# OCCURRENCE AND INTENSITY OF TICK INFESTATION ON STRAY DOGS IN KELANTAN AND SELANGOR

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## CERTIFICATION

This is to certify that we have read this research paper entitled '**Occurrence and intensity of tick infestation on stray dogs in Kelantan and Selangor**' by Lye Yi Yan, and in our opinion it is satisfactory in terms of scope, quality and presentation as partial fulfillment of the requirement for the course DVT 5436 – Research Project.

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My mother

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**Thank You**

## DEDICATIONS

I dedicate my dissertation work to my family and friends. A special thanks to my mother, Mei Yoong, who always accompanied my side and gave words of encouragement to me. Aside from her, I would like to express my gratitude to my childhood friend, Xiangjun who was always supporting me throughout the process of my Final Year Project.

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## ABSTRACT

An abstract of the research paper presented to the Faculty of Veterinary Medicine, Universiti Malaysia Kelantan, in partial requirement on the course DVT 5436 – Research Project

The close relationship of dogs with humans has raised concerns about the spread of potential zoonotic diseases that can be transmitted through ectoparasites such as ticks. Hence, the occurrence and intensity of tick infestation on stray dogs with risk factors of age, sex, neutering status, state, and body section of stray dogs were examined because of their high possibility to be in contact with human, or in contact with house dogs that have closer associations with humans thus facilitate the transmission of zoonotic disease to human via ticks. It was important to determine the predilection sites of the ticks in this study to develop a strategic tick control program for dogs to prevent or reduce tick-borne disease in Malaysia. A total of 64 stray dogs from Kelantan and Selangor were examined via integumentary examination and a total of 431 ticks, constituting *Rhipicephalus sanguineous* and *Haemaphysalis sp.* were collected. The overall occurrence of tick infestation was 82.81% (53/64) and the intensity of tick infestation from stray dogs examined was 8.13 ticks (1 – 17 ticks) per stray dog. Risk factors such as neutering status and states were associated with tick infestation in this study. The most preferred site for ticks attachment were the head, ear, and neck. In conclusion, this study revealed the high occurrence and tick intensity on stray dogs in Malaysia which highlights the importance of implementing tick control programs on stray dogs that can serve as the potential risk for public health on tick-borne diseases.

***Keywords:*** *Ixodidae, Occurrence, Tick intensity, Tick infestation, Risk factors, Stray Dogs*

## ABSTRAK

Abstrak daripada kertas penyelidikan dikemukakan kepada Fakulti Perubatan Veterinar, Universiti Malaysia Kelantan untuk memenuhi sebahagian daripada keperluan kursus DVT 5436 – Projek Penyelidikan.

Hubungan rapat antara anjing dengan manusia telah menimbulkan kebimbangan dalam kalangan masyarakat mengenai penyakit zoonosis yang boleh disebarkan melalui ectoparasit seperti kutu. Oleh itu, kajian ini direka untuk menentukan kelaziman dan intensiti kutu pada anjing liar yang berkeliaran di Selangor dan Kelantan, dengan menentukan faktor risiko seperti umur, jantina, status kemandulan, negeri dan bahagian badan anjing liar. Anjing liar mempunyai hubungan yang rapat dengan manusia, ataupun rapat dengan anjing yang dipelihara oleh manusia dan hal ini memudahkan penularan penyakit zoonosis kepada manusia melalui kutu. Tapak kecenderungan kutu hendaklah ditentukan oleh sebab kepentinganya untuk membangunkan program kawalan kutu yang strategik bagi mencegah atau mengurangkan penyakit bawaan sengkenit di Malaysia. Sebanyak 64 ekor anjing liar dari Selangor dan Kelantan telah diperiksa dan mempunyai sebanyak 431 ekor kutu yang mengandungi jenis kutu seperti Rhipicephalus sanguineus dan Haemaphysalis sp.. Hasilnya, kelaziman kutu pada anjing liar adalah 82.81% manakala intensiti kutu adalah 8.13 kutu (1-17 kutu) bagi setiap anjing liar. Status kemandulan dan negeri adalah faktor risiko yang paling signifikan mengenai kelaziman dan intensiti kutu pada anjing liar. Tapak yang paling disukai oleh kutu ialah kepala, telinga dan leher anjing liar. Kesimpulannya, kajian ini mendedahkan kelaziman dan intensiti kutu pada anjing liar adalah tinggi dan hal ini menonjolkan kepentingan untuk melaksanakan program kawalan kutu atas anjing liar yang berpotensi dijangkiti penyakit bawaan sengkenit.

