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PREVALENCE OF ZOONOTIC TREMATODES IN FISH FROM A VIETNAMESE FISH-FARMING COMMUNITY

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ABSTRACT: The prevalence of fish-borne zoonotic trematode (FZT) metacercariae was investigated in fish farmed by rural households in Nghe An Province, located in northern Vietnam. In total, 716 fish, including tilapia (*Oreochromis niloticus*) and 6 carp species, i.e., grass carp (*Ctenopharyngodon idellus*), bighead carp (*Aristichthys nobilis*), mrigal (*Cirrhinus mrigala*), common carp (*Cyprinus carpio*), silver carp (*Hypophthalmichthys molitrix*), and roha (*Labeo rohita*), collected from 53 fish farms were examined. The overall prevalence of FZT metacercariae was 44.6%, ranging from 12.5% to 61.0% in fish species collected from grow-out ponds, which are the production system for growing fish from fingerling size to market size. The overall prevalence was 43.6% in fingerlings cultured in nurseries, ranging from 7.4% to 62.8% for different fish species. The FZT species recovered were heterophyids and echinostomatids and included *Haplorchis pumilio*, *H. taichui*, *H. yokogawai*, *Centrocestus formosanus*, *Stellantchasmus falcatus*, and *Echinochasmus japonicus*, all of which are intestinal flukes in humans, other mammals, and birds. This is the first report of *H. yokogawai* and *E. japonicus* in fish in Vietnam, and the first record for *S. falcatus* in northern Vietnam. Although a previous cross-sectional survey of the people living in these fish farm households revealed a very low prevalence of FZTs (<1%), our results demonstrate that intestinal flukes are common in farmed fish in this area, suggesting that reservoir hosts such as dogs, cats, and pigs are more important in sustaining the life cycles of these flukes in fish farms than human hosts. This has implications for the effectiveness of control programs focused mainly on treatment of humans.

Fish-borne zoonotic trematodes (FZTs), including liver and intestinal flukes, are significant public health problems worldwide, especially in Asian countries (WHO, 1995, 2004; Chai, Murrell, and Lymbery, 2005; Keiser and Utzinger, 2005; Tesana, 2005; Yoshida, 2005). The number of people currently infected with FZTs was recently estimated by the World Health Organization (WHO) to exceed 18 million; however, the number of people at risk worldwide is more than 500 million (WHO, 2004). Recent figures suggest that about 1.5 million people in Korea, 6 million people in China, and over 5 million in Thailand are infected with liver flukes (*Clonorchis sinensis* or *Opisthorchis viverrini*) (Chai, Murrell, and Lymbery, 2005; Tesana, 2005; Yoshida, 2005). However, many of the numerous species of intestinal heterophyids and echinostomatids are also important, but less well recognized, compared to liver flukes, fish-borne zoonoses in China (Yu and Xu, 2005), Korea (Chai, 2005), Thailand (Waikagul and Radomyos, 2005), and Laos (Chai, Park et al., 2005). Although epidemiological data are scarce, the prevalence and species diversity of FZTs in Vietnam suggests that these parasites are also an important national public health problem (De et al., 2003). *Clonorchis sinensis* is reported from 9 northern provinces of Vietnam, with human prevalence ranging from 0.2% in Thai Binh Province to 26% in Nam Dinh Province. In contrast, *O. viverrini* has been reported only from 3 southern provinces, with prevalences ranging from 0.3% in Da Nang Province to more than 10% in Phu Yen Province (De et al., 2003). Only recently have FZT metacercariae been reported in Vietnamese fish (Hop et al., 2007; Thien et al., 2007; Thu et al., 2007).

