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RESEARCH & KNOWLEDGE

Research Article

Situation of liver fluke and cholangiocarcinoma among patients and risk groups of liver fluke in Maha Sarakham Province, Thailand

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Abstract - The objective of this point cross-sectional study was to explore the situation of liver fluke, cholangiocarcinoma, preventive behaviors and other health-risk behaviors among patients and risk groups of liver fluke at Nadoon Municipality, Nadoon District, Maha Sarakham Province, Thailand. A simple random sampling technique was used for selecting 1,148 villagers from 10 villages of Nadoon District, which was carried out from January 2014 to February 2014. All subjects were interviewed. Results showed that the incidence rate of liver fluke was 2.3% and the cholangiocarcinoma incidence rate was 2.8%. The subjects were female (53.2%), married (81.3%) and older (40-60 years) (68.3%). Most of them had a primary school education level (69.9%), were agriculturists (68.6%), families had 3-4 persons (41.9%) and had monthly family incomes lower than 5,000 Baht (72.9%). The largest source of information about liver fluke was 80.7% from public health personnel and 77.5% from television/radio. Raw fish dishes were among the favorite foods with a medium period of intake of 34.0 years, which comprise of more than 50% intake of bonnelville raw fish (jaew bong), orange little fish (som pla noi), food with raw pickled fish, raw fish consumed (mam) and raw pickled fish (pla jom), (range 53.7%-81.8%). The rest consumed it in the range from 9.2%-43.4%, such as raw fermented fish (som pla), minced fermented fish with chili paste and raw fish with spiced salad (koi pla). Stool exams were performed for the detection of liver flukes (27.4%). Liver fluke anthelmintic drug intake was by 31.3%, who obtained it manly from health services as 42.1%. Although many control programs have been used among them, the incidence of reinfection remained the same. Precise and regular main raw fish dishes, drug intake, hygienic defecation data directly from public health personnel and television/radio should be strong concerned with sufficient diagnosis and treatment.

Keywords: Liver fluke, cholangiocarcinoma, patients, risk group of liver fluke

1. Introduction

Opisthorchiasis caused by *Opisthorchis viverrini* (OV) remains a major public health problem in many parts of Southeast Asia, including Thailand, Lao PDR, Vietnam and Cambodia. The infection is associated with a number of hepatobiliary diseases. Moreover, both experimental and epidemiological evidence strongly implicates liver fluke infection as the major risk factor in cholangiocarcinoma (CCA) (Kaewpitoon *et al*., 2008). Throughout the Mekong River region, eating raw and or partially cooked fish causes CCA. The transmission is by native fish, but aquaculture fish also have high infection levels (Pitaksakulrat *et al*., 2013). Inadequate knowledge, misbeliefs and social, cultural and unhealthy behaviors are highly influential factors leading to the maintenance of risk behaviors. Moreover, unhygienic defecation and insufficient diagnosis and treatment were found to facilitate OV transmission.

Although a control program has been used, the incidence of OV infection remains the same. For the current status and control of liver fluke infections in the Mekong Basin countries, where OV is highly endemic, the prevalence and distribution were summarized in presentations from the "96 Years of OV International Congress of Liver Flukes". Despite treatment and control programs having been in place for decades, all countries of the Lower Mekong Basin are still highly endemic with OV as well as alarmingly high levels of CCA incidence (Sithithaworn *et al*., 2012). OV ecology, pathology and epidemiology have been considered more deeply than the socio-cultural dynamics with which the food-borne species complexes are associated. The Mekong region is characterized by strong livelihood and lifestyle associations within wetland ecosystems, which are inseparable from human eating habits. Within the fish-rice economies of the region there

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are many long-cherished food cultures based on eating raw, semi-cooked and fermented fish dishes, which are known to lead to opisthorchiasis and potentially CCA (Grundy-Warr *et al*., 2012).

