Surveillance Model of Parasitic Zoonosis in Cyprinoid Fishes in Northern Zone and Northeastern Zone of Thailand and Myanmar Using GIS

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Abstract

The parasitic zoonosis, opisthorchis viverrini has been an important public health problem in many parts of the globe. In Thailand, fish-borne parasitic zoonosis is highly spread in the northern and northeastern regions, where a large impact of cholangiocarcinoma occurs, a crucial source of the liver cancer. The rare occasions reports date published about the Opisthorchiasis in the middle zone of Myanmar. In our study, a total of a few species of fish borne trematode metacercariae i.e.; three kind of small intestinal flukes, the family of Heterophyidae; Haplorchoides sp., Haplorchis pumilio, Haplorchis taichi and one species of carcinogenic liver fluke, the family of Opisthorchiidae; ***Opisthorchis viverrini*** *have been detected from seven study areas from Thailand and Myanmar. The geographic information relevant with the rate of infection with vulnerable species of freshwater fishes was also posted from Thailand and Myanmar, and built a parasitic diseases combine with georeference for Geographical Information System (GIS) implementation. Furthermore, secondary descriptive analysis of the prevalence of fishborne trematodes metacercariae from countries of golden triangles (Southeast Asia) ie; Thailand, Myanmar, and Laos PDR have been created a GIS database for infection status of parasite infections. The outcome from this study may be helpful in strategies for protocol of the prevention of parasitic zoonosis in freshwater fishes reportin Thailand and Myanmar.*

1. Introduction

The parasitic zoonosis showed alarming the important problems in medical and public health in many Asian countries. Fish-borne zoonotic trematodes (FZT) especially minute intestinal flukes, the family of Heterophyidae and small carcinogenic liver flukes, the family of Opisthorchiidae have been endemic in that regions (Jeon et al., 2012, Dao et al., 2017, Saenphet et al., 2008 and Lovis et al., 2009).The life cycle of fish-borne trematode parasites involves three hosts which are definitive hosts i.e; Human and fish eating mammals, primary intermediate hosts i.e; snails in the family Bithyniidae, secondary intermediate hosts i.e; fresh water cyprinoid fishes. Stages of trematode can be identified as eggs in the feces of Human and fish eating mammals, cercariae in Bithyniidae snails' family and infective stage in cyprinoid fishes (Figure 1) (Anh et al., 2009, Do Trung Dung et al., 2007 and Toledo et al., 2012). The major source of transmission is due to the eating of metacercariae contaminated undercooked fresh-water fishes (Pumidonming et al., 2018).