Information on the status of FZTs in Vietnamese fish is highly important because of the importance of aquaculture to the economy and national nutritional needs. Freshwater fish pro-

duction in Vietnam has increased 9.3-fold, from 41,750 tons in 1962 to 390,000 tons in 2005 (Keiser and Utzinger, 2005). As part of the growing importance of aquaculture in Vietnam, various programs have been implemented to encourage the development of household-scale freshwater aquaculture. In Nghe An Province, for example, freshwater aquaculture is highly developed, and fish production has increased from 7,800 tons in 2000 to 13,382 tons in 2004 (data obtained from 2005 records of the Nghe An Province Fisheries Department). Expansion of aquaculture production may inadvertently increase the risk of FZTs in fish because of integration of animal production into fish rearing, which is very common in household-based culture systems. For economic reasons there is frequent reliance on the use of human and animal manure for pond fertilization, which could increase the risk of FZT egg contamination of the aquatic environment (WHO, 2004). This may also promote the proliferation of snail host populations and facilitate transmission of these trematode parasites to fish. Although surveys for human FZT infection in eastern Nghe An Province have indicated a low prevalence (De et al., 2003; Olsen et al., 2006), the status of FZT infection in farmed fish was of interest to assess the food safety risk of fish produced in this major aquaculture area.

MATERIALS AND METHODS

Study area and sampling design

A cross-sectional survey for zoonotic metacercariae in farmed fish was conducted from May 2005 to August 2005 in Nghe An Province, located about 300 km south of Hanoi. Five districts in the eastern section of the province (Tan Ky, Thanh Chuong, Yen Thanh, Nam Dan, and Hung Nguyen) were selected for fish sampling because of their highly developed aquaculture. Fifty farms were selected from a total of 1,281 fish farms present in these districts. Because the number of farms in the 5 districts was not equal, farms for sampling were selected by a random proportional sampling design. For example, Thanh Chuong district represented 20% of the total number of farms, and, therefore, 10 farms were randomly selected. The number of farms selected in each district was Thanh Chuong (10/263 farms), Nam Dan (15/384 farms), Tan Ky (8/199 farms), Hung Nguyen (8/196 farms), and Yen Thanh (9/239 farms). A previous human prevalence survey of the farm households in these districts identified 9 farms with cases of human FZT infection (Olsen et al., 2006), and these farms were also included in the study bringing the total to 59 study sites; however, only 53 farms were sampled because 6 of the 59 farms did not have fish in their ponds at the

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TABLE I. Number and species of fish examined in Nghe An Province.

Fish species	No. fish examined (%)	Origin of fish	
		Grow-out pond (%)	Nursery pond (%)
Bighead carp, <i>Aristichthys nobilis</i>	15 (2.1)	11 (1.5)	4 (0.6)
Common carp, <i>Cyprinus carpio</i>	135 (18.9)	89 (12.5)	46 (6.4)
Grass carp, <i>Ctenopharyngodon idellus</i>	148 (20.7)	105 (14.7)	43 (6.0)
Mrigal, <i>Cirrhinus mrigala</i>	38 (5.3)	22 (3.1)	16 (2.2)
Rohu, <i>Labeo rohita</i>	50 (7.0)	23 (3.2)	27 (3.8)
Silver carp, <i>Hypophthalmichthys molitrix</i>	107 (14.9)	72 (10.0)	35 (4.9)
Tilapia, <i>Oreochromis niloticus</i>	81 (11.3)	64 (8.9)	17 (2.4)
SRS*	142 (19.8)	122 (17.0)	20 (2.8)
Total (%)	716	508 (70.9)	208 (29.1)

* SRS = Self-recruiting species, includes crucian carp (*Carassius auratus*), anabas (*Anabas testudineus*), snakehead (*Ophiocephalus maculatus*), sailfish (*Istiophorus ssp.*), featherback (*Notopterus notopterus*), owsianka (*Leucaspis delineatus*), hemiculter (*Hemiculter leucisculus*), loach (*Mastacembelus armatus*), mai (*Rasbora lineatus*), macropodus (*Macropodus opercularis*), red-eyed carp (*Squaliobarbus curriculus*), and 3 unidentified species.

time of the survey. Two fish production systems, i.e., nursery ponds and grow-out ponds, were included in the study. Nursery ponds were used to grow fish from larvae (1 cm in length) or fry (30 days after hatch) to about 100 g in weight; fish density in these ponds was usually very high. After 2 to 4 mo, the fish were supplied to grow-out pond operators who cultured the fish at low density until harvesting and marketing. Fish were collected from each farm using a seine net or scoop net dragged through the pond up to 3 times.

