A SURVEY OF PARASITIC INFECTIONS IN WILD RATS FROM URBAN AREAS IN KUALA LUMPUR, MALAYSIA

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ABSTRACT. Wild rats are known as amajor reservoir and intermediate host for several pathogenic microbial species. Thus, the Veterinary Research Institute (VRI) conducted a survey to determine the presence of parasitic pathogens in local rats, such as blood protozoans, gastrointestinal parasites, as well as ectoparasites such as mites and lice. The study was conducted with the collaboration of Kuala Lumpur City Council Pest Control Unit, whereby a total of 105 wild rats were trapped at two urban areas of Kuala Lumpur; namely PasarPudu and Chow Kit. Autopsy was done on the rats to acquire the skin, organ and blood samples.. The skin scrapping was performed on skin samples to identify the common species of mites and lice, while the floatation technique was conducted on faecal samples to identify helminth eggs. Results showed thatspecies of *Tritrichomonas, Strongyloides, Nippostrongylus, Blastocystis, Rodentolepis*, Coccidia, *Trichuris*, *Capillaria*and*Ascarid*werefound in the faeces while *Trypanosoma sp.*was found in the blood samples taken from the animals.*Taeniataeniformis* was obtained from liver samples while theectoparasites found on skin were identified as*Radfordia,Polyplax,Linognathus*and *Hoploplura*sp. Control and eradication of rodent pests is crucial in combating emerging and re-emerging diseases which may be zoonotic as rodents are reservoirs to various pathogens.

Keywords: wild rats, ectoparasites, *Trypanosoma* sp.

INTRODUCTION

The first study conducted in Malaysia related to wild rats is on the prevalence of parasitic species in wild rats in 1933 by Adams followed by several other studies (Yeh, 1955; Sivanandamet al., 1965; Sinniah, 1979) highlighting the pathogens in wild rats in urban and rural areas. The common diseases carried by rats include leptospirosis, salmonellosis and tuberculosis. Rats also carry fleas, mites and ticks and can cause acute allergic reactions in humans. The wild rat is also known as one of the deadliest pests in Malaysia as it plays roles as the reservoir host and carrier at the same time, for several pathogenic microbial species. These microbial species is normally

associated with a wide range of zoonotic diseases such as hantavirus disease, rat bite fever, leptospirosis and cryptosporidiosis which pose a high health risk to human (Paramasvaran *et al.*, 2009). There are many studies on endoparasites of urban and wild rodents which were conducted in Malaysia since the early 1990s (Singh and Cheong, 1971; Leong *et al.*, 1979; Yap *et al.,* 1977; Krishnasamyet *et al.,* 1980; Ambuet *et al.*, 1996; Syed-Arnezet *et al.,* 2006; Mohd Zain, 2008; Premaalatha *et al.,* 2010). There are also several studies on ectoparasites of rodents in Malaysia which were carried out (Audy, 1957; Kohls, 1957; Nadchatram *et al.*, 1966; Lim, 1972; Zahedi *et al.*, 1984; Ho *et al.*, 1985; Ho and Krishnasamy, 1991; Shabrina, 1990; Mariana *et al.*, 1996; Salleh *et al.*, 2003; Chulan *et al.*, 2005). Despite numerous reports on parasites in rats, it is important in updating the parasitic species found across Malaysia, especially when rats act as reservoir to many parasites which are zoonotic. Therefore, the current study is aimed to determine the parasites from the wild rats caught in two regions in Kuala Lumpur which are PasarPudu and Chow Kit.

MATERIALS & METHODS

A total of 105 wild rats, of mixed ages and sex, were caught in PasarPudu and Chow Kit by using the conventional mouse trap placed by officers from Kuala Lumpur City Council Pest Control Unit. The rats were then transported to Veterinary Research Institute and euthanised with chloroform followed by immediate blood collection in EDTA tubes by intra cardiac route. All rats were identified as *Rattus norvegicus* or common brown rat based on descriptions by Medway (1983), and appeared clinically healthy. Visual appraisal was done to collect any ectoparasites seen. Following this, the autopsy was done and the skin and organ (liver and intestine) samples were collected from each individual wild rat and labelled accordingly before being despatched to the Parasitology Laboratory for further parasitological examinations. From the total of 105 rats, 45 were females and 60 were males.

Laboratory diagnostic techniques

This present study includes obtaining the skin, intestine and liver samples from autopsy of wild rats in order to determine the endoparasites such as helminths and protozoa as well as ectoparasites such as mites and lice.

