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Helminths and Helminthiosis of Pigs in the Mekong Delta, Vietnam with Special Reference to Ascariosis and *Fasciolopsis buski* Infection

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Abstract

To demonstrate the infection of pigs with helminths in the Mekong Delta, an investigation into the actual conditions was carried out in the area. A total of 87 pigs from 38 farms were examined for the presence of the helminths. Eggs of common nematodes (Ascaris suum, *Metastrongylus* spp., *Oesophagostomum* spp., *Trichuris suis* and *Strongyloides ransomi*) present in Japan, were detected. Especially, since the morbidity rate of A. suum infection was very high, the effect of the *Ascaris* infection on pig growth was investigated. As a result, economic loss was estimated at US$2.0/head. In addition, infection with *Fasciolopsis buski*, which dose not occur in Japan, was detected in 4 pigs from 4 small farms. These 4 pigs were given water spinach as a supplemental feed. It was suggested that this vegetable played an important role in *F. buski* infection and that the fluke infection was one of the zoonoses. Infection with this fluke in human can be anticipated, because most of the inhabitants ate this vegetable in the Mekong Delta.

Discipline: Animal health
Additional key words: *Ascaris suum*, zoonosis, anthelmintic, economic loss

Introduction

The Mekong Delta is an important region for the pig production industry in Vietnam[11]. The pig production is conducted in small farms in integrated farming systems or in the *Vuon-Ao-Chung* (VAC) system combining agriculture, animal husbandry and fisheries for feed from agricultural by-products. The economic efficiency of these farms is very low and the income of the farmers is also low[6]. To increase the productivity of pigs and farmers' income, it is important to prevent the occurrence of various problems relating to feeds and diseases[18]. It is considered that helminthic diseases are associated with the low productivity and cause economic loss in the VAC system[8].

On the other hand, helminthiosis may be an important zoonosis in the VAC system, because many intermediate hosts of helminths, such as snails, crabs or small fishes, occur in this area in which marshlands, small rivers, ponds and rice fields are located.

This paper reports the results obtained in the joint project on "Integrated Research on Farming Systems Combining Agriculture, Animal Husbandry and Fisheries in the Mekong Delta" among the Japan International Research Center for Agricultural Sciences, Japan, Can Tho University and Cuu Long Delta Rice Research Institute, Vietnam, during the period of October 1995 to September 1997.
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The objective of the present studies was to identify swine helminthiosis including zoonotic trematodes in the VAC system. Therefore, we investigated *Ascaris suum* infection and *Fasciolopsis buski* infection in the Mekong Delta.

Materials and methods

1) Surveys of helminths

Sampling sites: Eighty-seven fecal samples were collected from pigs in 38 farms in the VAC systems (Fig. 1) in the Mekong Delta area. Feces (50–100 g each) were packed in plastic bags and stored in a refrigerator until the examination. We also determined which feeds were given as supplement in the farms.

Detection of eggs: The Watanabe sedimentation method[19] was applied to detect eggs. Counts of eggs per gram (EPG) were carried out to estimate the degree of parasitism by using Dennis method[5] for *F. buski*. The effect of anthelminthics on *A. suum* was estimated using McMaster EPG counting plate, if necessary.

2) Use of scanning electronmicroscope (SEM) for morphological studies of F. buski

Several flukes collected from slaughterhouses were fixed in 10% phosphate-buffered formalin and washed 3 times in 0.1 M phosphate buffer. After dehydration in ethanol, the fluke materials were subjected to isoamyl acetate immersion, critical point drying and coating with gold. The fluke specimens were observed with a scanning electronmicroscope (SEM, JEOL, JSM-5300).

3) Experiment on effect of Ascaris *infection on pig growth*

Pigs: Six pigs weighing approximately 40 kg were used. They were littermates and crossbreds of *Ba Xuyen* and Yorkshire from the Experimental Animal Farm of Can Tho University. They were positive for the *A. suum* eggs.