Examination of fish for metacercariae

Fish species cultured in the farms sampled included grass carp (*Ctenopharyngodon idellus*), bighead carp (*Aristichthys nobilis*), mrigal (*Cirrhinus mrigala*), common carp (*Cyprinus carpio*), silver carp (*Hypophthalmichthys molitrix*), rohu (*Labeo rohita*), and tilapia (*Oreochromis niloticus*). Other species of fish not intentionally stocked by the farmer often occurred in ponds and were defined as self-recruiting species (SRS), a term used to describe aquatic animals that can be harvested from farmer-managed systems without intentional stocking. When these species were obtained during sampling, they were retained and examined for the presence of metacercariae.

After collection, the fish were placed on ice and transported to the laboratory of the Research Institute for Aquaculture no. 1 station in Cua Lo, Vietnam, 30–150 km from the study sites, where they were processed for recovery of metacercariae; transportation time was normally between 1 and 6 hr. Fish were held at 4 °C in a refrigerator until processed, within 1 wk of collection. In the laboratory the length and weight of each fish was measured and recorded. Because the prevalence of zoonotic metacercariae in fish from Thanh Chuong district farms was low, the entire fish was digested. For fish from the other 4 districts, however, fish weighing less than 100 g were digested whole; fish weigh-

ing more than 100 g were divided into 5 subsections (fish head, gill, muscles, fin, skin, and scales; 10–20 g/section). The fish tissue was subjected to 1% pepsin digestion to release metacercariae (WHO, 1995 [Annex 6]; Thu et al., 2007). The sediment from the digest was washed and allowed to resettle in 0.85% saline, and examined using a dissecting microscope. Any metacercariae present were separated, counted, and identified by mounting on a glass slide under a cover slip and viewing with a compound ($\times 100$) or a dissecting microscope. When encysted metacercariae were not readily identifiable, they were excysted by either physical pressure (pressing on the cover slip) or by placing them in trypsin digestion fluid (0.4% sodium hydrogen carbonate, 1.0% trypsin, 0.85% NaCl) until they emerged from the cyst and could be examined in an extended condition under a microscope. Identification of the metacercariae was according to morphological features detailed in Yamaguti (1971), Velasquez (1973), Pearson and Ow-Yang (1982), Scholtz et al. (1991), and Kaewkes (2003).

Data analysis

Data were entered into a Microsoft Excel datasheet. Microsoft Excel and Quantitative Parasitology software (Rozsa et al., 2000) were used for the descriptive statistics, following the terms recommended for parasite population and community ecology, e.g., prevalence, density, abundance, and intensity (Bush et al., 1997). Mean values for parasite prevalence and density are expressed as the mean, with 95% confidence intervals. Density of metacercariae is expressed as number of metacercariae/g fish digested. Fish weight and length averages are expressed as mean \pm SD. Sigmasat (3.00 SPSS Inc., Chicago, IL) was used to compare differences in prevalence and density using Kruskal Wallis 1-way ANOVA on ranks ($P \leq 0.05$ was considered significant).

RESULTS

Species of fish and fish sizes collected in each fish culture system

In total, 716 fish from the 53 fish farms, including 41 grow-out ponds (508 fish) and 12 nursery ponds (208 fish), were collected and examined for FZT metacercariae. The randomly caught fish from each pond type included both cultured and self-recruiting species (Table I). The mean \pm SD weight of the sampled fish was 41.1 ± 54.8 g for nursery ponds, and 97.4 ± 153.2 g for grow-out ponds. The mean length \pm SD was 13.0 ± 5.7 cm for fishes from nursery ponds, and 16.4 ± 9.7 cm for grow-out ponds.

