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Research Note

Survey on Larval *Echinococcus multilocularis* and Other Hepatic Helminths in Rodents and Insectivores in Hokkaido, Japan, from 1985 to 1992

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It is generally considered that the most suitable intermediate host of *Echinococcus multilocularis* in Hokkaido, Japan is the gray red-backed vole, *Clethrionomys rufocanus bedfordiae*. In addition, larval *E. multilocularis* infection was found in other rodents which act as intermediate hosts in nature (Kamiya *et al.*, 1977; Okamoto *et al.*, 1992; Yagi *et al.*, 1984, 1985, 1986; Yorozuya *et al.*, 1968). To investigate a role of rodents in the transmission of larval *E. multilocularis*, epidemiological survey was performed in Hokkaido, Japan.

Between May and November in 1985–1992, 2024 rodents and insectivores including following 9 species were collected on 12 areas in Hokkaido (Fig. 1): *Clethrionomys rufocanus bedfordiae, C. rutilus mikado, Apodemus argenteus, A. peninsulae, A. speciosus ainu, Rattus norvegicus, Mus musculus, Sorex caecutiens saevus* and *S. unguiculatus*. Animals which could not be identified were described in Tables as unknown species of each genus. Animals were killed under anesthesia, then immediately dissected. Parasites in abdominal viscera were examined under a dissection microscope after naked eye examination. Lesions hard to identify parasite species macroscopically were determined by histological examination.

The occurrence of helminths in the captured animals are shown in Tables 1 and 2. Larval *E. multilocularis* were found in 16 *C. rufocanus bedfordiae* (1.6%), 1 *A. argenteus* (0.2%), and 1 *R. norvegicus* (0.7%). In *C. rufocanus bedfordiae*, multilocular cysts were usually found in liver, spleen, kidney, mesentery, ureter and lung. In the large lesions, a lot of mature protoscoleces were observed in the cysts. In a case of a male *A. argenteus*, two multilocular cysts were in the liver and mesentery, 7×5 mm in size. But mature protoscoleces were observed only in the center of the liver cysts. In one case of a female *R. norvegicus*, 151g body weight and 4.2 month old, the lesion of liver was irregular in shape, 9×8 mm in size, and immature protoscoleces with no sucker or hook were observed inside the cysts.

Besides larval *E. multilocularis*, 5 species of hepatic helminths were also found: *Capillaria hepatica*, Dicrocoelidae gen. sp., *Taenia taeniaeformis*, and the larvae of unknown cestodes #1 and #2. The larvae of unknown cestode #1 were morphologically similar to *T. taeniaeformis* larvae, but taxonomical identification could not be done because they were in immature stage with no scolex. The larvae of unknown cestode #2 were found in round or oval cysts, 3–4 mm in size, were found

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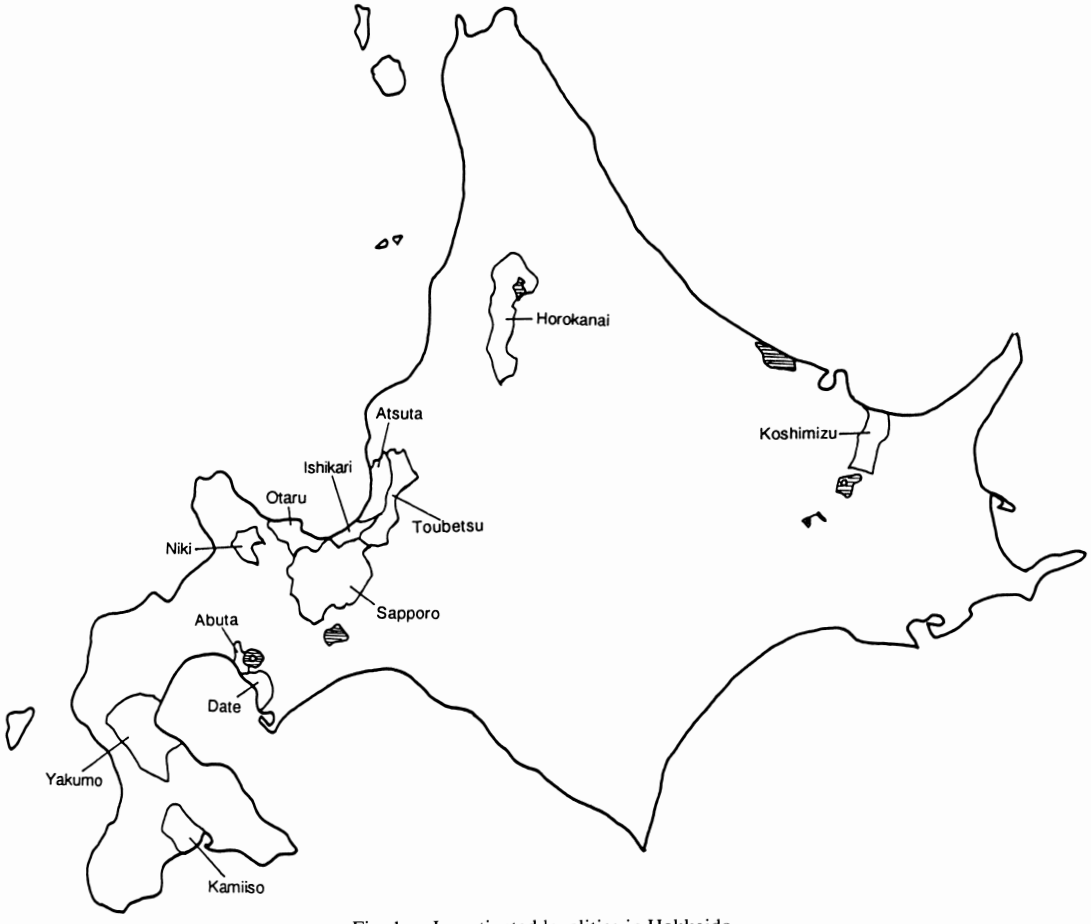