Liver fluke infection is induced by eating raw or uncooked fish products that are traditional and popular in the northeastern region, particularly in rural areas of Thailand (Kaewpitoon *et al*., 2008). OV infection due to eating freshwater fish dishes is the risk factor for CCA in the Northeast of Thailand where the infection is endemic and the incidence of primary liver cancer has been the highest in the world. The prevalence of OV infection was 37.2% and was highest in the 20-35 year age group. As many as 91.8% reported eating fish dishes, which lead them to be at risk of infection (Saengsawang *et al*., 2013). Unsuccessful eradication of the disease is probably due to the culture of eating raw and undercooked fish, especially from the endemic area. It is also interesting that villages with infection are mostly located in the vicinity of the Chi River (Aunpromma *et al*., 2012). The previous use of praziquantel (PZQ) and a lack of drug use as a protective effect against re-infection were the factors related to OV infection (Saengsawang *et al*., 2013). PZQ is the drug of choice for morbidity control, however, only a single drug, PZQ is available. Low dose comparisons have been made (Lovis *et al*., 2012).The infections are still prevalent in Northeastern Thailand. Thus, prospective epidemiologic information is needed for developing more effective public health interventions. A rural community in Thailand from December 2002-February 2004, reported the incidence rate of opisthorchiasis was 21.6/100 person-years. The independent factors associated with opisthorchiasis were an age > 60 years and consuming chopped raw fish salad (Koi pla) (Suwannahitatorn*et al*., 2013). Peak intensity in both males and females occurred at age 40 and above. A history of eating "koi pla", a symptom of liver fluke, occurred most frequently in the infected groups and was correlated with the intensity of infection (Upatham *et al*., 1982).

Among the various types of chronic helminthic infections, only liver diseases, with a long-stand in inflammation are found to be the causative agents of cancer. Opisthorchiasis and OV-associated bile duct cancer are prevalent among the residents in Northeastern Thailand, who prefer to consume raw or improperly cooked cyprinid fish. Although numerous studies have reported the presence of OV metacercariae in freshwater cyprinid fishes, none of the data has been derived directly from cyprinid fish dishes. Here, the metacercariae were determined in a number of cyprinid fish dishes that are among the favorite foods of Northeastern Thais. Traditional raw fish salad or cyprinid fish fermented in sticky rice for 69 hours were found to be not safe to consume (Prasongwatana *et al*., 2013). Regular main raw fish dishes, drug intake and other related data needs to be described for these further helminthic food-borne outbreak reduction strategies.

2. Materials and methods

2.1 Study design and population

This point cross-sectional study was carried out from January 2014 to February 2014 as part of a liver fluke survey that included CCA, preventive behaviors and other health-risk behaviors among patients and risk groups of liver fluke at Nadoon Municipality, Nadoon District, Maha Sarakham Province, Thailand. The proposal was reviewed and approved by the Ethics Committee for Research in Human Subjects of Mahasarakham University (Ref. No. MSU 184/2557) and informed consent was sought from the villagers. Villager consent was required for participation of those aged >40 years. A simple random sampling technique was used for selecting the 1,148 villagers from 10 villages in Nadoon District. First stage, these districts were randomly selected and represented the general characteristics and social structure of villagers in Maha Sarakham Province. Second stage, all villages were selected. All villagers in the villages were eligible to participate under the inclusion criteria of living in the study place at least 6 months, aged >40 years, patients and risk group of liver fluke, cooperate and agree to participate, relative got sick with liver fluke and have raw fresh fish intake. Exclusion criteria were that they were not from the place of study and did not agree to data collection. Data were collected using an interview questionnaire that consisted of four parts: general characteristics, media perception, risk of liver fluke behaviors and prevention of liver fluke behavior, which was administered by trained village health volunteers. Details of the study were explained and an informed consent form was signed by all the participants. The main reason for non-response was absenteeism on the day of the perception survey. Participant interviews took approximately 30 minutes to complete. Individual answers were kept confidentially.

2.2 Statistical analysis

General characteristics, media perception, risk of liver fluke behaviors and prevention of liver fluke behavior factors were given as numbers and a percentage.

