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SEROLOGICAL PREVALENCE OF LEPTOSPIRA INFECTION IN MALAYAN PORCUPINES (*HYSTRIX BRACHYURA*) IN CAPTIVITY

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Abstract

Leptospirosis is recognised as one of the important zoonotic diseases in the world including Malaysia and rodents are the major natural reservoir of this disease. A study was conducted to determine the serological prevalence of leptospiral infection among a rodent species, the Malayan Porcupine (*Hystrix brachyura*). A total of 50 serum samples were obtained from the porcupines kept in captivity at the Wildlife Conservation Centre, Sungai Dusun, Selangor. The microscopic agglutination test (MAT) was performed to detect the presence of agglutinating antibodies to a panel of 16 *Leptospira* serovars (Australis, Autumnalis, Ballum, Bataviae, Canicola, Celledoni, Djasiman, Hardjobovis, Hardjo, Hebdomadis, Hurstbridge, Icterohaemorrhagiae, Javanica, Pomona, Pyrogenes and Sejroe). From the serological test, 18% (9/50) of the porcupines had leptospiral antibodies to serovars Javanica (8%), Hurstbridge (4%), Ballum (2%), Celledoni (2%) and Hardjoprajitno (2%). Hence, this study disclosed that leptospiral infection was prevalent among the Malayan Porcupines and could be a potential source of infection to humans.

Keywords: leptospirosis, leptospira, Malayan Porcupines, MAT

INTRODUCTION

Leptospirosis is an acute septicaemic disease of domestic and wild animals as well as humans (zoonosis) that is caused by different serovars within the species *Leptospira interrogans*. Rats are believed to be the major natural reservoir for this bacterium (Bahaman and Ibrahim, 1988). Leptospire can survive for long periods of time in the environment under favourable conditions. Moisture, pH values of the soil and surrounding temperature will influence the survivability of the pathogenic leptospire which can lead to a high incidence of leptospirosis (Gordon, 1977). There are two modes

of transmission for leptospirosis namely by direct or indirect contact. Interactions between the maintenance host, contaminated environment and susceptible animals play a role in the epidemiology of leptospirosis.

Rodents such as rats and mice are known to be one of the most important reservoir hosts for *Leptospira* in the wild. Porcupines are also of rodents and ranked third largest. These mammals are commonly herbivores, nocturnal and terrestrial. The Malayan Porcupine (*Hystrix brachyura*) is one of the wildlife species proposed by the Department of Wildlife and National Parks (DWNP) of Malaysia to be commercialised as a meat product due to its high reproductive performance, large body size and easy management (Zainal, 1988).

Leptospirosis as mentioned earlier has zoonotic potential and a concern towards public health safety issues with this particular type of farming. The status of leptospirosis in the Malayan Porcupines is still not known. A previous study done by Mitchell *et al.* (1966) reported that *Leptospira pomona* was isolated from a porcupine's kidney among 15 species of wildlife trapped from a farm following an outbreak of leptospirosis in the farm studied. This certainly showed that porcupines are no different from rats or any other rodent species which are able to harbour leptospiral organisms and spread the infection.

Thus, this study was conducted to determine the serological prevalence of *Leptospira* infection in the Malayan Porcupines and to identify the leptospiral serovars infecting them. These would provide background information on leptospiral infection in porcupines and would facilitate the understanding of the epidemiology of leptospirosis in Malaysia.

MATERIALS AND METHODS

Animals

Fifty Malayan Porcupines (*Hystrix brachyura*) kept in metal wire pens at the Wildlife Conservation Centre, Sungai Dusun, Selangor, Malaysia were selected for this study. They were made up of 43 adults and 7 juveniles. All animals were judged to be in good health based on their general behaviour and physical conditions. They were fed twice a day with fruits, vegetables, tubers and concentrates.

Blood sampling

The Malayan Porcupines were restrained, anaesthetised, and 3 mL of blood were extracted from the femoral artery into a plain tube using a 23-gauge needle and a 5 mL syringe. Serum samples were kept at -20°C until analysis.

