



Ministry of Health  
Lao PDR

# The role of domestic dogs in the transmission of zoonotic helminthes in a rural area of Mekong river basin

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Acta Parasitologica, 2017, 62(2), 000–000; ISSN 1230-2821

# INTRODUCTION

Humans and dogs have close relationship - companionship, hunting aids, protection, shepherding, and laboratory animals (Wang et al. 2016)



HACHI



週間少年ジャンプ～流れ星銀



Dogs are the definitive hosts of a number of helminths (El-Shehabi et al. 1999 etc.)  
-dogs are commonly being infected by > one helminth species (Dai et al. 2009 etc.)



# INTRODUCTION



Some canine helminths infect to people and cause significant clinical diseases in humans; coenurosis, hydatidosis, trichinellosis, and ocular and visceral larva migrans (Ing *et al.* 1998 etc.)

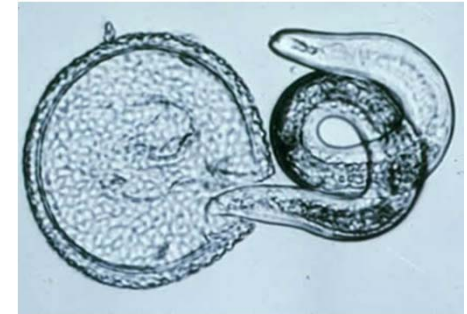
-*Toxocara canis*, *Ancylostoma* spp., *Giardia duodenalis*, and *Cryptosporidium parvum* have received great attention due to their zoonotic potential (Traub *et al.* 2005 etc.)

-Rural Peninsular Malaysia prevalence of helminths high >80% (Ngui *et al.*, 2014)

-Thailand shelter dogs helminthes prevalence 10% to 20% (Rojekittikhun *et al.*, 2013; 2014).

Zoonotic parasites associated with dogs in Laos are described in humans (Chai *et al.* 2005 etc.).

However, there are few studies evaluating the helminth infection in dogs and its role as a reservoir of human disease (Conlan *et al.* 2012).



## OBJECTIVE

In this study

- ✓ Parasite occurrence in dogs.
- ✓ Roaming area of the dogs in different villages.
- ✓ Potential risks for dogs transmitting helminth zoonosis.

were evaluated.



# MATERIALS AND METHODS

## Collection Time:

In 2010 and 2012, during the monsoon season

## Area:

Ban Lahanam area (coordinates N16 16.120 E105 16.077), Savannakhet Province, Lao PDR

## Samples:

- 34 dog fecal samples (Benkhamrai 6, Khohe 5, Lahako 12, Lahanam 5, Muanthan 6)
- 6 dogs' offal acquired from a dog butcher
- Questionnaires to dog owner about dog breeding
- GPS data of 10 roaming dogs

## Ethics Approval:

Lao Medical Ethics Committee (172/NECHR)



# MATERIALS AND METHODS



## Dog Fecal examination:

Modified formalin detergent (mFD) method (Waikagul et al. 1997) \* 70% EtOH was used. Fresh dog fecal samples were collected ASAP after defecation.

## Dogs offal examination:

All the organs were examined for parasites and the recovered worms were identified by morphology.

## Copro-DNA/ parasites DNA preparation and molecular identification:

Copro-DNA/ parasites DNA was extracted by Wizard® SV Genomic DNA Purification System (Promega, USA) after disruption of the eggs (for fecal samples).

Genus/ species specific PCR assays were conducted. \* PCR positive samples were sequenced.

Table I. Different helminth species analysed by copro-PCR in this study, the target gene, amplicom size and the respective references

Species	Target Gene	Size (bp)	References
<i>Opisthorchis viverrini</i>	DNA probe, pOV-A6	330	Wongratanacheewin <i>et al.</i> , 2001
<i>Toxocara canis</i>		380	
<i>Toxocara cati</i>	ITS-2	370	Jacobs <i>et al.</i> , 1997
<i>Toxascaris leonina</i>		300	
<i>Ancylostoma ceylanicum</i>			
<i>Ancylostoma caninum</i>	ITS-1, 5.8S, ITS-2	545	Traub <i>et al.</i> , 2008
<i>Spirometra</i> spp.	COI	444	Koonmee <i>et al.</i> , 2011
<i>Gnathostoma</i> spp.	COI	450	Ngarmamonpirat <i>et al.</i> , 2005

# MATERIALS AND METHODS

## Dogs GIS:

10 dogs were attached a wireless GPS logger (Holux M-241, Holux Taiwan) and dog roaming area was estimated.