Experimental design: Group 1 consisted of 3 pigs treated with 1 mL per 15 kg of Polystrongle (injectable form Tetramizole hydrochloride, France). Group 2 consisted of 3 untreated pigs. Their body weight was checked biweekly for 12 weeks. EPG count was carried out every day using McMaster egg counting plate for 10 days after treatment. *A. suum* eggs were also checked by the sedimentation method every week. All the animals were raised under the standard management applied in the animal farm of the university.

Pathological examination: Pigs from the 2 groups were necropsied 12 weeks after the onset of the experiment. At necropsy, middle parts of the small intestine were fixed in 10% formalin. For histological examination, the tissue samples were embedded in paraffin wax, sectioned and stained with hematoxylin and eosin (HE).

4) Ovicidal effect of biodigester

Samples containing *F. buski* eggs were collected from the inlet and outlet of the biodigester (Fig. 1). Each sample of the fluke eggs was washed in tap water and vibrated to remove the debris around the eggs. Then, the egg samples were transferred into Petri dishes with water and incubated at 29°C for 15 days. After the incubation, the eggs which did not develop to the next stage (or miracidium) were considered to be dead eggs. The eggs of *A. suum* were also incubated by the same method to examine their activity.

Results

1) Surveys of helminths in the Mekong Delta

Eggs of *A. suum* were detected in 51% (44) of 87 pigs, *Metastrongylus* spp. in 14% (12), *Trichuris suis* in 26% (23), *Oesophagostomum* spp. in 14% (12), *Strongyloides ransomi* in 5% (4) and *F. buski* in 5% (4). There were distinct differences between the regions surveyed in the infective rate of *A. suum*. The rate of this nematode in the pigs from 12 farms near Can Tho University was 100%, whereas about 30% in other regions.

Eggs of *F. buski* (Fig. 2) were detected in the feces of 4 pigs from 4 farms. All the pigs infected with this fluke had eaten water spinach (*Ipomoea aquatica*) as supplemental feed (Fig. 3). In 3 pigs, the EPG count was 10 or less. However, a sow with severe constipation had an EPG count of more than 1,000 in the feces. The snails which were intermediate hosts of the flukes (*Gyraulus* sp. and *Indoplanorbis* sp.) were detected in marshlands, ponds, small rivers and rice fields near the pig pens in the VAC systems. Furthermore, other snails (*Lymnaea* sp. and *Pila* sp.) also occurred in the same areas (Fig. 4).

2) Morphology of F. buski

The eggs of *F. buski* were yellowish ellipsoidal and 130–140 × 80–85 μm in size. They were very similar to the eggs of *Fasciola gigantica* (Japanese

Fig. 1. A tube type biodigester in a farming system combining pigpen, pond and orchard

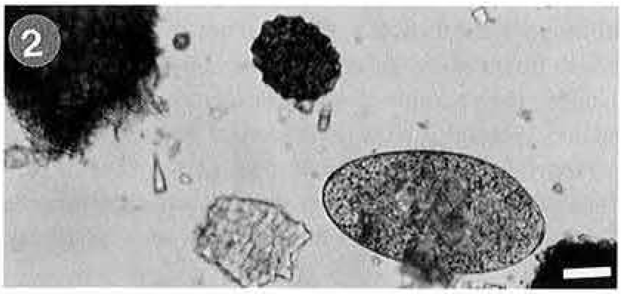

Fig. 2. *F. buski* (right) and *A. suum* (left) eggs
Bar shows 35 μm.

