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Research article

Detection for potentially zoonotic gastrointestinal parasites in long-tailed macaques, dogs and cattle at Kosamphi forest park, Maha Sarakham

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Abstract

Gastrointestinal parasite are the most important and common infectious agents of human and animals worldwide especially in tropical areas. Humans can be infected with the parasites as zoonoses by shedding their infective stage and eggs via feces of animal reservoirs resulting in environmental contamination of food and water. In Maha Sarakham, Kosamphi forest park is an interesting area which has close interaction and coexistence between wildlife, domestic animals and humans where it is possible to transmit some zoonotic pathogens including gastrointestinal parasites among them. This study was conducted to investigate the occurrence of gastrointestinal parasites present in feces of long-tailed macaques, dogs and cattle shed on the environment in and around Kosamphi forest park. A total of 134 fecal samples collected from 3 animal species were processed to detect helminth eggs using the formalin-ethyl acetate technique. Three species of parasitic helminthes including Strongyloides spp., Trichuris spp. and hookworm's eggs have been detected and assessed for their prevalence. Strongyloides spp., were the most common zoonotic parasite found in monkeys and cattle (52.24% and 45.71%) followed by hookworms (26.87% and 34.29%) while hookworms were the most common zoonotic parasite in dogs (31.25%) followed by Strongyloides spp. (25%). This study provides baseline information on potentially zoonotic gastrointestinal parasites in animal reservoirs at Kosamphi forest park and emphasizes the importance of both wildlife and domestic animals as reservoir hosts for zoonotic disease. A One Health Approach is necessary for prevention and control.

Keywords: Gastrointestinal parasite, Prevalence, Zoonosis, Kosamphi forest park, Reservoir

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INTRODUCTION

Gastrointestinal parasites are the most common infectious agents of human and animals including livestock, companion animals and also wildlife predominantly in rural areas in Southeast Asia (Odermatt et al., 2010). Many gastrointestinal (GI) parasites including Trichuris spp., Strongyloides spp., Ascaris spp., Schistosoma spp. and hookworms often infect both humans and animals. Infection by these GI parasites can cause sub clinical symptoms to clinical symptoms of long duration with abdominal pain, diarrhea, dysentery and weight loss (Hechenbleikner and McQuade, 2015). Most of them have been reported to be zoonotic agents which have the ability to transmit between humans, livestock, companion animals and wildlife especially in their larval stage of infection (Lavikainen, 2010). Domestic animals and wildlife play crucial roles as reservoirs that maintain zoonotic parasites under natural conditions from which humans can become infected (Youssef and Uga, 2014). Animal reservoirs often shed zoonotic parasites as oocysts, eggs and larvae in their feces into the environment. Humans are infected with the GI parasites by ingestion of contaminated water and food which contain oocysts or eggs as food or water-borne pathogens. In addition, direct transmission can occur by contact with feces of reservoir animals that contain larval infective stages of the parasites (Ayinmode et al., 2016). This neglected zoonotic transmission of parasites between animals and humans is the major concern for One Health problem worldwide (Thompson, 2013).

In Thailand, long-tailed macaques (Macaca fascicularis) occur widely including at Kosamphi Forest Park in Maha Sarakham, in the northeast of the country. A One Health problem is emerging because of increases in their population, habitat loss from urbanization and reduction of natural food sources (Malaivijitnond and Hamada, 2008). This is resulting in increased interaction and conflict between humans in the community around the forest park and the long-tailed macaques. The monkeys often invade houses, schools, temples and markets near the park for foraging and inhabiting, so that, they have a potential to shed zoonotic pathogens such as those parasites from their feces or excreta to humans via contaminated foodstuffs and drinking water. Moreover, the presence of domestic animals such as dogs and cattle around and inside the forest park areas suggests that it might be possible for them to acquire disease from the monkeys. Therefore, both domestic animals and wildlife seem to be potentially zoonotic reservoirs and spillover hosts which should be included as essential components of monitoring and surveillance programs.

This study was designed to investigate the occurrence of gastrointestinal parasites presented in animal reservoirs; long-tailed macaques, dogs and cattle feces shed on the shared environment at Kosamphi forest park in Maha Sarakham, Northeast Thailand. The study will provide useful provide baseline data on fecal parasites in those reservoirs which potentially cause zoonotic diseases to human including tourists, researchers, park staffs and people who live close to this area. The results will be beneficial for further developing suitable one health strategies to prevent and control zoonotic diseases.

