawakhom 1998), yet local factors may limit its proliferation in our study area.

In our study, four snail species, *B. s. siamensis*, *F. m. martensi*, *F. s. polygramma*, and *F. doliaris*, were infected with larval trematodes. These species, common in northern Thailand's agricultural areas, are important vectors of trematode transmission to humans and animals (Chantima *et al.* 2013; Chantima *et al.* 2018a; Chantima and Rika 2020; Phalee *et al.* 2018). Given their widespread distribution in rice paddies, managing aestivating snails requires an integrated approach that considers both their ecological role and their significance in parasitic transmission.

During the dry season, the snails encounter extreme environmental conditions. They must endure aestivation and burrowing into the mud, which is a physiological process like hibernation. However, aestivation occurs in times of drought and renders respiration difficult. This study found that almost all collected aestivating snails belonged to the family Bithyniidae, with *B. s. siamensis* as the most common species found at a soil depth of 0–5 cm. Our findings indicate that snails could bury themselves in depths up to 5 cm, in contrast to Brockelman *et al.* (1986) and Chaiyaseang *et al.* (2019), who noted that rapid habitat desiccation prevents snails from preparing for aestivation in deeper soil. The average survival rate of aestivating snails over a three-month period was high (67.7%), whereas Brockelman *et al.* (1986) reported a survival rate of under 10% during a 16-month dry period in a non-irrigated area. Our study, however, focused on a three-month aestivating period, yielding a survival rate exceeding 60%. This finding is consistent with the report by Chaiyaseang *et al.* (2019), which also indicated that bithyniid snails (*B. s. goniomphalos*) exhibited a higher survival rate during a shorter dry period of four months in irrigated rice plantations. Consequently, rice cultivation during the dry season in irrigated areas may enhance the survival of snail hosts, resulting in an increase in snail populations (Wang *et al.* 2015). However, results from other studies have demonstrated that effective water management in irrigation schemes can reduce snail populations (Chandiwana *et al.* 1998), highlighting the importance of manipulating snail habitats in combination with mass drug administration programs to more effectively reduce trematode transmission (Fenwick *et al.* 2009; Wang *et al.* 2009).

The ecology of aestivating snails is potentially influenced by land use practices, particularly in rice paddies. These environments undergo dynamic changes through different stages: irrigation, full growth, complete drying, and occasional burning throughout the rice planting seasons. Farmers in northern Thailand generally cultivate rice once or twice annually. After harvesting, the most common method for land clearing and rice straw removal is open burning. Dead snails have been found in rice paddy sampling locations where burning has occurred. This practice may impact snail populations and disrupt the trematode life cycle, consequently affecting the transmission of associated diseases. However, open burning does not completely eradicate snail intermediate hosts in rice paddies. Some snail species can burrow deep into the soil (Chaiyaseang *et al.* 2019) or re-enter paddies from adjacent habitats via irrigation systems (Wang *et al.* 2011, 2015).

The study on the relationship between soil properties and the survival of aestivating snails revealed a significant positive correlation with the survival of viviparid snails. Conversely, no significant relationship was observed for bithyniid snails. A previous study indicated that salinity is the most important environmental variable affecting the density and distribution of *Bithynia* intermediate host snails of *O. viverrini* in the Khorat basin (Suwannatrai *et al.* 2011). Currently, studies are underway to determine whether differences in the survival of snail intermediate hosts related to soil properties correspond to variations in trematode infection prevalence. However, due to the limited number of soil property parameters analysed, no conclusive results regarding the association between soil properties and the survival of aestivating snails could be established. Future investigations should focus on collecting a broader array of soil property parameters to achieve adequate data for analysing the survival of aestivating snails.

This study investigated trematode infections in aestivating snails from rice paddies during the dry season. Over 48 hours, no cercarial shedding was observed, aligning with Chaiyaseang *et al.* (2019) who also found no shedding after reactivating aestivating snails. The absence of cercarial shedding could be due to several factors. First, the dry season induces aestivation in snails, slowing metabolism and potentially interfering with parasite development and cercarial release. Second, the stress of limited nutrients and prolonged aestivation may lead to higher mortality rates in infected snails, delaying parasite development or causing parasite death. It is also possible that both the snails and trematodes enter dormancy during

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aestivation, with parasites ceasing cercarial production while the snails are metabolically inactive. Alternatively, the snails may have already reached an advanced stage of infection, with metacercariae encysted before the dry season, and cercarial shedding may have occurred earlier. These hypotheses suggest that environmental stress and metabolic changes during aestivation play a critical role in the dynamics of trematode infections, highlighting the need for further research to understand the timing and environmental influences on parasite development.