***Kata kunci:*** *Ixodidae, Occurrence, Tick intensity, Tick infestation, Risk factors, Stray Dogs*

# 1.0 Introduction

According to the World Health Organization (WHO), the total population of stray dogs makes up about 75% to 85% of the estimation of 200 million stray dogs worldwide (WHO, 2013). The presence of uncontrolled stray dogs nowadays has emerged as a public health problem in Malaysia (Ahmad et al., 2021). Due to the living conditions of the stray dogs that lack proper care and treatment, the stray dogs are always facing high mortality due to several factors such as malnutrition, starvation, and zoonotic diseases (Beck 2000; Reece 2005). Stray dogs can be regularly found anywhere in all human populations, or in contact with house dogs that have closer associations with humans. Hence, the close relationship of dogs with humans has raised concerns about the spread of potential zoonotic diseases that can be transmitted through ectoparasites such as ticks (Otranto et al., 2009).

Ticks are blood-sucking ectoparasites for most terrestrial vertebrates. It raises public health concerns due to its role of being a vector of various tick-borne pathogens, and it can result in life-threatening health issues to its main host, dogs (Liu et al. 2013; Low et al. 2017). There have been reports proving that high tick prevalence and intensity will lead to a high disease rate (Prakash et al, 2018). Among Southeast Asia countries, the Philippines had an overall prevalence of tick infestation rate of 36.29% in dogs (Galay et al., 2018). Besides that, the prevalence rate of tick infestation on stray dogs in Thailand was over 90%, whereby most of the ticks were morphologically identified as *Rhipicephalus sanguineus* (Thom et al., 2021). In another study that is conducted in Indonesia, the prevalence of dogs infested by ticks was 67.90%

(Pratomo, 2014). The result was considered similar to another study conducted in Malaysia, whereby the prevalence rate of tick infestation on stray dogs was 78.18% (Sipin et al., 2020). In an aspect of intensity of tick infestation in Philippines, the overall mean intensity of infestation in one dog is 25.75% out of the 97 dogs sampled for the study, and the toes (6.85%) were the most preferred attachment site (Bartolome-Cruz, 2018).

Ticks are arthropod vectors of many infectious diseases to animals and humans, especially the known tick-borne diseases such as Babesiosis, Anaplasmosis, and Ehrlichiosis in Malaysia (Jing et al., 2017). Thus, the purpose of the present study was to safeguard public health by investigating the occurrence and intensity of tick infestation on stray dogs, thereby avoiding explosive outbreaks that could result in difficulties in disease transmission control.

## 2.0 Research problem

Various studies have been done on ticks detection of tick-borne diseases in Malaysia. To the best of our knowledge, however, efforts into investigating the occurrence and intensity of tick infestation on stray dogs in Malaysia were limited. The knowledge of determining the occurrence and intensity of tick infestation among stray dogs is a prerequisite for the prevention of tick-borne disease (Shimada et al., 2003) to develop a strategic tick control program against tick infestation in stray dogs in Malaysia.

## 3.0 Research questions

3.1 What is the occurrence of tick infestation in stray dogs?

3.2 What is the tick infestation intensity in stray dogs?

3.3 What are the risk factors for tick infestation in stray dogs?

## 4.0 Research hypothesis

4.1 The overall occurrence of tick infestation in stray dogs is more than 80%.

4.2 The overall tick infestation intensity in one stray dog is more than 25.75%.

4.3 The risk factors for tick infestation in stray dogs are the ages, sex, neutering status, and states.

## 5.0 Objectives

5.1 To determine the occurrence of tick infestation in stray dogs.

5.2 To determine the tick infestation intensity in stray dogs (map preference body section)

5.3 To determine the risk factors (age, sex, neutering status, status) of tick infestation in stray dogs

# 6.0 Literature review

## 6.1 Public Health Concerns of Stray Dogs

According to World Organisation for Animal Health (OIE), stray dogs were free-roaming dogs that were not under the direct control or restriction of the owner (Otranto et al., 2017). As a result of the current situation such as the increasing growth of the human population, absence of responsible dog ownership policies, poor waste management, as well as lacking awareness of animal welfare and disease issues in the human society, increased attention is being given to the problem of the stray dogs (Jackman and Rowan, 2007).

