

Current status and its epidemiological consideration of *Fasciola* and *Eurytrema* infections in beef cattle of Japan

Jungo OKAJIMA¹⁾, Kazuhiko SHIBATA¹⁾, Eiichi TAKAHASHI¹⁾, Tsuneyuki NAGAFUCHI²⁾, Kazue OKAJIMA³⁾ and Nariaki NONAKA^{4,5)*}

¹⁾Animal Care and Consultation Center Jonanjima branch office, Bureau of Social Welfare and Public Health, Tokyo Metropolitan Government, 3-2-1 Jonanjima, Ota-ku, Tokyo 143-0002, Japan

²⁾Wholesale Market Sanitary Inspection Station, Bureau of Social Welfare and Public Health, Tokyo Metropolitan Government, 5-2-1 Tsukiji, Chuo-ku, Tokyo 104-0045, Japan

³⁾Shibaura Meat Sanitary Inspection Station, Bureau of Social Welfare and Public Health, Tokyo Metropolitan Government, 2-7-19 Kounan, Minato-ku, Tokyo 108-0075, Japan

⁴⁾Laboratory of Veterinary Parasitic Diseases, Department of Veterinary Sciences, Faculty of Agriculture, University of Miyazaki, 1-1 Gakuen-Kibanadai-Nishi, Miyazaki, Miyazaki 889-2192, Japan

⁵⁾Center for Animal Disease Control, University of Miyazaki, 1-1 Gakuen-Kibanadai-Nishi, Miyazaki, Miyazaki 889-2192, Japan

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ABSTRACT. To elucidate current status of fasciolosis and eurytremosis in beef cattle of Japan, inspection data of Tokyo Metropolitan Shibaura Slaughterhouse where beef cattle were shipped from all over Japan were analyzed, and questionnaire to farmers was conducted to assess the relationship between recognition of the disease occurrence in one's own farm and attention to the diseases. The occurrence of fasciolosis and eurytremosis in beef cattle gradually decreased from 18.6% to 0.06% and from 0.58% to 0.02% during the period of 1964 to 2010, respectively. When the current data from 2009 to 2012 were analyzed, the occurrence of fasciolosis was recognized in cattle produced and fattened all over Japan, indicating the disease was prevalent nationwide. While, 97.5% of *Eurytrema* infection were detected in cattle produced in Okinawa, Shimane and Kagoshima, indicating the disease was endemic in these regions. Higher occurrence (>0.7%) of fasciolosis was observed in minor breeds, such as Japanese Shorthorn. Japanese Black showed 0.09% and 0.05% of occurrence for fasciolosis and eurytremosis, respectively, but F1 crossbred with Japanese Black showed lower occurrence (0.007% and 0.002%, respectively). No tendency of occurrence in the age of cattle at slaughter was recognized, indicating the infections may have occurred at the growing and early fattening stage of cattle. The questionnaire survey revealed that farmers experiencing fasciolosis had more knowledge about the disease, however, factors, such as testing parasite infections and use of anti-*Fasciola* dewormers, were not affected by the recognition of occurrence.

KEY WORDS: cattle, epidemiology, *Eurytrema*, *Fasciola*, Japan

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Fasciolosis caused by *Fasciola* spp. parasitizing in the bile duct is an important parasitic disease of cattle, accompanied with chronic cholangitis and liver malfunction. Cattle are infected by ingesting its metacercariae attached on aquatic vegetation [3]. In Japan, it is popularly conducted to feed confined cattle with rice straws. Since cattle manure is scattered in rice fields as a fertilizer and straws mowed from the fields are given to cattle, the life cycle of the parasite has been maintained by the practice of raising stock [2, 14]. In addition, in the grazing areas such as in Hokkaido, infection occurs at watersides in pasturelands, and wild animals, such as deer, may actively be involved in the maintenance of the life cycle [14].

Eurytremosis accompanied with nutritional disorder is caused by *Eurytrema pancreaticum* or *E. coelomaticum* parasitizing in the pancreatic duct [6, 13]. The parasites require two intermediate hosts, terrestrial snails, such as *Bradybaena* spp. and *Acusta* spp., as the first one and grasshopper, such as *Conocephalus* spp., as the second one. Cattle are infected by ingesting the second intermediate host having metacercariae [7]. *Eurytrema* is considered to distribute widely in Japan, especially in isolated islands and Kyushu area where grazing of beef cattle is commonly conducted [6]. However, the actual distribution has not been evaluated in detail.