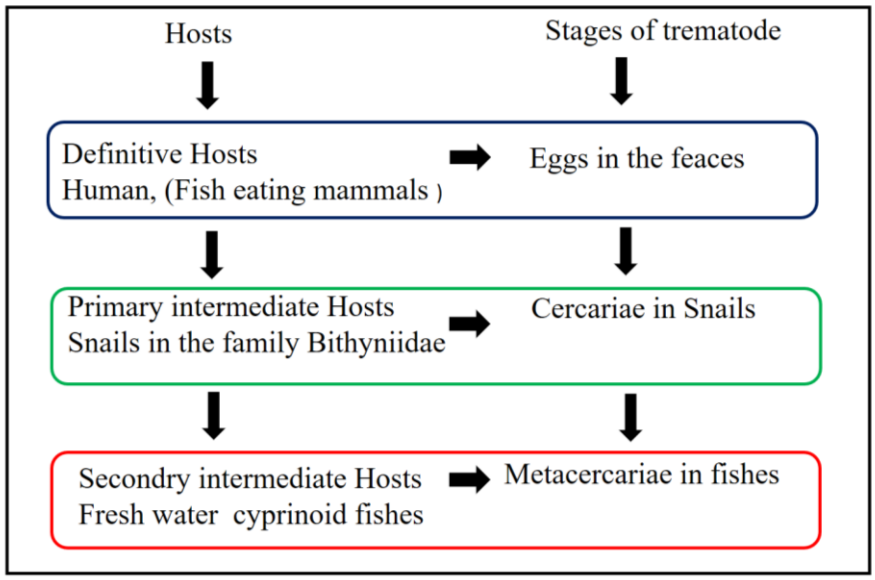

Figure 1: Hosts and Stages of Fish borne trematode parasites

The main local behavior of ingestion of fresh or uncooked fish is a well know as a risk factor for liver fluke infections (Sithithaworn et al., 2012). When a human consumes underdoneor rare fish that may contain infective stage of the parasite, and then mature stage placed the biliary tract of liver (Sripa et al., 2011). The main complications of chronic human opisthorchiasis, dis-ease caused by Opisthorchis viverrini are associated with cholangiocarcinoma a main primary carcinoma of the liver cancer with a very impropability (Honjo et al., 2005). The northern and North-eastern regions of Thailand have been the epicenter of opisthorchiasis together with a high incidence of cholangiocarcinoma (Sripa et al., 2011). Bago, one of the cities of Myanmar, is the main production source of popular local food, Nyar lay chin which has been distributed to the whole country. Nyar lay chin is made with raw freshwater fishes mixed with rice and is packaged with local banana leave consumed by mixing with raw onion and oil. Consumption of this raw salad is very famous and popular in this region as well. According to literature reviews about fishbone trematode infections from Myanmar, only a few have been published. In 2017, Aung et al., (2017) investigated fishbone trematodes infections in human stool from Bago Region, Mon State and Yangon Region. Before that there have been no previous records about human fishbone trematodes infections from Myanmar (Aung et al., 2017). Among the three survey areas, the result demonstrated that 18.9% of parasites in fish already posted in Bago Region and showed the highest prevalence among the three survey areas. In addition, Han et al., (2019) investigated about the neglected helminthiasis in four villages of Shwegyin Township, Bago-East Region, Myanmar. They found that 51 out of 698 human stool samples are infected with fish-borne trematode infections (Htwe et al., 2019). All of these literature findings have been indicating that people living in Bago region have been effecting by human fish -borne trematode infections. In 2018, Sanpool et al detected fish-borne liver fluke infection in central Myanmar (Sanpool et al., 2018). More information needs to be investigated about fish borne trematode infection secondary intermediate host to halt the cycle and for more intervention required for local public health issues and for public awareness of fish-borne parasite. Consequently, we performed the investigation about the trematodes metacercariain the fresh water fishes from north and northeast of Thailand and the central region of Myanmar. In addition, study about the metacercarial infections in the secondary intermediate host of trematodes could become a useful information on the parasitic zoonosis epidemiology. We also conducted the geographic information of the prevalence of fish-borne parasitic infections in freshwater fishes from Bago, the Central area of Myanmar, north of Thailand, and northeast of Thailand. Furthermore, secondary descriptive analysis of the frequency of trematodes metacercariae from Countries of golden triangles (Southeast Asia) ie; Thailand, Myanmar and Laos PDR were created a GIS data-base to study about the infection status of fish borne parasitic zoonosis in secondary intermediate hosts.

2. Materials and Methods

2.1 Fishes Study and Study Areas

A overall of 2195 cyprinoid fishes were brought from seven locations from reservoirs in northern zone and northeastern zone of Thailand and Bago, Middle region of Myanmar and collected fishes were transferred on ice to Faculty of Medicine, TPDRU, MSU, Mahasarakham, Thailand. The total number from fresh water cyprinoid fishes and species of fishes collected from seven study areas are shown in Table 1. Taxonomic classification of the fishes was determined with the characterize of the fish base (http://www.fishbase.org/search.php).

2.2 Metacerearіae Preparation by Pepsin Digestion Technique and Analysis

To observe the prevalence of fish borne trematode metacercariae, the sample were identifiedfor morphology of the metacercaria stage by solution of 1% pepsin digestion (Nithikathkul and Wongsawad, 2008) and the prevalence of fish-borne trematode metacerearіae was calculated by using the following equation.

$$\text{Prevalence (\%)} = \frac{\text{Number of infected fish} \times 100}{\text{Total number of fish examined}}$$

Equation 1

2.3 Geographic Information Systems (GIS) of Fish-borne trematodes Metacercariae

In this study, after fishes were examined for the presence of metacercariae by the digestion technique, observed results were then summarized as cumulative prevalence for fish-borne trematodes metacercariae infections in a tubular format. To create a GIS database for fish borne trematode infections, each record in the prevalence and intensity tables were associated with geographic coordinates where fishes were taken from using Global Positioning System (GPS) and converted into a shape file with ArcGIS Desktop Program from ESRI Company.