Fig. 1 Investigated localities in Hokkaido.

through transparent cyst wall. Three types of the larvae were recognized by the length of hooks: (1) the larvae with 38–44 rostellar hooks which were 16–23 μm in length; (2) the larvae with 44–50 hooks which were 26–32 μm; (3) the larvae with 48–52 hooks, consisting of 64–70 μm large hooks and 50–52 μm small hooks. However, classifications of these larvae were not determined.

In this study, larval *E. multilocularis* infection were detected from rodents in Kamiiso, Koshimizu, Yakumo and Toubetsu, and the infected cases were found from rodents in former 2 areas for the first time. The cases of pigs in these areas were already reported by Hokkaido prefectural government (1992) (Table 3), thus the spread of endemic area of *E. multilocularis* is confirmed by these data. The examination of pigs have the advantages, because the field survey of small animals takes a lot of time and effort.

While high susceptibility of *C. rufocanus bedfordiae* to *E. multilocularis* eggs were confirmed experimentally (Yamashita *et al.*, 1958), the prevalence of *E. multilocularis* in the voles is relatively low in general (1.6% in this study), in comparison with the high prevalence in red foxes, the definitive host of *E. multilocularis* in Hokkaido (18.3%) (Table 3). But it is thought that the foxes eat enough number of voles necessary to maintain the life cycle of *E. multilocularis*, because the foxes prefer voles as the main foods (Yoneda and Nakata, 1984).

Natural infection of larval *E. multilocularis* in *A. speciosus ainu* or *A. peninsulae* have not been identified neither in this study nor other studies (Ishimoto, 1974; Inaoka *et al.*, 1984; Yagi *et al.*, 1984, 1985,

Table 1 Occurrence of larval *E. multilocularis* in captured animals*

Location	Rodentia†								Insectivora†		
	Crb	Crm	Aa	Ap	As	*Apodemus* sp.	Rn	Mm	Sc	Su	*Sorex* sp.
Kamiiso	3/57(5.3)	–	0/86	–	0/22	0/15	1/78(1.3)	–	–	–	0/3
Yakumo	2/30(6.7)	–	0/7	–	0/5	0/9	0/4	0/2	–	0/4	–
Abuta	0/79	–	0/57	0/2	0/13	0/34	0/35	–	–	0/3	0/1
Date	0/50	–	0/9	–	–	0/2	0/10	–	–	–	–
Niki	0/5	–	0/13	–	0/4	–	0/1	–	0/5	0/2	–
Otaru	0/1	–	0/7	–	0/1	–	–	–	–	–	–
Sapporo	0/194	–	0/51	0/4	0/18	0/58	0/4	–	–	–	–
Ishikari	–	–	0/16	–	0/1	–	–	–	–	–	–
Atsuta	0/1	–	0/12	0/1	–	–	–	–	–	–	–
Toubetsu	3/383(0.8)	–	0/16	0/1	0/34	0/6	–	–	0/1	0/1	0/1
Horokanai	0/10	–	0/65	–	–	0/23	–	–	–	–	–
Koshimizu	8/199(4.0)	0/14	1/90(1.1)	0/41	0/86	–	0/4	–	0/9	0/23	0/1
Total	16/1,009(1.6)	0/14	1/429(0.2)	0/49	0/184	0/147	1/136(0.7)	0/2	0/15	0/33	0/6

*Positive/examined animals (%).
†Crb: *Clethrionomys rufocanus bedfordiae*, Crm: *C. rutilus mikado*, Aa: *Apodemus argenteus*, Ap: *A. peninsulae*, As: *A. speciosus ainu*, Rn: *Rattus norvegicus*, Mm: *Mus musculus*, Sc: *Sorex caecutiens saevus*, Su: *S. unguiculatus*.