Stray dogs can be regularly found anywhere in all human populations, and they can cause public health concerns in a community such as zoonotic disease transmission, inflicting wounds on people, as well as noise pollution (Saree et al., 2021). Dogs are social animals, by being in contact with housed dogs which have closer associations with humans, the disease can be transmitted from the stray dog to humans via the pet dog. Rabies (Rahaman, 2017) and Echinococcosis (Avcioglu, 2021) are the most common diseases of stray dogs that possess a potential danger to humans, but tick-borne diseases such as Babesiosis (Vishwakarma et al., 2019), Anaplasmosis, Ehrlichiosis (Ismail et al., 2010) and so forth also is not an uncommon matter among human.

Ectoparasite infestation is very common in stray dogs worldwide, with ticks ranked for the second greatest percentage of infestation in stray dogs compared with other ectoparasites (Lefkaditis et al., 2016). In addition, dogs are known to be infested with ticks at different degrees depending on the potential host

risk factors including the dog's age, sex, neuter status, body weight, and breed (Raghavan et al., 2007). Thus by having close contact, ticks from stray dogs can be easily transmitted to the house dog and humans.

## 6.2 Common ticks and tick-borne diseases in dogs

In Malaysia, the most common tick found in stray dogs was *Rhipicephalus sanguineus* (98.36%), followed by *Haemaphysalis sp*. (1.64%) (Anurddin et al., 2010). Ticks are arthropod vectors of disease that can transmit an extensive range of viral, bacterial, and protozoan pathogens to vertebrate hosts (Otranto et al., 2009). Examples of tick-borne diseases that have been reported in Malaysia were Ehrlichiosis caused by *Ehrlichia canis,* anaplasmosis caused by *Anaplasma platys*, as well as Babesiosis caused by *Babesia gibsoni* and *Babesia vogeli*. (Sipin et al., 2020).

When a dog is infested with ticks frequently, it will bear a relatively high risk of tick-borne infection (Eng et al., 1988; Day, 2011). For instance, according to Prakash, a high infestation of *R. sanguineus* leads to the high occurrence of canine babesiosis. The same ticks may transmit infectious disease-causing agents to both non-human and human species at the same time. To illustrate, the American dog tick and the wood tick are considered the major vectors of Rocky Mountain spotted fever caused by *Rickettsia rickettsii* in both humans and dogs. (Dryden and Payne 2004).

## 6.3 Occurrence and intensity of tick infestation

In recent years, we have seen a significant increase in interest in ticks and tick-borne diseases both from a medical point of view and among the public, highlighting the importance of the 'One health' approach to the problem (Theo et al., 2020). Thus, data on the occurrence and intensity of tick infestation on stray dogs need to be determined to solve the problem.

The occurrence of tick infestation on stray dogs indicates the proportion of stray dogs that are having tick infestation in a given period. For instance, in Thailand, all the ticks collected from dogs in the study were *Rhipicephalus sanguineus*, which has been reported as the most common tick species infesting dogs in Thailand with a prevalence of over 90% (Do et al., 2021). Besides that, the prevalence of tick infestation among dogs in Malaysia was 78.18%. In another study that is conducted in Indonesia, the prevalence of dogs infested by ticks was 67.90% (Pratomo, 2014). In general, the prevalence rate of tick infestation on dogs in Southeast Asia countries such as Philippines, Indonesia, Thailand, and Malaysia all fall within the range of 26% to 90% with similar tropical climates respectively.

The intensity of tick infestation on stray dogs indicates the measurable amount of tick infestation on stray dogs. According to the study conducted in the Philippines by Kathlyn, the overall mean intensity of infestation in one dog is 25.75% out of 97 dogs sampled, while the toes (6.85%) were the most

preferred attachment site. The occurrence and intensity of tick infestation on stray dogs all depended on tick risk factors including environmental factors such as ambient air temperature and relative humidity (Sahu et al., 2013), as well as the sex and developmental stage of ticks (Dantas-Torres, 2010)

## 7.0 Materials and methods

### 7.1 Study area

The study was conducted in Subang, Selangor, and Wakaf Bharu, Kelantan. Subang is a township located in Petaling Province, Selangor, Malaysia. It is located about 17km west of downtown Kuala Lumpur at a latitude of 3.1282° N and a longitude of 101.5523° E. For Wakaf Bharu, it is a satellite town in Tumpat district, northern Kelantan, Malaysia, lying at a latitude of 6.1189° N and a longitude of 102.1987° E. These two places were chosen as they are surrounded by a high number of stray dogs which possess the risk of zoonotic diseases to the public. The samples were collected randomly in areas occupied by humans such as restaurants, housing, and factory areas in Selangor, as well as temples, housing, and roadside area in Kelantan.