3. Results

In all, 1,148 villagers (58.9% of the total sample) were self-identified as current participants. Most of them were female (53.2%) and the mean age was 54.5 years (range 40-89). Most of them were 40-60 years old (68.3%), married (81.3%), agriculturists (68.6%), had family members of ≥5 (39.0%) and 3-4 person (41.9%). Monthly family income was from lowest of <5,000 Baht (72.9%) to highest >7,001 Baht (22.6%). Their educational level ranged from 4.3% - 8.7% with no education to under graduate/diploma. Most of the group had primary school level education (69.9%). The majority of study subjects had enough money (68.0%). There were underlying diseases of 26.5%, past CCA (2.8%), liver diseases (21.5%) and signs of CCA which they reported as only flatulence (17.4%), dyspepsia at xiphoid process (5.7%) and dull pain at right ribs (3.1%). Their relatives had liver fluke of 11.5%, and relative/family member with known CCA (6.2%), as shown in Table 1.

Table 1. General characteristics of participants (N = 1,148).

Variables	Number	Percentage
Gender		
Male	537	46.8
Female	611	53.2
Age (yrs)		
40-60	784	68.3
>60	364	31.7
($\bar{x}$) = 54.5,± S.D. = ±11.2		
Min. = 40 Max. = 89		
Marital status		
Married	933	81.3
Widowed/divorced/separated	155	13.5
Single	60	5.2
Education		
No education	49	4.3
Primary school	803	69.9
Secondary school	196	17.1
Under graduate/diploma	100	8.7
Occupation		
Agriculturist	787	68.6
Employee	183	15.9
Merchant	101	8.8
Government officer/state enterprise	77	6.7
Monthly family income (Baht)		
<5,000	838	72.9
5,001-6,000	40	3.5
6,001-7,000	11	1.0
>7,001	259	22.6
Monthly family income expenses		
Enough income	780	68.0
Not enough income	261	22.7
Needs to borrow money	91	7.9
No report	16	1.4
Family members (persons)		
1-2	219	19.1
3-4	481	41.9
≥5	448	39.0
Underlying disease		
No	844	73.5
Yes	304	26.5
Cholangiocarcinoma		
No	1,116	97.2
Yes	32	2.8
Liver diseases		
Do not know	901	78.5
Know but no cure	222	19.3
Receiving treatment	9	0.8
Recovered	16	1.4
Signs of cholangiocarcinoma		
None	872	76.1
Do not want to report	295	25.7
Flatulence	200	17.4
Dyspepsia at xiphoid process	65	5.7
Dull pain at right ribs	36	3.1
Relative with liver fluke		
No	1,016	88.5
Yes	132	11.5
Relative/family member with CCA		
No	965	84.0
Do not know	112	9.8
Yes	71	6.2
(71 person)		
1	67	94.4
≥ 2	4	5.6

For the perception of liver fluke data, the majority of villagers received liver fluke data (77.1%), with the main source being from public health personnel (80.7%), follow by village health volunteers (56.0%). Approximately 77.5% of villagers got data from television/radio, 31.4% from publish material and14.8% from the internet as shown in Table 2.

Table 2. Perception of liver fluke data (N = 1,148).

Variables	Number	Percentage
Receive data (answer >1 item)		
No	263	22.9
Yes	885	77.1
Person who give data (answer>1 item)		
Public health personnel	926	80.7
Village health volunteers	643	56.0
Family member	342	29.8
Neighbor	255	22.2
Other	75	6.5
Data from media (answer >1 item)		
Television/radio	890	77.5
Publish material	360	31.4
Internet	170	14.8
Other	119	10.4

Liver fluke risk behaviors were namely the period that freshwater fish was consumed and the majority of study samples were similar and consumed <20 years (21.3 %) and 31-40 years (20.2%). The median period was 34.0 years (range 1-80 years). They had consumed undercooked/raw food (84.9%), with 77.2% pork/beef/buffalo meat and fish 51.7 %. Most of them consumed fresh water scale fish/ crustacean-clad body more than 50%, such as bonnelville raw fish (jaew bong), raw shrimp, fermented little fish (som pla noi), food with raw pickled fish, raw fish eggs (mam) and raw pickled fish (pla jom) (range 53.7%-81.8%). The rest consumed in the range from 9.2%-43.4%, such as raw fermented fish (som pla), minced fermented fish with chili paste, raw fish with spiced salad (koi pla), whole body raw fish, raw shellfish and one solid sun fish (pla salid). Raw freshwater fish consumption at present was 62.2% as shown in Table 3 and Figures 1.

