Analysis of Risk Areas of *Opisthorchis viverrini* in Rural Communities by Using SUT-OV-001

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Background: *Opisthorchis viverrini is still a serious problem in rural areas of Thailand particularly Northeastern and Northern region. Active surveillance is required to determine the update data for further prevention and control planning.*
Objective: *To determine the population at risk and analyze the risk areas for O. viverrini in rural communities of Nakhon Ratchasima province, Thailand.*
Material and Method: *A cross-sectional survey was conducted between October 2015 and March 2016 at Kang Sanam Nang district, Nakhon Ratchasima province, Thailand. The population at risk for O. viverrini was screened by SUT-OV-001 with Cronbach' alpha coefficient, 0.724. O. viverrini infection was examined by using Kato thick smear. The risk areas were analyzed by using geographic information system.*
Results: *Three hundred ninety seven people were recruited for this study. The majorities were female (53.15%), age group 41 to 50 years old (35.01%), educated with primary school (59.45%), agriculture (85.64%), and of income of 2,000 baht (47.36%). The majorities of them were high-risk (49.62%), followed by moderate risk (36.02%), and low-risk (7.3%). Risk areas were classified as very-high-risk areas, found in Beng Samrong (11.44 km^2), followed by Keang Sanam Nang (5.21 km^2). High-risk areas were found in Bueng Phalai sub-district (70.16 km^2), followed by Bueng Samrong (30.45 km^2), and Non Samran (27.33 km^2). O. viverrini infection was 3.02%, and distributed in the moderate risk areas (four cases), high-risk areas (three cases), low-risk areas (three cases), and very-high-risk areas (two cases).*
Conclusion: *The present study indicates the population at risk for O. viverrini and risk areas in the rural communities by using SUT-OV-001 and GIS. These tools are useful to display the risk areas for further prevention and control planning and monitor.*

Keywords: *Opisthorchis viverrini, SUT-OV, Geographic Information System, Nakhon Ratchasima, Thailand*

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Liver fluke infection caused by *O. viverrini* remains a major public health problem in Thailand particularly Northeastern and Northern region[1-3]. A nationwide survey in Thailand has been done and found that the prevalence was 5.1%. The highest prevalence was found in the northeast (9.2%) and followed by the north region (5.2%)[4]. The *O. viverrini* infection in Nakhon Ratchasima province has been analyzed from the 1,168 stool samples obtained from 516 males and 652 females. The stool examination showed that 2.48% were infected with *O. viverrini*[5]. Recently, we reported the re-examination of *O. viverrini* infection in the three districts of Nakhon Ratchasima province, Thailand. Three hundred fifty five participants were included from a 194,152 population. We found that *O. viverrini* infection was 2.25%[6]. This figure

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indicated that this province still has a problem with *O. viverrini*, particularly in the rural communities.

The *O.viverrini* infection is associated with hepatobiliary diseases including hepatomegaly, cholangitis, cholecystitis, and gallstones[7-9]. It has been classified as Type 1 carcinogens by the International Agency for Research on Cancer, World Health Organization (WHO)[10]. Mortality rate of liver cancer and cholangiocarcinoma in Nakhon Ratchasima province has been reported at 13.67 to 16.2 per 100,000 populations[11]. Eradication of the fluke and identification of high-risk populations are urgently needed.

Material and Method

A cross-sectional survey was conducted between October 2015 and March 2016 at Kang Sanam Nang district, Nakhon Ratchasima province, Thailand. Kang Sanam Nang district is located in Nakhon Ratchasima. It is one of the northeast provinces of Thailand. Neighboring districts are (from the north clockwise) Mueang Chaiyaphum and Khon Sawan of Chaiyaphum province, Waeng Noi of Khon Kaen province, Bua Yai, and Ban Lueam of Nakhon Ratchasima province. The district is further subdivided into five sub-districts (Keang Sanam Nang, Non Samran, Bueng Phalai, Si Suk, and Bueng Samrong) and 48 villages. The district covers an area of 107.3 km^2 (41.4 sq mi), with a population of 37,174. The study protocol was approved by Suranaree University Ethical Review Committee, EC58-64.

