

**ENDOPARASITIC INFECTIONS IN WILD  
RATS AND THEIR DISTRIBUTION USING  
GEOGRAPHIC INFORMATION  
SYSTEM (GIS) IN PENANG, MALAYSIA**

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by

**PUTRI WULAN DARI BINTI PERISON**

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## LIST OF SYMBOLS

%	Percentage
°	Degree
μl	Microlitre
ml	Mililitre
g	Gram
Kg	Kilogram
Km	Kilometres
Cm <sup>3</sup>	Cubic centimetre
sp	Species

## **LIST OF ABBREVIATIONS**

SPSS            Statistical Package for Social Science

GIS             Geographic Information System

MBSP          ‘Majlis Bandaraya Seberang Perai’

MBPP          ‘Majlis Bandaraya Pulau Pinang’

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**JANGKITAN PARASIT DALAMAN DARIPADA TIKUS LIAR DAN  
TABURANNYA MENGGUNAKAN SISTEM MAKLUMAT  
GEOGRAFI (GIS) DI PULAU PINANG, MALAYSIA**

**ABSTRAK**

Spesies rodensia terkenal sebagai haiwan pembawa penyakit. Rodensia terutamanya tikus dapat menyesuaikan diri dengan pelbagai ekosistem dan keadaan. Selain daripada itu, mereka juga dapat berkembang dengan baik di kawasan bandar. Oleh kerana jarak yang dekat antara tikus dan masyarakat, kajian ini bertujuan untuk mengkaji prevalens jangkitan endoparasit daripada tikus liar di Pulau Pinang, Malaysia serta mengetengahkan parasit zoonotik daripada tikus bagi memahami lebih lanjut mengenai tahap jangkitan parasitik yang membawa risiko kepada penduduk setempat. Sejumlah 150 tikus liar telah ditangkap dari kawasan komersial ( $n=86$ ) dan kawasan kediaman ( $n=64$ ) di sekitar Pulau Pinang dan Seberang Perai. Penangkapan tikus liar dilakukan dengan kerjasama bersama Majlis Bandaraya Pulau Pinang (MBPP) dan Majlis Bandaraya Seberang Perai (MBSP). Dalam kajian ini, empat spesies tikus liar telah dikenal pasti sebagai *Rattus rattus* (58%), *Rattus norvegicus* (15.33%), *Bandicota indica* (24%) dan *Bandicota bengalensis* (2.67%). Endoparasit daripada sistem pencernaan, darah dan najis dianalisa melalui pemeriksaan salur pencernaan, calitan darah, kultur *in-vitro* dan teknik apungan najis. Sebanyak 12 spesies endoparasit dikenal pasti dan terdiri daripada kumpulan cacing pita (*Hymenolepis diminuta*, *Hymenolepis nana*, dan *Raillietina* sp.), cacing gelang (*Heterakis spumosa*, *Gongylonema neoplasticum*, *Syphacia muris*, dan *Pterygodermatites* sp.), akantosefala (*Moniliformis moniliformis*), cacing pipih (*Echinostoma* sp.) dan protozoa (*Tritrichomonas muris*, *Trypanosoma lewisi*, dan

*Blastocystis* sp.). Daripada 12 spesies endoparasit yang dikenal pasti, terdapat lapan spesies (*H. diminuta*, *H. nana*, *Raillietina* sp., *G. neoplasticum*, *M. moniliformis*, *Echinostoma* sp., *T. lewisi* dan *Blastocystis* sp.) yang mempunyai potensi zoonotik dan kepentingan perubatan kepada masyarakat setempat. Dengan bantuan Sistem Maklumat Geografi (GIS), peta visual telah dihasilkan untuk pemahaman yang lebih baik mengenai taburan tikus liar serta kawasan titik panas bagi penyakit parasit zoonotik. Berdasarkan peta GIS, kawasan kediaman Mak Mandin di Butterworth, Pulau Pinang dikenalpasti sebagai kawasan panas dengan taburan tikus liar yang tinggi. Manakala hotspot bagi penyakit parasit zoonotik adalah Batu Ferrenghi, Balik Pulau, Gelugor dan Mak Mandin Butterworth Pulau Pinang, Malaysia. Akhir sekali, data ini dapat memberi bantuan untuk melaksanakan langkah kawalan dan pencegahan yang berkesan bagi mengawal tikus liar bagi pihak berkuasa.