3. Results

2195 fishes in total of freshwater fishes (36 species) were collected from seven study areas from central region of Myanmar, north of Thailand and northeast of Thailand ie; Bago, Chom Thong, Mae Ngat, NikhomKham Soi, Sakon Nakhon, Sisaket, Kalasin. 1040 fishes in total (18 species) were positive with infective stage or metacercaria. In our study, a total of several species of metacercariae i.e.; some species of small intestinal parasite, such as; *Haplorchoides* sp., *Hap-lorchis pumilio*, *Haplorchis taichi* and one species of small liver fluke, the family of Opisthorchiidae; *Opisthorchis viverrini* were detected. The prevalence of fish-borne trematode metacercariae was 44.80% (56/125) in Bago, the central region of Myanmar. The highest prevalence fish-borne trematode metacercariae was 57.89% (165/285) in Sakon Nakhon, Northeast of Thailand. The lowest is in Sisaket, Northeast of Thailand, 27.38% (46/168). The GIS georeference (location position) related to the fish borne trematode incidence have been reported and establish a geoinformation for GIS application. Geographic mapping of study locations from North and Northeast of Thailand and cen-tral region of Myanmar was created by using ArcGIS 10.5 (Figure 2).

Table 1: Total Number of Fresh water cyprinoid fishes and species of fishes collected from Myanmar and Thailand

No	Location of Fish Collection	Latitude	Longitude	Total Number of Fishes examined	Total Fish Species
1	Bago, Central region of Myanmar.	17.3221	96.4663	125	4
2	Chom Thong District, Chiang Mai, North of Thailand.	18.4205	98.6724	530	8
3	Mae Ngat reservoir, Chiang Mai, North of Thailand	19.1741	99.0307	318	5
4	Nikom KomeSoi, Northeast of Thailand.	16.3356	104.5597	335	5
5	Sakon Nakhon, Northeast of Thailand	17.1546	104.1348	285	3
6	Sisaket, Northeast of Thailand	15.1186	104.322	168	6
7	Kalasin, Northeast of Thailand	16.4385	103.5061	434	5
	Total			2195	36

The size and scale of the blue circles correspond to the cumulative prevalence of fish-borne trematode metacercariae and the background is showing population density. The GIS could be useful in the establishment of prevention strategies for transmission of fish borne trem-atode diseases from infected freshwater fishes (the secondary intermediate hosts). Figure 2 demonstrates the prevalence of detected metacercariae from North and Northeast of Thailand and Bago, central region of Myanmar. The diagrams show a proportion of four detected metacercariae; small liver parasite, *Opisthorchis viverrini*, minute intestinal worms; *Haplorchoides* sp., *Haplorchis taichui*, *Haplorchis pumilio* (*O. viverrini* in Dark green, *H. taichui* in light green, *H. pumilio* in orange, Haplorchoides sp in red). Figure 3 shows the secondary descriptive analysis of fish-borne trematode infections in Countries of golden triangles (Myanmar, Thailand, Laos PDR) with the population density background. The data were collected from Google Scholar (https://scholar.google.com/) Table 2 and created a GIS database for the prevalence of Fish-borne Parasitic zoonoses. The summarized data were converted into a shape file with ArcGIS 10.5 Desktop Program from ESRI Company and analyzed the infection status of fish bone trematode infections in freshwater in that region. The size and scale of the green circles correspond to the cumulative prevalence of fish-borne trematode metacercariae with the population density background.

4. Discussion

Zoonotic trematode infected fishes have been reported throughout many parts of ssoutheastAsia countries, especially in Thailand (Wongsawad et al., 2012 and Pinlaor et al., 2013), Laos (Sripa et al., 2010, Sithithaworn et al., 2007 and Wongratanacheewin et al., 2003), Cambodia (Sohn et al., 2012 and Touch et al., 2013), Vietnam (Phan et al., 2010) and Myanmar (Chai et al., 2017). In the crucial public health problem of the fish are being dominant endemic in the countries of the Mekong basin (Sithithaworn and Haswell-Elkins, 2003, Wattanayingcharoenchai et al., 2011 and Jeon et al., 2012). In our study, 2195 fishes in a total of freshwater fishes (36 species) were investigated from seven locations from the central region of Myanmar, north of Thailand and northeast of Thailand ie; Bago, Chom Thong, Mae Ngat, NikhomKham Soi, Sakon Nakhon, Sisaket, Kalasin.

