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Potential Risks of Zoonotic Diseases of Ascariasis and Hookworm Infection in Rural Villages in Indonesia

Blego SEDIONOTO[1,3*], Wigati WILUJENG[2], Witthaya ANAMNART[1]

[1]*Doctoral Program in Biomedical Sciences, School of Allied Health Sciences Walailak University, Thailand 80160*

[2]*Department of Parasitology, Central Developing Research of Zoonotic Diseases B2P2VRP Salatiga, Ministry of Health Republic Indonesia*

[3]*Department of Environmental Health, Faculty of Public Health Mulawarman University, Indonesia 75123*

(Corresponding author's email: blego_kesling@yahoo.com)

Abstract

Ascariasis and hookworm infection are a neglected helminth infection which can potentially cause zoonotic diseases. This study explained potential zoonotic diseases from cat and rat which are infected by *Ascaris lumbricoides* and hookworm in rural areas. Stool samples were collected from 27 participants, 15 cats and 7 rats at rural area Magelang district Central Java, Indonesia. Two methods of diagnosis of stool samples were used in this research including; Kato katz thick smear and Koga agar plate culture. The result of study showed human infected by *Ascaris lumbricoides* and hookworm were 44.4% and 3.7% respectively. Cats infected by *Ascaris lumbricoides*, hookworm and double infections (*Ascaris lumbricoides* and hookworm) were 93%, 66,67% and 80% respectively. In this study rats have multiple infections (*Ascaris lumbricoides*, hookworm, and *Ascaris lumbricoides* and double infections (*Ascaris lumbricoides* and hookworm) were 86% and 14% respectively. The zoonotic diseases identified in this research caused by *Ascaris lumbricoides* and hookworm infection demonstrates possible transmission from cat and rat to humans via contact with soil around houses in the villages. The incidence of ascariasis and hookworm infection may be reduced if control measures of personal hygiene, environmental sanitation, and protection from animal infected by *Ascaris lumbricoides* and hookworm will be instituted.

Keywords: Ascariasis, Hookworm Infection, Zoonotic Diseases

Introduction

Worldwide, there is a significant variation in the prevalence of gastrointestinal zoonotic helminths in dogs and cats [1,2]. Soil transmitted helminth infections (STHs) are the most common infections among humans and domestic animals such as dogs, cats and pigs, particularly in the rural areas of Southeast Asia. Infection of zoonotic helminths has previously been researched in Thailand. Where has a high prevalence of hookworm *Ancylostoma ceylanicum* was reported among dogs in temple communities in Bangkok [3]. Chronic infections with one or several of the most common soil-transmitted helminths (STHs), *Ascaris lumbricoides, Trichuris trichiura* and hookworms, might account for a global burden of 39 million disability-adjusted life years lost annually [4]. Another STHs, *Strongyloides stercoralis*, is often neglected in helminth surveys [5,6], yet previous studies show high *S. stercoralis* infection rates in Cambodia [7]. School-aged children in the developing world are at highest risk of morbidity due to STHs and intestinal protozoan infections [8]. Many of the IPIs in animals, especially those with the larval stages of hookworms, *Gnathostoma spp.* and *Toxocara spp.*, may result in zoonotic diseases such as eosinophilic enteritis [9], cutaneous larval migrans, and toxocariasis.

However, mass treatment only focuses on three major STHs (*Ascaris*/hookworm/*Trichuris*). Other nematodes like *S. stercoralis*, trematodes and protozoan infections are not addressed. In rural Southeast Asia, little is known about the zoonotic potential of STHs in humans and animals. Therefore of domestic animals, such as cats, dogs and pigs, as contributors to human STHs and as reservoir hosts for zoonotic parasites remains

unexplored and/or the data are inaccessible.
Although surveys of zoonotic gastrointestinal helminths in dogs and cats had been done in Thailand, most of the studies have focused on the Central or Northeastern region [10, 11, 12, 3]. This was the first study to investigate prevalence of zoonotic helminth infection in humans, cats and rats in Magelang district Central of Java Indonesia that is introduction potential risk zoonotic diseases of soil transmitted helminth infections.

