PREVALENCE OF *Salmonella* SP. IN WILD RATS IN KELANTAN

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ABSTRACT. *Salmonella* is known as one of the important food-borne pathogen that causes salmonellosis in human and animals worldwide. A prevalence study on salmonellosis was conducted on wild rats in Kelantan. From April to June 2015, a total of 36 rats and house shrews sent to the Regional Veterinary Laboratory in Kota Bharu, Kelantan were examined for the presence of *Salmonella*. These animals were caught from various locations in the state and were sent to the laboratory either as live or dead specimens. Post mortem was conducted and intestines were taken for detection of salmonellosis. Results showed that of the 32 rats and 4 shrews examined, 5 (15.6%) rats and 3 (75%) shrews were found positive and on serotyping four serotypes of *Salmonella* were identified which are *Salmonella* ser. Kalamu (62.5%), *Salmonella* ser. Thyphimurium (12.5%), *Salmonella* ser. Weltevreden (12.5%) and *Salmonella* ser. Brancaster (12.5%). In conclusion, positive identification of *Salmonella* in wild rats indicates that there is a possible transmission of the pathogen to humans due to constant contact between the two. Thus appropriate measures are needed to control these pests population to prevent spread of diseases to the humans and animals.

Keywords: *Salmonella*, wild rats, Kelantan

INTRODUCTION

Salmonellosis, caused by the gram negative bacteria *Salmonella,* is one of the major foodborne diseases that infects both human and animals and had caused some 93.8 million cases of gastroenteritis and 155,000 deaths in humans around the world every year (Majowicz *et al.,* 2010). Salmonellosis occurs via ingestion of food contaminated by the bacteria, which may be transmitted by feces of the infected animals. Animals that carry *Salmonella* play an important role in the spread of salmonellosis (Phan *et al.,* 2005). Wild rodents not only cause damage and spoilage of feed, they are also known to be the reservoir and vector of a number of agents that cause disease in animals and humans including *Salmonella* spp. (Meerburg and Kijlstra, 2007). These mammals carry the bacteria in their intestinal tracts, mostly without showing

any clinical signs, and later transmit the bacteria to other animals or to human (Meerburg and Kjilstra, 2007).

Not much study on salmonellosis in rats was done in Malaysia. Joseph *et al.* (1984) had conducted a study on the occurrence of *Salmonella* in rats and house shrews in Ipoh, Perak from July 1978 to December 1979. The study found that of the 55 shrews and 8 rats examined, 39 (71%) shrews and 2 (25%) rats were found positive of *Salmonella* and the serotypes obtained were *S*. Weltevreden, *S*. Bareilly, *S*. Stanley, *S*. Augustenborg, *S*. Hvittingfoss, *S*. Emek, *S*. Paratyphi B, *S*. Ohio and *S*. Matopeni in order of frequency of isolation. A surveillance study on animal *Salmonella* in Peninsular Malaysia by the same authors on 1981 to 1985 had isolated *S*. Weltevreden from house rats (*Rattus rattus diardii*).

No specific study on *Salmonella* in rats in Kelantan had been recorded so far. Thus, this study was conducted to determine the prevalence of *Salmonella* sp. in wild rats in the state of Kelantan.

MATERIALS AND METHOD

Trapping of Rats

Trapping of rats was conducted using wire traps by Pejabat Kesihatan Negeri Kelantan at locations where leptospirosis cases were reported which included Tumpat, Kota Bharu, Pasir Puteh, Tanah Merah, Gua Musang dan Machang.

Collection and Processing of Samples

This study was conducted from April to June 2015. All rats and house shrews sent to the Regional Veterinary Laboratory in Kota Bharu, Kelantan for detection of leptospirosis were examined for the presence of *Salmonella*. Live rats and shrews were euthanised with cotton wool soaked with chloroform and post mortem was conducted. Intestines were collected and incubated in buffered peptone water (BPW) at 37 °C for 24 hours (pre-enrichment). Then 0.1 ml of the cultured BPW was transferred into Rappaport-Vassiliadis broth (enrichment media) an incubated for another 24 hours at 42 °C. Later, loopfuls of the cultured broth were spread on brilliant green agar (BGA) and xylose lysine deoxycholate agar (XLD) and incubated at 37 °C for 24 hours. If typical Salmonella-like colonies appear, which shown as pink colonies on BGA (Figure 1) and blackish colonies on XLD agar (Figure 2), subcultures were done on BGA and XLD to get the pure colonies before identification using biochemical tests, which included TSI, Indole and Urea. If the results of biochemical tests showed typical characteristics of *Salmonella*, further confirmation test with Polyvalent O and Polyvalent H was performed (Figure 3). Samples with positive *Salmonella* were then cultured onto nutrient agar and sent to Veterinary Research Institute (VRI) for serotyping.