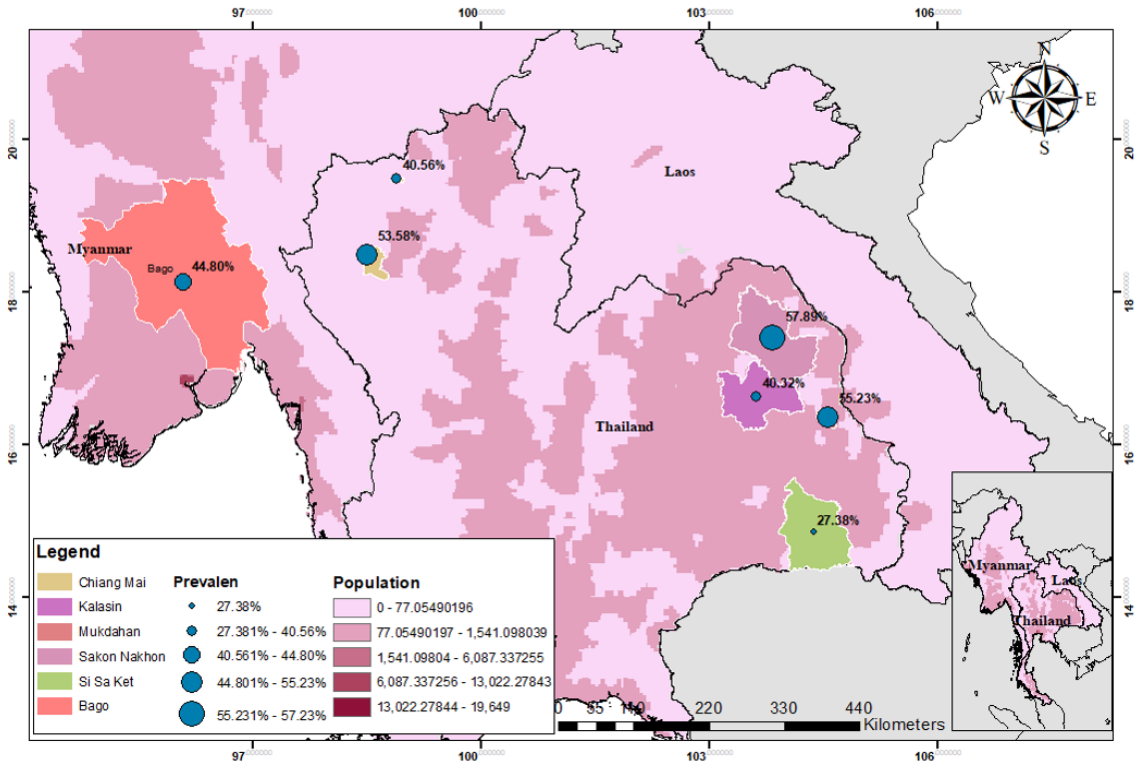

Figure 2: Geographic map showing Prevalence of fish-borne Trematode Metacercariae from North and Northeast of Thailand and central region of Myanmar with the population background by ArcGIS 10.5