Materials and methods

Study design and area

The study was carried out in July to August 2017 in seven villages in rural area of Magelang district, Central of Java province, Indonesia). The climate is tropical, with cold temperatures all year round and alternating dry and wet seasons. Households from these villages were randomly selected. Stool samples from humans and animals (cats and rats) were assessed for STHs using a single diagnostic test. Risk factors for infection of humans and animals were assessed based on information collected through interviews and observations.

Field procedures and sample collection

On the day of the first visit, informed consent was obtained from all household members and interviews were conducted with enrolled participants. Interviews with young children were conducted with the help of a parent or legal guardian. All enrolled participants received a prelabelled stool container. Participants were asked to fill the container with feces passed the following morning. Upon collecting the first sample, a second stool container was given to participants for filling. The collected stool samples were transported to a laboratory in Department of Parasitology Central Zoonotic Diseases Health Ministry of Republic Indonesia. Stool samples from each human and cat present at the time of the visit and belonging to the household were obtained. For each animal, approximately five grams of fecal samples were collected from around houses, placed into a sterile plastic fecal container and chilled immediately in a box containing ice. For each human, one stool sample given on consecutive day was analyzed and for each animal, one sample was analyzed.

Laboratory procedures

For each human stool sample, the following tests were performed: Kato Katz [13], Koga Agar culture [14], As they arrived in the laboratory, human samples were processed as follows: First, duplicate Kato Katz smears were prepared. Stool was filtered using a nylon mesh and then placed on the standard Kato Katz template, leaving 41.7 mg of stool for examination on a microscopic slide. Examination was performed at 100x magnification [13] for hookworm and S. stercoralis. Second, a Koga Agar test was prepared by placing a piece of stool (3–5 g) on a freshly produced Agar plate. The plates were then incubated for 48 hours at 28 °C. Larvae were washed from the plate into a tube, the liquid was centrifuged and the entire sediment was read at 40x magnification [14] for hookworm and S. stercoralis larvae.

Results and discussion

In this research we reported the prevalence soil transmitted helminth in human,cats and rats such as human infected ; 12/27(44,4%) *Ascaris lumbricoides,* and 1/27(3,7%) hookworm, cats have infected; 14/15(93%) Ascaris spp, 10/15(66,67%) hookworm, 12/15 (80%) double infected (*Ascaris lumbricoides* and hookworm), rats had multiple infected *Ascaris lumbricoides,T. Trichiura*, hookworm 86%(6/7) and double infected *Ascaris lumbricoides,* hookworm 14%(1/7),

Table 1. Prevalence Soil Transmitted Helminths Infection in Human

No	Parasite	Prevalence/Diagnostical Methods		
		DWS	Kato Katz	Agar Culture
1.	*Ascaris l*	11/27(40,7%)	12/27(44,4%)	Un-examination
2.	*T. Trichiura*	-	-	Un-examination
3.	*S. stercoralis*	-	-	Un-examination
4.	*Hookworm*	1/27(3,7%)	1/27(3,7%)	Not found
5.	*Ascaris+hookworm*	1/27(3,7%)	1/27(3,7%)	Not found

The zoonotic soil transmitted helminth infection in this research showed, this study had deference with one studied at northern Thailand that research reported the prevalence of zoonotic intestinal helminths in lower Northern Thailand as 40.1 % (79/197) in dogs and 33.9 % (61/180) in cats, respectively. Zoonotic helminths found included hookworms, *Spirometra* spp., *Toxocara*, *O. viverrini*, *Taenia* spp. *Strongyloides* and *Trichuris* [15,16] showed dogs and cats are important reservoir hosts of various zoonotic helminths [17, 18, 2], many of which cause serious public health problems. We researched found 14% hookworm infection in human, but maybe not yet potential zoonotic hookworm, like others researcher that was found to have low infection rates in both dogs and cats for zoonotic hookworm. Similar to other areas, prevalence of *A. caninum* was lower than that of *A. ceylanicum* [19, 20]. Although its infection rate was low, this hookworm can result in eosinophilic enteritis and chronic abdominal pain in human [8,3]. Other zoonotic hookworm, such as *A. braziliensis*, was not found in this area.