Table 2 Occurence of hepatic helminths except larval *E. multilocularis* in captured animals*

	Rodentia†								Insectivora†		
	Crb	Crm	Aa	Ap	As	*Apodemus* sp.	Rn	Mm	Sc	Su	*Sorex* sp.
No. of examined	1,009	14	429	49	184	147	136	2	15	33	6
Trematoda											
Dicrocoelidae gen. sp.	1(0.1)	0	0	0	0	0	0	0	0	0	0
Cestoda											
Taenia taeniaeformis	16(1.6)	0	12(2.8)	0	2(1.1)	3(2.0)	57(41.9)	0	0	0	0
unknown cestode #1	25(2.5)	0	5(1.2)	0	2(1.1)	3(2.0)	33(24.3)	0	0	0	0
unknown cestode #2	77(7.6)	3(21.4)	10(2.3)	0	9(4.9)	6(4.1)	0	0	0	0	0
Nematoda											
Capillaria hepatica	117(11.6)	0	52(12.1)	1(2.0)	5(2.7)	10(6.8)	100(73.5)	0	1(6.7)	0	0

*No. of positive animals (%).
†Crb: *Clethrionomys rufocanus bedfordiae*, Crm: *C. rutilus mikado*, Aa: *Apodemus argenteus*, Ap: *A. peninsulae*, As: *A. speciosus ainu*, Rn: *Rattus norvegicus*, Mm: *Mus musculus*, Sc: *Sorex caecutiens saevus*, Su: *S. unguiculatus*.

1986). *A. speciosus ainu* was experimentally confirmed to be resistant to oral infection of *E. multilocularis* eggs. On the other hand, *A. argenteus* is known to be susceptible to natural and experimental infections (Yamashita *et al.*, 1958; Yagi *et al.*, 1984, 1985, 1986; Ooi *et al.*, 1992). In this study, no larval *E. multilocularis* infection was found in *Sorex* spp. Ohbayashi *et al.* (1971) noted that larval *E.*

Table 3 Occurrence of larval and adult *E. multilocularis* in mammals in Hokkaido (After Hokkaido prefectural government, 1992, and this study)

	Hokkaido prefectural government (1992)*				This study	
	Adult		Larva		Larva	
Location	Dogs†	Foxes†	Rodents†	Pigs‡	Rodents†	Shrews†
Kamiiso	0/27	–	0/53	43	4/258(1.6)	0/3
Yakumo	0/5	22/78(28.2)	2/45(4.4)	68	2/57(3.5)	0/4
Abuta	–	4/30(13.3)	0/49	8	0/220	0/4
Date	–	6/31(19.4)	–	35	0/71	–
Niki	–	5/10(50.0)	0/28	1	0/123	0/7
Otaru	–	6/18(33.3)	0/85	–	0/9	–
Sapporo	–	2/58(3.4)	0/971	5	0/329	–
Ishikari	0/2	0/40	0/196	–	0/17	–
Atsuta	–	0/11	0/184	–	0/14	–
Toubetsu	–	4/30(13.3)	1/344(0.3)	17	3/440(0.7)	0/3
Horokanai	–	1/9(11.1)	0/19	–	0/98	–
Koshimizu	1/25(4.0)	27/93(29.0)	0/111	148	9/434(2.1)	0/33
Hokkaido	97/9,742(1.0)	2,468/13,457(18.3)	555/32,621(1.7)	4,788	18/1,970(0.9)	0/54

*Investigation during 1966–1991 in Hokkaido.
†Positive/examined animals (%).
‡No. of positive pigs. No. of slaughtered pigs during fiscal 1966–1991 in Hokkaido were 23,787,818.

multilocularis were found in *Sorex jacksoni* collected on St. Lawrence Island, however, only few immature protoscoleces were observed in the hosts. Experimental infection with *E. multilocularis* eggs to *Suncus murinus* was confirmed not to be successful. (Kamiya *et al.*, unpublished data).

The present case of larval *E. multilocularis* infection in *R. norvegicus* is the second one in Japan, following the first case report by Okamoto *et al.* (1992). In general, Norway rats are considered to be resistant to oral infection with *E. multilocularis* eggs. Webster and Cameron (1961) failed to experimentally infect to rats with *E. multilocularis* eggs. However, natural cases of larval *E. multilocularis* infection in *R. norvegicus* have been reported in 4 of 50 rats in west Siberia by Lukashenko and Zorikhina (1961), and in 1 of 42 rats in Hokkaido, Japan by Okamoto *et al.* (1992). Norway rats may not significantly contribute to the maintenance of the life cycle of *E. multilocularis*. But the risk of infection to human is getting higher if Norway rats play the intermediate host, because the rats inhabit the region of human activity.

Further study is necessary to investigate the biological characteristics of *E. multilocularis* such as morphology and genetics, and to clarify the role of rats which may participate in the life cycle of *E. multilocularis*.

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