

Assessment Of Wild Rodents Endoparasites In Kirimiri Forest In Embu County, Kenya

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Abstract: Rodents are reservoirs and hosts of zoonotic diseases. Rodents' pathogenic parasites can be introduced onto soils, water supplies, vegetables and fruits thus playing significant role in human infection. Though studies on rodents and their parasites are necessary to understand and manage zoonotic disease cycle, knowledge gap of endoparasite composition of wild rodents that interact with domestic animals and human still exists in Kenya. This study was carried out to determine the prevalence of rodents' endoparasites in Kirimiri forest, Embu County in Kenya between January and May 2016. Wild rats were caught by laying traps in 100 m x 100 m grid of 50 Sherman and 50 victor traps. Rodents' morphometric data was used for their identification. Necropsy was performed for gastrointestinal tract (GIT) and endoparasites extracted, counted and prevalence determined. Three species of rats totaling to 355 rats comprising of 199 males and 156 females were captured and identified. A total of 533 endoparasites extracted. Rate of endoparasite prevalence was significantly higher in *Rattus* spp a peri-domestic rodent than forest rodents' species ($\chi^2 = 57.791$, $P < 0.05$). *Asyphalia obvelata* (44.79 %) had higher prevalence while the *H. dinimuta* had lowest prevalence (6.20 %). Prevalence based on forest patches, GIT and was different. The current study highlights the importance of rodents as potential vectors for intestinal parasitic infections.

Keywords: Rodents, Endoparasites, Kirimiri Forest, Kenya

Introduction

Rodents are reservoirs and are carriers of zoonotic diseases (Luis et al., 2013; Chaisiri et al., 2015). Commensal rats may spread many diseases to human, cattle and pets. Zoonotic diseases of medical significance among others include plague, murine typhus, scrub typhus, hantavirus hemorrhagic fever (Kosoy et al., 2015). Other diseases transmitted by rodents are leishmaniasis (Davami et al., 2014), trichinosis (Ryan and Ray, 2004), Hantavirus pulmonary syndrome (Peters, 2006). Rodents' pathogenic parasites can be introduced onto soils, water supplies, vegetables and fruits leading to human infections (Wood and Johnson, 2015; Hamidi, 2018). Mechanisms of disease transmission include contact with saliva, urine and feces of rodents. Majority of rodent species express opportunistic behavior and high fecundity rates (Geffen et al., 2011). In a new ecosystem, rodents may accelerate spread and invasions by spillover of parasites into new hosts (Schmid et al., 2015). Spillover of parasites accelerates local acquisition and spread of parasites to new rodent hosts (Hulme 2014). Infested rodents' reservoirs maintain parasites in a habitat through spill-back (Reusken et al., 2011; Meerburg and Reusken, 2011). Parasite's spill over and spill-back negatively impact ecological systems including wildlife, domestic species and have implication to human health (Wood et al., 2012; Hatcher et al., 2012). Humans infection with parasites have clinical symptoms that epitomized by diarrhea, abdominal pain and anorexia (Karuna and Khadanga, 2013; Kim et al., 2014). Parasites that infest small mammals can be taxonomically be grouped into: Cestodes, nematodes and finally, acanthocephalans (Gibson et al., 2014). Majority of these parasites require invertebrate intermediate hosts for the

development of their larvae in their life cycles (Gibson et al., 2014). Parasitic fauna of the rodents in each ecological setting is different (Seifollahi et al., 2016). Studies on rodents and their parasites have medical and veterinary importance to prevent transmission of diseases to human and domestic animals. Studies on endoparasite of wild rodents have gotten attention in several countries; China (Chaisiri et al., 2015), Lao People's Democratic Republic (Pakdeenarong et al., 2014), Malaysia (Mohd et al., 2012), Philippines, Thailand (Chaisiri et al., 2012) and Indonesia (Prasetyo, 2016). In Kenya, studies on rodents have focused largely on; taxonomy (Corti et al., 2005; Demos et al., 2014); Effect of land use on rodent biodiversity; ectoparasite diversity (Mugatha, 2004; Young et al., 2016) and Agricultural losses (Taylor, 1968). Most of the studies on rats pathogens have concentrated in dwelling places and have involved; *Leishmania* parasites the causative agent of visceral leishmaniasis (Kinuthia et al., 2011); *Bartonella* spp. (Halliday et al., 2015; Wainaina et al., 2018). Thus, information of rodents endoparasites particularly rats and their significance in disease transmission is scarce in Kenya. Such studies are essential for public health given their role in diseases cycle. This study was carried out to assess the occurrence of wild rats endoparasites and create understanding of their zoonotic significance. Availability of this information is crucial in zoonotic disease management, wildlife conservation, policy formulation and for monitoring of variation in the ecosystem through comparative studies.