Table 2. Prevalence Soil Transmitted Helminths Infection in Cats

No	Parasite	Prevalence/Diagnostical Method		
		DWS	Kato Katz	APC
1.	*Ascaris l*	14/15(93%)	14/15(93%)	Un-examination
2.	*T. Trichiura*	-	-	Un-examination
3.	*S. stercoralis*	-	-	Not found
4.	*Hookworm*	8/15(53,3%)	10/15(66,67%)	Not found
5.	*Single ascaris*	3/15 (20%)	2/15 (13,3%)	Un-examination
6.	*Single hookworm*	-	-	Not found
7.	*Ascaris+hookworm*	10/15(66,67%)	12/15 (80%)	Not found

Among zoonotic helminth infections in dogs in the lower Northern area of Thailand, hookworms were the most prevalent helminth, and *Spirometra* was the second most prevalent. Our results confirmed that hookworm infection in dogs is common in Thailand [12]. The high prevalence of hookworm infections in dogs can contribute to the occurrence of zoonotic ancylostomiasis in human [21]. Zoonotic helminth infections in cats were different from dogs. *Spirometra* was the most prevalent, while hookworms were the second most prevalent helminth. High rates of *Spirometra* infection might be a reflection of the fact that most cats roam freely and had access to small prey as a food source. High infection rates of *Spirometra* spp. in cats might indicate a high infection rate of plocercoid and plerocercoid in intermediate hosts in the area. The infection of *Spirometra* spp. in cats and dogs can lead to a high risk of sparganosis in humans who have the habit of eating undercooked meat [22, 23]. However, human sparganosis in Thailand is rare. In the period 1943-2010, only 53 cases had been reported [22, 23]. Dogs are associated with more than 60 zoonotic parasites worldwide, many of which pose serious public health concerns [24]. Compared with some other studies in South Asian countries, the overall prevalence of IPIs in dogs in Cambodia was higher (81.9%) than previously reported for dogs in rural India, for example [25].

This research in contrast with Combodian research that dogs in rural Cambodian villages such as Dong village are largely kept as guard dogs and allowed to roam freely, especially during the day. The dogs are also allowed inside the house and around rice and vegetable fields and ponds. At night-time, the dogs then often stay in or around the house. Dogs, therefore, pose a serious zoonotic risk as they have the potential to transmit zoonotic parasites through their close association with household members as well as through heavy contamination of the environment, including soil, fresh produce and waterways, with parasite eggs and oocysts, in our observation sew behavior of cats almost all day and night stayed around houses and rare contact with ponds and did not stay around rice and vegetable fields.

The research at Indonesia had contrasted with our research at Thailand where showed prevalence of Soil Transmitted Helminths/STHs infection in Humans such as, hookworm (52%), *S.stercoralis* (13%), *T. trichiura* (14%) and *Ascaris* (7%), in this research ascariasis is low while the studied at Magelang district prevalence Ascarisis 44%, these data could have association with environmental factors such as characteristic of soil, climate/humadity, also behavior human and facilitate of sanitation in that area.

Table 3. Prevalence Parasite in rats

No	Parasite	Prevalence
1.	*Ascaris l, T. Trichiura, hookworm*	86%(6/7)
2.	*Ascaris, hookworm*	14%(1/7)
3.	*S, stercoralis*	-
4.	*Single Hookworm*	-
5.	Single Trichuris trichiura	-

The present study showed in contrast with patterns of IPIs in humans compared to previous surveys conducted in Cambodia [4,19,20]. The major IPIs found in humans were hookworms (63.3%), Entamoeba spp. (27.1%), *S. stercoralis* (24.3%), *G. duodenalis* (22.0%) and *Blastocystis* (18.4%) . In total, 14 different parasite species were diagnosed, including eight helminthic and six protozoan parasites. Of the 218 participants, 27 (12.8%) were negative in all examinations. More than a quarter of the human participants (64, 29.4%) were infected with one parasite and a third (72, 33.0%) with two or more parasites. Three (1.4%) and one (0.5%) participant(s) harboured five and six parasites, respectively, the prevalences of parasites (those with the highest infection rates) are given for the different age-groups. For hookworm, the prevalence increases from less than 50.0% in children up to the age of ten to more than 60.0% in adolescents and then remains above 60.0% in all subsequent age-groups. For *S. stercoralis*, the prevalence also increases over age, reaching its peak in age-groups 30 years and older. It shows that the average number of helminthic co-infections increases over age, whereas the average number of protozoan co-infections is highest in children and lowest in adults older than 51 years.