Both diseases not only reduce the productivity of cattle, but also require public health attention as zoonosis [1, 8, 9, 15]. Light infection with the parasites, however, produces little clinical signs, and thus, most infections are detected at the slaughtering meat inspection. In Shibaura Meat Sanitary Inspection Station, severe cases of infection had been detected routinely in previous years, but the number of cases has decreased drastically in recent years and it became rare to encounter severe ones. Along with the situation, epidemiological studies for the parasite prevalence in cattle have not been conducted in recent years, and thus, it became difficult to comprehend the present situation in the cattle production

*CORRESPONDENCE TO: NONAKA, N., Laboratory of Veterinary Parasitic Diseases, Department of Veterinary Sciences, Faculty of Agriculture, University of Miyazaki, 1-1 Gakuen-Kibanadai-Nishi, Miyazaki, Miyazaki 889-2192, Japan.
e-mail: nnonaka@cc.miyazaki-u.ac.jp

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field. It can be predicted that farmers tend to lose their attention to the prevention of the parasitic infections due to the reduced prevalence.

Tokyo Metropolitan Shibaura Slaughterhouse deals with cattle from all over Japan, and thus, it can collect nationwide data. Using the merit of this, the present study was conducted for the epidemiological analysis of *Fasciola* and *Eurytrema* infections in beef cattle by using the inspection data for fasciolosis and eurytrema in cattle stored at Shibaura Meat Sanitary Inspection Station and by conducting a questionnaire survey to farmers on the occurrence and the awareness of the parasitic diseases.

MATERIALS AND METHODS

Meat inspection data of Fasciola and Eurytrema infections in cattle: The occurrence of *Fasciola* and *Eurytrema* infections in cattle slaughtered at Tokyo Metropolitan Shibaura Slaughterhouse on the basis of detection of the corresponding worms in the routine meat inspection procedure was evaluated using the data of the annual inspection reports of Shibaura Meat Sanitary Inspection Station and the database of cattle diseases recorded by a touch-panel system at the same station. The routine meat inspection was conducted by the procedure stipulated by Tokyo prefecture. Briefly, for the liver, the surface was examined first for the pathological lesions, and then, the left hepatic duct and the parenchyma of the liver that are adjacent to the portal vein were cut and examined for the lesions and existence of the parasites. When pathological changes potentially relating to *Fasciola* infection, such as hypertrophy, enlargement and calcification of bile ducts, were observed, the liver was cut further and some of the bile duct branches in the parenchyma were examined. For the pancreas, after the surface was examined for pathological lesions, the parenchyma and pancreatic duct were cut at the two sites adjacent to the duodenum and examined for the existence of *Eurytrema*.

Change in annual occurrence of the parasites' infection was evaluated using the annual data from 1964 to 2010. During this period, the fiscal year system accounting for one year from April to March of the next year was introduced in 1973, and therefore, annual data stored before 1973 were from January to December, those after 1973 were from April to March of the next year, and those in 1973 were from January 1973 to March 1974. The difference in the occurrence among the regions (prefectures) of shipment, breeds and age in month of cattle slaughtered was analyzed using the data from April 2009 to December 2012. The cattle breeds examined included Japanese Black, Japanese Shorthorn, Holstein, F1 crossbred with Japanese Black (described as F1 crossbred thereafter), and other breeds and crossbred (described as Others thereafter). For the analysis of age difference in occurrence, Japanese Black cattle aging from 25 to 36 months old were examined, because a total number slaughtered in the period was beyond 1,000. In addition, because most beef cattle are raised at two different regions; they are bred until 5 to 12 months of age at production sites and then moved to shipment sites where they are fattened until about 30 months

of age, production sites of cattle was also considered in analysis.

Questionnaire survey to farmers: The results of meat inspection data analysis were informed to the beef cattle producers and shippers who used Tokyo Metropolitan Shibaura Slaughterhouse in order to have them pay attention to fasciolosis and eurytrema, and then, a questionnaire survey was conducted to attendants at the competitive exhibition and the prize-giving ceremony held by Tokyo Meat Market Co., Ltd. in November to December 2012 in order to clarify the relationship between the recognition of occurrence of the parasitic diseases in one's own farm and the attention level of the diseases (knowledge about the diseases, examination of parasite infections and treatment of the infections).