5. Materials and Methods

Area of study

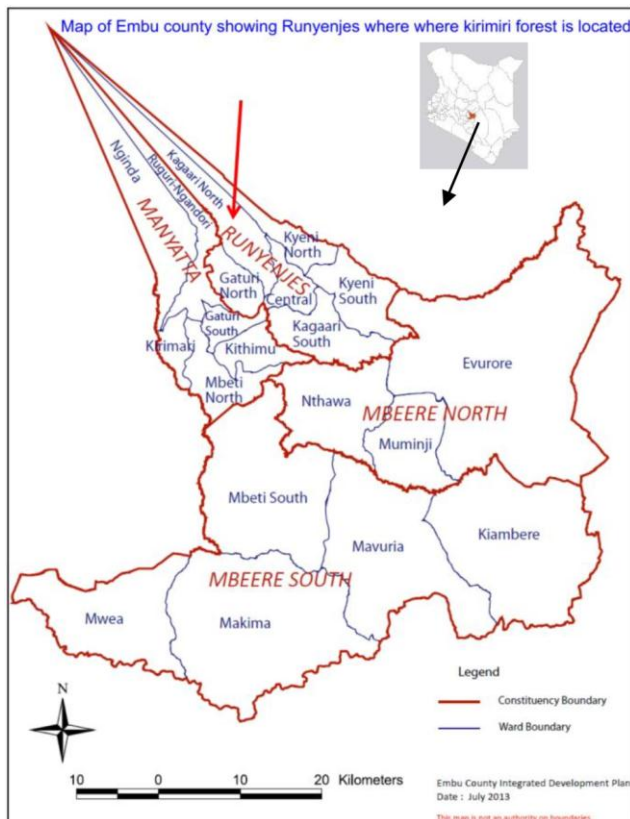


Figure 1: Map of Embu county (CGoK, 2014)

The study was conducted in Kirimiri Forest situated in Mukuuri - Runyenjes in Embu County in Kenya. It is regarded as an Ecologically Sensitive Site by the International Union for the Conservation of Nature. It is dominated by tree vegetation including rare variety of indigenous and medicinal trees which are faced by threat of deforestation elsewhere in the country. The area has an elevation of 1520 meters above sea level with its center lying at the latitude of -0.41667 and a longitude of 37.55 (Longitude/ latitudes S $0^{\circ} 25' 22.30''$ E $37^{\circ} 32' 41.42''$). The average annual rainfall ranges from less than 600 mm to 2500 mm. Temperatures range from 12°C in July to a maximum of 30°C in March with a mean average of 21°C (CGoK, 2014). There is settlement and agricultural activities in the immediate surrounding of Kirimiri forest where tea among other crops are grown as well as rearing of animals. The forest was divided into four zones; farmland comprising of tea and maize, forest edge comprising of pine tree with tree logging, mixed Indigenous with pine trees and an intact Indigenous forest zones with mixture of tree species. In each zone, a trap line was established.

Rodent capturing and identification

Traps were laid in 100 m x 100 m grid of 50 Sherman and 50 victor traps. Two trap lines were established in each grid 10 m away from the boundaries. In a trap line, the distance gap of 10 m was set between one trap to the next. The distance between one grid to the next was 150 m apart. A total of 16

traps were laid in each trap line comprising of Victor traps and Sherman traps. The traps were laid every evening at 1800 hrs and checked every morning at 0800 hrs. Peanut butter was used as bait in all cases. Trapped rats were tagged and placed in Zip lock bags prior to transportation to Chuka University zoology laboratory in cool box for analysis. Rodent morphometric data recorded such as weight, total length, and lengths of the tail, hind foot, forearm and ear. Morphometric data were used to identify the rodent's specimen using mammalogy guide book (Stuart and Tilde).