**ENDOPARASITIC INFECTIONS IN WILD RATS AND THEIR  
DISTRIBUTION USING GEOGRAPHIC INFORMATION SYSTEM (GIS)  
IN PENANG, MALAYSIA**

**ABSTRACT**

Wild rat species are renowned for their capacity to act as reservoir hosts and carriers of diseases. Wild rats have effectively acclimated to various ecosystems and living conditions, even thriving in urban areas. Due to the close interaction between wild rats and the community, this study aims to determine the prevalence of endoparasite infections in wild rats from Penang, Malaysia as well as highlight zoonotic parasitic to understand more the severity of the parasitic infections that pose a risk to the public. A total of 150 wild rats were collected from commercial areas ( $n=86$ ) and residential areas ( $n=64$ ) around Penang Island and Seberang Perai. The sample was collected by collaborating with the Pulau Pinang City Council (MBPP) and Seberang Perai City Council (MBSP). In this study, four species of wild rats collected and identified as *Rattus rattus* (58%), *Rattus norvegicus* (15.33%), *Bandicota indica* (24%) and *Bandicota bengalensis* (2.67%). The endoparasites from gastrointestinal, blood and fecal was examined through necropsy, blood smear, *in vitro* cultivation and fecal floatation. A total of 12 species of endoparasites were identified comprising groups from cestode (*Hymenolepis diminuta*, *Hymenolepis nana* and *Raillietina* sp.), nematode (*Heterakis spumosa*, *Gongylonema neoplasticum*, *Syphacia muris* and *Pterygodermatites* sp.), acanthocephalan (*Moniliformis moniliformis*), trematode (*Echinostoma* sp.) and protozoan (*Tritrichomonas muris*, *Trypanosoma lewisi* and *Blastocystis* sp.). From the 12 species

of endoparasites identified, there are eight species of endoparasites (*H. diminuta*, *H. nana*, *Raillietina* sp., *G. neoplasticum*, *M. moniliformis*, *Echinostoma* sp., *T. lewisi* and *Blastocystis* sp.) had zoonotic potential and medical significance to the neighbouring community. With the help of a Geographic Information System (GIS), visuals map was generated for a better understanding about the distribution of the wild rats and the hotspot area for zoonotic parasitic diseases. According to the GIS map, Mak Mandin residency, Butterworth Penang is the hotspot for high distribution of wild rats and hotspot area for parasitic zoonotic disease among all of the areas in Penang, Malaysia. This data will serves as preliminary insight for the local authorities to implement effective control and prevention measures for wild rats.

# CHAPTER 1

## INTRODUCTION

### 1.1 Background of the study

Rodents, which encompass over 1,700 species are a significant group of mammalian animals that have adeptly adapted to diverse environments worldwide. They make up more than 42% of all mammalian species (Mustapha *et al.*, 2019; Raja- Azizi *et al.*, 2023). Not only are they recognised for their remarkable capacity to adjust and thrive in various environmental settings, but also being acknowledged as pests and significant contributors to the transmission of zoonotic diseases (White & Razgour, 2020; Azhar & Bakar, 2021).

Rodents or wild rats are well known for their potential as reservoir hosts and disease carriers. Cestode, nematode, trematode, acanthocephalans, and protozoa are the main group of zoonotic parasites infecting rodents that jeopardise human health (Tijjani *et al.*, 2020; Hardgrove *et al.*, 2021).

Within the zoonotic cycle, wild rats endoparasite plays a significant role in the transmission of various diseases such as angiostrongylosis and schistosomiasis (Paramasvaran *et al.*, 2009). In Malaysia, commonly found zoonotic endoparasites in rodents are *Balantidium*, *Capillaria*, *Giardia*, *Trichinella*, *Moniliformis* and *Hymenolepis* (Rahdar *et al.*, 2016; Coomansingh-Springer *et al.*, 2019).