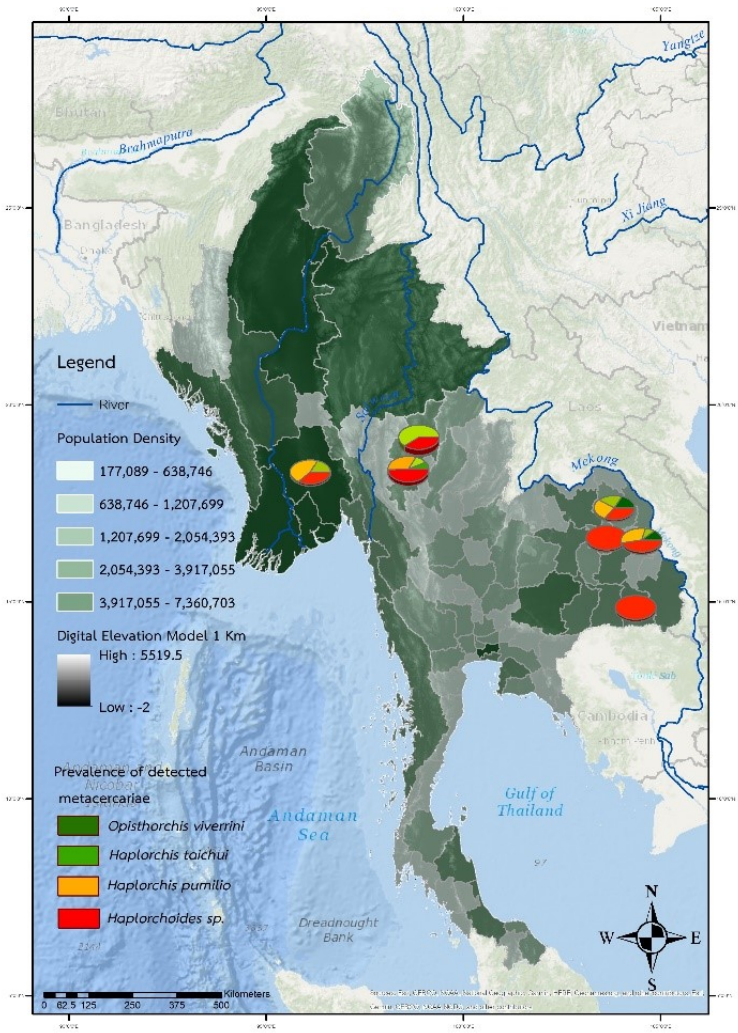

Figure 2: Geographic map showing Prevalence of detected metacercariae from North and Northeast of Thailand and Myanmar with the population background by ArcGIS 10.5

We found that 1040 fishes in total (18 species) were positive with metacercariae infections. A majority species from metacercariae i.e.; *Haplorchoides* sp., *Haplorchis pumilio* and *Haplorchis taichi*. *Opisthorchis viverrini* have been reported in this study. In Myanmar, fish borne trematode infection is first reported in 2017 by Chai et al., (2017). In 2019, Sohn et al., (2019) reported about *O. viverrini* metacercariae in freshwater fishes and human infection in Yangon, Myanmar (Sohn et al., 2019). In our study, from Bago, three species of minute intestinal flukes, members of the Heterophyidae, *H. taichui*, *H. pumilio*, *Haplorchoides* sp. were reported in secondary intermediate hosts, fresh water fishes. In this study demonstrates the phenomenal of the life cycles showed three species of fish borne trematode infections around Bago region of Myanmar. Moreover, our findings showed a new report about the existence of the life cycles of 3 members of the Heterophyidae around Bago, the Central region of Myanmar. In 2017, Aung et al., (2017) first investigated fishbone trematodes infections in human stool from Bago Region. Before that there have been no previous records about human fishbone trematodes infections from Myanmar (Aung et al., 2017). Furthermore, Han et al., (2019) investigated about the neglected helminthiasis in four villages of Shwegyin Township, Bago-East Region, Myanmar (Htwe et al., 2019). All of these literature findings have been indicating that people living in Bago region have been effecting by human fish borne trematode infections. Only one report that Sanpool et al., (2018) has been published about fish borne trematode infection in second intermediate hosts from Bago region.

Table 2: Reference and studied areas for the prevalence of fish borne parasite infections in Countries of Golden Tringle collected data from reported articles