Rodents' examination and endoparasite identification

In the laboratory, rodent specimen was placed on a tray and necropsy performed. Gastrointestinal tract was removed and placed on petri dishes containing physiological saline. Stomach was opened longitudinally using scissors and observed for parasites infestation. Parasites were extracted and placed on a separate clean petri dish for identification and counting. Dissecting microscope was used for this purpose. Preservation of parasites extracted was done in ethanol (70%). After the visible parasites had been removed, further examination of colon contents was done using simple floatation technique. Assessment of helminth eggs and coccidia oocysts in intestinal contents was done using a simple qualitative method. In this method, 2 g of the gastrointestinal contents were mixed with 30 ml saturated sodium chloride (NaCl) solution and the suspension was filtered into a beaker. Strained gastrointestinal content suspension was transferred into a test tube filled up and covered with coverslip and left to stand for 20 minutes. After 20 minutes, a cover slip was removed placed on inverted LSD microscope and examined for parasite eggs and oocysts.

Data analysis

Prevalence of parasite infestation was calculated following Bush et al. (1997);

Prevalence (P) =

$$\frac{\text{Number of hosts parasited with a particular parasite group}}{\text{Total number of hosts examined for that parasite group}} \times 100$$

Prevalence was calculated for each group of endoparasite. Data analysis was performed with STATA 6 (Stata Press, College Station, Texas, USA).

Results

Infectivity of captured rodents with endoparasites

A total of 355 rodents belonging to three genera were captured, among which *Rattus* spp 180 (54 %) were the highest *Mastomys* spp 112 (32%) while *Hylomyscus* 63 (18 %) were the lowest. The infection rates of these species with endoparasites are shown in Table 1. *Rattus* spp had higher prevalence rates (61.67%) followed by *Mastomys* spp (59.82%) while *Hylomyscus* spp had the lowest prevalence rates (36.51%) in Table 1. Rate of endoparasitic infections was significantly higher in peridomestic rodents than forest rodents' species ($\chi^2 = 57.791$, $P < 0.05$).

Table 1: Infectivity of captured rodents with endoparasites

Host species	Total Rodent (n)	Number infected	Endoparasite Species	Endoparasites Number	Prevalence (%) by Endoparasite	Prevalence rodent spp	by
Rattus spp	180(54%)	96	Asyphalia obvelata	116	53.33	61.67%	
		70	Heterakis spimosa	86	38.89		
		49	Aspiculuris tetraptera	65	27.22		
		22	Moniliformis sp.	59	12.22		
		17	Syphacia Spp.	52	9.44		
		17	Trichris muris	44	9.44		
		47	Echinostoma Spp.	85	26.11		
		15	Hymenolepis diminuta	66	8.33		
		19	Hymenolepis nana	41	10.56		
		33	Gongylonema	43	18.33		
Mastomys spp.	112(32%)	54	Asyphalia obvelata	61	48.48	59.82 %	
		11	Heterakis spimosa	20	9.82		
		9	Aspiculuris tetraptera	14	8.08		
		13	Moniliformis sp.	17	11.61		
		7	Syphacia Spp.	15	6.25		
		16	Trichris muris	25	14.29		
		22	Echinostoma Spp.	36	19.64		
		6	Hymenolepis diminuta	13	5.36		
		8	Hymenolepis nana	30	8.04		
		9	Gongylonema	15	8.03		
Hylomyscus spp	63(18%)	9	Asyphalia obvelata	21	26.98	36.51 %	
		1	Heterakis spimosa	2	1.59		
		11	Aspiculuris tetraptera	31	17.46		
		1	Moniliformis sp.	4	1.59		
		2	Syphacia Spp.	6	3.17		
		3	Trichris muris	11	4.76		
		5	Echinostoma Spp.	33	7.94		
		1	Hymenolepis diminuta	1	1.59		
		2	Hymenolepis nana	6	3.17		
		2	Gongylonema	8	3.17		

Endoparasite counts were higher in tea plantation with mean prevalence of 71.67 % and lower in pine forest with a mean prevalence of 47.09%. In all the forests types studied, infection prevalence was higher in Rattus spp. Prevalence in

Rattus spp was higher in tea plantation with 83.72 % and the lowest in the pine area recording 60 % (Table 2).