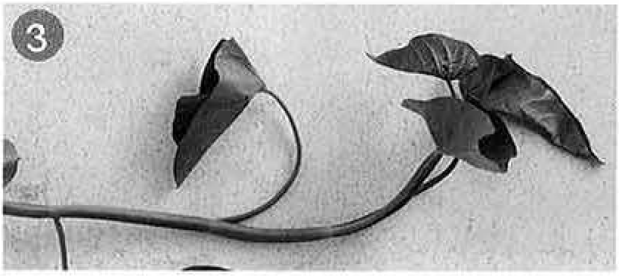

Fig. 3. Water spinach used as supplement for feed in a farm

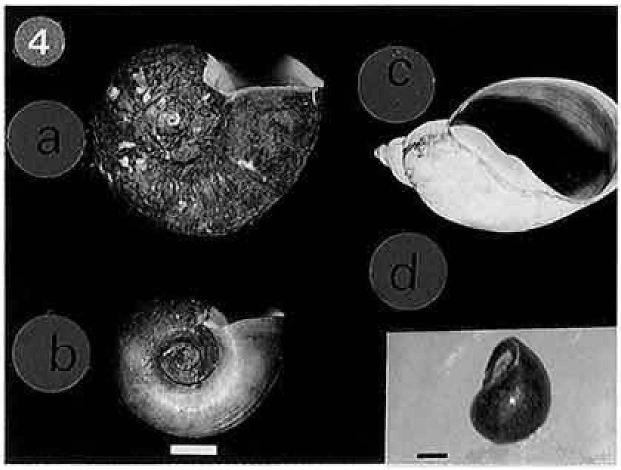

Fig. 4. Intermediate hosts detected in VAC system
a; *Indoplanorbis* sp., b; *Gyraulus* sp.,
c; *Lymnaea* sp., d; *Pila* sp.
Left bar indicates 3 mm and right one 15 mm.

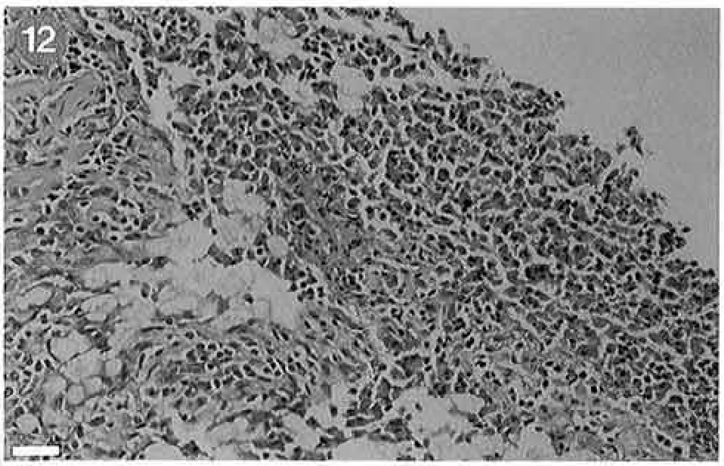

Fig. 12. Histological specimen of middle part of duodenum in a pig infected with 4 worms
Severe desquamation of epitelium and cellular infiltration of lamina propria.
HE staining × 150, bar indicates 35 μm.

type). The *F. buski* flukes in pigs from slaughterhouses were 2.0–4.4 × 0.5–2.0 cm in size. Several adult flukes were detected in the intestine (Fig. 5). Under the scanning electronmicroscope an oral sucker, genital atrium and ventral sucker were observed (Fig. 6). The middle part of the ventral surface of the body showed a crocodile skin appearance (Fig. 7). In the tail of the fluke, the excretory bladder opened near the posterior extremity of the body (Fig. 8).