Participants were randomly sampled and they completed the SUT-OV-001 with Cronbach' alpha coefficient, 0.724, containing general characteristic data and eight questions for *O. viverrini* screening; (1) raw fish consumption; raw spicy minced fish salad (Lahb Pla) or raw fish in spicy condiment (Koi Pla) (0.689), (2) undercooked fish consumption; raw pickled fish (Pla Som) or raw pickled small fish (Pla Jom) or raw fermented fish (Pla Ra) (0.691), (3) histories with opisthorchiasis (0.685), (4) histories with relative family with cholangiocarcinoma (0.719), (5) intend to raw consume (0.721), (6) family members had a histories with opisthorchiasis (0.694), (7) family member consumed raw fish dish (0.658), (8) household located near natural water reservoirs (<10 kilometers) 0.705). Questionnaires were weighted by parasitological expertise for *O. viverrini*, briefly, No.1; 0.2, No.2; 0.2, No.3; 0.2, No.4; 0.2, No.5; 0.05, No.6; 0.05, No.7; 0.05, and No.8; 0.05, respectively. Population at risk was calculated following 1+2+3+4+5+6+7+8, and then interpreted to very-high-risk (0.9-1.0 point), high-risk (0.7-0.8 points), moderate-risk (0.4-0.6 point), low-risk (0.1-0.3 point), and no risk (0 point), respectively.

Stools were collected and then kept in labeled plastic bags and transported in an icebox to the laboratory at the Parasitic Disease Research Unit, Institute of Medicine, Suranaree University of Technology, Thailand, within a day after collection. Stool specimens were examined the *O. viverrini* egg by the Kato thick smear procedures according to the method of Kato and Miura[12]. Briefly, the materials used were prepared in accordance with standard laboratory in-house procedures. Thus, the glycerin-malachite green solution was mixed with 1 ml of 3% malachite green, 100 ml of 6% phenol and 100 ml of pure glycerin. The cellophane strips, each 22x40 mm, were soaked in this solution for at least 24 hours before use. Stool was transferred to slides covered by the cellophane soaked cover slips and allowed to stand for 30 minutes. All preparations were initially screened with a low-power (10x) objective lens. Suspected parasitic objects were subsequently examined under a high-power (40x) objective. *O. viverrini* positive case was confirmed by two parasitologists before a definitive diagnosis was established. Patients who infected with other known parasitic were treated with anti-parasitic drugs and attended the health education.

ArcGIS 10.2 was used for created map displayed. Spatial database of Keang Sanam Nang district was created containing point of villages, sub district (Tambon) boundary, district (Amphoe) boundary. Attribute database was created containing of personal data and risk points of each person. Data were transferred to WGS84 UTM ZONE 48. The point with the populations at risk for *O. viverrini*, was used to analyze and calculate, according procedures; ArcToolbox >Spatial Analyst Tools >Interpolation >Inverse Distance Weighted, and then Reclassify for classified the areas. Map lay out was created by image management, geographic coordinated, symbolic, and data Framed. Descriptive and analytical statistical data were analyzed with SPSS software.

Results

Three hundred ninety seven participants completed the SUT-OV-001 for this study. The majority were females (53.15%), age group 41 to 50 years old (35.01%), educated with primary school (59.45%), agriculture (85.64%), and income of 2,000 baht (47.36%). Population at risk was screened and calculated. It was found that the majority of them were high-risk (49.62%),