Although no cercarial infections were found, this study identified four types of larval trematodes: echinostome metacercariae, *T. anastomusa* metacercariae, unidentified metacercariae, and rediae in bithyniid and viviparid snails. Previous reports have also found echinostome metacercariae in these snail families in Thailand (Burch and Lohachit 1983; Chantima *et al.* 2013, 2018a, 2018b; Chantima and Rika 2020; Mard-arhin *et al.* 2001). Additionally, these snail species have been reported as second intermediate hosts for Echinostomatidae in various areas of Southeast Asia (Chai *et al.* 2011; Madsen and Hung 2014).

Metacercariae of *T. anastomusa* were found exclusively in *F. doliaris*, consistent with previous reports in viviparid snails (*Filopaludina* spp.) from Thailand (Chantima *et al.* 2018a, 2018b; Chantima and Rika 2020; Phalee *et al.* 2018). Earlier studies also recorded *T. anastomusa* in *Bellamya bengalensis* (Viviparidae) in India (Agrawal *et al.* 2002; Rai and Pande 1967; Srivastava 1953), with its adult stage found in the oesophagus of birds (Agrawal 1958; Prudhoe 1957). The occurrence of *T. anastomusa* in multiple snail species suggests a broader host range, with birds likely serving as definitive hosts. Since birds commonly consume these snails, they may play a key role in its transmission and geographic spread. Further research is needed to clarify its host specificity, life cycle, and potential avian hosts in the region.

This study recorded a low number of infected aestivating snails with larval trematodes. Previous research has shown that the prevalence of larval trematode infections in snails is closely linked to snail population density (Lively 2001). Recent studies suggest that anthropogenic factors such as pollution, habitat modification, and the introduction of non-native species significantly influence larval trematode infections in freshwater snails. These factors can alter the dynamics of trematode infections within snail populations, as well as affect the availability of potential hosts or environmental conditions in their habitats. (Bachtel *et al.* 2019; Kuris and Lafferty 1994). Understanding these interactions is crucial for assessing the ecological implications of trematode infections in freshwater ecosystems. To gain further insights into the relationship between larval trematodes and their snail intermediate hosts during the aestivated state, additional experimental studies are needed. Such studies should focus on controlling the timing of infection and aestivation. It is also recommended that future research explore the experimental aestivation of snails, both with and without parasitic infections, to enhance our understanding of this phenomenon.

Conclusion

This study focused on the diversity of aestivating snails and their potential role in the transmission of trematodes in rice paddies during the dry season. Our findings highlight that burrowing behaviour played a crucial role in the survival of bithyniid snails, leading to high abundance of aestivating individuals. Additionally, we identified various larval trematodes in bithyniid and viviparid snails, suggesting that aestivation may facilitate the persistence of trematode life cycles in seasonal rice paddy ecosystems. Given the high survival rate of aestivating snails, recolonization after the rainy season may contribute to the continued transmission of trematodes. While habitat modification and targeted snail control have been proposed as potential strategies for trematode management, their effectiveness in seasonal rice paddy ecosystems remains uncertain. Future research should prioritize field-based experimental studies on aestivation dynamics and their impact on snail population regulation and trematode transmission.

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Competing interest. The authors declare that there are no conflicts of interest.

Ethical standard. We followed the guidelines for animal care in the International Guiding Principles of Biomedical Research Involving Animals (Council for International Organizations of Medical Sciences: CIOMS) including the relevant document (U1-03137-2559 to KC; U1-09340-2564 to KS).

References

Agrawal SM (1958) Further observations on *Thapariella anstastomusa* Srivastava, 1953 (Trematoda: Thapariellidae). *Indian Journal of Helminthology* **10**, 40–45.

Agrawal N, **Pandey KC and Kiran GB** (2002) Record of metacercaria of *Thapareilla anastomusa* Srivastava, 1958, from *Bellamya bengalensis* (L.). *Indian Journal of Helminthology* **20**, 35–38.