The data were collected individually and analyzed using Holux logger utility software and Google Earth Pro version 7.1.5.1557.

## Questionnaires for Dog Breeding Situation:

Data regarding dog breeding attitude in hygiene, directed to the relationship with dogs were collected.



# RESULTS

## Dogs ownership pattern and demography

### Dog owner 48.8% (165/338)

- ✓ Free roaming and being fed at home, usually with the same food as the owners.
- ✓ 1dog-61.2% 2 dogs-25.1% 3 dogs- 8.5% >4 dogs-1.8%
- ✓ None owners use anthelmintic for their dogs.

### GPS data: successfully obtained 8 of 10 dogs

- ✓ Dogs walking range: 1.4 to 13.3 km/day (ave. 6.5 km).
- ✓ Walking in the neighborhood + to water sources one or more/day.
- ✓ Dog walking area varied 882 to 7041 sq m (ave. 3356.38 sq m).

All the dogs accessed at least one time per day a source of water including getting into the water source.



# RESULTS



## Parasite infection in Dogs

From Dog faces examination, [6 helminth species](#) were found.

microscopic		Genus/species specific PCR		
<i>O. viverrini</i> -like eggs	17.6% (6/34)	<i>O. viverrini</i>	8.8% (3/34)	
Ascarid eggs	44.1% (15/34)	<i>T. canis</i>	11 samples	*90% mixed
		<i>T. cati</i>	10 samples	
Hookworm eggs	11.7% (4/34)	<i>Ancylostoma</i> spp.	61% (21/34) 8 <i>A. caninum</i> 13 <i>A. ceylanicum</i>	
<i>Spirometra</i> spp.	44.1% (15/34)	<i>Spirometra</i> spp.	10 samples <i>S. erinaceieuropaei</i>	
<i>Gnathostoma</i> eggs	ND	<i>Gnathostoma</i> eggs	ND	

From the dogs necropsied, [14 helminth species](#) were found.

(Nematode) *A. caninum*, *A. ceylanicum*, *Dirofilaria immitis*, *T. canis*, *T. cati*, *Spirocera lupi*

(Trematode) *O. viverrini*, *Echinochasmus* sp., *H. taichui*, *H. pumilio*, *Centrocestus* sp., *Procerovum* sp.

(Cestode) *Dipylidium caninum*, *Spirometra* sp.

## DISCUSSION

### Dogs are important as reservoir host of zoonotic infections;

- Moving freely, a pattern of walking to the water-side and in vegetation areas.
- Contribute to contaminating the environment and water resources
  - spreading helminthic infections.
  - cohabit with people sharing the areas, drinking water sources, vegetable farming plots and houses
  - high potential for active transmission of dog-human zoonoses.
- A dog broker frequently comes to buy or exchange dogs to sell for restaurants. These dogs may defecate on the way, contaminating the environment and spreading their parasites near markets and areas with agglomeration of people.



## DISCUSSION



From the dogs' materials examined, 14 helminth species were found.

*O. viverrini* 8.8% (3/34)

In **small water bodies** area cohabitate human and dog may be contaminated with dogs feces.

*Ancylostoma* spp. 61% (21/34) 8 *A. caninum* and 13 *A. ceylanicum*

**Human cases of *A. caninum* and *A. ceylanicum*** infection have been reported previously (Sato et al. 2010).

*Toxocara* spp. 44.1% (15/34) *T. canis* and *T. cati* \* Serum examination of the people (MS in preparation)

**Adult dog infections** may contribute significantly to the contamination of the environment.

*Spirometra erinaceieuropaei* 44.1% (15/34)

In the same area, several plerocercoids (*S. erinaceieuropaei*) were found **in the frogs** (Yoonuan *et al.* in preparation).

Dogs may be infected by eating frogs, or by drinking proceroid infected copepod contaminated water. Human consumption of snakes and frogs is very common, and the infection with *Spirometra* may occur when the frogs are not properly cooked.

# DISCUSSION



For the control of zoonotic diseases.....(WHO 1987; FAO 2014; Perry 1993)

- Study of dog ecology and related anthropological aspects of pet ownership.
- Understanding the epidemiology of canine infectious diseases.
- Making decisions in the planning and implementation of dog population management schemes.

Human first?