and followed by moderate risk (36.02%), low-risk (7.3%). Data is shown in Table 1. The majorities of participants were habitat in Bueng Phalai (25.69%), followed by Si Suk (21.16%), Non Samran (20.15%), and Bueng Samrong (20.41%) (Table 2). Risk areas were screened by SUT-OV-001 and then calculated with weighted scores. High-risk areas covered 162.56 km^2, followed by moderate risk areas (98.2 km^2), low risk areas (26.29 km^2), and very-high-risk areas (23.7 km^2), respectively. Very-high-risk areas were found in Beng Samrong (11.44 km^2), followed by Keang Sanam Nang (5.21 km^2). High-risk areas were found in Bueng Phalai sub-district (70.16 km^2), followed by Bueng Samrong (30.45 km^2) and Non Sanam (27.33 km^2), respectively (Table 3). *O. viverrini* infection was 3.02%, and distributed in the moderate risk areas (four cases), high-risk areas (three cases), low-risk areas (three cases), and very-high-risk areas (two cases). Meanwhile, no risk areas were found uninfected *O. viverrini* (Table 4). Map display for risk areas is shown in Fig. 1. Red color is very-high-risk areas and found in all sub-districts. The majorities were found in Beng Samrong followed by Keang Sanam Nang.

Table 1. General characteristic data of participants who completed SUT-OV-001

Characteristics	Number (397)	%
Gender		
Male	186	46.85
Female	211	53.15
Age (years)		
≤40	45	11.34
41-50	139	35.01
50-60	160	40.3
61-70	43	10.83
>70	10	2.52
Education		
Primary school	236	59.45
Junior high school	57	14.36
Senior high school	92	23.17
No data	12	3.02
Occupation		
Agriculture	340	85.64
Not agriculture	15	3.78
Employee	32	8.06
No data	10	2.52
Income		
≤2000	188	47.36
2001-4000	92	23.17
4001-6000	63	15.87
>6000	54	13.6
Risk level		
No risk	5	1.26
Low risk	29	7.3
Moderate risk	143	36.02
High risk	197	49.62
Very high risk	20	5.04
No data	3	0.76
Total	397	100

Table 2. Distribution of participants, classified by subdistrict

Subdistrict	Number	%
Keang Sanam Nang	48	12.09
Non Samran	80	20.15
Bueng Phalai	102	25.69
Si Suk	84	21.16
Bueng Samrong	81	20.41
No data	2	0.50

Discussion

SUT-OV-001 is a screening test developed based on the risk factors associated with *O. viverrini* infection[2,3,7,9,11]. This is the first report that population at risk is screened by SUT-OV-001. It showed that *O. viverrini* infection was 3.02%, and distributed in the moderate risk areas (four cases), high-risk areas (three cases), low-risk areas (three cases), and very-high-risk areas (two cases). Meanwhile, no risk areas were found uninfected *O. viverrini*.

We previously have applied the geographic information system for distribution mapping of *O. viverrini* infection. It is suitable in the liver fluke management strategy[13]. The combination of GIS and statistical analysis can help simulate the spatial distribution and risk areas of liver fluke. Therefore, this may be an important tool for future planning of prevention and control measures[14]. In addition, GIS displayed the population at risk for cholangiocarcinoma in Bua Yai district[15], and Mueang Yang district[16] of Nakhon Ratchasima province, Thailand. This data is useful for surveillance, monitoring, and long-term care for the high-risk areas.

Presently, population at risk was screened and calculated. It was found that the majority of the population lived in high-risk (49.62%), followed by moderate risk (36.02%), and low-risk (7.3%). High-risk areas cover 162.56 km^2, followed by moderate risk areas (98.2 km^2), low-risk areas (26.29 km^2), and very-high-risk areas (23.7 km^2). Very-high-risk areas were found in

Table 3. Risk areas of opisthorchiasis were screened by SUT-OV-001, and then calculated with GIS

Subdistrict	Risk areas (Km^2)						
	Very high risk	High risk	Moderate risk	Low risk	No risk	Total	%
Si Suk	1.85	25.95	22.51	0.01	0	50.32	15.98
Bueng Samrong	11.44	30.45	4.15	0.81	0	46.86	14.89
Bueng Phalai	2.95	70.16	19.25	0.15	0	92.5	29.38
Non Samran	2.25	27.33	27.8	9.77	0.96	68.12	21.64
Keang Sanam Nang	5.21	8.67	24.49	15.55	3.09	57.01	18.11
Total	23.7	162.56	98.2	26.29	4.05	314.8	100