Endoparasites in wild rats are categorised into two groups, which are protozoan and helminths. Commonly known rodent-borne zoonotic protozoan infections are toxoplasmosis, babesiosis, cryptosporidiosis, giardiasis, amoebiasis, leishmaniasis and Chagas disease (Mustapha *et al.*, 2019). The protozoan parasitic infection may happen through various pathways. It is known that humans and

animals may be infected through consuming infected water or food that is contaminated by infective oocysts and also direct contact with the infected individuals (e.g., cryptosporidiosis, toxoplasmosis, amoebiasis) (Pierce & Kirkpatrick, 2009; Tijjani *et al.*, 2020).

On the other hand, wild rats can serve as either the definitive host or intermediate host for helminthic infection (Singla *et al.*, 2008; Hamdan *et al.*, 2016). Various parasitic helminths identified as *Taenia taeniaeformis*, *Hymenolepis nana*, *Hymenolepis diminuta* and *Heterakis* sp. are recognised for their significance in public health (Paramasvaran *et al.*, 2009; Hamdan *et al.*, 2016).

Due to the close interaction between wild rats and a community in certain areas, zoonotic parasitic infection potential is concerning as some of the infections may cause severe human diseases and mortality and the transmission of rodent-borne diseases can be transmitted directly or indirectly (Meerburg *et al.*, 2009). Hence this study aims to identify the prevalence of endoparasites in Penang, Malaysia and highlighting their zoonotic potential to the surrounding community.

Additionally with the aids of GIS, it provides a visual map for a better understanding of the hotspot area for the distribution of wild rats and the hotspot area for zoonotic potential area association. This data aims to serves as preliminary insight for the local authorities to implement effective control and prevention measures for wild rats control measure.

## **1.2 Objectives**

This study embarks on the following objectives:

- 1) To determine the prevalence of wild rats and the endoparasites infections in wild rats at Penang, Malaysia.
- 2) To determine the association of endoparasite infection with the host species, age groups, sexes, and geographical settings.
- 3) To provide data visualisations on the distributions of wild rats and the zoonotic endoparasites infection in Penang, Malaysia.

## CHAPTER 2

### LITERATURE REVIEW

#### 2.1 Rodents

Approximately 42% of the mammalian world biodiversity order is categorised as Rodentia. Antarctica is the only place where rodents are nowhere to be found. Order Rodentia comprises 33 families with 481 genera, where 2,277 are known species (Chakma *et al.*, 2018). The order Rodentia is further divided into commonly known suborders such as rat-like rodents in Myomorpha suborders, Sciuromorpha consisting of squirrel-like rodents, and lastly, the Hystricomorpha comprising rodents like porcupine (Shomer *et al.*, 2015).

In Southeast Asia, four significant families are commonly found namely, Muridae (Figure 2.1), Rhizomyidae (Figure 2.2), Sciuridae (Figure 2.3), and Hystricide (Figure 2.4). The family Muridae (rat and mouse) consists of two-thirds of rodents and 1,350 recognised species found in Africa, Australia, and Eurasia (Aplin *et al.*, 2003; Chakma *et al.*, 2018). Within the four rodent families, Muridae, specifically rats and mice, exhibits the highest species diversity, with 27 species documented in Peninsular Malaysia (Lim, 2015).

Rodents usually are small in size ranging from 10g to around 70kg (e.g., capybaras). They are nocturnal and exhibit various adaptations such as cheek pouches for food transport, specialised limbs, and claws for activities like burrowing or climbing, and unique sensory organs like vibrissae. They rely on a strong sense of smell and a well-developed vomeronasal organ, which is crucial for reproduction and social behaviour through pheromone detection. Since rodents lack sweat glands, they are prone to overheating (Delaney *et al.*, 2018).

Rodents can be identified by their teeth, featuring large, curved incisors in both upper and lower jaws, no canines, and a toothless gap. In Southeast Asia, all rodents have five well-developed toes on their hind feet but there are only four of them are long. The toes are clawed on each front foot, with a short thumb bearing a nail instead of a claw (Francis, 2019).

Additionally, the order Rodentia has multiple lifespans. A relatively long lifespan exceeding 20 years was recorded in tree squirrels, porcupines, naked mole-rat and beavers. Meanwhile, the family Muridae predominantly consists of short-lived species. However, there is considerable variability within this group. Old World mice, rats, and hamsters typically have maximum lifespans of less than five years (Gorbunova *et al.*, 2008).