Comparisons by districts

The prevalence of metacercariae was significantly different among the 5 districts (Table II). Prevalence of metacercariae in

TABLE II. Comparison of FZT prevalence in fish sampled from different districts and fish culture systems of Nghe An Province.

District and culture system	No. fish infected/ no. fish examined	% Prevalence (95% CI)
District sampled		
Thanh Chuong	28/228	12.3 (8.5–17.3)
Nam Dan	109/197	55.3 (48.2–62.2)
Yen Thanh	77/97	79.4 (70.2–86.7)
Tan Ky	33/91	36.3 (26.8–46.7)
Hung Nguyen	73/103	70.9 (61.2–79.2)
Culture system		
Grow-out pond	221/508	43.5 (39.2–47.9)
Nursery pond	99/208	47.6 (40.9–54.6)
Total	320/716	44.7 (41.1–48.4)

TABLE III. Prevalence and infection density of FZTs in different species of cultured fish, excluding SRS.

Fish species	No.	% Prevalence (95% CI)	Mean density (95% CI)	Trematode species recovered*
Nursery pond (total)	188	43.6 (36.7–50.8)	1.8 (0.2–3.4)	Ht, Hp, Sf, Cf, Ej
Bighead carp, <i>Aristichthys nobilis</i>	4	75.0 (24.9–98.7)	0.2 (0.1–0.4)	Ht, Hp, Cf
Common carp, <i>Cyprinus carpio</i>	46	54.3 (39.8–68.6)	0.4 (0.6–1.0)	Hp, Cf
Grass carp, <i>Ctenopharyngodon idellus</i>	43	62.8 (47.7–75.9)	7.1 (0.5–13.7)	Ht, Hp, Sf, Cf
Mrigal, <i>Cirrhinus mrigala</i>	16	56.3 (30.6–79.2)	0.4 (0.1–0.6)	Ht, Hp, Cf
Rohu, <i>Labeo rohita</i>	27	7.4 (1.3–23.7)	0.0 (0.0–0.1)	Ht, Hp, Ej
Silver carp, <i>Hypophthalmichthys molitrix</i>	35	40.0 (25.0–57.2)	0.2 (0.1–0.4)	Ht, Hp, Cf
Tilapia, <i>Oreochromis niloticus</i>	17	11.8 (2.1–35.0)	0.0 (0.0–0.0)	Hp, Cf
Grow-out pond (total)	386	44.6 (39.6–49.6)	0.7 (0.4–1.1)	Ht, Hp, Sf, Cf, Hy
Bighead carp, <i>Aristichthys nobilis</i>	11	45.5 (20.0–73.5)	0.1 (0.0–0.3)	Hp, Cf
Common carp, <i>Cyprinus carpio</i>	89	42.7 (32.5–53.4)	0.1 (0.1–0.2)	Hp, Sf, Cf
Grass carp, <i>Ctenopharyngodon idellus</i>	105	61.0 (51.0–70.1)	1.9 (0.6–3.1)	Hp, Hy, Cf
Mrigal, <i>Cirrhinus mrigala</i>	22	59.1 (38.3–77.8)	0.5 (0.7–0.2)	Ht, Hp, Cf
Rohu, <i>Labeo rohita</i>	23	39.1 (21.3–61.1)	0.1 (0.0–0.2)	Ht, Hp, Cf
Silver carp, <i>Hypophthalmichthys molitrix</i>	72	48.6 (36.9–60.4)	0.8 (0.1–1.4)	Ht, Hp, Cf
Tilapia, <i>Oreochromis niloticus</i>	64	12.5 (5.9–23.2)	0.1 (–0.1–0.3)	Hp

* Hp = *Haplorchis pumilio*; Ht = *H. taichui*; Hy = *H. yokogawai*; Cf = *Centrocestus formosanus*; Sf = *Stellantchasmus falcatus*; Ej = *Echinochasmus japonicus*.