- Lao national scheme for school deworming: a round of treatment at the beginning of the 1st semester (Sep-Dec) and in the 2nd semester (Jan–Apr). Mebendazole (single 500 mg oral dose) is distributed to all school-aged children since 2005 (Phommasack *et al.* 2008; Conlan *et al.* 2012).
- But there is **no regulation or recommendation for the treatment of domestic animals to control zoonotic parasites in Laos** (Conlan *et al.* 2012).
- **Usually the hygiene efforts are focused on controlling human defecation behavior** (Saowakontha *et al.* 1993).



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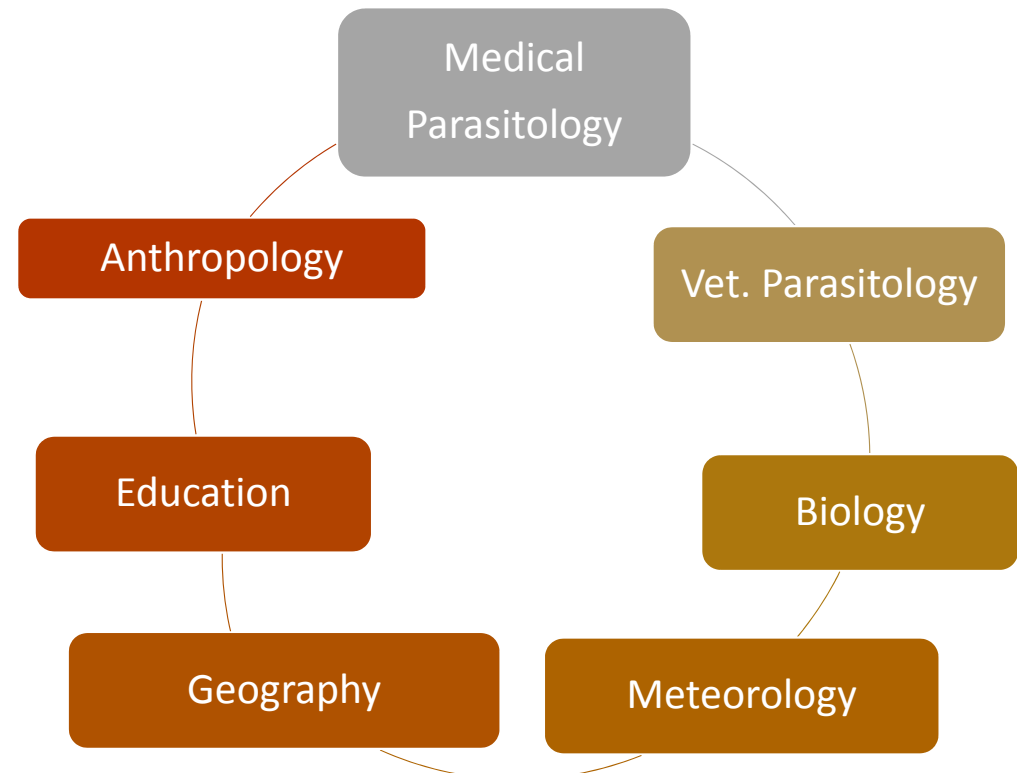
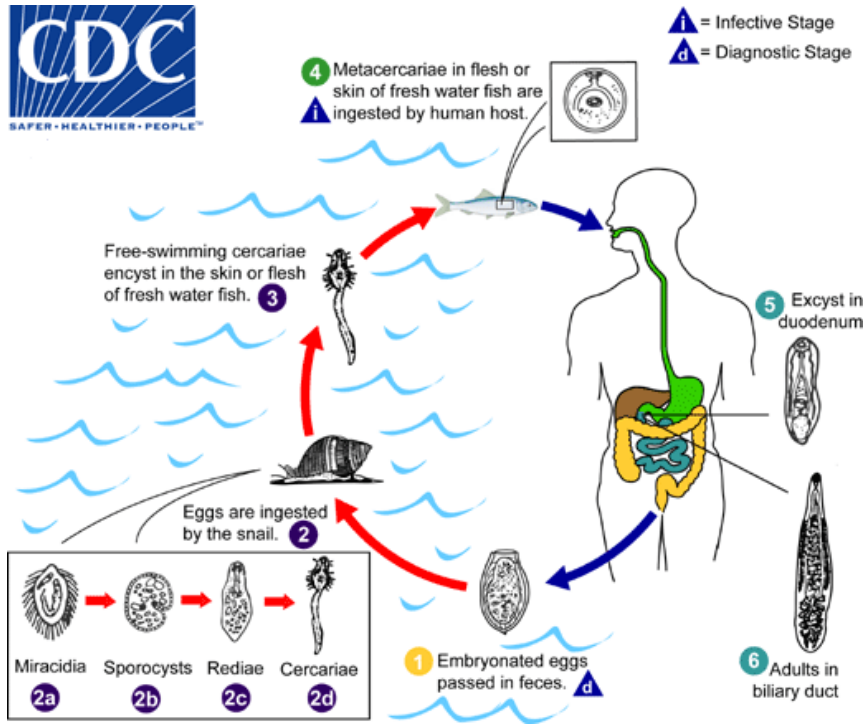
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## DISCUSSION