Furthermore, the Rodentia order exhibits a remarkable level of diversity, boasting a considerable species richness. The relatively brief lifespan characteristic of this order has played a pivotal role in the swift proliferation of its population, thereby facilitating the group's adaptation and evolution within a novel environment over an extended period. Rodents possess a remarkable capacity for dispersal and a remarkable ability to acclimate to the anthropogenic activities associated with human presence (Paramasvaran *et al.*, 2009).

Behavioural adaptations are essential for managing environmental fluctuations. For instance, the wood mouse (*Apodemus sylvaticus*) transitions from territorial foraging in open landscapes during summer to communal use of forest shelters in winter. Similarly, the African striped mouse (*Rhabdomys pumilio*) modifies its home range based on food availability. This often involve seasonal regulation of reproduction, with food quality or availability serving as key triggers, allowing rodents to respond rapidly to environmental changes (Auffray *et al.*, 2009).



**Figure 2.1:** *Rattus norvegicus* species from the Muridae family (Mabbett, 2014).



**Figure 2.2:** *Cannomys badius* species from the Rhizomyidae family (Philips, 1983).



**Figure 2.3:** *Tamiasciurus hudsonicus* species from the Sciuridae family (Niese, 2015).



**Figure 2.4:** *Hystrix brachyura* species from the Hystricidae family (Ongin, 2018).

### 2.1.1 Wild rats

The family Muridae, which is generally known as rats and mice, encompasses a vast array of over 1,500 recognised species, with 70 of these species currently acknowledged within mainland Southeast Asia (Francis, 2019). The terms rat and mouse are commonly used globally based on their size. Generally, the term rat is used for larger species in the *Rattus* genus, while the term mouse is used for smaller sizes from the genus *Mus*. Nevertheless, these names have subsequently been assigned to various genera worldwide, primarily based on their size, often without significant consideration for their actual relationships (Francis, 2019).

The wild rat is notably diverse, with a significant abundance of species. The relatively short life span inherent in this group has been crucial in the rapid expansion of its population, enabling the group to adapt and evolve quickly in a new environment over an extended period. They can inhabit different environments, including aquatic, semi-aquatic, or dry biotopes (Rabiee *et al.*, 2018). They can disperse and readily adjust to human-related activities in their surroundings (Paramasvaran *et al.*, 2009).

Despite the environmental changes caused by human activities, *R. rattus diardii* (black rat) and *R. norvegicus* (brown rat) are a few examples of species that are dispersed and distributed around the world (Auffray *et al.*, 2009). Not only are they recognised as mammals that are known for their ability to adjust to new surroundings but wild rats are also known as vectors for many zoonotic diseases, parasitic diseases and destructive agricultural pests (Chakma *et al.*, 2018). They play an essential role in agricultural and urban environment pests, and approximately 20 species are known as

agricultural pests (Claveria *et al.*, 2005; Paramasvaran *et al.*, 2009).

Furthermore, it is imperative to note that wild rats, as classified pests, are primarily characterised by their gnawing behaviour, leading to substantial economic loss, structural deterioration, and contamination of food sources, as highlighted in a study conducted by Brown *et al.* (2017). Additionally, comprehensive studies, such as those undertaken by Stenseth *et al.* in 2003, have elucidated that wild rats compete directly with humans for food resources, particularly during the pre-harvest season. In essence, the multifaceted impact of rodents as pests encompasses broader implications for economic sectors, health and the sustenance of human livelihoods (Lim, 2015).

On top of that, wild rats are potentially serving as reservoirs for pathogens, leading to the emergence of debilitating diseases affecting both humans and livestock (Singleton *et al.*, 2003). The gravity of the situation is underscored by the staggering fact that approximately four million rats are born daily globally. These prolific rodents harbour the capability to propagate over 60 diseases posing a significant global health threat with the potential to infect human populations worldwide (Singleton *et al.*, 2003). This alarming scenario not only emphasises the role of wild rats as vectors of infectious diseases but also underscores the urgency of implementing effective measures to mitigate the associated risks to public health and agriculture.

### **2.1.2 Wild rats in Malaysia**

Malaysia is recognised as a biodiverse hotspot within the tropical expanse of Southeast Asia. The diverse array of habitats present there leads to a significant abundance of varied fauna, including a wide range of wild rat species (Ramli & Hashim, 2009; Azhar & Bakar, 2021). Malaysia has documented and reported the diversity of about 440 mammal species of which 66 species (15%) are exclusively found within Malaysia (Francis & Barret, 2008).