Hung Nguyen, Nam Dan, and Yen Thanh was significantly greater than from Tan Ky (36.3%) and Thanh Chuong (12.3%) districts ($P \leq 0.001$). In contrast, there was a significant difference in infection density of metacercariae in fish from Tan Ky compared to fish from the other 4 districts ($P \leq 0.000001$).

The species of FZT metacercariae, their prevalence, and intensities in cultured fish

The FZT prevalence and intensity data for cultured fish (excluding SRS) from both nurseries and grow-out ponds are presented in Tables II and III. The overall prevalence of metacercariae in all fish collected was 44.7% (CI: 41.1–48.4). All species of metacercariae recovered were potentially zoonotic intestinal flukes belonging to either the Heterophyidae or the Echinostomatidae. The prevalence of *Stellantchasmus falcatus*, *Echinochasmus japonicus*, and *Haplorchis yokogawai* was very low ($\leq 0.5\%$); the 3 most numerous species were *H. pumilio*, *H. taichui*, and *C. formosanus*. *Haplorchis pumilio* was significantly more common than the other 2 species in both nursery and grow-out ponds ($P \leq 0.05$) (Table IV). The prevalence of *C. formosanus* was significantly higher in fish from nursery

ponds than from grow-out ponds ($P \leq 0.05$). Neither the prevalence nor density of FZT metacercariae was significantly different between nursery ponds and grow-out ponds (Tables II, IV). The density of *H. pumilio* was significantly greater than that of the other FZT species recovered ($P \leq 0.000001$).

Comparisons of FZT infections between species of cultured fish

The FZT prevalence and density data for all cultured fish species are shown in Table III. The prevalence of metacercariae varied significantly between the species of fish examined ($P \leq 0.000001$). Grass carp from both nursery and grow-out ponds had the highest FZT prevalence and infection densities. The apparent higher prevalence in bighead carp from nursery ponds was based on only 4 fish.

Prevalence and intensity of FZT in self-recruiting species of fish

Fourteen SRS fish species ($n = 142$ fish) were collected from 53 ponds in the 5 districts in Nghe An Province (Table I). The

TABLE IV. Comparisons of FZT species prevalence and mean densities in fish from nursery and grow-out systems including SRS.*

FZT species	Culture system			
	Nursery		Grow-out	
	% Prevalence (95% CI)	Mean density (95% CI)	% Prevalence (95% CI)	Mean density (95% CI)
<i>Haplochis taichui</i>	1.9 (0.7–4.9)	0.0	1.0 (0.4–2.3)	0.0
<i>H. pumilio</i>	45.2 (38.4–52.2)	2.2 (0.7–3.7)	41.1 (36.9–45.5)	0.7 (0.4–1.0)
<i>H. yokogawai</i>	0.0 (0.0–1.8)	0.0	0.2 (0.0–1.1)	0.0
<i>Stellantchasmus falcatus</i>	0.5 (0.0–2.8)	0.0	0.2 (0.0–1.1)	0.0
<i>Centrocestus formosanus</i>	16.3 (11.7–22.1)	0.1 (0.0–0.1)	8.5 (6.3–11.2)	0.1 (0.0–0.3)
<i>Echinochasmus japonicus</i>	0.5 (0.0–2.8)	0.0	0.0 (0.0–0.7)	0.0
Total	47.6 (40.9–54.6)	2.3 (0.8–3.8)	43.5 (39.2–47.9)	0.9 (0.6–1.2)

* Results to 1 decimal place.

TABLE V. Prevalence of FZTs from different species of SRS fish.