### 7.2 Stray dogs sampling

The ticks were collected from the stray dogs in Selangor and Kelantan, Malaysia to detect the presence of ticks. A convenient sampling method was adopted. The stray dogs were sampled randomly based on manageable handling and information regarding age, sex, and neutering status of stray dogs were collected. With respect to age, the stray dogs were categorized into young

dogs (less than 6 months) and adults (more than 6 months) based on the state of their teeth.

Firstly, the stray dog was restrained properly by the restrainer. The stray dogs were restrained either in a sitting position or standing position. To restrain the stray dog in a sitting position, one arm was placed under the stray dog's neck so that the forearm holds the head securely against the restrainer's body. Next, the other arm was placed around the hindquarters to prevent the dog from standing or lying down during the procedure. To restrain the stray dog in a standing position, one arm was placed under the dog's neck so that the forearm holds the dog's head securely. The head was positioned so that the dog cannot bite. The other arm was placed under the abdomen to prevent the dog from sitting or lying down during the procedure of tick collection.

### 7.3 Tick collection

In this research, a 5-body section method for sampling ticks on stray dogs was applied. The sections were divided into head, ears, and neck, dorsal, abdomen, axillary and inguinal, legs and feet, as well as tail and perianal. The 5 sections are illustrated in Figure 1.

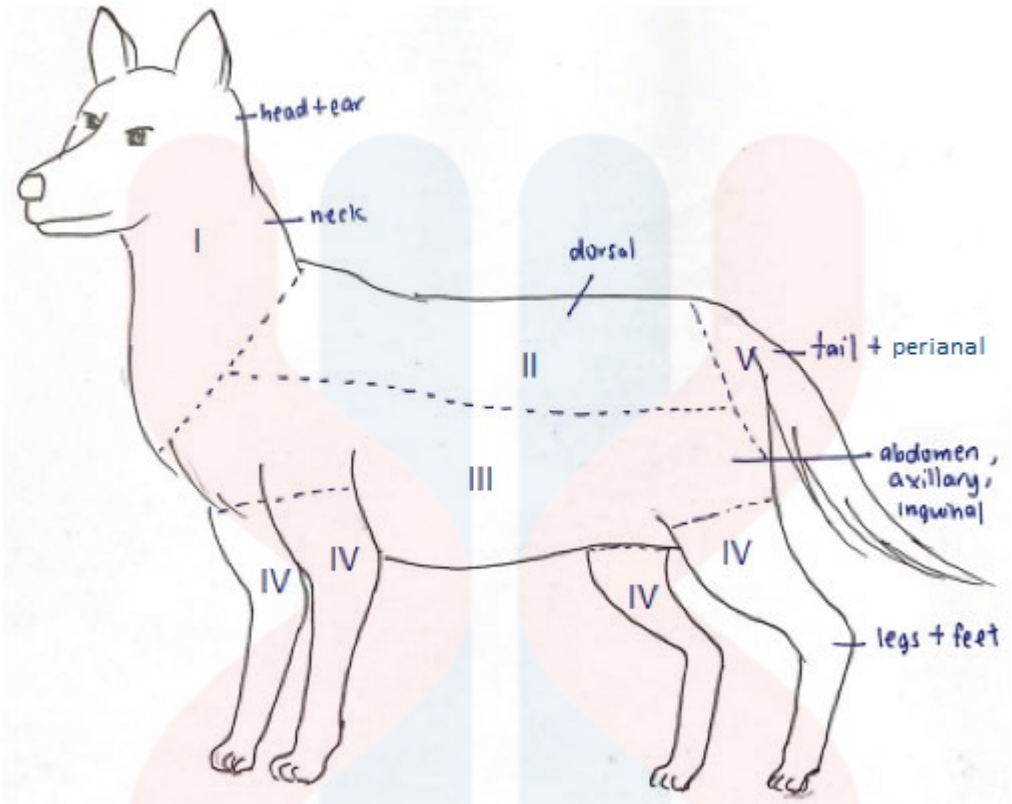



***Figure 1: 5-body section on a stray dog***

The method of Poh (2020) was modified and used in this study. Firstly, the entire process of collecting ticks on the infested dog was 125 seconds, with each section being checked for a period of 20 to 25 seconds. All of the ticks were collected carefully from each infested stray dog. The time was kept with a stopwatch and as many ticks are removed as possible with forceps and a flea comb. After the removal of ticks, the ticks were all placed in a hard-plastic container filled with 70% ethanol and closed with a tightly fitting lid.