Serological Examination

A microscopic agglutination test (MAT) was performed according to Cole *et al.* (1973) for the detection of agglutinating antibodies against a panel of 16 leptospiral serovars (Australis, Autumnalis, Ballum, Bataviae, Canicola, Celledoni, Djasiman, Hardjobovis, Hardjo, Hebdomadis, Hurstbridge, Icterohaemorrhagiae, Javanica, Pomona, Pyrogenes and Sejroe). The leptospiral cultures (antigens) were adjusted to a cell density of 1.5×10^8 leptospores/mL based on MacFarland standard. Serum positive at titre 1:20 was further titrated until 1:5120. A serum sample was considered positive when more than

50% agglutination was observed compared to the negative control and at a titre of 1:40 or higher. A negative MAT would demonstrate live and active leptospiral antigen with few or without any agglutination.

RESULTS

From the serological test (MAT), out of 50 serum samples, 18% (9/50) were positive to one or more of the five serovars including Javanica, Ballum, Celledoni, Hardjoprajitno and Hurstbridge (Table 1). All the seropositive results were among the adult Malayan Porcupines in which the highest serum titre was found to be 1:40. The highest prevalence of leptospiral infection among the Malayan Porcupines was serovar Javanica at 8% (4/50), followed by Hurstbridge at 4% (2/50) and as for Ballum, Celledoni and Hardjoprajitno was at 2% each (1/50).

DISCUSSION

Based on the results, the serological prevalence of *Leptospira* in the Malayan Porcupines was determined to be 18%. This value is considerably high and comparable to rats. Mohamed-Hassan *et al.* (2010) reported a serological prevalence of 17.9% (30/168) of leptospiral infection in wild rats in Kelantan and Terengganu with 17.3% (14/81) and 18.4% (16/87) in the respective states. Similarly, a previous study by Siti-Aminah (2006) also reported a similar serological prevalence of 18.1% (6/33) of leptospiral infection among wild rats caught from Kuala Lumpur areas and almost 4% (1/27) were from wild rats from the Serdang area.

From this present study, Javanica was the major serovar detected at 8% (4/50) in the serum samples. The same serovar was found in majority of the rats from the study done by Siti-Aminah (2006). The positive serum samples were found only among the adult Malayan Porcupines. No positive MAT was seen in the juvenile porcupines tested, possibly because they have not yet been exposed to *Leptospira*. The serum titre of leptospiral antibodies was found to be 1:40. This low titre may mean the start of antibody production in an early acute infection, or a low level of antibodies persisting long after infection. There is also a possibility that the serovar infecting the Malayan Porcupines was not covered by the serovars in the test antigens.

From the findings, it is suggestive that the Malayan Porcupines could become carriers of leptospiral organisms and thus, farmers as well as other handlers are at risk of contracting leptospirosis from the Malayan Porcupines. Precautions should be taken during handling porcupines or their carcasses in preparation of their meat for human consumption. Farmers and handlers are advised to wear protective clothing such as boots, gloves and masks whenever handling such animals or during cleaning of their cages or pens. It is also important to wash or shower after exposure to urine splashes from these porcupines or from contaminated soil or water.

Table 1. The prevalence rate (%) of *Leptospira* infection in the 50 Malayan Porcupines

Serovars	No. of Positives	Prevalence Rate (%)
Javanica	4	8.0
Ballum	1	2.0
Celledoni	1	2.0
Hardjoprajitno	1	2.0
Hurstbridge	2	4.0
Total Positives	9	18.0
Total Serum Samples		50

From this study, the Malayan Porcupines are possibly carriers of leptospiral organisms which is expected as porcupines are of rodent species, similar to rats. To date, no study has been carried out on porcupines to implicate them as the source of leptospirosis to humans in Malaysia. Thus, further research is required to look into its association with human leptospirosis. Exposure of porcupine farmers and handlers to urine-contaminated environment put them at risk at contracting leptospirosis.

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