The order Rodentia found in Peninsular Malaysia comprised four families namely, Muridae which includes mice and rats; Sciuridae, encompassing tree and ground squirrels; Hystricidae, representing porcupines and Rhizomidae which consists of bamboo rats (Paramasvaran *et al.*, 2009). Within the group of four rodent families, the family Muridae stands out for having the greatest variety of species with Peninsular Malaysia alone having a documented count of 27 distinct species of rats and mice (Lim, 2015). The success and dominance of species within the Muridae family of wild rats can be attributed primarily to their adeptness at swiftly adapting to a new environment (Singleton, 2003).

Habitat selection and preference are vital factors in the population dynamics of wild rats as they dictate the essential elements for their survival. While certain species inhabit particular and distinct environments, others are distributed across various areas. Many species exhibit an inclination for modified habitats in urban areas and natural habitats such as forests, mountains, and grasslands (Azhar & Abu Bakar, 2021).

Some endemic species such as *Tachyoryctes macrocephalus*, *Lophuromys melanonyx*, *Leopoldamys sabanus* and *Maxomys whiteheadi* can only be found in the forested areas as in Kelantan, Sabah, Sarawak, and Selangor (Jayaraj *et al.*, 2012). Meanwhile, the *Leopoldamys sabanus* and *Maxomys whiteheadi* species are prevalent in various forest types across Malaysia. These indigenous rodent species are abundant in forested areas due to their specialised diet, which includes forest resources such as shrub fruits, mushrooms, and small invertebrates. The food source can only be found in forested areas hence, these species will not be available in the urban habitats (Azhar & Abu Bakar, 2021).

Additionally, several species of wild rats preferred modified habitats such as houses, commercial areas, scrub, and cultivated areas. In Penang Island, primarily due to the availability of food resources, *Rattus rattus* were predominantly captured within the residential and commercial areas (Amni *et al.*, 2019). In Tasik Bera, Malaysia's natural habitat, a study by William-dee *et al.* in 2019 found that the location exhibited greater biodiversity than Penang Island, an urban area. Penang Island demonstrated lower diversity than Tasik Bera which showcased a richer array of species with a recorded 21 caught species (William-dee *et al.*, 2019).

In urban and suburban regions of Malaysia, the rodent species that are commonly found within households and industrial settings as shown in Figure 2.5 are *Rattus rattus diardii* (house rat), *Rattus norvegicus* (Norway rat), *Mus musculus* (house mouse), and *Rattus exulans* (Burmese rat) (Francis, 2019). These species are known to scavenge in trash heaps and residential areas. They are also frequently spotted exhibiting rapid movements at night or

occasionally during the day. They can be observed in various locations such as markets, food courts, coffee shops, street stalls, residences, kitchens, and the drainage systems of urban areas. These species are known to be omnivorous and commonly feed on household waste, garbage, animal matter and plant material (Lim, 2015).



**Figure 2.5:** Common rodents found in urban areas ((a) *Rattus rattus*; (b) *Rattus norvegicus*; (c) *Rattus exulans*; (d) *Mus musculus*) (Francis, 2019).

Meanwhile, in field area and plantation as in Figure 2.6 the *Rattus argentiventer* (rice field rat), *Mus caroli* (rice field mouse), *Bandicota indica* (greater bandicoot rat), *Bandicota bengalensis* (lesser bandicoot), and *Rattus tiomanicus* (Malaysian wood rat) are the major pest for oil palm estate and rice field. *Rattus argentiventer* is widely found in Malaysia while *Mus caroli* and *Bandicota indica* are solely found in the northern region of Peninsular Malaysia whereas *Bandicota bengalensis* is endemic to Penang Island (Lim, 2015; Francis, 2019).

Field rats and mice are classified as omnivores. Their natural diet consists of plant material, insects, earthworms, molluscs, and crustaceans. In rice fields, their diet includes insects and young rice plants. In oil palm estates, they primarily feed on insects, snails, slugs, and young oil palm shoots (Lim, 2015).