Fig. 5. Adult of *F. buski* worms collected from a slaughterhouse

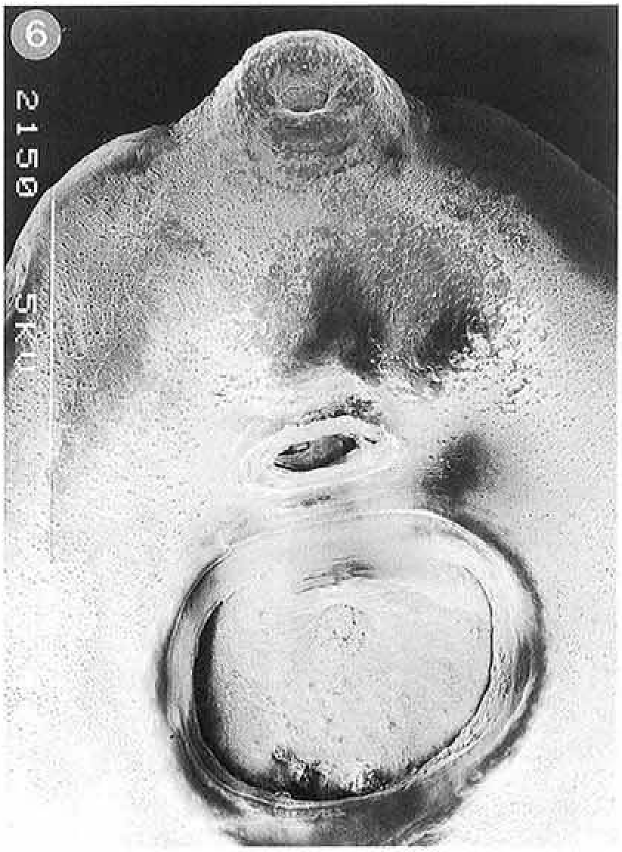

Fig. 6. Posterior extremity of the mature *F. buski* worm (SEM × 20)

3) Effect of A. suum *infection on pig growth*

In 6 pigs of the 2 groups, only *A. suum* eggs were detected in the feces. In 3 pigs of the treated group, adults of *A. suum* were eliminated within a few days after treatment with Tetramizole, and the EPG of this nematode reached a zero value (Table 1). However, since *A. suum* eggs were found again 6 weeks later, treatment was resumed. While the growth rate of the untreated pigs was low, the growth rate of the treated pigs improved (Fig. 9). In the untreated group, the small intestine of an emaciated animal contained 12 *Ascaris* (Fig. 10). The pig infected with *A. suum* showed a low weight gain (Table 2). The growth of the untreated pigs which reached a weight of 80 kg was delayed by 2 weeks compared with the treated pigs. Anorexia and

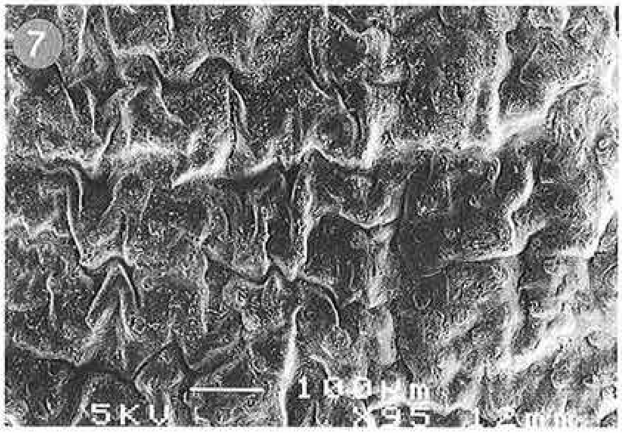

Fig. 7. Middle part of the ventral surface of mature *F. buski* worm (SEM × 100)

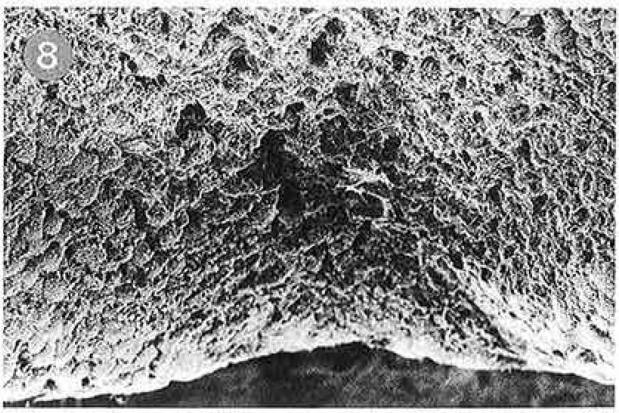

Fig. 8. Posterior extremity of mature *F. buski* worm (SEM × 100)