Blood sample

The blood in ethylenediaminetetraacetic acid (EDTA) was subjected to two diagnostic tests; namely, the buffy coat technique and stained thin blood smear technique. The buffy coat technique was done in order to identify the presence of microfilaria or trypanosomes, whereas the thin blood smear was conducted in order to identify the blood protozoans. The buffy coat technique includes taking the blood sample up in a microhaematocrit tube to ¾ of the tube. The tube is then centrifuged and the buffy coat was examined under 100x magnification for evidence of motile microfilaria or trypanosomes. Before the capillary tube was centrifuged for buffy coat technique, a drop of blood was put at the end of a glass

slide to make a thin blood smear. A glass slide which acts as spreader touches the drop of blood, at 45º. Then, the spreader was pushed forward along the glass slide to obtain a thin blood smear. Thin blood smear on the glass slide was allowed to dry and fixed with methyl alcohol. The alcohol was tipped off before 8% Giemsa staining for 30 minutes. Lastly, the Giemsa stain was tipped off, rinsed with tap water and the smear was allowed to dry for observation under microscope at 100x magnification under immersion oil. All the above protocols were followed as outlined by MAFF (1986).

Cysts from Liver sample

The liver had several cysts with fluid. On incising the cysts, the parasite larval stage was obtained.

The larval stages from the cyst were stained by aceto-alum-carmine staining in order to identify the species of worms collected. This protocol was done by first preparing the stain prior to staining. Carmine was boiled in excess in a saturated solution of potassium alum. The solution was allowed to cool before the glacial acetic acid was added to make up 10% solution. This staining solution was keptfor 24 hours in a dark bottle before being used for staining. The staining steps were initiated by pressing the worm between two glass slides before fixing in formalin-acetic-alcohol or 10% formal saline overnight. Following this, the specimen was washed in running water for onetoeight hours. The slide was stained in aceto-alum-carmine overnight. The specimen was washed again in distilled water before it was dehydrated in ascending strength of alcohol. Lastly, the stain was cleared in Beechwood creosote or xylene and mounted in Canada Balsam or Depex. All the above protocols were conducted as mentioned by MAFF (1986).

Intestinal contents

Faecal samples were collected from the large intestines and floatation technique was performed to identify the helminth ova present. This protocol was initiated by mixingthe faecal sample obtained from intestinal sample with saturated salt. The faecal sample was then filtered through a tea sieve into a test tube and allowed to stand erect in a rack. The tube later was topped up with saturated salt until the meniscus was formed on the top of the tube. After that, a coverslip was lowered gently on the meniscus and allowed to stand for 15-30 minutes. The coverslip was lifted vertically up and put onto a glass slide. Lastly, the slide was examined under microscope with low magnification (40×) for parasite eggs or oocysts. The above protocol was conducted following MAFF (1986).

Skin sample

The ectoparasite species from the wild rats were collected using the deep skin scrapping technique. This was initiated by performing deep scrapping on the skin lesions by using a scalpel blade until the subject showed slight-bleeding. The skin scrapping was transferred into a clean container with a drop of glycerol, and observed under microscope for mites or lice. The above protocol is performed as in MAFF (1986). Identification of parasites was

based on keys and references by Soulsby (1982).

All procedures performed on the rats were done by a certified veterinarian to comply with animal welfare protocols.

RESULTS

Tables 1 and 2 summarises the results of parasites detected in various samples; blood, liver, intestinal content and skin from wild rats.

From Table 1, three species of intestinal protozoans, one tapeworm species, and six helminth species were diagnosed from faecal examination.

The protozoan *Tritrichomonas spp.* and helminth *Nippostrongylus* eggs were found in 80% and 63% of the rats making it the most common parasitic infection from faecal examination. Followed closely by *Strongyloides* eggs and *Blastocystis* infections (59 and 40% respectively). A total of 12% of the blood samples showed the presence of Tryopanosomes and 9% of the rats harboured cysts of *Taenia taeniformis.*

Table 2 shows four common ectoparasites found from gross observation and skin scrappings. Notably, 5% had *Radfordia* sp., followed by *Polyplax* sp. (2%) and others.

DISCUSSION

Results from this present study showed that they are several parasitic species which can be found in urban wild rats of Kuala Lumpur. The most prevalent endoparasite protozoan found in wild rats in this study is *Tritrichomonas*, followed by parasites of zoonotic importance — *Nippostrongylus*, *Strongyloides*, *Blastocystis* and *Rodentolepis*. Out of 105, 84 (80%) rats were positive for *Tritrichomonas*. It is a flagellated protozoan parasite which is transmitted easily via the faecal-oral route. *Tritrichomonas muris*is is a common *Tritrichomonas* species that occupies the intestine of rats and other rodents. However, the pathogenicity of infection on animal-host by this parasite is not clear and this parasite is not considered to be zoonotic (Kashiwagi *et al.,* 2009). *Nippostrongylus* spp. eggs was the second most prevalent parasite found in the intestinal content of the examined rats. *Nippostrongylus brasiliensis*, previously known as *N. muris*, is a gastrointestinal roundworm or nematode that infects rodents, primarily Malayan rats (Mohd Zain *et al.,* 2012; Paramasvaran *et al.,* 2012; Siti Safiyyah *et al.,* 2012). Detection of *Strongyloides* spp. in this study is in agreement with Sinniah (1979) who reported that *Strongyloides ratti*is is one of the most prevalent nematode infections in rat in Peninsular Malaysia. This finding is also in agreement with findings by Premaalatha *et al.* (2010) which reported *Strongyloides* eggs from rats in the Veterinary Research Institute, Malaysia. *Strongyloides* spp. can be found in the large intestine of wild rats and could remain in the large intestine of wild rats for 80 days (Shintoku *et al.,* 2005). *Blastocystis*, a protozoan parasite was found in 40% of the examined rats. This parasite inhabits the intestinal tract of a wide range of hosts which includes reptiles, birds, fish, amphibians and also humans (Yoshikawa *et al.,* 2007). Several clinical cases have been found to be related to this parasite presented as vomitting, diarrhoea and other