Distribution of endoparasites in different rodent hosts

All the rodent endoparasites common with Rattus species were present in all other rodents (Table 1)

Effect of Forest type on rodents and endoparasite infection prevalence

Table 2: Forest type, number of individual host species captured and endoparasite infection prevalence

Habitat type	Host species	Number trapped	Endoparasite	Number infected	Prevalence (%)	Prevalence Mean (%)
Indigenous Forest	Rattus spp.	92	167	73	79.35	60.72
	Mastomys spp.	64	87	29	45.31	
	Hylomyscus spp.	40	72	23	57.50	
Mixed Indigenous and Pine Tree Forest	Rattus spp.	25	28	18	72.00	49.19
	Mastomys spp.	31	60	14	45.56	
	Hylomyscus spp.	10	24	3	30.00	
Tea Plantation	Rattus spp.	43	42	36	83.72	71.67
	Mastomys spp.	22	59	14	63.63	
	Hylomyscus spp.	9	22	6	66.67	
Pine Forest	Rattus spp.	20	33	12	60.00	47.09
	Mastomys spp.	16	28	5	31.26	
	Hylomyscus spp.	4	5	2	50.00	

Prevalence of endoparasite according to rodent's gender
Rodent's gender had significant effect on their endoparasites prevalence ($\chi^2 = 5.208, P < 0.05$). Males had higher mean prevalence (59.92 %) than in females (40.08 %) (Table 3).

Males in Rattus spp had prevalence of 62.16 % being the highest. Nonetheless, prevalence observed in all male rodents were above mean prevalence of 59.92 % while those of female rodents were below the mean prevalence total male rodents trapped were higher than female (Table 3)

Table 3: Prevalence of endoparasite in according to rodent's gender

Rodent species	Total endoparasite	Male Rodent trapped	Male Prevalence n (%)	Female Rodents trapped	Female Prevalence n (%)
Rattus spp	270	99	69 (62.16%)	81	42 (37.84 %)
Mastomys spp.	240	61	38 (56.72%)	51	29 (43.28 %)
Hylomyscus spp	123	39	14 (60.87 %)	24	9 (39.13 %)
Total Mean Prevalence		199	40.33 (59.92)	156	26.67(40.08)

Prevalence of endoparasite according to gastrointestinal tract section (GIT)

Gastrointestinal tract affected rodents endoparasites prevalence. Higher prevalence was recorded in the small intestine (44.79%) and lowest in the stomach (3.94%) (Table 4).

Table 4: Prevalence of endoparasite in according to gastrointestinal tract

GIT	Endoparasite	Number infected	Prevalence n (%)
Stomach		14	3.94 %
Intestine		159	44.79 %
Caecum		48	13.52 %

Number of rats examined = 355

Prevalence according to Endoparasite species

Rodents infection prevalence differed with different species of endoparasites. *Asyphalia obvelata* was the most prevalent endoparasite with prevalent mean of 44.79 % while *H. dinimuta* was the lowest in terms of its prevalence (6.20 %). *Asyphalia* spp, *Heterakis* spp, *Echinostoma* spp and *Aspicularis* spp had their prevalence higher than the prevalence mean of 14.51 while the rest were below the prevalence mean (Figure 2)