Statistical analysis: For comparison of annual occurrence of fasciolosis and eurytrema from 1994 to 2010, ninety-five percent confidence intervals (95% C.I.) of occurrence were calculated on the basis of binomial distribution using the software program R [12]. Difference in the occurrence among breeds and between sexes of slaughtered cattle and difference between the occurrence of fasciolosis in respondents' farms and responses in questionnaire were analyzed by the Fisher's exact test, and correlation between the occurrence and the age in month of slaughtered cattle was analyzed by Pearson's test using the same program.

RESULTS

Change in annual occurrence: The change in annual occurrence of *Fasciola* infection in beef and dairy cattle at Tokyo Metropolitan Shibaura Slaughterhouse from 1964 to 2010 is shown in Fig. 1A. The annual number of slaughtered beef cattle ranged from 20,180 to 94,347 (61,478 in average) during the whole period, while that of slaughtered dairy cattle ranged from 12,775 to 41,032 (25,215 in average) during 1964 to 1992 and then decreased to range from 2,202 to 9,130 (5,056 in average) during 1993 to 1999 and from 279 to 1,828 (1,010 in average) during 2000 to 2010. The occurrence of the parasite infection in both cattle decreased gradually during the period and changed from 18.70% (6,683/35,733) in 1966 to 0.06% (58/94,347) in 2010 in beef cattle and from 33.11% (5,117/15,453) in 1966 to 0% (0/2,679) at the period between 2007 and 2010 in dairy cattle. The occurrence in dairy cattle was significantly higher than that in beef cattle during 1964 to 1998, except for 1986, 1987 and 1989, and especially, the difference from 1964 to 1973 was obviously larger.

The change in annual occurrence of *Eurytrema* infection in beef and dairy cattle during the same period is shown in Fig. 1B. The occurrence is lower than that of *Fasciola* infection. The highest occurrence was observed in 1964 (0.58%) in beef cattle and in 1965 (0.24%) in dairy cattle, but the values decreased sharply during 1970s and then were kept at low level (<0.03%) after 1983. The significant difference was observed during 1964 to 1982, except for 1965, 1976, 1978, 1979 and 1980.

Regional difference in occurrence: During the study period, cattle were shipped to Tokyo Metropolitan Shibaura

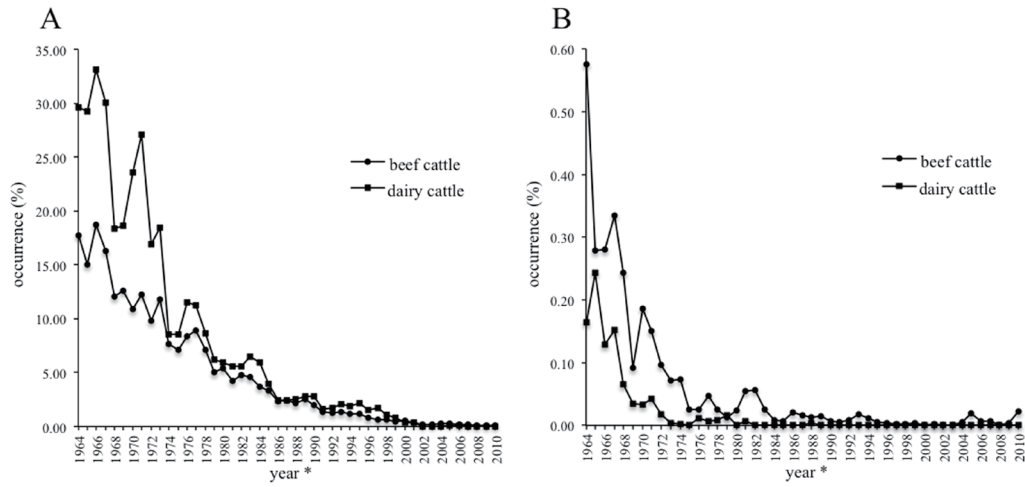


Fig. 1. Change in the annual occurrence of *Fasciola* (A) and *Eurytrema* (B) infections in beef and dairy cattle that were slaughtered at Tokyo Metropolitan Shibaura Slaughterhouse during 1964 to 2010. *: Annual data before 1973 were from January to December, those after 1973 were from April to March of the next year, and those in 1973 were from January 1973 to March 1974.