## 7.4 Tick identification

The ticks were then brought to the Parasitology Laboratory, Faculty of Veterinary Medicine, UMK to be examined under a dissecting microscope to

determine the species and stages (female, male, nymph, larva) of each tick collected. Ticks were identified up to the genus level based on the keys (Walker et al., 2003).

### 7.5 Tick Occurrence, Risk Factors, and Tick Infestation Intensity

The occurrence of tick infestation was calculated based on the number of stray dogs infested by ticks with the aspect of age, sex, neutering status and states as the identified risk factors. The mean intensity of tick infestation in this study was calculated by dividing the number of total ticks collected by the total number of infested dogs.

### 7.6 Data management and statistical analyses

Data management was conducted using Microsoft Excel® and all recorded data were analyzed using Statistical Package for Social Science (SPSS®) version 26. Descriptive statistics, percentages, and 95% confidence interval were used to summarize the proportion of infested and non-infested animals. The association with different risk factors on the occurrence and distribution of ectoparasites was analyzed using the Chi-square test. The differences were considered as significant when at 95% confidence intervals while statistical significance was set at $p < 0.05$. Moreover, ANOVA test was used to make a comparison on the difference between the 5 body sections of tick infestation on stray dogs.

## 8.0 Results

A total of 431 ticks are collected from 64 stray dogs, with 44 from Selangor and 20 from Kelantan respectively. Among the 64 stray dogs, the overall mean intensity of tick infestation was 8.13 ticks per dog, ranging from 1 to 17 tick infestations per stray dog. The stray dogs in Selangor and Kelantan were identified to be most frequently infested with *Rhipicephalus sanguineous* (430/431; 99.77%) (Figure 2A), as well as *Haemaphysalis* spp. (1/431; 0.23%) (Figure 2B). The stages of the ticks were mostly mature (380/431; 88.17%) and nymph (51/431; 11.83%). No larvae were collected in this study. Among the 64 stray dogs, there were 3 stray dogs co-infested with fleas, *Ctenocephalides canis.*