Table 1. Changes in egg counts in feces of pigs after Tetramizole treatment

Group	Pig No.	Days after treatment									
		1	2	3	4	5	6	7	8	9	10
Treatment	1	800[a)]	0	0	0	0	0	0	0	0	0
	2	900	0	0	0	0	0	0	0	0	0
	3	300	100	0	0	0	0	0	0	0	0
Control	4	700	800	900	800	800	1,000	900	1,100	900	1,100
No treatment	5	800	900	800	800	800	800	800	900	1,000	1,000
	6	400	400	400	300	400	400	400	400	400	600

a): Number of eggs per g in feces.

Table 2. Body weight of treated and untreated pigs

(Unit: kg)

Group	Pig No.	Weeks after treatment						
		0	2	4	6	8	10	12
Treatment	1	37	44	51	60	79	81	89
	2	40	45	52	60	70	82	89
	3	39	44	51	58	68	80	90
Control	4	41	46	52	59	66	74	82
No treatment	5	39	44	50	57	64	72	81
	6	42	46	51	58	66	69	72

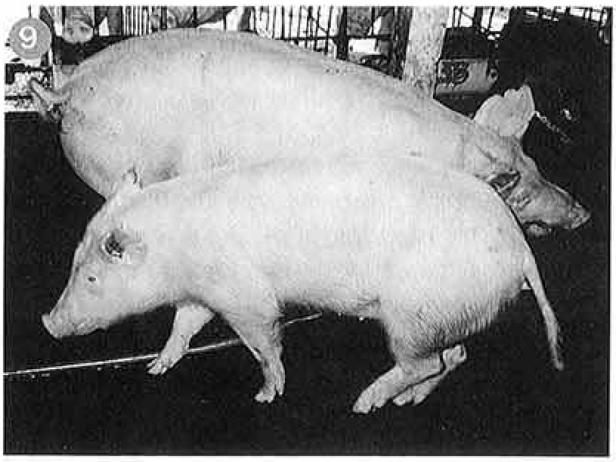

Fig. 9. Treated (back) and untreated (front) pigs in the experimental farm

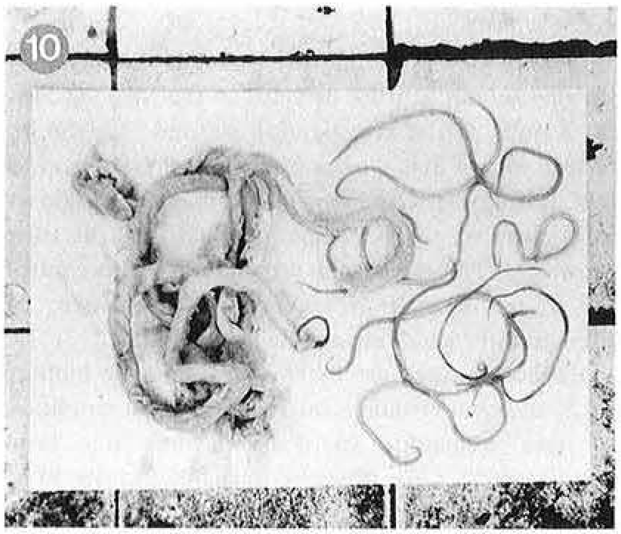

Fig. 10. Small intestine and *Ascaris* worms from No. 6 pig (untreated group)

diarrhea were not observed in pigs of both groups during the examination.

Macroscopically, there were a few petechial hemorrhage on the pulmonary pleura and white spots on the liver's capsule in the untreated pigs. These changes were less conspicuous in pigs of the treated group. One (No. 4), two (No. 5) and four (No. 6) adult worms were found in the middle part of the duodenum from untreated pigs and the lumen of the intestine became enlarged due to the presence of *A. suum* (Fig. 11). Microscopically, the intestinal villi were depressed by *A. suum* infection and epithelial cells desquamated in the duodenum where the worms occurred. Eosinophils infiltrated the lamina propria (Fig. 12).