Studied location in Countries of Golden Tringle	**Reference**
Yangon, Myanmar (2017)	(Chai et al., 2017)
North Dagon township, Yangon, Myanmar (2019)	(Sohn et al., 2019)
Bago Region, Myanmar	(Sanpool et al., 2018)
Tachileik, Mekong region of Myanmar	(Phyo Myint et al., 2020)
Chiang Mai, North of Thailand	(Saenphet et al., 2008)
Reservoir in , Nan Province, Thailand	(Boonmekam et al., 2016)
Agricultural Basin, Chiang Mai province, Thailand	(Wongsawad et al., 2013)
Chom Thong reservoirs district, Chiang Mai province, Thailand	(Kumchoo et al., 2005)
Mae Ngad Reservior, Chiang Mai province, North of Thailand	(Kumchoo et al., 2005)
Huay Luang dam, Northeast of Thailand	(Prakobwong et al., 2017)
Nakhon ratchasima, Northeast Thailand	(Nithiuthai et al., 2003)
Pasak Cholasid Reservoir, Thailand	(Krailas et al., 2016)
Savannakhet Province, Lao PDR	(Rim et al., 2008)
Vientiane Municipality, Lao PDR (2008)	(Rim et al., 2008)
Vientiane Municipality, Lao PDR (2015)	(Eom et al., 2015)
Champasack Province, Lao PDR	(Eom et al., 2015)
Khammouane Province, Lao PDR (2009)	(Manivong et al., 2008)
Khammouane Province, Lao PDR (2013)	(Rim et al., 2013)
Saravane Province, Lao PDR	(Rim et al., 2013)
Luang Prabang, Lao PDR	(Rim et al., 2013)

In 2018, Sanpool et al., (2018) detected fish borne liver fluke infection in central Myanmar (Sanpool et al., 2018). Myanmar is one of the southeast countries and borders with Thailand and Lao PDR and is named as countries of Golden Triangle (Southeast Asia). In Thailand, the highest reported of fish borne parasitic infections occur in the North and Northeast of Thailand (Sripa et al., 2011 and Jongsuksuntigul and Imsomboon, 2003). Estimated number of Opisthorchaisis are more than eight million people reported in Thailand (Sripa et al., 2010). In Lao PDR, the middle ans south parts are the highest prevalence of fish borne trematode infections (Forrer et al., 2012 and Sayasone et al., 2009). Approximately two million were infected with liver fluke infection in Lao PDR (Sripa et al., 2011). Liver fluke (*O. viverrini*) are classified as Group 1 carcinogens especially in Southeast Asia (Bouvard et al., 2009).

According to prevoius reported, the liver fluke shoed the crucial risk factors for cholangiocarcinoma (Haswell-Elkins et al., 1994). In our study from Mae Ngat reservoir, Chiang Mai province, Thailand, we collected 318 fishes (5 species) and found 129 fishes (3 species) are infected with 2 species metacercariae *H. taichui, Haplorchoides* sp. The overall prevalence is 40.56%. From water reservior, Chiang Mai province, Thailand, we investigated 530 fishes (8 species) and occurred 284 fishes (4 species) are contaminated with four types of metacercaria. The overall prevalence is 53.58%. In 2013, Wongsawad et al., (2013) found five species of metacercariae from Fang-Mae Ai Basin, Chiang Mai province (Wongsawad et al., 2013). They occurred four species of small intestinal flukes; *H. taichui*, *Hap* sp., *Centrocestus caninus*, and *Stellantchasmus falcatus* and one species of liver fluke.