Fish species	No. infected/ no. examined	FZT species recovered*
Nursery pond (total)	17/20	Hp, Cf
Crucian carp, <i>Carassius auratus</i>	3/3	Hp, Cf
Snakehead, <i>Ophiocephalus maculatus</i>	1/2	Hp
Sailfish, <i>Istiophorus</i> spp.	2/2	Hp
Featherback, <i>Notopterus notopterus</i>	5/5	Hp
Hemiculter, <i>Hemiculter leuciscus</i>	4/5	Hp, Cf
Loach, <i>Mastacembelus armatus</i>	1/1	Hp, Cf
Unidentified species	1/2	Hp
Grow-out pond	47/122	Ht, Hp, Cf
Crucian carp, <i>Carassius auratus</i>	9/12	Hp, Cf
Snakehead, <i>Ophiocephalus maculatus</i>	2/5	Hp, Cf
Sailfish, <i>Istiophorus</i> spp.	1/1	Hp, Cf
Featherback, <i>Notopterus notopterus</i>	14/47	Hp, Cf
Hemiculter, <i>Hemiculter leuciscus</i>	10/27	Ht, Hp, Cf
Anabas, <i>Anabas testudineus</i>	0/1	
Owsianka, <i>Leucaspis delineatus</i>	0/1	
Mai, <i>Rasbora lineatus</i>	3/9	Hp, Cf
Macropodus, <i>Macropodus opercularis</i>	0/1	
Red-eyed carp, <i>Squaliobarbus curriculus</i>	1/1	Hp
Unidentified species	7/17	Hp, Cf

* Hp = *Haplorchis pumilio*; Ht = *Haplorchis taichui*; Cf = *Centrocestus formosanus*.

FZT prevalence and density data for all SRS are presented in Table V. The prevalence and density of FZT metacercariae in SRS fish were similar to that for the cultured fish species from the same ponds. Statistical comparisons of prevalence between species of SRS fish were not undertaken because of the small sample sizes for most species.

DISCUSSION

The results from this investigation reveal a high prevalence of intestinal fish-borne zoonotic trematodes (FZTs) in farm-raised fish from a community of fish farmers with a very low human prevalence (0.6%) (Olsen et al., 2006). For the first time, *Haplorchis yokogawai* and *Echinochasmus japonicus* metacercariae were recovered from fish in Vietnam, although *H. pumilio*, *H. taichui*, *C. formosanus*, and *S. falcatulus* were only recently reported from Vietnamese fish (Arthur and Te, 2006; Hop et al., 2007; Thien et al., 2007; Thu et al., 2007). While the fish-borne liver fluke *Clonorchis sinensis* has been reported from 9 northern provinces of Vietnam (De et al., 2003), the recent discoveries of potential zoonotic intestinal trematode metacercariae in fish may reflect an emerging public health problem for Vietnam (Thu et al., 2007). More than 50 species of fish-borne intestinal flukes belonging to the Heterophyidae and Echinostomatidae are widespread and emergent in Southeast Asia (De et al., 2003), Laos (Chai, Murrell, and Lymbery, 2005), Thailand (Waikagul and Radomyos, 2005), Cambodia (Stauffer et al., 2004), China (Yu and Xu, 2005), and Korea (Chai, 2005). The fish-borne intestinal flukes are much less well characterized clinically than the liver flukes, but are reported to cause significant pathology in the heart, brain, and spinal cord of humans (Africa et al., 1940; WHO, 1995; Chai, Murrell, and Lymbery, 2005). With expanding freshwater and marine/brackish water fisheries in Asia, the economic impact on commercial

aquaculture due to FZT food quality and safety issues will become more burdensome (WHO, 2004; Chai, Murrell, and Lymbery, 2005; Murrell and Crompton, 2006; Duarte et al., 2007).

The relatively high prevalence of intestinal flukes in fish from Nghe An Province in the present study and the low prevalence in their human residents suggests reservoir hosts, such as dogs, cats, and pigs, may be important in sustaining the life cycles of these flukes in fish farms. This possibility deserves further investigation because the role of reservoir hosts is not regarded as important in many proposed control strategies (reviewed in Chai, Murrell, and Lymbery, 2005). If animal reservoir hosts are capable of maintaining the transmission of FZTs in the absence of environmental contamination with fluke eggs from infected humans, the long-term impact of control efforts focused on drug treatment of people in rural communities alone is unlikely to remove the threat from FZTs. Therefore, there is a need to improve the management of domestic animals on fish farms to help achieve FZT control.