Table 4. Risk areas and *O. viverrini* infection

Level risk	Risk area (Km^2)	*O. viverrini* positive	%	Case/ Km^2
Very high risk	23.70	2	16.67	0.08
High risk	162.56	3	25.00	0.02
Moderate risk	98.20	4	33.33	0.04
Low risk	26.29	3	25.00	0.11
No risk	4.05	0	0.00	0
Total	314.80	12	100	0.04

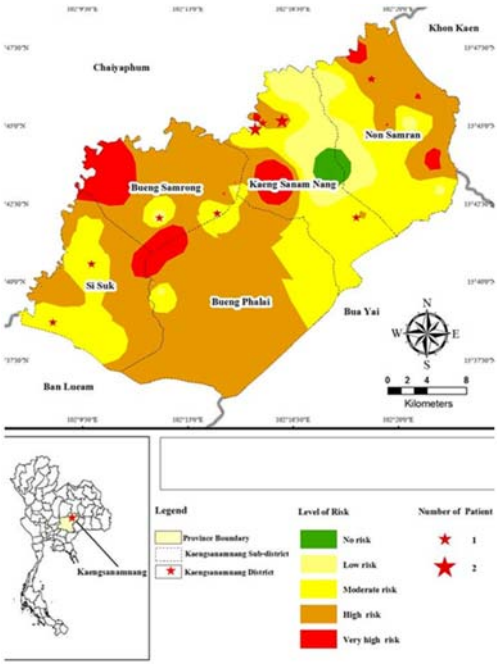

Fig. 1 Map display the risk areas for *O. viverrini* in Keang Sanam Nang district, Nakhon Ratchasima province, Thailand by using SUT-OV-001 and Geographic Information System.

Beng Samrong (11.44 km^2), followed by Keang Sanam Nang (5.21 km^2). High-risk areas were found in Bueng Phalai sub-district (70.16 km^2), followed by Bueng Samrong (30.45 km^2) and Non Samran (27.33 km^2). Map display for risk areas is shown in Fig. 1. The red color is very-high-risk areas and found in all sub-districts. The majorities were found in Beng Samrong, followed by Keang Sanam Nang.

Conclusion

The present indicates the population at risk for *O. viverrini* and risk areas in the rural communities by using SUT-OV-001 and GIS. These tools are useful for display the risk areas, prevention, control planning, and monitor.

What is already known on this topic?

O. viverrini is a problem in rural communities of Nakhon Ratchasima province particularly the areas located along Khon Kaen and Chaiyaphum province.

What this study adds?

Population at risk for *O. viverrini* was classified as high-risk (49.62%), followed by moderate risk (36.02%), low-risk (7.3%). Risk areas were classified

as very-high-risk areas, found in Beng Samrong (11.44 km^2), followed by Keang Sanam Nang (5.21 km^2). High-risk areas were found in Bueng Phalai sub-district (70.16 km^2), followed by Bueng Samrong (30.45 km^2) and Non Sanam (27.33 km^2). *O. viverrini* infection was 3.02%, and distributed in the moderate risk areas (four cases), high-risk areas (three cases), low-risk areas (three cases), and very-high-risk areas (two cases).

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Potential conflicts of interest

None.