Figure 1. Colonies appear pink on BGA agar

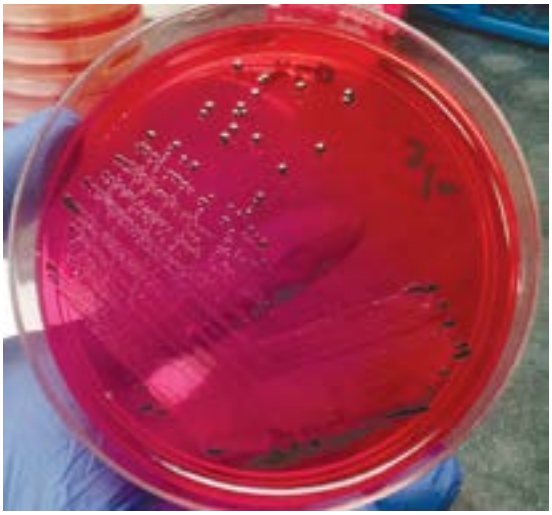

Figure 2. Colonies appear blackish on XLD agar

RESULTS

A total of 32 rats and 4 shrews were examined for salmonellosis. Eight out of 36 (22.2%) animals were positive for *Salmonella*. Of the 32 rats and 4 shrews examined, 5 (15.6%) rats and 3 (75%) shrews were found positive. *Salmonella* was isolated from three districts which were Tumpat (prevalence rate = 50%), Kota Bharu (prevalence rate = 40%) and Pasir Puteh (prevalence rate = 7.14%). Tumpat showed the highest *Salmonella* isolation rates (50%) (Table 1).

Four serotypes were identified from the isolates which were *Salmonella* ser. Kalamu (62.5%), *Salmonella* ser. Thyphimurium (12.5%), *Salmonella* ser. Weltevreden (12.5%) and *Salmonella* ser. Brancaster (12.5%) (Table 2).

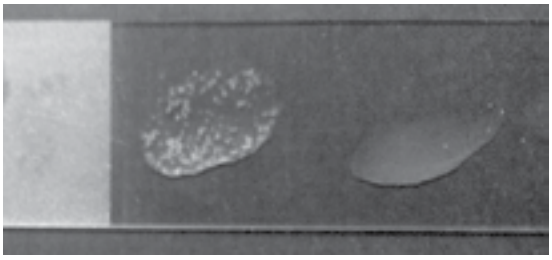

Figure 3. Confirmation test by Polyvalent O and Polyvalent H: positive Polyvalent O (left) and negative Polyvalent H (right)

Table 1. Prevalence of *Salmonella* in wild rats in Kelantan.

Location (District)	No. of *Salmonella*-positive samples/No. of samples examined (%)
Tumpat	5/10 (50%)
Kota Bharu	2/5 (40%)
Pasir Puteh	1/14 (7.14%)
Tanah Merah	0/5 (0%)
Machang	0/1 (0%)
Gua Musang	0/1 (0%)
Total	**8/36 (22.2%)**

Table 2. Serotypes of *Salmonella* isolates in wild rats in Kelantan

Serotypes	No. of isolate (%)
S. Kalamu	5 (62.5%)
S. Typhimurium	1 (12.5%)
S. Weltevreden	1 (12.5%)
S. Brancaster	1 (12.5%)

DISCUSSION

Salmonella in rats and shrews was isolated at considerably high rate in Kelantan with prevalence rate of 22.2%, compared to other reports found in other countries such as 6.0% in France (Seguin *et al*., 1985), 32% in Nigeria (Oboegbulem and Okoronkwo, 1990), 16.2% in the USA (Henzler *et al*., 1992), 19.3% in Vietnam (Phan *et al*., 2003), 10.0% in the UK (Hilton *et al*., 2004), 28.7% in Japan (Lapuz *et al*., 2007) and 2.0% in Trinidad and Tobago (Nkogwe *et* al., 2011). Besides, the fact that *Salmonella* was detected from rats in three out of six districts tested suggests that the pathogen is widely prevalent in wild rats in Kelantan. This study also showed that from 32 rats and 4 shrews examined, 5 (15.6%) rats and 3 (75%) shrews were found positive. Compared to the rats, almost all of the house shrews examined were *Salmonella*-positive, which was the same as the study conducted in Ipoh, Malaysia by Joseph *et al.* (1984) where 39 out of 55 shrews (prevalence rate of 71%) and only 2 out of 8 (25%) rats were positive for *Salmonella*.

From this study, four serotypes of *Salmonella* were obtained. All of these serotypes are of *Salmonella enterica* species. *S.* Kalamu is the most serotype that infects both shrews and rats. There is not much information about this serotype on rats, but in animal, *S.* Kalamu was the fourth serotype mostly isolated in the study of salmonellosis in stray dogs in Central Taiwan (Chang *et al*., 2011). The other three serotypes (*S.* Typhimurium, *S.* Weltevreden and *S.* Brancaster) isolated from this study were similar to the serotypes found in humans and other food animals. According to Global Monitoring of *Salmonella* Serovar Distribution by the World Health Organization (WHO) that collected data on human salmonellosis from 2001 to 2007, *S.* Weltevreden and *S.* Typhimurium were listed as the most frequent serotypes that cause human salmonellosis in Malaysia (Hendriksen *et al.,* 2001). Several cases of rodent-borne human salmonellosis by *S.* Typhimurium had also been reported (Swanson *et al*., 2007). *S.* Typhimurium and *S.* Weltevreden were also found in the poultry farm (Ong *et al*., 2014), clinical livestock samples (Thenamutha *et al*., 20013, Hanani *et al*., 2014), poultry and meat products (Thong *et al*., 2002, Roseliza *et al*., 2011, Marina *et al*., 2013). Outbreak of human salmonellosis by *S.* Brancaster had been reported in London (Hinden *et al*., 1952 and Cardinale *et al.,* 2006).

CONCLUSION

In conclusion, positive identification of *Salmonella* in wild rats indicates that the rodents may play an important role as the reservoir for the pathogen and cause possible transmission of the pathogen to human due to constant contact between the two. Thus appropriate measures are needed to control these pests population to prevent spread of diseases to the humans and animals.

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