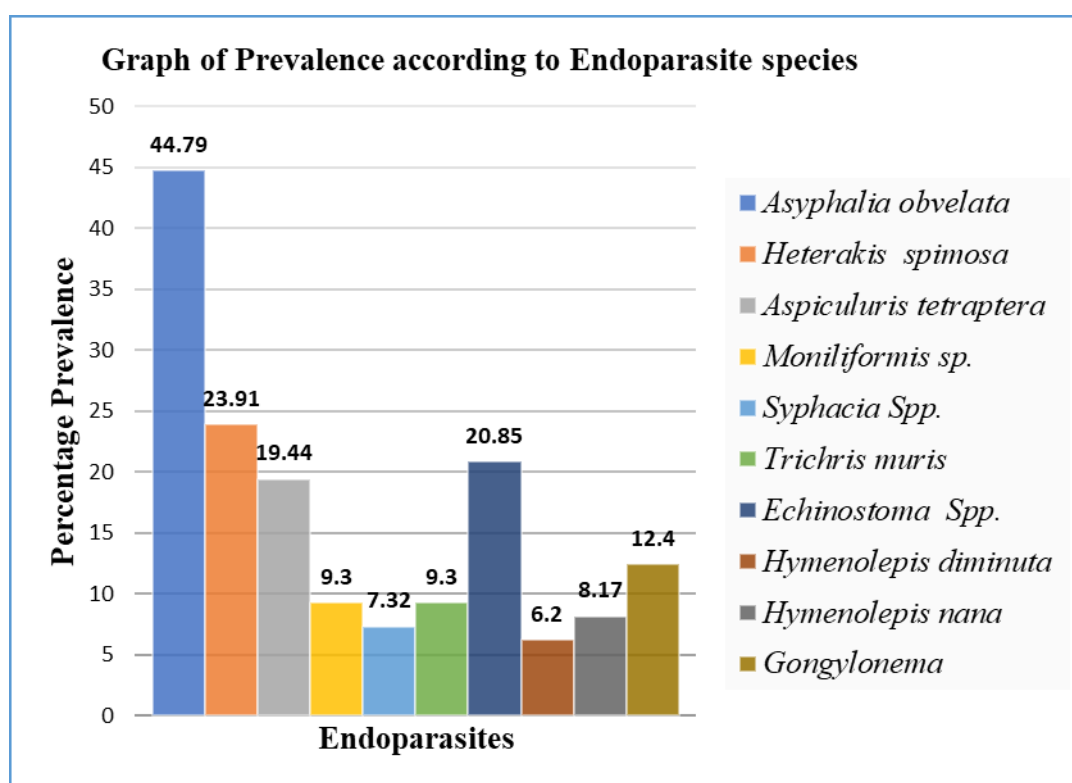


Figure 2: Graph of Prevalence according to Endoparasite species

Discussion

Peri-domestic rats (*Rattus* spp) were captured in all forest patches studied including pine forest which recorded low number of rodents generally. Occurrence of commercial rats and their endoparasites deep in the forest is due to their dispersal from homes and farmland the forest edge as a result of ecological destruction (Guttinger 1998). Wide occurrence confirms that rodents are highly opportunistic and can quickly easily adapt to new conditions, take advantage of temporarily suitable environmental and multiply rapidly

(Löhmus et al., 2013). Different endoparasites were found to infect different rat species captured. The most affected rat species was *Rattus* spp (Table 3). *Rattus* spp was found to be the main host of most endoparasites observed. This finding is in line with those of (Krishnasamy et al., 1980; Ambu et al., 1996; Chaisiri et al., 2010). High vulnerability of *Rattus* spp to most endoparasites is a pointer to their diet on intermediate insect hosts (Singh et al., 1987; Jeffery et al., 2003). Additionally, higher infection of *Rattus* spp is partly due to their foraging behaviour and indiscriminate feeding

nature. This exposes them to a variety of endoparasites and ultimate infection. In this study, low endoparasite load was observed in *Hylomyscus* spp. The moderate endoparasite prevalence in *hylomyscus* spp. is attributed to low consumption of endoparasites intermediate hosts as well as wide home range which minimized interactions (Nunn and Dokey 2006). Rodents infection with endoparasites was observed to differ depending on forest type (Table 2). Intact forest had higher prevalence of endoparasites. Artificial forest comprising majorly of cypress and pine had lower endoparasite prevalence. The effect of forest type on rodents' endoparasite prevalence reported in this study corroborates with the finding of Chaisiri et al., (2012) and Archer et al., (2017). Higher endoparasites prevalence in tea planta was attributed to presence of open domestic waste dumpsites at the edge of the farm that borders dwelling houses. The prevalence was also higher in the intact forest area. Location of tea farm near dwelling houses at Kirimiri forest facilitates high interaction between domestic animals (cats and dogs) faecal material and the rodents thus high prevalence observed. Intact forest area had many invertebrates including and not limited to snails, cockroaches, millipedes and centipedes. These insects and invertebrates which are fed on by rodents hosts infective stages of these endoparasites thus higher prevalence. Higher endoparasite prevalence was also partly attributed to higher number of peri-domestic rats captured in tea planta and intact forest patches. Generally, there was low number of rodents and their endoparasites in the pine area. Pine (forest edge) area appeared to have had a lot of human activities since it borders the tea and maize planta. There was constant grazing activity observed. Besides pine forest area had reduced vegetation height and associated reductions in predation cover. Limited insects and rodent food source were available. This fact confirms report by Guttinger et al., (1998) that vegetation structure, food availability and human activity influence distribution of rodents. Endoparasites with the highest prevalence was *Asyphalia obvelata*. *H. diminuta*. was the least prevalent (Figure 2). Higher infection by *Asyphalia obvelata* demonstrates its high adaptability and persistence to its host who moves from one habitat to the next thus it's rapid transmission rate (Warner, 1998). Ecologically, this parasite is having cosmopolitan distribution in and around Kirimiri forest. The finding is similar to those of Moradpour et al., (2018) who reported *Asyphalia obvelata* as the most prevalent endoparasite in Iran. Results differ with those of De Leon, (1964) who reported *Hymenolepis diminuta* as the most prevalent; Coomansingh et al., (2009) reported *Nippostrongylus brasiliensis* as the most prevalent in wild rats in Grenada. Reasons for the difference in results of endoparasites studies published from different countries are due to geographical difference and environmental factors (Lotfy, 2014). Generally, the result confirms the argument by Seifollahi et al., (2016) that parasitic fauna of the rodents is different in each ecological setting. Most of the endoparasites observed in *Rattus* spp. were shared with other rats across the forest (Table 1). This points at possible transmission of the pathogens either by common food source, water source or from common host. Common hosts such as *Rattus* spp can facilitate spread to new niches and infection of new host through parasite spill over (Seifollahi et al., 2016) and spill-back (Wood et al., 2012; Hatcher et al., 2012). Common food rats' food source contamination with rat faeces by spills account for the continuity of infection