References

1. Kaewpitoon N, Kaewpitoon SJ, Pengsaa P. Opisthorchiasis in Thailand: review and current status. World J Gastroenterol 2008; 14: 2297-302.
2. Sithithaworn P, Andrews RH, Nguyen VD, Wongsaroj T, Sinuon M, Odermatt P, et al. The current status of opisthorchiasis and clonorchiasis in the Mekong Basin. Parasitol Int 2012; 61: 10-6.
3. Kaewpitoon N, Kootanavanichpong N, Kompor P, Chavenkun W, Kujapun J, Norkaew J, et al. Review and current status of *Opisthorchis viverrini* infection at the community level in Thailand. Asian Pac J Cancer Prev 2015; 16: 6825-30.
4. Wongsaroj T, Nithikathkul C, Rojkitikul W, Nakai W, Royal L, Rammasut P. National survey of helminthiasis in Thailand. Asian Biomed 2014; 8: 779-83.
5. Kaewpitoon SJ, Rujirakul R, Kaewpitoon N. Prevalence of *Opisthorchis viverrini* infection in Nakhon Ratchasima province, Northeast Thailand. Asian Pac J Cancer Prev 2012; 13: 5245-9.
6. Kaewpitoon SJ, Rujirakul R, Loyd RA, Panpimanmas S, Matrakool L, Tongtawee T, et al. Re-examination of *Opisthorchis viverrini* in Nakhon Ratchasima province, Northeastern Thailand, indicates continued needs for health intervention. Asian Pac J Cancer Prev 2016; 17: 231-4.
7. Harinasuta C, Vajrasthira S. Opisthorchiasis in Thailand. Ann Trop Med Parasitol 1960; 54: 100-5.
8. Thamavit W, Bhamarapravati N, Sahaphong S, Vajrasthira S, Angsubhakorn S. Effects of dimethylnitrosamine on induction of cholangiocarcinoma in *Opisthorchis viverrini*-infected Syrian golden hamsters. Cancer Res 1978; 38: 4634-9.
9. Harinasuta T, Riganti M, Bunnag D. *Opisthorchis viverrini* infection: pathogenesis and clinical features. Arzneimittelforschung 1984; 34: 1167-9.
10. The International Agency for Research on Cancer (IARC). Infection with liver flukes (*Opisthorchis viverrini*, *Opisthorchis felineus* and *Clonorchis sinensis*). IARC Monogr Eval Carcinog Risks Hum 1994; 61: 121-75.
11. Sripa B, Kaewkes S, Intapan PM, Maleewong W, Brindley PJ. Food-borne trematodiases in Southeast Asia epidemiology, pathology, clinical manifestation and control. Adv Parasitol 2010; 72: 305-50.
12. Kato K, Miura M. Comparative examinations. Jpn J Parasitol 1954: 3: 35. [in Japanese].
13. Rattanasing W, Kaewpitoon SJ, Loyd RA, Rujirakul R, Yodkaw E, Kaewpitoon N. Utilization of google earth for distribution mapping of cholangiocarcinoma: a Case study in Satuek District, Buriram, Thailand. Asian Pac J Cancer Prev 2015; 16: 5903-6.
14. Rujirakul R, Ueng-arporn N, Kaewpitoon S, Loyd RJ, Kaewthani S, Kaewpitoon N. GIS-based spatial statistical analysis of risk areas for liver flukes in Surin Province of Thailand. Asian Pac J Cancer Prev 2015; 16: 2323-6.
15. Kaewpitoon SJ, Rujirakul R, Sangkudloa A, Kaewthani S, Khemplila K, Cherdjirapong K, et al. Distribution of the population at risk of cholangiocarcinoma in Bua Yai District, Nakhon Ratchasima of Thailand using google map. Asian Pac J Cancer Prev 2016; 17: 1433-6.
16. Kaewpitoon SJ, Rujirakul R, Joosiri A, Jantakate S, Sangkudloa A, Kaewthani S, et al. GIS database and google map of the population at risk of cholangiocarcinoma in Mueang Yang District, Nakhon Ratchasima Province of Thailand. Asian Pac J Cancer Prev 2016; 17: 1293-7.