Table 1. Occurrence of *Fasciola* and *Eurytrema* infections in cattle from different regions of shipment detected at Tokyo Metropolitan Shibaura Slaughterhouse during 2009 to 2012

Region of shipment	No. inspected	<i>Fasciola</i>		<i>Eurytrema</i>	
		No. infected	Prevalence (%)	No. infected	Prevalence (%)
Hokkaido	25,882	10	0.04	0	0
Aomori	12,891	7	0.05	0	0
Iwate	35,215	37	0.11	0	0
Miyagi	30,849	9	0.03	0	0
Akita	2,011	2	0.10	0	0
Yamagata	8,971	13	0.14	14	0.16
Fukushima	39,206	10	0.03	0	0
Gunma	15,790	4	0.03	0	0
Ibaraki	32,251	20	0.06	0	0
Tochigi	61,886	23	0.04	26	0.04
Saitama	9,270	11	0.12	0	0
Chiba	11,546	4	0.03	16	0.14
Tokyo	983	1	0.10	0	0
Kanagawa	2,180	2	0.09	0	0
Niigata	45,919	3	0.07	0	0
Gifu	672	1	0.15	0	0
Mie	8,097	18	0.22	1	0.01
Shiga	1,662	2	0.12	0	0
Tottori	3,301	2	0.06	0	0
Shimane	6,162	5	0.08	19	0.31
Yamaguchi	1,197	5	0.42	0	0
Kagawa	621	2	0.32	0	0
Fukuoka	615	5	0.81	7	1.14
Saga	4,234	9	0.21	4	0.09
Nagasaki	1,724	3	0.17	0	0
Oita	1,787	1	0.06	0	0
Kagoshima	13,172	25	0.19	31	0.24
Okinawa	59	1	1.69	3	5.08
Total	378,153	235	0.06	121	0.03

Slaughterhouse from 44 regions (prefectures). Infected cattle with *Fasciola* were found in 28 regions with regional occurrence from 0.03% to 1.69% (Table 1). The highest occurrence was found in Okinawa (1.69%), followed by Fukuoka (0.81%) and Yamaguchi (0.42%). In contrast, infected cattle with *Eurytrema* were found only in 9 regions with regional occurrence from 0.01% to 5.08%. The highest occurrence was found in Okinawa (5.08%), followed by Fukuoka (1.14%) and Shimane (0.31%). On the other hand, the cattle infected with *Fasciola* were produced at 26 regions, while those with *Eurytrema* were produced at 6 regions (Table 2). Unfortunately, since a total number of cattle produced at each production region were not obtained, the regional occurrence of the parasites among production regions could not be compared. Nevertheless, 46.8% of the infected cattle with *Fasciola* were produced in Iwate (56 cattle), Kagoshima (28 cattle) and Hokkaido (26 cattle), and 97.5% of the infected cattle with *Eurytrema* were produced in Okinawa (84 cattle), Shimane (20 cattle) and Kagoshima (14 cattle).

Breed and sex differences in occurrence: The cattle breed that most inspected during the study period was Japanese Black, followed by F1 crossbred. The occurrence of the parasite infections among cattle breeds was significantly different (Table 3). For fasciolosis, Japanese Shorthorn and others showed the highest occurrence. For eurytrema, Japanese Shorthorn showed the highest occurrence. Japanese Black showed higher occurrence than F1 crossbred, while there was no infected cattle in Holstein. There was no significant sex difference in the occurrence, except for *Eurytrema* infection in Japanese Black.

Change in occurrence by age in month of cattle: Among cattle aged from 25 to 36 months, *Fasciola* infection was detected in cattle aged from 26 to 35 months with the occurrence from 0.03% to 0.08%, and *Eurytrema* infection was detected in cattle aged from 25 to 36 months with the occurrence from 0.03% to 0.11%. Obvious tendency of occurrence in the age of cattle at slaughter was not recognized (correlation coefficient for *Fasciola* infection was $r=-0.47$, and that for *Eurytrema* infection was $r=0.60$).

Responses to questionnaire: A total of 118 famers gave responses to a questionnaire. Thirteen respondents (11.1%) recognized that fasciolosis occurred in their own farms. When this occurrence was compared to the factors relating to the attention of the disease, there was a significant relationship between the occurrence and the knowledge about fasciolosis (Table 4). However, a significant relationship was not obtained either with testing parasite infections or with use of anti-*Fasciola* dewormers. For eurytrema, only 2 respondents (1.7%) recognized the occurrence in their own farms. Since the number of respondents experiencing the disease is small, correlation analysis with the attention factors was not performed.

