RESEARCH ARTICLE

Infection with *Opisthorchis viverrini* and Use of Praziquantel among a Working-age Population in Northeast Thailand

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Abstract

Infection with *Opisthorchis viverrini* (OV) due to eating certain traditional freshwater fish dishes is the principal risk factor for cholangiocarcinoma in Northeast Thailand where the infection is endemic and the incidence of this form of primary liver cancer has been the highest in the world. This paper is the second report of a prospective research project to monitor the impacts of a national liver fluke control programme in a rural community of Northeast Thailand. A sample of 684 villagers aged 20-65 years completed an interview questionnaire and were tested for infection using the Kato thick smear technique. The questionnaire was designed for the exploration of associations between OV infection, previous treatment with praziquantel, and knowledge and beliefs about the drug. The data were analysed using descriptive statistics and multiple logistic regression. The overall prevalence of OV infection was 37.2% and was highest in the 20-35 year age group, in those with a university degree and in those employed in the government sector. As many as 91.8% reported eating fish dishes known to place them at risk of infection. In the multiple regression analysis, previous use of praziquantel and lack of knowledge about whether or not the drug has a protective effect against re-infection were the only factors related to OV infection (OR_{adj}= 2.31, 95%CI =1.40-3.79 and OR_{adj}= 1.95, 95%CI= 1.24-3.05). The findings were discussed in terms of the possibly unwise dependency on praziquantel as a primary element in a control programme.

Keywords: *Opisthorchis viverrini* - praziquantel - working-age population - Northeast Thailand

Asian Pacific J Cancer Prev, **14 (5)**, 2963-2966

Introduction

Opisthorchis viverrini (OV) is a liver fluke parasite, and OV infection is the most important risk factor for cholangiocarcinoma (CHCA) in the northeast region of Thailand where the infection is endemic (Watanapa et al., 2002; Poomphakwaen et al., 2009). In Thailand, approximately 10% of the population or about six million people are infected with this liver fluke (Bureau of Epidemiology, 2008), and in the period 2004 to 2008 the OV infection rate was higher in the northeast than in any of the other regions (Bureau of Epidemiology, 2009). However, there is a large geographic variability among the provinces in this region with infection rates reported to vary between 4.6% and 60% (Sithithaworn et al., 2012).

In Khon Kaen, one of the provinces in the northeast region with historically very high rates of OV infection, the incidence rate of CHCA has been higher than in any other part of the world (Sripa et al., 2007). Every year over 1000 new cases of CHCA present at the university hospital in Khon Kaen, and this incidence rate appears to have shown no sign of declining over the decades since the association with OV infection was known (Sripa et al., 2012). Data retrieved from the Khon Kaen Cancer registry shows that in the period 1998-2009 over twice as many men than women presented with CHCA, and, whereas 45.2% were 65 years of age or older, 52.5% were 20-65 years old with most of this group aged between 50 and 65 years (Kamsa-ard et al., 2011). These findings underscore the observation that a large proportion of deaths due to CHCA occurs in the male heads of households who are the principal sources of family income (Andrews et al., 2008; Sripa et al., 2012).

The major risk factor for OV infection in the northeast of Thailand has been the consumption of raw, undercooked or freshly pickled freshwater cyprinid fish in local, traditional dishes, such as *koi pla* (Upatham et al., 1984; Rangsin et al., 2009) and *pla som* and *pla ra* (Rhongbutsri and Kitvatanachai, 2002; Sithithaworn and Haswell-Elkins, 2003). Metacercariae are the source of OV infection in a definitive host such as man, and the presence of viable metacercariae has been directly demonstrated in a variety of differently prepared traditional cyprinid fish dishes (Prasongwatana et al., 2012).