4) Ovicidal effect of biodigester

After processing by the biodigester, eggs of *A.*

Fig. 11. Transverse section of the middle part of the duodenum of 3 infected pigs
Left; intestine containing with 1 worm, middle; 2 worms, right; 4 worms.

suum which were collected from the outlet of the biodigester survived in the incubator. On the other hand, all the eggs of *F. buski* were inactivated by biodigester processing and they were not hatched.

Discussion

Survey of the helminths disclosed that eggs of various helminths were detected in the Mekong Delta. Results of the investigation showed that the infection rate of lung worms (*Metastrongylus* spp.) was low. However, swine metastrongylosis is an important cause of swine pneumonia and disturbs their growth[9]. Therefore, it is very important to control this disease in order to increase the productivity of pigs and farmers' income in this area.

Although there are many studies on the biology of *F. buski* in Asian countries, the actual condition of this trematode in Vietnam has not been determined[10]. We observed this fluke under SEM and the morphological characteristics of *F. buski* were demonstrated. It is interesting to note that 4 pigs which ate water spinach given as a supplemental feed were infected with *F. buski*. This vegetable may thus play an important role in *F. buski* infection. Since many people eat water spinach in the Mekong Delta, human may possibly become infected with the fluke. In the small VAC system, a snail of *Pila* sp. which is the intermediate host of *Angiostrongylus cantonensis* and a snail of *Lymnaea* sp. which is the intermediate host of *Fasciola* sp. were detected[16]. Judging from the disappearance of fluke eggs in the outlet of a biodigester in VAC systems, it is likely that the biodigester contributes to the control of zoonoses, such as fasciolopsiosis, paragonimiosis or fasciolio-sis. It was considered that the inactivation of *F. buski* eggs may be due to the temperature and also to the effect of ammonia in the biodigester, since fluke eggs were susceptible to a low concentration of ammonia[12,22]. The highest temperature in the tube type of biodigester was approximately 45°C[2] and *F. buski* eggs were killed at 50°C after 4 hours in an experiment[21].

As *A. suum* eggs were detected again at 6 weeks after Tetramizole treatment, the treatment was resumed. After ingestion of *A. suum* eggs, about 9 weeks were required for the development of the adult stage[13]. Therefore, it is considered that this anthelminthic was effective in the adult stage of the nematode in the intestine, but not in the immature stage. Comparison of treated and untreated pigs showed that the growth rate of the untreated pigs was very low and this nematode infection might be a cause of heavy economic losses for the farmers. Andersen[3] reported that the growth rate decreased to 20% after *Ascaris* infection in Denmark. Calculations showed that the economic loss was about 25,000 Don Vietnam (about US$2.0) per head in an experimental farm[17]. Low nutrition level including deficiency in protein and vitamin A may exert more deleterious effects on the infected hosts[7]. If farmers use anthelminthic therapy for their pigs, the growth period of the pigs may be shortened at least by 2 weeks. Total cost for the treatment was about 5,000 Don Vietnam. It may be necessary that the farmers use anthelminthics in order to increase productivity.

Bossaler et al.[4] pointed out that the treatment of parasites contributed to the production of good quality meat.

One untreated pig which was clinically thin and infected with 12 *A. suum* showed moderate pathological changes. The main pathogenetic mechanism of ascariosis may involve physical stimuli[20] and the effects of inhibitors of host pepsin, trypsin, chymotrypsin, and carboxypeptidases which are contained in the extract of *A. suum*[1]. Intestinal ascariosis directly interferes with the absorption of protein, fat, and carbohydrates[14]. Larval migration caused "white spots in the liver" and "thumps in the lungs." In the primary phase of severe infection, the growth rate of the pigs decreased[15].

Further examinations should be carried out to identify the presence of other zoonotic parasitoses and to determine the relation between the presence of parasites and the economic losses in the VAC system.

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