***Figure 2:(A) Male Rhipicephalus sanguineus. (B) Male Haemaphysalis sp. was collected from the stray dogs.***

***Figure 5: Female Ctenocephalides canis was detected from the stray dogs***

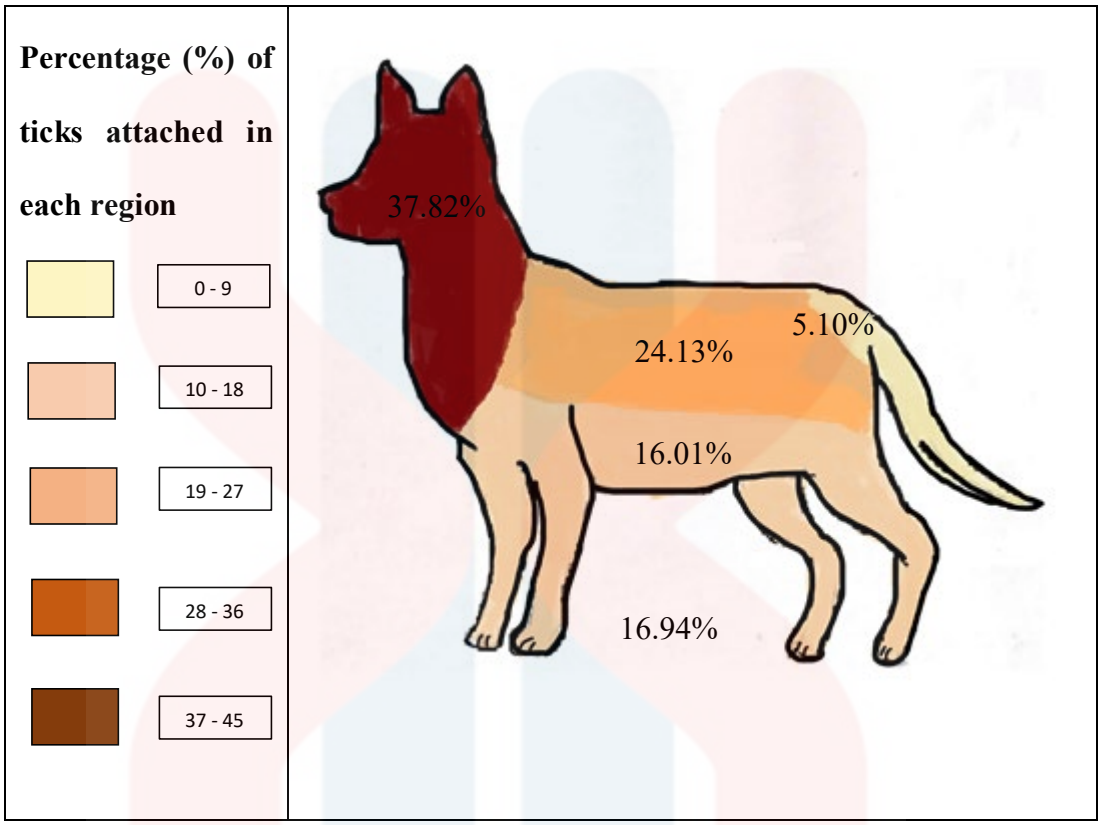
The overall occurrence of tick infestation from Selangor and Kelantan was 82.81% (53/64), as shown in Table 1. In Selangor, there are 33 stray dogs out of 44 stray dogs that were infested by ticks, indicating an occurrence tick infestation rate of 75% (33/44) of stray dogs in Selangor. As for Kelantan, the occurrence of tick infestation in stray dogs was 100%, whereby the encountered stray dogs in Kelantan were all infested by ticks (20/20).

*Table 1: Occurrence of ticks infestation on stray dogs in Selangor and Kelantan*

| States | Stray dogs with ticks | Stray dogs without ticks | Total stray dogs sampled | Occurrence rate (%) |
|---|---|---|---|---|
| Selangor | 33 | 11 | 44 | 75 |
| Kelantan | 20 | 0 | 20 | 100 |
| Total | 53 | 11 | 64 | 82.81 |

Based on the present study, a 5-section method for sampling ticks on stray dogs was applied. The section was divided into head, ears, and neck, dorsal, abdomen, axillary and inguinal, legs and feet, as well as tail and perianal. From the data collected, ticks were more attached at sites of the head, ear, and neck, which accounted for 37.82% compared to other sites as shown below. This is followed by dorsal (24.13%), legs and feet (16.94%), abdomen, axillary and inguinal (16.01%), and finally tail and perianal (5.10%) (Figure 6).

*Figure 4: Number of ticks (percentage) attached to different body sections on stray dogs and tick infestation intensity in stray dogs*




Various risk factors were examined for their association with apparent tick infestation on stray dogs. From a total of 64 individuals of stray dogs examined, Table 2 shows the association of risk factors (age, sex, neutering status, states) with tick infestation in this study by using Chi-square test. The Chi-square test of independence showed that there was a significant association between neutering status and tick infestation with a P-value of 0.032, indicating the occurrence of tick infestation is higher in neutered dogs rather than in unneutered stray dogs. In addition, the Chi-square test of independence also showed that there was a significant association between the states and tick infestation with a P-value of 0.013, whereby stray dogs in Kelantan were more likely to be infested with ticks. However, the Chi-square test of independence showed that there was no significant association between the age of stray dogs and tick

infestation with a P-value of 0.848. The same result goes for the other risk factors, whereby the Chi-square test of independence indicated that there was no significant association between the sex of stray dogs and tick infestation with a P-value of 0.752.