The Thai Ministry of Public Health currently advocates three strategic guidelines for the prevention and control of

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OV infection: (i) stool examination, (ii) health education, and (iii) the treatment of positive cases of OV infection with praziquantel (Jongsuksuntigul and Imsomboon, 1997; Bureau of Epidemiology, 2009). The anthelmintic, praziquantel, has been more widely used than any other drug for treatment of OV infection because of its apparent effectiveness and the short time for the healing of damage done by OV following its eradication (Boonmars et al., 2007). However, while praziqantel seems effective against an existing infection with cure rates reported to be as high as 96% and 100% (Soukhathammavong et al., 2011), there is no evidence of any protective immunity against re-infection (Sirisinha et al., 1983; Upatham et al., 1988) and repeated dosing with the anthelmintic is necessary for those re-exposed to infection.

The purpose of this study was to investigate the associations between OV infection, past use of praziquantel and knowledge and beliefs about the drug in people of working-age who are exposed to the risks of infection and is the second report of a prospective research project to monitor the impacts of a national liver fluke control programme in a rural community of Northeast Thailand. The findings of this and the first report (Saengsawang et al., 2012) are expected to provide information for the improvement of control programmes and the reduction of risks for CHCA.

Materials and Methods

This cross- sectional analytical study was conducted during January- March, 2012, in Hua Mueang, a subdistrict of Maha Chana Chai, which is located in Yasothon Province, Northeast Thailand. The study area was one of the two subdistricts previously selected by multistage sampling for a cross-sectional survey in Yasothon Province (Saengsawang et al., 2012) and was chosen by simple random sampling. The subdistrict is composed of 14 villages and 1,635 households. The population between 20 and 65 years of age is 2,459, and most (80%) are involved in agriculture.

All villagers aged 20-65 years, who had resided in the study area for at least six months, were invited to participate and agree to provide samples for a stool examination. Stools were examined within two days of collection using the Kato thick smear technique (WHO, 1991). Villagers (n=5), who reported taking praziquantel in the week prior to stool collection, were excluded. Information about socio-demographic characteristics, knowledge and beliefs about praziquantel and previous use of praziquantel were collected on interview using a structured-questionnaire.

The data were analysed with descriptive statistics, and chi-square tests were used to investigate the crude associations with OV infection. Multiple logistic regression with backward elimination of the variables was used to control for confounding factors. The candidate variables were those found to have a statistically significant association with OV infection in the crude analysis and those with no significant association but likely to be related on the basis of previous studies. Statistical significance was set at $p<0.05$.

The research was approved by the Khon Kaen University Ethics Committee for Human Research (reference number HE542213).

Results

Interview questionnaires were completed for 684 eligible participants, and 255 (37.2%) were found to be infected with OV. The results are shown in Table 1.

The infection rate was slightly higher in males than in females, and most of the infected participants were found in the 20-35 year age group (39.3%).

While those who reported eating the fish dishes known to contain viable metacercariae were slightly more likely to be infected (37.7%) than those denying this (32.1%), a large majority (91.8%) had been consuming them.

The only factors found to be associated with OV infection in the multivariate analysis (Table 2) were reported past use of praziquantel (OR_{adj}=2.31, 95%CI=1.40-3.79, p-value=0.001) and ignorance about whether or not praziquantel can prevent OV infection (OR_{adj}=1.95, 95%CI=1.24-3.05, p-value=0.004).

Table 1. Characteristics of Participants

Characteristics (n=684)	OV Infection				Total	
	Positive	%	Negative	%	N	%
Gender						
Male	127	40.9	183	59.1	310	45.3
Female	128	34.2	246	65.8	374	54.7
Age-years						
20-35	24	39.3	37	60.7	61	8.9
36-55	138	37.6	229	62.4	367	53.7
56-65	93	36.3	163	63.7	256	37.4
Education						
Primary education	200	37.3	336	62.7	536	78.4
Certificate(diploma)	3	30.0	7	70.0	10	1.5
Secondary education	46	36.5	80	63.5	126	18.4
Bachelor's degree or higher	6	50.0	6	50.0	12	1.8
Occupation						
Agriculture	234	37.9	383	62.1	617	90.2
Government service	6	50.0	6	50.0	12	1.8
Trade	12	48.0	13	52.0	25	3.7
Other employment	3	10.0	27	90.0	30	4.4
Consumption of raw fish in past year						
Yes	237	37.7	391	62.3	628	91.8
No	18	32.1	38	67.9	56	8.2