DISCUSSION

In meat inspection, whole organs were not completely examined for parasites infections, and therefore, the occurrence reported in this study could be underestimated.

Table 2. Number of *Fasciola* and *Eurytrema* infected cattle detected at Tokyo Metropolitan Shibaura Slaughterhouse during 2009 to 2012, classified by the regions of production

Region of production	No. infected with	
	<i>Fasciola</i>	<i>Eurytrema</i>
Hokkaido	26	0
Aomori	5	0
Iwate	56	0
Miyagi	4	0
Akita	5	0
Yamagata	11	0
Fukushima	5	0
Gunma	2	1
Ibaraki	6	0
Tochigi	5	0
Chiba	2	0
Nagano	1	0
Gifu	1	0
Hyogo	6	0
Okayama	1	0
Hiroshima	7	0
Shimane	4	20
Yamaguchi	5	0
Fukuoka	3	1
Saga	3	0
Nagasaki	11	1
Kumamoto	3	0
Oita	12	0
Miyazaki	17	0
Kagoshima	28	14
Okinawa	6	84

However, data accumulated at the slaughterhouse have been obtained by the same procedure, and thus, the quality of data is valid enough for analyzing the time trend in occurrence and for evaluating the difference in occurrence among various factors.

Compared to the mid 1960s, the occurrence of bovine fasciolosis and eurytrema decreased to about 1/100 and 1/10, respectively, and thus, a trend of decrease in occurrence was recognized in a recent half-century. Especially, pronounced decreases were recognized in 1960s and 1970s, presumably due to the modernization of farm rearing system for cattle in Japan conducted during this period. In this period, a confined cattle-rearing system in a barn had been popularized, leading to the reduction of chance for cattle eating rice plant and freshwater plants. Especially for fasciolosis, reduction in the number of rice plantation around cattle farms, use of imported rice straw that had been sterilized for the metacercariae by steam and development of effective anthelmintic [4] against *Fasciola* may have also affected.

Although the occurrence of fasciolosis has been less than 0.2% in the recent five years (2006 to 2010), we sometimes encountered the calcification of bile duct, but worms were not detected. Since this pathological change is characteristic in *Fasciola* infection, we assumed that the observed patho-

Table 3. Breed- and sex-classified prevalence of *Fasciola* and *Eurytrema* infections in cattle that were slaughtered at Tokyo Metropolitan Shibaura Slaughterhouse from 2009 to 2012

Breed	No. inspected			No. (%) infected with <i>Fasciola</i>			No. (%) infected with <i>Eurytrema</i>		
	Total	Male ^{b)}	Female	Total	Male ^{b)}	Female	Total	Male ^{b)}	Female
Japanese Black	254,059	148,629	105,430	222 (0.087) ^{c)}	124 (0.083)	98 (0.093)	119 (0.047) ^{c)}	45 (0.030) ^{d)}	74 (0.070) ^{d)}
F1 crossbred ^{a)}	88,555	49,542	39,013	6 (0.007) ^{c)}	1 (0.002)	5 (0.013)	2 (0.002) ^{c)}	1 (0.002)	1 (0.003)
Japanese Shorthorn	269	162	107	1 (0.372) ^{c)}	1 (0.617)	0 (0)	0 (0) ^{c)}	0 (0)	0 (0)
Others	609	379	230	6 (0.985) ^{c)}	4 (1.055)	2 (0.870)	0 (0) ^{c)}	0 (0)	0 (0)
Holstein	1,925	1,841	84	0 (0) ^{c)}	0 (0)	0 (0)	0 (0) ^{c)}	0 (0)	0 (0)

a) F1 crossbred with Japanese Black. b) All males were castrated. c) Significant difference ($P < 0.01$) was observed among cattle breeds in *Fasciola* and *Eurytrema* infections. d) Significant difference ($P < 0.01$) was observed among sexes of Japanese Black in *Eurytrema* infection.