Bachtel RZ, **Rittenhouse M**, **Sandland GJ and Koop JAH** (2019) Infection patterns of trematodes across size classes of an invasive snail species using field and laboratory studies. *Parasitology* **146**(**4**), 438–444.

Badger LI and Oyerinde JPO (1996) *Schistosoma mansoni*: Effect of aestivation on the intra-molluscan stages and the survival rate of infected *Biomphalaria pfeifferi*. *Annals of Tropical Medicine and Parasitology* **90**(**6**), 617–620.

Brandt RAM (1974) The non-marine aquatic mollusca of Thailand. *Archiv fur Molluskenkunde* **105**, 1–423.

Brewer R (1994) *The Science of Ecology*, 2nd edn. New York: Saunders College.

Brockelman WY, **Upatham ES**, **Viyanant V**, **Ardsungnoen S and Chantanawat R** (1986) Field studies on the transmission of the human liver fluke, *Opisthorchis viverrini*, in northeast Thailand: Population changes of the snail intermediate host. *International Journal of Parasitology* **16**(**5**), 545–552.

Burch JB and Lohachit C (1983) Snail of medical importance in Thailand. *Walkerana Transactions of the POETS Society* **1**, 395–398.

Chai JY, **Sohn WM**, **Na BK and De NV** (2011) *Echinostoma revolutum*: metacercariae in *Filopaludina* snails from Nam Dinh Province, Vietnam, and adults from experimental hamsters. *Korean Journal of Parasitology* **49**(**4**), 449–455.

Chai JY, **Murrell KD and Lymbery AJ** (2005) Fish-borne parasitic zoonoses: Status and issues. *International Journal of Parasitology* **35**(**11–12**), 1233–1254.

Chai JY, **Shin EH**, **Lee SH and Rim HJ** (2009) Food-borne intestinal flukes in Southeast Asia. *Korean Journal of Parasitology* **47**(**Suppl**), 69–102.

Chaichana R and Sumpan T (2015) Environmental tolerance of invasive golden apple snails (*Pomacea canaliculata* (Lamarck, 1822)) and Thai native apple snails (*Pila scutate* (Mousson, 1848)). *Tropical Ecology* **56**(**3**), 347–355.

Chaiyasaeng M, **Pechdee P**, **Sereewong C**, **Suwannatrai A**, **Laha T and Tesana S** (2019) Effects of aestivation on survival of *Bithynia siamensis goniomphalos* snails and the infection of *Opisthorchis viverrini* in the irrigation area of wet- and dry-season rice paddy. *Acta Tropica* **192**(**Suppl. 4**), 55–60.

Chandiwana SK, **Taylor P**, **Chimbari M**, **Ndhlovu P**, **Makura O**, **Bradley M and Gondo P** (1998) Control of schistosomiasis transmission in newly established smallholder irrigation schemes. *Transactions of The Royal Society of Tropical Medicine and Hygiene* **82**(**6**), 874–880.

Chantima K and Rika C (2020) Snail-borne zoonotic trematodes in edible viviparid snails obtained from wet markets in Northern Thailand. *Journal of Helminthology* **94**, E198.

Chantima K, **Chai JY and Wongsawad C** (2013) *Echinostoma revolutum*: Freshwater snails as the second intermediate hosts in Chiang Mai, Thailand. *Korean Journal of Parasitology* **51**(**2**), 183–189.

Chantima K, **Suk-ueng K and Kampan M** (2018a) Freshwater snail diversity in Mae Lao agricultural basin (Chiang Rai, Thailand) with a focus on larval trematode infections. *Korean Journal of Parasitology* **56**(**3**), 247–257.

Chantima K, **Phromkhong K**, **Sonyod N and Chaichana N** (2018b) Occurrence of larval trematode in freshwater snail from Pa Ko Dam Sub-district, Mae Lao District, Chiang Rai Province. In McCormick K, Mungthaisong S, Khittasangka M, Chomchalow N, Walsh JC and Lisec A (eds), *ASIAN Community Knowledge Networks for the Economy, Society, Culture, and Environmental Stability*. Chiang Rai: Chiang Rai Rajabhat University, 84–88.

Chantima K, **Lekpet S**, **Butboonchoo P and Wongsawad C** (2020) Diversity and abundance of gastropods in relation to physio-chemical parameters in rice paddies, Chiang Rai Province, Thailand. *Agricultural and Natural Resources* **54**(**3**), 295–300.