(Fagir and EL-Rayah, 2009). This fact appeared to be the case in Kirimiri forest where *rattus* spp were found to be present at almost all forest patches. Higher endoparasite prevalence was observed in male than in female rodents (Table 3). Similarly, Archer et al., (2017) observed different endoparasite load in male and female rodents in South Africa. The result differs with those of Gurler et al., (2011) who observed no difference in endoparasite load in male and female. The higher parasite infection in males than in female is factor of hormonal difference (Klein, 2000). Males have weaker immune responses than females and hence less resistant to helminth parasites (Luong et al., 2009). Males are more susceptible to infection than females because androgens reduce immunocompetence. Also sex steroid hormones affect disease resistance genes and behaviors making males more vulnerable to infection. In this study, more males (199) were captured in traps than the female (156) rats (Table 3). High number of males caught was attributed to their wide home range as compared to females which mostly spend their time around the nests (Hooker and Innes, 1995). According to Odhiambo and Ogue (2003), males rodents make wider field excursion than females. Females remain in their nests nursing their litters; this limits their movement across the habitat where traps are laid. Gastrointestinal (GIT) section affected endoparasites count in rodents studied (Table 4). Small intestine was the most preferred area of GIT by the endoparasites. These findings are supported by those of Okorafor et al., (2012). Preference of the small intestine by most of endoparasites is due to the presence of digested absorbable nutritive food materials as opposed to crude hard food particles in the stomach and nutrient-less food remains in the caecum. Digested soluble foods products are readily available for absorption by most endoparasites which cannot use the crude food in the stomach. Zoonotic potential of most of the parasites extracted from rats in this study have been reported (Salehabadi et al., 2008). For instance, cases of human infection by *moliniformis* spp has been reported in Nigeria (Ikeh et al., 1992) and Iran (Berenji et al., 2007; Rokni, 2008). *Hymenolepis nana* which is common helminth occurring both in man and rodent plays significant role in the prevalence of some of the essential human parasites (Flynn, 1973). *Hymenolepis nana* needs just one host to complete its lifecycle in their host (Beaver et al., 1984). Both *H. nana* and *H. diminuta* pathogens have been isolated in stools of children in Mexico (Quihui et al., 2006; Martínez-Barbabosa et al., 2010). *Hymenolepis diminuta* was reported in a child living in the urban area of Rome Italy (Marangi et al., 2003). Human infection with *Gongylonemagongylonemosis* has been reported in Japan (Haruki et al., 2005). Intestinal parasite such as *Heterakis spumosa* genus *Heterakidae* (Šnábel et al., 2014) is common parasite of rats, mice and occasionally hedgehogs (Ito and Itagaki 2003; Ribas et al., 2013) with no reported zoonotic cases. Endoparasites such as *Trichinella spirallis* reported in this study has been flagged as major zoonotic helminth of public health concerned (Stojcevic et al., 2004).

Conclusion

Occurrence of zoonotic endoparasites in wild rats elucidates the public health implication of interaction between the wild rats, peri-domestic rats and human habitat. The current study highlights the importance of rodents as potential vectors for parasitic intestinal infections around and in forest

environment. Build-up of these parasites may negatively impact ecological systems, wildlife, domestic species and have implication to human health. Thus, studies on rodents and their parasites with environmental, medical and veterinary importance are necessary to prevent spillover for conservation purpose and to minimize disease occurrence.

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