Table 3. Liver fluke risk behavior (N = 1,148).

Variables	Number	Percentage
Period of freshwater fish intake (yrs)(n=782)		
<20	244	21.3
21-30	168	14.6
31-40	232	20.2
41-50	138	12.0
>51	71	6.2
Median = 34.0, Q.D 10.0 ± Min= 1 Max = 80		
Undercooked/raw food intake		
No	172	15.1
Yes	976	84.9
Kind (answer >1 item)		
- Pork/beef/buffalo meat	886	77.2
- Fish	593	51.7
- Others	81	7.1

Table 3. Liver fluke risk behavior (N = 1,148). (Cont.)

Variables	Number	Percentage
Fresh water scale fish/clustacean-clad body intake (answer>1 item)		
Bonnelville raw fish (jaew bong)	939	81.8
Raw shrimp	735	64.0
Fermented little fish (som pla noi)	691	60.2
Food with raw pickled fish	654	57.0
Raw fish (mam)	640	55.7
Raw pickled fish (pla jom)	617	53.7
Raw fermented fish (som pla)	498	43.4
Minced fermented fish with chili paste	429	37.4
Raw fish with spiced salad (koi pla)	340	29.6
Whole body raw fish	182	15.9
Raw shellfish	130	11.3
One solid sun fish (pla salid)	106	9.2
Raw freshwater fish consumption at present		
No	434	37.8
Yes	714	62.2

Figure 1. (1) Spiced salad (koi pla), (2) fermented little fish (som pla noi), (3) raw fermented fish (som pla), (4) raw fish eggs (mam), (5) raw fish (jaew bong) and (6) pickled fish (pla jom).

For liver fluke preventive behavior, the villager protect themselves with a stool exam 27.4%. Approximately 19.7% of them had a history of being examined more often than twice and 80.3% of them had a history being examined only once. Only 2.3 % have got sick due to liver flukes while 96.3% have not. Only 31.3% of them have taken a liver fluke anthelmintic drug and the places to receive the liver fluke anthelmintic drugs were community hospital (district) (15.5%) and medical center (15.3%). The places with less contacted were health promotion hospital (7.8%) and general hospital/medical center hospital that were contacted only 1.7%, as shown in Table 4.

Table 4. Liver fluke preventive behavior (N=1,148).

Variables	Number	Percentage
Stool exam		
No	833	72.6
Yes	315	27.4
Number (episode)		
1	922	80.3
2	161	14.0
3	52	4.5
5	10	0.9
10	3	0.3
Liver fluke cases		
No	1122	97.7
Yes	26	2.3
Liver fluke occur		
1	1106	96.3
2	42	3.7
Liver fluke anthelmintic drug intake		
No	789	68.7
Yes	359	31.3
Places for liver fluke anthelmintic drugs		
None	665	57.9
Community hospital	178	15.5
Medical center	176	15.3
Health promotion hospital	90	7.8
General hospital/ medical center hospital	19	1.7
Other	20	1.8

4. Discussion and conclusions

Our findings demonstrated that the incidence rate of liver fluke was 2.3% and CCA incidence rate was 2.8%. This result may be an underestimate. One of the reasons may be that they were afraid of CCA but were not concerned about liver fluke, despite the fact that they had been assured that the liver fluke anthelmintic drug could be a cure and that no hygienic raw fish dishes information would be accepted. Villagers facilitated the start of the risk behaviors, for instance, consuming raw fish dishes and stool exams were not performed for the detection of liver fluke (72.64%), while the liver fluke anthelmintic drug intake was 31.3% as well as other parasitic drugs. This study confirmed that older villagers had been consuming raw fish dishes longer than the younger ones. For the poor socioeconomics-related behaviors, most of them that had a primary school education (69.9%), were agriculturists (68.6%), families with 3-4 persons (41.9%) and had monthly family incomes lower than 5,000 Baht were at higher risk of consuming low cost/preserved/naturally found food, such as freshwater fish to make pickled fish dishes. In addition, those who consumed raw fish dishes were at a higher risk of serious CCA in their lives. This corresponds with the results of previous studies (Sithithaworn *et al*., 2014; IDRC, 2015). These behaviors may lead to high parasitic infection from raw fish dishes. This study was subject to a few limitations. First, the present study was a point cross-sectional survey, so it was difficult to make statements about the cause and effect relationships between the health-risk behaviors among patients, the risk groups and infected liver fluke. Second, these data applies only to those aged >40 years in the study samples and, therefore, are not representative of all the population. Moreover, the data collection during the day might exclude samples who went out from the village, or those who could not be free from their work places. Finally, all data were based on the villagers' self-reporting, which may lead to a variation in the number of raw fish dishes consumed and

other related risk/protective/media perception behaviors.