การวิเคราะห์พื้นที่เสี่ยงต่อการติดเชื้อพยาธิใบไม้ตับชนิด ออพิสทอร์คิส วิเวอร์รินี ในชุมชนชนบทโดยประยุกต์ใช้แบบคัดกรอง SUT-OV-001

สรญา แก้วพิทูลย์, สุดาพร สวัสดิ์ผล, มัตติกา ชัยมีแรง พันธ์ดี, วิชาญ พันธุ์ดี, วาสนา ภานุรักษ์, รัตนา รุจิรกุล, ปาริชาติ วัคคุวัทพงษ์, ลิขิต มาตระกูล, ทวีศักดิ์ ทองทวี, สุกิจ พันธุ์พิมานมาศ, เฟื่องฟ้า เบญจโอพาร, นิวัฒน์ชัย นามวิชัยศิริกุล, ดาราวรรณ จอมเกาะ, อภิญญา จูศิริ, ณัฏฐวุฒิ แก้วพิทูลย์

ภูมิหลัง: *พยาธิใบไม้ตับออร์พิสทอร์คิส วิเวอร์รินียังคงเป็นปัญหาที่สำคัญของประเทศไทยโดยเฉพาะภาคตะวันออกเฉียงเหนือและภาคเหนือ การเฝ้าระวังเชิงรุกเป็นสิ่งจำเป็นในการตรวจหาและการได้ข้อมูลปัจจุบันเพื่อสำหรับการควบคุมป้องกันในอนาคต*

วัตถุประสงค์: *เพื่อค้นหาประชากรกลุ่มเสี่ยงและวิเคราะห์พื้นที่เสี่ยงต่อโรคพยาธิใบไม้ตับออร์พิสทอร์คิส วิเวอร์รินีในชุมชนชนบทของจังหวัดนครราชสีมา ประเทศไทย*

วัสดุและวิธีการ: *การสำรวจแบบภาคตัดขวางระหว่างช่วงเดือนตุลาคม พ.ศ. 2558 ถึงเดือนมีนาคม พ.ศ. 2559 ในพื้นที่อำเภอแก้งสนามนาง จังหวัดนครราชสีมา ประเทศไทย ประชาชนกลุ่มเสี่ยงตรวจคัดกรองด้วย SUT-OV-001 มีค่าสัมประสิทธิ์อัลฟาของครอนบาค 0.724 ตรวจการติดเชื้อพยาธิใบไม้ตับออร์พิสทอร์คิส วิเวอร์รินี ด้วยวิธีคาโตดิคสเมียร์และวิเคราะห์พื้นที่เสี่ยงด้วยสารสนเทศภูมิศาสตร์*

ผลการศึกษา: *กลุ่มตัวอย่างจำนวน 397 ส่วนใหญ่เป็นเพศหญิง (53.15%) กลุ่มอายุ 41-50 ปี (35.01%) จบการศึกษาระดับประถมศึกษา (59.45%) เกษตรกรรม (85.64%) รายได้ 2,000 บาท (47.36%) ส่วนใหญ่จัดอยู่ในกลุ่มเสี่ยง (49.62%) รองลงมาคือเสี่ยงปานกลาง (36.02%) เสี่ยงต่ำ (7.3%) พื้นที่เสี่ยงสูงมากพบในตำบลบึงสำโรง (11.44 ตร.ม.) และตำบลแก้งสนามนาง (5.21 ตร.ม.) พื้นที่เสี่ยงสูงพบในอำเภอบึงพะไล (70.16 ตร.ม.) รองลงมาคือบึงสำโรง (30.45 ตร.ม.) และโนนสำราญ (27.33 ตร.ม.) ตามลำดับ อัตราการติดเชื้อพยาธิใบไม้ตับคิดเป็น 3.02% กระจายตามพื้นที่เสี่ยงปานกลาง (4 ราย) เสี่ยงสูง (3 ราย) เสี่ยงต่ำ (3 ราย) และเสี่ยงสูงมาก (2 ราย)*

สรุป: *การศึกษานี้ชี้ให้เห็นกลุ่มเสี่ยงต่อพยาธิใบไม้ตับออร์พิสทอร์คิส วิเวอร์รินิ และพื้นที่เสี่ยงในชุมชน ชนบท โดยการใช้ SUT-OV-001 เครื่องมือนี้เป็นประโยชน์ในการแสดงพื้นที่เสี่ยงและสามารถใช้เป็นข้อมูลในการวางแผนป้องกันควบคุมและการติดตามในการดำเนินงานในอนาคตได้*