Chanyapeth CH and Achawakhom T (1998) Golden apple snails. In *Proceedings of a Seminar on the Golden Apple Snail*. Bangkok: Entomology and Zoology Division, Department of Agriculture, and the Entomology and Zoology Association of Thailand. 3.1–3.15.

Clarke KR and Warwick RM (1994) *Change in Marine Community: An Approach to Statistical Analysis and Interpretation*. Plymouth: Plymouth Marine Laboratory.

Eastham J, **Mpelasoka F**, **Mainuddin M**, **Ticehurst C**, **Dyce P**, **Hodgson G**, **Ali R and Kirby M** (2008) *Mekong River Basin Water Resources Assessment: Impacts of Climate Change*. Australia: CSIRO: Water for a Healthy Country National Research Flagship.

Fenwick A, **Webster JP**, **Bosque-Oliva E**, **Blair L**, **Fleming F**, **Zhang Y**, **Garba A**, **Stothard J**, **Gabrielli AF and Clements A** (2009) The Schistosomiasis Control Initiative (SCI): Rationale, development and implementation from 2002-2008. *Parasitology* **136**(**13**), 1719–1730.

Geo-Informatics and Space Technology Development Agency (Public Organization) (GISTDA) (2011) *L05_AdminBoundary_Province_2011_50k_FGDS_Beta* [Shapefile]. Bangkok: Geo-Informatics and Space Technology Development Agency (Public Organization).

Github (2024) World_Countries [Shapefile]. Available at https://github.com/prasertcbs/thailand_gis/tree/main/world_map Accessed 16 April 2024.

Hill D, **Fasham M**, **Tucker G**, **Shewry M and Shaw P** (2005) *Handbook of Biodiversity Methods: Survey, Evaluation and Monitoring*. Cambridge: Cambridge University Press.

Hillebrand H (2008) Dominance. In Jorgensen SE and Fath B (eds), *Encyclopedia of Ecology*. Oxford: Elsevier, 938–944.

Krailas D, **Namchote S**, **Koonchornboon T**, **Dechruksa W and Boonmekam D** (2014) Trematodes obtained from the thiarid freshwater snail *Melanoides tuberculata* (Müller, 1774) as vector of human infections in Thailand. *Zoosystematics and Evolution* **90**(**1**), 57–86.

Kuris A and Lafferty KD (1994) Community structure: Larval trematodes in snail hosts. *Annual Review of Ecology, Evolution, and Systematics* **25**, 189–217.

Lively CM (2001) Trematode infection and the distribution and dynamics of parthenogenetic snail populations. *Parasitology* **123**(**7**), 19–26.

Mard-arhin N, **Prawang T and Wongsawad C** (2001) Helminths of freshwater animals from five provinces in Northern Thailand. *Southeast Asian Journal of Tropical Medicine and Public Health* **32**(**Suppl. 2**), 206–214.

Madsen H and Hung NM (2014) An overview of freshwater snails in Asia with main focus on Vietnam. *Acta Tropica* **140**(**Pt B**), 105–117.

Min F, **Wang J**, **Liu X**, **Yuan Y**, **Guo Y**, **Zhu K**, **Chai Z**, **Zhang Y and Li S** (2020) Environmental factors affecting freshwater snail intermediate hosts in Shenzhen and adjacent region, South China. *Tropical Medicine and Infectious Disease* **7**(**12**), 426.

Ng TH, **Annate S**, **Jeratthitikul E**, **Sutcharit C**, **Limpanont Y and Panha S** (2020) Disappearing apple snails (Caenogastropoda: Ampullariidae) of Thailand: A comprehensive update of their taxonomic status and distribution. *Journal of Molluscan Studies* **86**(**4**), 290–305.

Osborne TR and Wright JC (2018) Seeking refuge in subsurface microhabitats during aestivation aids avoidance of lethally high temperature and desiccation in the snail *Helminthoglypta tudiculata* (Binney, 1843) (Pulmonata: Helminthoglyptidae). *Journal of Molluscan Studies* **84**(**2**), 132–140.

Phalee W, **Phalee A and Wongsawad C** (2018) New record of *Thapariella anastomusa* (Trematoda: Thapariellidae) metacercariae in Northern Thailand. *Korean Journal of Parasitology* **56**(**1**), 49–52.

Prudhoe S (1957) Trematoda: Exploration du Parc National de l'Upemba. *Institut des Parcs Nationaux du Congo Belge* **48**, 1–28.