This study described the risk/protective/media perception behaviors and liver flukes among villagers. Therefore, a reduction in the raw fish dishes consumed should be performed. The effective intervention strategies, such as regular perception of liver fluke data from public health personnel and from television/radio, increased screening of disease by stool exam, reduced exposure to anthelmintic drug advertising and marketing, implementation of comprehensive community-based programs to prevent raw fish dishes being consumed and development of a national media campaign to reduce risky consumption of food, etc. to reduce liver flukes should be targeted to the new generation (IDRC, 2015; Sithithaworn *et al*., 2014). Family members, public health personnel and neighbors are the first key factor to promote immunity against raw fish dishes being consumed (Sripa *et al*., 2007). Not only family member roles but also good practice models could be done. Poor advice, family dissolution and negative perception influence have all been shown to be linked with liver flukes. In addition, a policy for the development of prevention programs aimed to prevent/reduce raw fish dishes consumed and integrate the programs into informal education should be initiated. Moreover, a policy advocating food safety in multiple settings against fish dishes, such as the selling of safe ready make/package food should be promoted. The health promoting village model should be utilized to develop the effective village healthy food programs to relieve these problems. These measures could go a long way in not only reducing the number of patients/risk groups but also make a positive overall contribution to public health. However, prevention of raw fish dishes consumption cannot be successful by focusing on villagers alone, the efforts strongly need to focus on youths, local teachers, their family members, their neighbors and adults (Sithithaworn, 2014).

CCA prevention and control is through preventing liver fluke infection. The campaign against liver fluke, particularly against raw fish consumption, has met with little success, although the fluke itself was discovered over a century ago. Even though drug treatment with Praziquantel (PZQ) is effective and has been available for more than 30 years. It is believed that disruption of the liver fluke life cycle is necessary to achieve parasite control, but health education is always thought to be the best strategy. Now, the target is the younger generation, particularly primary school children, with school-based health education programs. The ultimate aim of the program is to educate these children so they are aware of liver fluke infection, thus lowering the risk of CCA 20-30 years later in life. The key method is a systematic training program for school teachers. In addition to local public health workers, the teachers can play roles in transferring the health message to the children, their parents at home and also the communities (Sithithaworn, 2014).

The Ministry of Public Health used PZQ to treat infected community members and their animals. Village health volunteers were trained on the life cycle and transmission of the liver fluke and how villagers can protect themselves. These volunteers, village leaders, local health officers and school teachers were the keys to the change. They organized public exhibits to raise awareness. For example, they used microscopes to show villagers infected liver specimens and the flukes themselves. They used folk songs and videos to explain transmission, health volunteers made home visits and organized liver fluke campaigns in local schools, educating the community on the risks of eating raw fish and safe defecation practices (IDRC, 2015). The combination of treatment and community education has been highly effective because of the parasite's complex life cycle. By following treatment and sanitation measures, community members have broken this cycle, as the host fish have a life span of two to three years. The health education programs to prevent and control opisthorchiasis are still required in high-risk areas (Kaewpitoon *et al*., 2008). Thus, avoiding raw fish salad should be emphasized in the national control program (Suwannahitatorn *et al*., 2013).

By using approaches to infectious disease and socio-economic research, ecological and systems science perspectives, researchers have been able to strengthen protection by looking at local dietary and sanitation practices that might be fostering transmission (IDRC, 2015). For disease prevention and health education approaches to be most effective, they must be sensitive to the culture, livelihood economics, gender and age. Further international research must incorporate the complex dynamics of parasite ecology, human behavior, socio-economics and public health awareness (Grundy-Warr *et al*., 2012).

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