Table 2. Multivariate Analysis of Factors Associated with OV Infection

Factors	OV		Crude OR	P-value	Adj. OR*	P-value
	ve+	ve-	(95%CI)		(95%CI)	
Knowledge about side effects of praziquantel						
Not known	219	338	1.00		1.00	
Known	36	91	0.61 (0.40-0.93)	0.022	0.84 (0.50-1.40)	0.506
Past use of praziquantel						
No	176	275	1.00		1.00	
Yes	79	154	0.80 (0.43-0.81)	0.190	2.31 (1.40-3.79)	0.001
Belief that praziquantel can prevent infection						
No, it cannot	68	151	1.00		1.00	
Yes, it can	72	159	1.01 (0.67-1.49)	0.978	0.96 (0.61-1.51)	0.875
Do not know	115	119	2.14 (1.46-3.15)	0.000	1.95 (1.24-3.05)	0.004

*Adjusted for gender, age, consumption of raw fish, occupation, income, and toilet use. OR=Odds Ratio; 95% CI=95%Confidence Interval

Discussion

The prevalence rates of OV infection were not very different across different age-groups and the various occupational and educational backgrounds, and the overall rate (37.3%), which embraces a wide range of working age groups, is similar to that found in the previous study (38.7%), which was based on a larger sample (Saengsawang et al., 2012). These rates are higher than that reported from a 2009 survey (22.5%) by Sithithaworn et al. (2012), but this survey result was for Yasothon Province as a whole and apparently included data for the dryer northern plains which are areas of less risk for infection.

The positive association between past use of praziquantel and infection raises questions about the extent to which the availability of praziquantel and the belief in its effectiveness may encourage people to continue eating the traditional raw or undercooked freshwater fish dishes. Furthermore, the lack of knowledge about whether or not the medication protects against a future infection suggests a 'devil-may-care-attitude', and the finding that the self-reported consumption of raw fish was so high supports this. In general terms, the results present a somewhat gloomy picture of resistance to change in the eating of traditional fish dishes and hence an ongoing exposure to OV infection and the risk of CHCA.

Even if regular testing of all the community in OV endemic areas occurred, the dependence on praziquantel is probably unwise for several reasons. Firstly, the standard method of screening for OV infection has relied on the use of a single Kato-Katz thick smear test which has low sensitivity, especially when the intensity of an infection is light (Lovis et al., 2012). Secondly, a continued extensive use of praziquantel may eventually result in drug resistance, just as has happened with so many antibiotics and antimalarial drugs. In vivo and in vitro tests have shown that praziquantel resistance can develop in another potentially life-threatening trematode, *Schistosoma mansoni* (Melman et al., 2009). Thirdly, it is not known how many people given praziquantel for OV infection actually take it as recommended. Medication non-adherence has been a common problem worldwide in developed countries and is assumed to be an even more serious issue in the developing regions (WHO, 2003). Lastly, and perhaps most importantly, there are now serious concerns that repeated cycles of re-infection and treatment with praziquantel may increase the risk of CHCA (Boonmars et al., 2007; Sripa et al., 2012).

This study has a number of limitations, the most serious of which is that the subjects constituted only a 'convenience' sample, leaving room for many potential sampling biases. One major probable bias was the availability of respondents, which were those present in the villages at the time of data collection. Another is the validity of the information provided by respondents in the absence of independent checks or attempts to minimize errors of recall. These kinds of research limitations are difficult to avoid in areas where funding is often largely unavailable or inadequate. While the findings of studies conducted under these conditions can only be tentative, they can nevertheless raise key issues which justify better resourced investigations.

Acknowledgements

This study was supported by Khon Kaen University (KKU) Graduate School, the KKU Faculty of Nursing Research and Training Centre for Enhancing Quality of Life of Working-Age People, the National Research University Project of Thailand through the Center of Excellence in Specific Health Problems in the Greater Mekong Sub-region (SHeP-GMS), and a KKU research grant (no. NRU542005).

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