Table 4. Relationship of the occurrence of fasciolosis in respondents' farms with (A) knowledge about fasciolosis, (B) testing parasite infections and (C) use of anti-*Fasciola* dewormers

A. Knowledge about fasciolosis (n=116)		$P=0.022$		
Element	Occurrence of fasciolosis			
	yes	no	total	
infection route and pathogenicity to cattle	7	24	31	
only name of disease	6	52	58	
not well	0	27	27	
total	13	103	116	

B. Testing parasite infections (n=114)		$P=1.000$		
Element	Occurrence of fasciolosis			
	yes	no	total	
regularly	0	4	4	
irregularly	1	11	12	
not at all	12	86	98	
total	13	101	114	

C. Use of anti- <i>Fasciola</i> dewormers (n=117)		$P=0.222$		
Element	Occurrence of fasciolosis			
	yes	no	total	
yes	4	15	19	
no	9	89	98	
total	13	104	117	

logical change was caused by *Fasciola* infection and that actual infection may have occurred in higher number of cattle.

Recent *Fasciola* infection seems to have occurred in a wide area of Japan. It is difficult to determine where and when the cattle were infected, because the infection of *Fasciola* lasts several years in cattle. Considering that *Fasciola* infection was detected in cattle from variable regions of shipment (28 regions including most-northern prefecture, Hokkaido and most-southern one, Okinawa) as well as those of production (26 regions), the infection may take place in both regions of shipment and of production. It is supported by the facts that a suitable intermediate host of *Fasciola*, *Austropeplea ollula*, distributes widely in Japan [17] and feeding of rice straw is popularly conducted all over Japan.

Among the cattle breeds, Japanese Shorthorn showed the higher occurrence of fasciolosis. Probably, this is because cattle of this breed are commonly put in pasture during spring and autumn for their growing period [10, 11], and they would have been infected during this period. Cattle breed categorized as others also showed higher occurrence. However, all of the infected cattle of others were originated from the same shipper, and thus, it seemed that the occurrence simply reflected the situation of the shipper's farm. When comparing Japanese Black and F1 crossbred, the former showed higher occurrence. This may suggest that Japanese Black is fed with rice straw more frequently and has higher chance of being pastured than F1 crossbred.

It was reported that higher occurrence of fasciolosis was recognized in cattle aged from 5 to 8 years [16, 18]. However, since our slaughterhouse does not process old animals, cattle with age between 25 to 36 months old have been investigated in this study. Among those cattle, an obvious relationship between month in age at slaughter and the occurrence of *Fasciola* infection was not recognized. This may indicate that *Fasciola* infection may have mainly occurred in their younger age.

For *Eurytrema* infection, the current occurrence at our slaughterhouse became less than 0.03%. Since cattle are infected by ingesting a grasshopper that is infected with the metacercariae, *Eurytrema* infection to cattle is generally considered to occur during pasturing. *Eurytrema* infection was detected in cattle, mostly in Japanese Black, from the restricted regions of production and shipment and thus showed a local endemicity. The infection was detected in cattle from 9 shipping regions, but 97.5% of infected cattle were produced in 3 regions of Shimane, Kagoshima and Okinawa where *Eurytrema* was reported to be endemic [6]. In those regions, it is commonly conducted to put Japanese Black cattle in pasture under warm climates. It was reported that hares could serve as the definitive host of *E. coelomaticum* in Kagoshima region, maintaining the pasture-cycle of the parasite [19]. Therefore, it is reasonable to consider that most of the infection took place in those production regions, and then, cattle were moved to shipping regions. Supporting this, it is commonly conducted in shipping regions that Japanese Black cattle are fattened in confined barns without pasturing, and thus, it is hard for fattening cattle to ingest an intermediate host of grasshoppers. Moreover, an obvious

difference in occurrence was not observed among different ages of cattle at slaughter, and this can support that *Eurytrema* infection would mainly occur in their growing age in the production regions [5].

The questionnaire survey revealed that farmers experiencing fasciolosis had more knowledge about the disease. This indicates the importance of an active feedback of inspection data. However, unfortunately, other factors relating to the attention, such as testing parasite infections and use of anti-*Fasciola* dewormers, were not affected by the recognition of occurrence. It may indicate that farmers' attention to fasciolosis was low presumably due to the current low occurrence. Nevertheless, the infection with *Fasciola* as well as *Eurytrema* has still been encountered at the slaughterhouse. Therefore, veterinarians in meat sanitary inspection stations should continue to provide update information of the occurrence of the parasite infections to farmers and local veterinarians in order to raise the attention of production field to the parasitic diseases.

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