***Table 2: The Chi-square test to determine the association of risk factors with tick infestation in this study.***

| Risk factors | With ticks | Without ticks | Chi-square | P-value | Significance |
|---|---|---|---|---|---|
| < 6 months | 13 | 3 | 0.04 | .848 | No |
| > 6 months | 40 | 8 | | | |
| Male | 31 | 7 | 0.10 | .752 | No |
| Female | 22 | 4 | | | |
| Neutered | 12 | 6 | 4.59 | .032 | Yes |
| Unneutered | 41 | 5 | | | |
| Selangor | 33 | 11 | 6.04 | .013 | Yes |
| Kelantan | 20 | 0 | | | |

A Chi-square test was also done to determine the association of body parts with tick infestation as shown in Table 4. The Chi-square test of independence showed that there was a significant association between head, ear, and neck, as well as tail and perianal with tick infestation with a P-value of 0.014. Thus, different body parts of the dogs have a significant association with the tick infestation intensity.

*Table 3: The Chi-square test to determine the association of body sections with tick infestation in this study.*

| Body sections | With ticks | Without ticks | Chi-square | P-value | Significance |
|---|---|---|---|---|---|
| Head, Ear, Neck | 41 | 12 | 6.04 | .014 | Yes |
| Dorsal | 36 | 17 | | | |
| Abdomen, Axillary, Inguinal | 27 | 26 | | | |
| Legs and Feet | 28 | 25 | | | |
| Tail and Perianal | 13 | 40 | | | |

A one-way ANOVA was conducted to compare the effects of tick infestation intensity between the body sections of stray dogs. As a result, there was a significant difference in terms of tick infestation intensity at different body parts with a P-value of $< 0.001$. The result determines that body sections such as head, ear, and neck, as well as tail and perianal were the significant body parts that influence the tick infestation on stray dogs in this study. The predilection site for tick infestation on stray dogs were the head, ear, and neck whereby the least predilection site for tick infestation on stray dogs was tail and perianal area.

***Table 4: ANOVA test to determine the association of body sections of stray dogs with tick infestation in this study***

| Body sections | Post Hoc |
|---|---|
| Head, Ear, Neck | $3.08 \pm 0.38^{a}$ |
| Dorsal | $1.96 \pm 0.26^{b}$ |
| Abdomen, Axillary, Inguinal | $1.30 \pm 0.24^{bc}$ |
| Legs and Feet | $1.38 \pm 0.24^{bc}$ |
| Tail and Perianal | $0.42 \pm 0.12^{c}$ |

***Mean in the column with similar alphabets' are not significantly difference at p <0.05***

## 9.0 Discussion

This study reported the occurrence and intensity of tick infestation on stray dogs in Selangor and Kelantan. A total of 64 individuals of stray dogs were checked thoroughly for their tick infestation in Selangor and Kelantan. The data collected showed that the stray dogs were mostly infested with ticks, with an occurrence rate of 82.81%. Among the ASEAN countries, the occurrence rate was similar to Thailand, which were having a tick prevalence of over 90% (Do et al., 2021), as well as another study conducted in Malaysia, which was having a tick prevalence of 78.18%. (Galey et al.,2020). However, the occurrence was higher in Indonesia, which was having tick prevalence of 67.90% (Pratomo, 2014), and Philippines which was having tick prevalence of 36.29%. The prevalence of tick infestation on dogs in the study conducted in Indonesia and Philippines were much lower compared to the present study as they are sampling pet dogs instead of stray dogs. Pet dogs were having lesser tick infestation as they were living in a cleaner environment and the ticks were removed manually by the owner or through a tick bath in the clinic (Odeniran et al., 2021). For the intensity of tick infestation on stray dogs in Selangor and Kelantan, it was 8.13 ticks per stray dog, among the 64 stray dogs samples. This was relatively lower than the Philippines, which was 25.75 ticks per dog, out of 97 dogs sampled (Bartolome-Cruz, 2018). The data on intensity of tick infestation was different for each study probably because of the difference in total time used to collect ticks, whereby the total time allocated to examine and collect ticks from different tick attachment sites in a study conducted in Philippines was much longer (10 – 15 minutes) which induces the differences in data collected. The reason behind the reduced total time allocated in the entire process of collecting ticks on infested dogs in the present study was due to

the aggressive behavior exhibited by stray dogs. Hence, it was necessary to reduce the time allocated in handling the stray dogs to reduce the stress exerted on them.