However, in this study, there cases of human *Ascaris spp*. infection were detected by microscopy. This coincides with the findings of Park and colleagues [26]. We demonstrated that in all age-groups, the average number of co-infections is about the same, yet helminthic co-infections accumulated over time, with a peak in 30–50year old individuals. The trend for protozoan co-infections is reversed, with the highest number of protozoan co-infections occurring in children. This pattern might reflect higher exposure of children. Alternatively, it could be because of higher infection intensities rather than prevalence in children, as microscopy can miss low-intensity protozoan infections [27], although this applies also for helminthic infections.

DNA Analysis is very important for analyzing characteristic of species of worm but this has not yet been completed in our research. Species of hookworm is *N. americanus*, but cats usually could be infected by *Ancylostoma caninum*, condition of data this research not similar with research in Northern Thailand that it has significant zoonotic hookworms include *A. ceylanicum, A. braziliensis* and *A. caninum* [28, 21, 29]. Molecular analysis revealed that the most prevalent hookworm (over 80%) found in dogs and cats in the lower Northern area was *A. ceylanicum. A. ceylanicum* is highly prevalent in many areas in Asian countries [29, 19,20] and is known to produce potent infections in humans. *A. ceylanicum* is the second most common hookworm infection in humans that can lead to anemia [28, 21]. PCR and sequencing were used for detection and identification of parasites in various specimens with high sensitivity and specificity [3, 29]. In our survey, molecular analysis was applied for two significant helminths infection, hookworms and *O. viverrini*. Morphological identification of hookworm larvae or eggs to species is difficult, and molecular identification provides great results in this regard [16].

This research has deference with research in Cambodia which it showed that in humans about half of the infections (51.6%) were *Ancylostoma ceylanicum* and the remaining *Necator americanus* infections. In dogs over 90% were *A. ceylanicum* indicating that most probably dogs are the source of infection. We hypothesize that regular deworming in community lead to a replacement of *N. americanus* by *A. ceylanicum*. Parallel deworming of the dog population is likely to reduce the incidence in humans [30]

Zoonotic Risk factors, in this research could be seen via behavior of defecation of cats and rats, environmental factors and personal hygiene that also facilitate sanitation can contribute to the spread of STHs infection from animal to human. Defecation behavior of cats upon observation posted little risk for infection as most cats would cover up their waste with soil. Further, the exposure of the cat feces to the sun is also a deterrent to transmission. Studied in Northern Thailand showed that Zoonotic hookworm, *A. caninum*, was found to have low infection rates in both dogs and cats. Similar to other areas, prevalence of *A. caninum* was

lower than that of *A. ceylanicum* [3, 16]. Although its infection rate was low, this hookworm can result in eosinophilic enteritis and chronic abdominal pain in human [31,19,20, 32].

Environmental factors have potential zoonotic determinants for hookworm infection and strongyloidiasis. This research location has poor sanitation, without wastewater drainage that could easily facilitate the spread of *Ascaris lumbricoides* and potential risk of zoonotic diseases by contamination from cats and rats to human via feces of cats and rats could spread soil transmitted helminth infection.

Conclusions

Results of this study showed that cats and rats had similar infections with human soil transmitted helminth infections only *Ascaris lumbricoides* infection, and hookworm, *Strongyloides stercoralis*, and *T. trichiura* but did not have equal zoonotic potential as observed with the cat defecation behavior and other [redisposing factors where animal feces may be left exposed to the elements and be potentially inactivates the interferes with the completion of the life cycle to infective filariae form larvae especially hookworm. *Strongyloidisis strercoralis* and *Trichuris* from rats did not show infective potential to humans because rats usually defecated on wet and moist areas where there is little to no human activity. Further environmental epidemiological studies of soil transmitted helminth infection are important for analysis of zoonotic diseases especially in communities with differerent geographical profile.

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