Pratumchart K, **Suwannatrai K**, **Sereewong C**, **Thinkhamrop K**, **Chaiyos J**, **Boonmars T and Suwannatrai AT** (2019) Ecological niche model based on maximum entropy for mapping distribution of *Bithynia siamensis goniomphalos*, first intermediate host snail of *Opisthorchis viverrini* in Thailand. *Acta Tropica* **193**(**5**), 183–191.

Qiu J, **Li R**, **Zhu H**, **Xia J**, **Xiao Y**, **Huang D and Wang Y** (2020) The effect of ecological environmental changes and mollusciciding on snail intermediate host of *Schistosoma* in Qianjiang city of China from 1985 to 2015. *Parasites Vectors* **13**(**1**), 397.

Rai DN and Pande BP (1967) On the metacercariae of *Thapariella anastomusa* Srivastava, 1955. *Indian Journal of Helminthology* **19**, 64–69.

Rubaba O, **Chimbari MJ and Mukaratirwa S** (2016) The role of snail aestivation in transmission of schistosomiasis in changing climatic conditions. *African Journal of Aquatic Science* **41**(**2**), 143–150.

Schell SC (1962) *Parasitology Laboratory Manual*. New York: Wiley.

Sparks DL, **Page AL**, **Helmke PA**, **Loeppert RH**, **Soltanpour PN**, **Tabatabai MA**, **Johnston CT and Sumner ME** (1996) Methods of Soil Analysis. Part *3* - Chemical Methods. Washington, DC: American Society of Agronomy, Inc.

Sri-Aroon P (2011) *Freshwater Snails of Medical Importance in Thailand*. Thailand: Mollusk Museum, Applied Malacology Center, Department of Social and Environmental Medicine, Mahidol University, 6–8.

Srivastava NN (1953) On the morphology and systematic position of a new avian trematode *Thapariella anastomusa* n.g., n.sp., with a suggestion on the creation of a new family Thapariellidae. *Indian Journal of Helminthology* **5**, 101–108.

Suwannatrai A, **Suwannatrai K**, **Haruay S**, **Piratae S**, **Thammasiri C**, **Khampoosa P**, **Kulsantiwong J**, **Prasopdee S**, **Tarbsripair P**, **Suwanwerakamtorn R**, **Sukchan S**, **Boonmars T**, **Malone JB**, **Kearney MT and Tesana S** (2011) Effect of soil surface salt on the density and distribution of the snail *Bithynia siamensis goniomphalos* in northeast Thailand. *Geospatial Health* **5**(**2**), 183–190.

Upatham ES, **Sornmani S**, **Kitikoon V**, **Lohachit C and Burch JB** (1983) Identification key for fresh-and brackish-water snails of Thailand. *Malacological Review* **16**, 107–136.

U.S. Salinity Laboratory Staff (1954) *Diagnosis and Improvement of Saline and Alkaline Soils* (Agriculture Handbook No. 60). Washington, DC: United States Government Printing Office.

Wang LD, **Chen HG**, **Guo JG**, **Zeng XJ**, **Hong XL**, **Xiong JJ**, **Wu XH**, **Wang XH**, **Wang LY and Xia G** (2009) A strategy to control transmission of *Schistosoma japonicum* in China. *The New England Journal of Medicine* **360**(**2**), 121–128.

Wang YC, **Feng CC**, **Sithithaworn P**, **Feng Y and Petney TN** (2011) How do snails meet fish? Landscape perspective needed to study parasite prevalence. *Ecohealth* **8**(**3**), 258–260.

Wang YC, **Ho RC**, **Feng CC**, **Namsanor J and Sithithaworn P** (2015) An ecological study of *Bithynia* snails, the first intermediate host of *Opisthorchis viverrini* in northeast Thailand. *Acta Tropica* **141**(Pt B), 244–252.

Washington HG (1984) Diversity, biotic and similarity indices. *Water Research* **18**, 653–694.

Woodruff DS and Upatham ES (1993) Snail transmitted diseases of medical and veterinary importance in Thailand and the Mekong valley. *Journal of Medical and Applied Malacology* **4**, 1–12.

Yamaguti S (1975) *A Synoptical Review of Life Histories of Digenetic Trematodes of Vertebrates*. Kyoto: Keigaku Publishing.

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