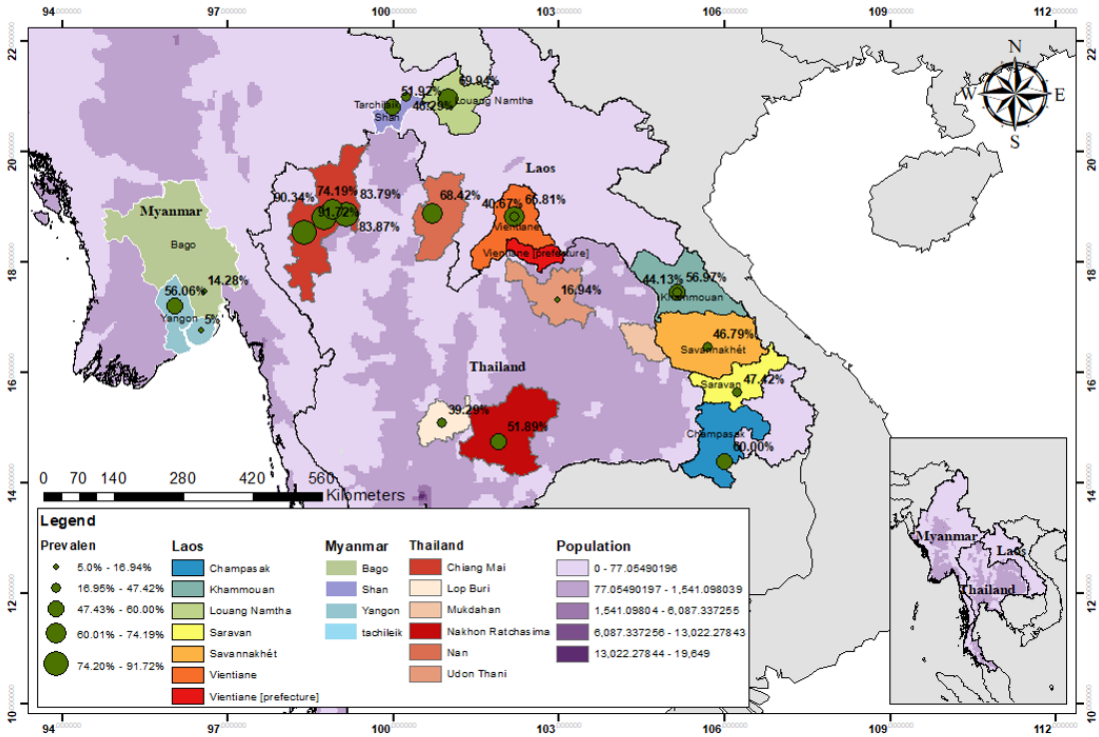

Figure 3: Geographic map showing Prevalence of fish-borne trematode infections in Countries of Golden Triangle (Myanmar, Thailand, Laos)

Several species of fresh water fishes placed as secondary intermediate hosts of parasitic infections. *H. taichui* are reported in several parts of Thailand (Srisawangwong et al., 1997 and Wongsawad et al., 2000). In our study from North east of Thailand, a total 1222 samples of freshwater fishes (19 different species) were randomly collected from four locations ie; Nikom KomeSoi, Sakon Nakhon, Sisaket and Kalasin. The 571 cyprinoid fishes samples (eight groups of fishes) were infected with infective stage of parasite with 46.72% prevalence (571/1222). In our study, the geographic information (latitude and longitude) associated with the prevalence of parasite infections from multi study areas were also investigated and built a combination between parasite and category of GIS and application The GIS database was overlaid with population density and precipitated GIS datasets. Geographic combine with each area are identified with a Global Positioning System. We also conducted the geographic information of the secondary descriptive analysis of parasitic infection from fish in Countries of golden triangles, Southeast Asia (Thailand, Myanmar and Laos PDR). Secondary data of the prevalence of fish-borne trematodes metacercariae from countries of golden triangles (Southeast Asia) ie; Thailand, Myanmar and Laos PDR were collected from reported articles from Google Scholar (https://scholar.google.com/) and created a GIS database for prevalence of fish-borne parasitic zoonoses and analyzed the status of the prevalencein that region. In conclusion, the distribution of fish-borne trematode infections is mainly relying on the prevalence of fish-borne parasitic infections in the freshwater fishes. Our findings could improve public health awareness about the fish borne parasitic zoonosis in Thailand and Myanmar. A better understanding of the geographically and incidence status of the fish-borne parasite could implemented to useful information for prevention programs of fish borne parasitic zoonosis for the endemic area. Moreover, this current report might bring situation comfimation leading to support for the campaign prevention and control of fish-borne parasitic zoonosesin Me kong basin area. In addition, study about geographic Information of parasitic zoonosis in freshwater fishes could be valuable in the establishment of elimination and strategies for transmission of diseases in the tropical countries. Further research studies on the geographic information of human e infections in definitive hosts and the primary intermediate hosts might be considered for comprehensive intervention to halt the parasitic life cycle.

5. Conclusion

Our finding will support for increasing public health awareness about the fish borne trematode infections. The study of the distribution and infection status of the fish-borne trematode metacercariae could contribute to valuable information for prevention and control programs of human liver fluke and intestinal fluke infections for the community. Moreover, this study might provide evidence leading to improve public health awareness for surveillance and control of the fish-borne trematode contamination in Thailand and Myanmar.

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