Additionally, the identification of wild rats encompasses a comprehensive assessment of their physical attributes, including their fur colouration and morphometric measurements such as tail length and overall body length (Lim, 2015; Francis, 2019). The critical criteria for distinguishing various species of wild rats commonly found in a household and field area have been succinctly compiled in Table 2.1.



**Figure 2.6:** Common rodents found in plantation and field areas ((a) *Rattus argentiver*; (b) *Mus caroli*; (c) *Rattus tiomanicus*; (d) *Bandicota indica*) (Francis, 2019).

**Table 2.1:** Summary of the key identification to identify common wild rats found in household and field areas (Lim, 2015; Francis, 2019).

Wild Rat Species	Physical Appearance		Weight (g)	Mammary Tits (Adult Female)	Morphometric Measurements (Mm)			
	Colour	Other Description			Head Body	Skull	Tail	Hindfoot
<i>Rattus rattus</i>	Dull brown back upperpart and Light greyish brown underpart	Dark tail. Similar length of tail and body.	100-200	10 (2+3)	105-215	33-43	120-230	26-40
<i>Rattus norvegicus</i>	Greyish brown-black upperpart with grey belly	Bicolored tail, dark above and lighter below	200-500	12 (3+3)	160-265	41-56	170-250	35-50
<i>Rattus exulans</i>	Grey to greyish brown upperpart with grey belly	Long facial whiskers	25-60	8(2+2)	21-26	27-30	105-160	21-26
<i>Rattus argentiver</i>	Olive brown body with dark soft guard hairs and silvery belly	Entirely dark tail	85-240	12(3+3)	140-210	35-45	130-205	30-40
<i>Rattus tiomanicus</i>	Brownish body with soft spiny guard hairs and pure white to yellowish belly	Entirely dark brownish tail	55-150	10(2+3)	140-190	34-45	150-200	28-35
<i>Bandicota indica</i>	Blackish grey belly with long black guard hairs and grey belly	Very dark brown tail	400-900	12(3+3)	190-330	49-64	190-280	46-60
<i>Bandicota bengelensis</i>	Brownish body with long dark guard hairs with greyish white to light brown belly	Feet with dark brown or brownish grey hair	200-400	5-17	160-210	36-44	110-160	27-38
<i>Mus musculus</i>	Uniformly brownish body	Tail entirely dark brown	9-17	10(2+3)	65-90	18.5-22	67-92	14.5-18.5
<i>Mus caroli</i>	Brownish grey body with whitish belly	Bicolored tail, dark above and whitish below	8-14	10(2+3)	65-85	19-22	65-95	16.5-19.2

## 2.2 Parasitic infection

Parasites are commonly known as organisms that benefit from other organisms and cause harm to the host. However, parasites can nourish the host and obtain more excellent fitness than the host. Modes of parasitism are the most successful mode of life due to evolution and the richness and diversity of existing species (Poulin & Morand, 2000). There are two (2) parasitism relationships which can occur within the host namely, obligatory parasitism, which refers to the parasitic interaction that is entirely dependent on the host and facultative parasitism, which refers to the independent parasitic interaction with the host (Wall & Shearer, 2008).

Additionally, parasites can be divided into two groups: endoparasites and ectoparasites. Ectoparasites or external parasites infest the host's skin or outgrowth of the skin periodically. There are six ectoparasites, specifically mites, lice, ticks, flies, and fleas (Wall & Shearer, 2008). They shared the same features, such as chitinous exoskeletons, segmented bodies, and jointed appendages. Fleas and lice were classified within the Order Hemiptera and class Insecta that possessed six-legged. Meanwhile, mites, comprising chiggers and ticks, are eight-legged organisms categorised within the class Arachnida, subclass Acari (Diaz, 2015).

The infection caused by ectoparasites presents a direct and imminent threat to the host by infiltrating, feeding, residing within, and reproducing in the host skin and various orifices. Additionally, this parasitic infestation may manifest as a perilous menace by extracting blood or tissue from the host, exacerbating the potential harm of parasitic interactions (Diaz, 2015).

Meanwhile, endoparasites are classified into two major categories which are protozoans and helminths. In general, a protozoan is a unicellular organism that possesses at least one nucleus and is divided into five categories, which are sporozoans, ciliates, flagellates, amebae, and pseudopods (Deplazes *et al.*, 2016).