### Multidisciplinary Approach for Eco-Health

Control programs of human NTDs, particularly parasitic diseases, should be done in parallel with parasitic diseases control programs in animals, especially dogs, in the Mekong River basin area.

# Collaborative structure for Eco-health approach of *O. viverrini*



## DISCUSSION

### Multidisciplinary Approach for Eco-Health



DRAGON QUEST, SQUARE ENIX CO., LTD.

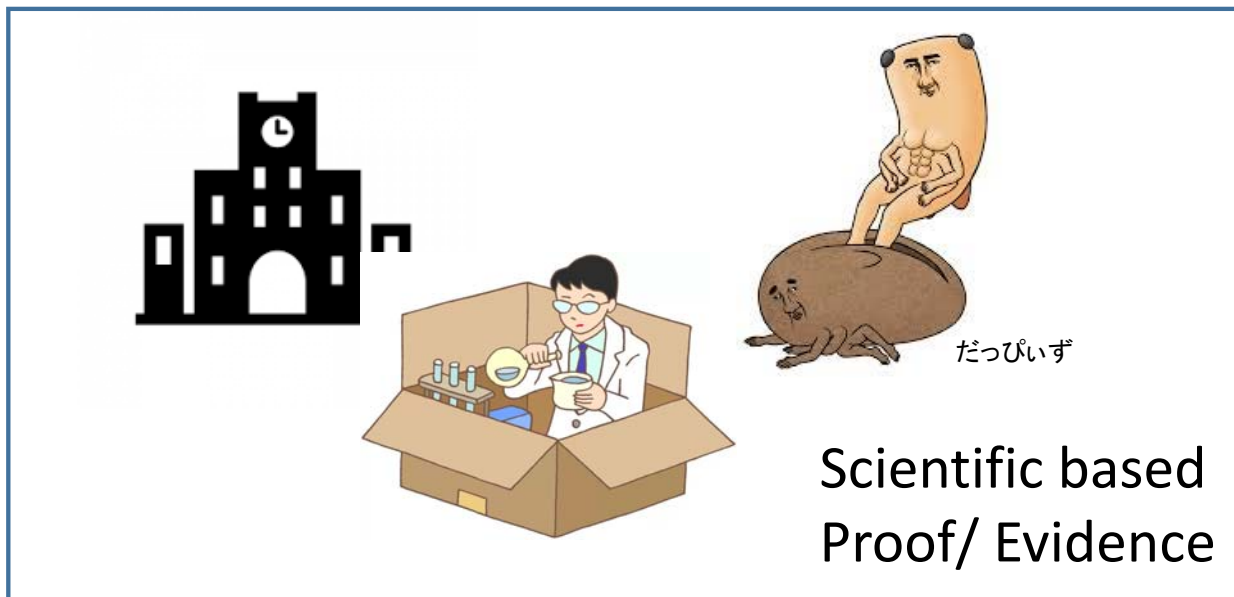
- Get good companions who have different weapons/ back ground.
- Gain experience, and level up each persons skills.
- Get Items/ equipment.

Go toward the final dungeon.

Combat against final boss!

# DISCUSSION

## Role of Academia



Food and Agriculture Organization of the United Nations



World Health Organization



Policy Making

## ACKNOWLEDGEMENT

**Mr. Nirandorn Homsuwan** of the Department of Helminthology, Faculty of Tropical Medicine, Mahidol University

**Mrs. Vongphaka Boutsyhalath** of the Health Centre, Lahanam Village, Sonkon District, Savannakhet Province, Laos

**Mr. Takuya Ishida**, alumni of Niigata University

**Dr Carol Bain**, Indiana University

**Dog Owners** in Lahanam Area

**Dogs** Cooperate to us



This research was partially supported by the **RIHN research project on "Environmental Changes and Infectious Diseases in Tropical Asia"** (for Kazuhiko Moji), and the **Thailand Research Fund (BRG 5180005)** (for Jitra Waikagul).



Thank you very much for  
your attention.