MATERIALS and METHODS

The sample collection was performed without disturbing or contact with the animals. Fresh stool (feces) samples were collected, using gloves from 67 long-tailed macaques, 32 dogs and 35 cattle in Kosamphi Forest Park, Kosum Phisai district, Maha Sarakham, Northeast Thailand at N16º 15' 12.6" E103º 04' 02.0" (Figure 1) and selected locations close and around the park during November, 2015 to April, 2016. All feces samples were collected in labelled plastic universal bottles and rapidly transported to the laboratory where they were preserved in 10% formalin.

Fecal material was processed for detecting the presence of GI nematode eggs by formalin-ethyl acetate procedure and the identification of parasitic eggs was carried out by microscopic examination (Gracia, 2011). Different diagnostic procedure for detection of *Strongyloides stercoralis* larvae was done by fresh stool examination under a microscope. Results were recorded in Windows Excel (Microsoft, USA). The occurrence of GI parasitic eggs which referred to infected animals was used to calculate an occurrence using the descriptive statistics on MS Excel version 2013 (Microsoft, USA).

All data were represented as means ± standard errors (SE). Data were statistically evaluated by a one-way analysis of variance. Significant difference ($p \leq 0.05$) among the groups was analyzed by Duncan's multiple range tests. All experiments were repeated 3 times, each with 5 replicates of the designed concentration.

Figure 1 Geographical location of sampling points; Kosamphi forest park in Kosum Phisai district, Maha Sarakham province, Northeast Thailand (from Google Earth). Yellow line: a forest park area (a habitat of long-tailed macaques), blue line: urban and rural areas (habitats of dogs and cattle)

RESULTS

Out of total 134 fecal samples collected from long-tailed macaques, dogs and cattle, parasitic eggs were identified in 79 (58.96%) indicating the prevalence of parasitic infection. Gastrointestinal parasite eggs were found in 42 out of 67 long-tailed macaque samples (62.69%), 14 out of 32 dog samples (43.69%) and 23 out of 35 cattle samples (65.71%). Three species of parasitic helminthes including *Strongyloides* spp., *Trichuris* spp. and hookworm's eggs which have been reported to be potentially zoonotic helminthes were identified in all three animal species as indicated in Table 1. Among the positive samples, multiple parasitic infections (infection with more than one species of parasite) were found in most long-tailed macaques, dogs and cattle (68.66%, 56.25% and 57.14%, respectively). In addition, the study revealed the highest prevalence of *Strongyloides* spp. infection was in long-tailed macaques and cattle, while, hookworm infection had the highest prevalence in dogs. However, *Trichuris* spp. was the least prevalent of GI parasitic infections in all animal species. Besides, 12 feces samples (17.91%) from long-tailed macaques were found with rhabditiform larvae of *Strongyloides stercoralis* as shown in Figure 2. Overall prevalence of parasitic infection and the prevalence of each parasite species in each animal reservoirs are also showed in Table 1.

Table 1 Prevalence of gastrointestinal parasite eggs in feces of animal reservoirs in Kosamphi Forest Park, Maha Sarakham, Thailand.

Animal species (No. examined)	Species of parasite eggs identified	Number of positive (Prevalence %)
Long-tailed macaques	*Strongyloides* spp.	35 (52.24)
(67 samples)	*Trichuris* spp.	9 (13.43)
	Hookworms	18 (26.87)
	Total	42 (62.69)
Dogs	*Strongyloides* spp.	8 (25.00)
(32 samples)	*Trichuris* spp.	3 (9.38)
	Hookworms	10 (31.25)
	Total	14 (43.69)
Cattle	*Strongyloides* spp.	16 (45.71)
(35 samples)	*Trichuris* spp.	7 (20.00)
	Hookworms	12 (34.29)
	Total	23 (65.71)
Total 134 samples		

Figure 2 Rhabditiform larvae of *Strongyloides stercoralis* in the feces sample of long-tailed macaques under 10X under microscope.

DISCUSSION

There is concern about the prevalence of gastrointestinal helminthes infection in many animal reservoirs. This study highlights and emphasizes the presence of zoonotic potential of these parasites which can transmit infection between animals and humans as well between animal reservoirs such as monkeys, dogs and cattle inhabiting this study area as has been mentioned in many previous reports (Inpankaew et al., 2015; Parmar et al., 2012; Squire et al., 2012). The result clarified the view that all of these parasitic infections (Strongyloidiasis, Trichuriasis and hookworm) are predominant diseases of tropical and humid areas especially in Southeast Asia (Puthiyakunnon et al., 2014; Bethony et al., 2006).