In the present study, *H. pumilio* and *C. formosanus* were the most common FZTs in fish from Nghe An Province. Although common in other Southeast and East Asian countries, the reasons for the high prevalence of *H. pumilio* in this area, and in the Mekong Delta (Thu et al., 2007; Thien et al., 2007), may include better adaptation to cultured species of fish, or the availability of preferred definitive hosts (such as cats, dogs, pigs, and fish-eating birds) associated with aquaculture activities. There may also be differences in ecological conditions required by suitable vector snail populations in fish ponds; however, important ecological and epidemiological aspects of these parasites and their hosts are too poorly characterized to draw any conclusions.

The factors responsible for the observed differences in prevalence among different fish species are also difficult to explain, and we can only speculate that certain specific behaviors and innate differences may be important determinants. Grass carp, for example, which had a high prevalence of FZT infection in both nursery and grow-out culture systems, also had a highly diverse parasite species fauna (5 FZT species), including *H. pumilio*, *H. taichui*, *H. yokogawai*, *C. formosanus*, and *S. falcatulus*. In this instance, the habitat preferences of grass carp may render it more exposed to infection because it is normally associated with the littoral zone of the pond where vegetation favored by snails is most abundant and possibly exposure to cercariae. In contrast, species with low prevalence such as rohu and tilapia frequent deeper zones of the water column, perhaps reflecting a less suitable habitat for the snail vector. Other host-specific factors also merit consideration, especially innate and acquired host characteristics, which are known to have an influence on susceptibility to cercariae infection. These include skin thickness, scale structure, protective properties of superficial mucus, and the fish's immune status (Chun, 1964; Rhee et al., 1988; Lun et al., 2005). For example, fish epidermal mucus cells have been shown to produce attractants that influence host-finding and host-specificity in monogenean trematodes (Kearn, 1967).

The comparable prevalence between nursery and grow-out ponds was unexpected and raises an important issue for the aquaculture industry. Obviously the wide distribution of infected fingerlings negatively affects whatever efforts are made by grow-out pond operators to control FZT infection. Several fac-

tors could have contributed toward the prevalence observed in nursery and grow-out ponds. These include susceptibility of the fish, density, or, more likely, surface area of fish in the pond and number of infective organisms. Each one of these variables could be subdivided; for example, susceptibility could be physiological or behavioral, and the number of infective organisms could vary in both time and space. We do not have the data to explain the higher prevalence in nursery ponds, but this is an area for further study.

The high prevalence of SRS fish in these farm ponds suggests that improved pond management could have an impact on the occurrence of FZTs. The presence of SRS fish indicates that the ponds are not well prepared before fish restocking. Management practices such as removing noncultured fish species and snails from ponds before restocking and restricting access of fish and snails during the production cycle could have a significant effect on reducing FZT infections.

In conclusion, the present study has demonstrated the occurrence of FZTs in Vietnamese cultured fish and the potential risk posed to human health. At present they do not appear to represent a significant public health problem in these particular Nghe An districts, presumably because of local eating habits that do not favor consumption of raw fish. Elsewhere in Vietnam, however, the habit of consuming improperly cooked fish is widespread and may be increasing (De et al., 2003; Chai, Murrell, and Lymbery, 2005); therefore, fish moved to other parts of Vietnam for consumption may pose a greater threat, emphasizing the need to control these parasites in food for human use. A pilot project employing a HACCP (Hazardous Critical Control Points) approach was carried out in Vietnam in 1996–1997 to control liver flukes in Vietnamese aquaculture systems (Lima dos Santos, 2002). This project had initial success, but there has not been any follow-up to assess its long-term effects. The results of the study reported here strongly suggest that any efforts to control transmission will require controlling infections in reservoir hosts and the thorough control of snails in ponds.

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