Among 64 individual stray dogs, adult tick-infested stray dogs (40/48; 83.33%) were greater than young tick-infested stray dogs (13/16; 81.25%). However, there were no significant association between the different ages in this study. It could be attributable to the allocation of a broad range of adult age (more than 6 months), resulting in more adult tick-infested stray dogs compared to younger tick-infested stray dogs. This is in contrast with the results done in other studies (Mosallanejad et al., 2012; Lefkaditis et al., 2016; Sahu et al., 2013), where younger animals were more susceptible to tick infestation due to lacking acquired immunity among puppies. Thus, more research with precise age allocation (less than 6 months old for puppies, 6-12 months old for adolescents, more than 12 months old for adults) need to be done to prove the result in the present study. The same goes to the other risk factor such as sex, whereby there was also no significant association between both sexes even though female stray dogs (22/26; 84.62%) was greater than male tick-infested stray dogs (31/38; 81.58%). According to Aldemir, female stray dogs were more susceptible to tick infestation because they always tend to stay in certain areas while nursing their offspring, thereby undergoing frequent re-infestations, especially in a heavily tick-infested area. (Aldemir, 2007).

On the other hand, there were significant association between neutered stray dogs (12/18; 66.67%) and unneutered stray dogs (41/46; 89.13%). This may be related to the mating behavior of dogs. According to McGreevy, neutered dogs tend to wander

less in search of mating opportunities (McGreevy et al., 2018), and this will reduce the probability of tick infestation from the other dogs. Between Selangor (33/44; 75%) and Kelantan (20/20; 100%), Pearson Chi-Square shows that there are significant association between both states. Kelantan was having more tick-infested stray dogs as Kelantan is more to rural areas. According to Smith, the tick infestation in a rural area would be much higher and evenly spread. This is because the surroundings are much more homogenous and the possibility of the dogs being infested by ticks will be higher (Smith et al., 2011). Instead, Selangor is more to urban areas. The tick infestation is much lower and aggregated in certain areas such as parks and gardens that create a hotspot to facilitate the tick infestation on stray dogs in urban areas (Smith et al., 2011). The examples of hotspots in the present study would be restaurant and factory areas, whereby there were more tick-infested stray dogs compared to other areas.

There was a significant association between the 5 different body sections. Out of 5 body sections, the head, ear, and neck were the most preferred predilection sites for ticks to be infested on stray dogs in Selangor and Kelantan, whereby the finding agrees with earlier reports in the USA (Saleh et al., 2019). These sections were the top choices for the tick as self-grooming by biting is impossible, and the skin is thinner which allows for better feeding and infestation of ticks (Aziz et al., 2017). The present study results were also on par with other previous studies on tick distribution in different parts of the body, whereas the head, neck, ear, and dorsal had the highest infestation compared to other parts. (Khovand et al., 2022; Saleh et al., 2019; Tinoco-Gracia et al., 2009). The dorsal part is the second-highest tick infestation body part of stray dogs in this study as the dorsal part of stray dogs was considered a less accessible place for

them to remove the ticks with their paws. (Khovand et al., 2022). The tail and perianal was the body sections that had the lowest tick infestation among stray dogs. This could be due to their behavior, whereby the dog will be chewing their butt whenever there is presence of irritation caused by the ticks (Taylor, 2021), making the ticks easily to be dropped off or removed by the stray dogs themselves.

## 10.0 Conclusion

In conclusion, the overall occurrence and intensity of tick infestation on stray dogs in Selangor and Kelantan were relatively high. Of all the associated risk factors investigated, it was confirmed that the neutering status of stray dogs and states were the most significant risk factor that influences the occurrence and intensity of tick infestation on stray dogs. As strays are closely linked to human habitats, the data of tick prevalence and intensity in stray is vital for the tick control for being strategically implemented to safeguard these common zoonotic infections from spreading to humans.

## 11.0 Recommendations and future work

Several limitations were identified in this study. Firstly, the convenient sampling method, as well as the limited small sample size, may lead to bias which can result in producing inaccurate data. Among the four developmental stages of ticks, the adults and nymphs were the only stages encountered in this study, which suggests there may be a risk of overlooking larvae tick infestation due to its smaller body size compared to the other 2 stages. This may indirectly influence the result of the data. Furthermore, the variable lengths of the hair coats on each stray dog could also affect the success of the examination. It could be affected when the small, flat ticks that have not been fed

attach to the long hair coats of the stray dogs, making them unlikely to be located and removed. Some of the body parts, such as the abdomen and interdigital area of stray dogs were not easily touched and checked thoroughly during the examination, thus making limitations in collecting the data accurately. Finally, it may be useful to study the potential vector-borne disease agent carried by the ticks collected by using serological and molecular approaches.

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