Many previous studies also mentioned a possibility of zoonotic diseases at high risk interfaces between humans and animals especially where humans, domestic animals and wildlife coexist, as is the case at Kosamphi forest park (Malaivijitnond et al, 2006; Klaus et al., 2017). At the present, The potential risk factors which promote the transmission of zoonotic helminthes in the studied area are variables associated with human-wildlife-domestic animals coexistence leading to close interaction among them, an increasing number of monkeys, expanding human populations and urbanization, a behavioral changing from food restriction leads to people community invasion or peri-urban environment, increasing of physical contact to macaques from tourists who attempt to feed them by hand and lack of knowledge in parasitic zoonoses as well as their transmission. Also, severe flooding occur regularly because the forest park is located very close to Chi-river. Therefore, disaster is also a vital factor that affects the problem by decreasing their living space and natural food.

All of these factors can contribute to close relationship (Figure 3) and conflict between human-animal reservoirs and also influence the transmission of zoonotic diseases by increasing the potential risk of zoonotic parasite diseases spreading in ways similar to many previous studies (Priston and McLennan, 2013; Malaivijitnond et al., 2005; Parmar et al., 2012). Therefore, it is important to pursue some further studies on the dynamics between human wildlife and domestic animal species in this area in order to prepare proper preventions and controlling strategies.

Figure 3 Long-tailed macaques were invading the human community near Kosamphi forest park. Monkeys were invading outdoor cooking areas near the forest park and foraging for food (A, B). Some monkeys lived in a school in front of the forest park (C, D).

Interestingly, this examination found rhabditiform larvae of *Strongyloides stercoralis* in 12 feces samples of long-tailed macaques. They can be passed along with the feces, contaminate soil or water, and live as a free-living parasite in the environment. On the other hand, the rhabditiform larvae can develop to be an infective stage as filariform larvae which is a significant cause of autoinfection within the animal reservoirs. It usually penetrates either the skin of perianal region (external autoinfection) or the intestinal mucosa (internal autoinfection) of the hosts (Toledo et al., 2015). In the case of rhabditiform larvae, this autoinfection provides an opportunity for persistent long-term infection of long-tailed macaques in the forest park. It is crucial that this knowledge be used assessing a chronic strongyloidiasis and fatal *Strongyloides* hyperinfection that affect

animals and also human health in the study area (Kassalik and Mönkemüller, 2011).

The detection of hookworm and whipworm eggs implies the current dynamics and consequences of their infections in domestic animal and wildlife. Hookworms (Ancylostomatoidae) are blood-feeding parasitic nematodes that penetrate through the digestive system of mammalian (Popova, 1964). Previous studies mentioned that most hookworms usually parasitize in the small intestine of the host and cause blood loss, inflammation in the mucous membrane, impairing digestion and absorption, secondary bacterial infections. Domestic animals, wildlife and human whom infected with the hookworms will have various significant adverse effects as anemia, poor growth, secondary bacterial infection, tissue damage and also mortality (Seguel et al., 2017). Likewise, Whipworm (*Trichuris* spp.) is known globally as one of the most common soil-transmitted helminth which causes trichuriasis, neglected tropical diseases. The infection can lead to chronic diarrhea, anemia, retarded growth (Stephenson et al., 2000) and also potentially transmit to human as zoonotic trichuriasis which has occasionally been reported such as *Trichuris trichuira* that usually found in human and other non-human primates in many regions around the world (Yao et al., 2018).

This finding revealed the significance of hookworm and whipworm infection which are some of the most common helminth infection in domestic animals and wildlife. In addition, they could be potential important zoonotic pathogens as a previous study (Smout et al., 2013), while, those domestic animals and wildlife play an essential role as maintaining source of zoonotic infections in this area, as well.

CONCLUSION

This study provides clear evidence that there is a high prevalence of zoonotic GI parasites detected in feces sampled from wildlife (long-tailed macaques) and domestic animals (dogs and cattle) living in or near Kosamphi Forest Park. They seem to be the important reservoir hosts for zoonotic GI parasites found in the study area including *Strongyloides* spp., *Trichuris* spp. and hookworms that spillover among many different animal hosts and contribute a potential risk to health status of humans living near the forest park with regard to be health hazardous to humans as zoonoses. Therefore, it is necessary to plan suitable strategies of prevention and a control program for such parasitic diseases in animal reservoirs. Improvements a sanitation are also required to inhibit transmission of these zoonoses and improved future education to increase risk awareness and knowledge of parasitic zoonoses. An important point is that one health collaboration between veterinarians, physicians, public health staff, forest park staff and also environmental health staff is quite appropriate for better operational approach. However, follow-up studies of GI parasitic prevalence in people living near Kosamphi forest park are needed.

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