Table 1. Number of male and female urban wild rats (%) infected by endoparasites in faeces, liver and blood. Total number of rats examined was 105.

Endoparasites	Samples	Number of rats (%)		
		Male N=60	Female N=45	Total N=105
Tritrichomonas spp. Protozoan.	Faeces	49 (47)	35 (33)	84 (80)
Strongyloides. Egg. Helminth.	Faeces	36 (34)	26 (25)	62 (59)
Nippostrongylus spp. Egg. Helminth.	Faeces	36 (34)	30 (29)	66 (63)
Blastocystis sp. Protozoan.	Faeces	24 (23)	18 (17)	42 (40)
Rodentolepis spp. Egg. Helminth.	Faeces	4 (4)	7 (7)	11 (10)
Coccidia oocysts. Protozoan.	Faeces	0 (0)	1 (1)	1 (1)
Trichuris spp. Egg. Helminth.	Faeces	0 (0)	1 (1)	1 (1)
Capillaria spp. Egg. Helminth.	Faeces	4 (4)	2 (2)	6 (6)
Ascarid. Egg. Helminth.	Faeces	6 (6)	1 (1)	7 (7)
Trypanosoma sp. Protozoan.	Blood	9 (9)	4 (4)	13 (12)
Taenia taeniaformis larvae. Tapeworm.	Liver	4 (4)	5 (5)	9 (9)

N = TOTAL NO

Table 2. Number of male and female urban wild rats (%) infected by ectoparasites. Total number of rats examined was 105.

Types of ectoparasite	Number of rats		
	Male	Female	Total
Radfordia sp.	3 (3)	2 (2)	5 (5)
Polyplax sp.	2 (2)	0 (0)	2 (2)
Hoploplura sp.	1 (1)	0 (0)	1 (1)
Linognathus sp.	0 (0)	1 (1)	1 (1)

gastrointestinal symptoms in humans (Puthia *et al.,* 2006). However, no significant signs or symptoms has been reported in animals particularly rats. Only 10% of the examined rats were positive for *Rodentolepis* which has also been reported previously in Malayan rats by Syed-Arnez *et al.* (2006) and Mohd Zain *et al.* (2008, 2012). *Hymenolepis nana* and *Hymenolepis diminuta* are the two commons species of *Rodentolepis* which can cause a human disease called hymenolepiasis presenting symptoms of restlessness, irritability, diarrhoea and abdominal pain. *Trypanosoma*, *Taenia*, *Ascaris*, *Capillaria*, *Trichuris* and coccidia were also found in the examined rats but with the least number infected.

It was found that very few examined rats were infected with ectoparasites namely, *Radfordia* sp., *Polyplax* sp., *Linognathus* sp., and *Hoplopleura* sp. *Radfordia* is a myobiids mite with no medical and economical significance (Mohd Zain *et al.,* 2015) whereas, *Haploplura* and *Polyplax* are rat louse that can be found on rats in Peninsular Malaysia (Mohd Zain *et al.,* 2015). The anoplura parasitic sucking louse; *Linognathus* sp. was also found on one of the examined rats.

From this study, infections by parasites such as *Nippostrongylus, Strongyloides, Blastocystis, Rodentolepis*as well as ectoparasites on rats may have the potential to cause illnesses to humans as these rats were caught in urban areas where there is high dense population. Coupled with poor sanitation and hygiene, it could be a disastrous situation in an outbreak of disease. The information obtained from this study will help authorities from the city council, health departments and housing area management to evaluate current control methods for pest control and carry out preventive awareness programmes for people residing in these areas to improve on cleanliness. The results of this current study reported the risk that humans face in urban areas where rats as pests harbour dangerous zoonotic pathogens. With this information, authorities will be able to formulate and evaluate better programmes for rat pest control as well as disease transmission to humans and livestock or pet animals. The benefits of this will translate into savings in time and treatment costs for humans as well as productivity of animals as rats as pests contaminate the environment with disease-causing pathogens.

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