The term helminth encompasses a diverse group of parasitic metazoans from the phyla Platyhelminthes (flatworms), Acanthocephala (thorny-headed worms), and Nematoda (roundworms). The flatworms (Platyhelminthes) are then further classified into Cestode (tapeworm), Trematode (flukes), and Tubercularia. The Tubercularia possesses a ciliated body and is a free-living aquatic organism, whereas the rest are exclusively parasitic (Bowman *et al.*, 2002).

The transmission of protozoa usually occurs through a faecal-oral route, while those residing in human blood or tissue are typically transmitted to other humans through an arthropod vector (Ogbera & Anaba 2021). Meanwhile, helminths infect the subcutaneous tissues, gastrointestinal tract, blood, and lymphatic system (CDC, 2022). Alternatively, helminths may attach themselves to the mucosal layer within the intestine or the trachea, engaging in blood-sucking or epithelial cell consumption. Additionally, certain helminths exhibit a more localised distribution, inhabiting specific organs or particular segments within organs (Ogbera & Anaba 2021).

### 2.2.1 Ectoparasitic infection of wild rats

According to Paramasvaran *et al.*, (2009), five groups of ectoparasites are commonly found in rodents are ticks (Acarina), mites (Mesostigmata), chiggers (Prostigmata), fleas (Siphonaptera), and lastly, lice (Phthiraptera).

Ticks are categorised in order Acarina and they infest mammals, birds, reptiles, and amphibians. They are obligate parasites that fully depend on their host (Keirans & Durden, 2005). Ticks can be visibly seen and primarily feed on the blood, lymph, or digested tissues of vertebrates. The capitulum, body, and legs compose the exterior body sections, and there is a specialised hold-fast organ called a hypostome that plays a critical role in the feeding process. Ticks cause indirect injury to humans by spreading dangerous organisms while feeding, such as protozoa, viruses, and bacteria (Anderson, 2008).

Mites are also categorised in the order Acarina and Arachnida class. Mites differ significantly morphologically from insects, the most noticeable is the lack of segmentation. Mite's body parts are divided into two which are the gnathostome (mouthpart) and body (idiosome). The mite's integument, or exterior coat, is made of chitin. Meanwhile, chiggers are the larval stage of mites from the Trombiculidae family. The parasitic larva is bloodsucking and infests vertebrates such as mammals, birds and even humans. Chigger is approximately 100–300µm, smaller than the nymphs and adults (Arlan, 2009).

In addition, fleas belong to the order Siphonaptera and are commonly found in mammals. Fleas have a number of very distinctive traits, including being wingless, having a bilaterally compressed body, having decreased eyes and antennae, and having an enlarged hind femur adapted for leaping. In contrast with lice, fleas did not fully depend on their host (Bourne *et al.*, 2018).

On the other hand, lice are categorised in Phthiraptera. Lice are obligatory ectoparasites that feed on the host's blood (Bonilla *et al.*, 2013). Unlike other insects, lice have no wings and have flattened, elongated bodies with oval heads. They have a three-stage life cycle, which includes the stages of egg, nymph, and adult (Burgess, 2004).

In addition, rodent ectoparasites are significant vectors of pathogenic microorganisms, responsible for transmitting various diseases to humans. Several studies on rodent ectoparasites have been conducted in Malaysia (Madinah *et al.*, 2011; Mohd-Zain *et al.*, 2015). Additionally, fleas have been identified as vectors for numerous diseases, including Salmonellosis, Tularemia, Leishmaniasis, Trypanosomiasis, and relapsing fever (Tijjani *et al.*, 2019).

*Trypanosoma lewisi* particularly is commonly transmitted by various species of rat fleas, including *Ceratophyllus fasciatus*, *Nosopsyllus fasciatus*, and *Xenopsylla cheopis*. Infection with *T. lewisi* can occur through contact with the faeces of infected fleas or by ingesting infected fleas. Although primarily affecting rodents, this organism can sporadically infect other mammals. *T. lewisi* is distributed worldwide in both urban and wild rats, with a particularly high prevalence in Asia (Kamaruzaman *et al.*, 2021).

Meanwhile, tick species, *Haemaphysalis* sp. and *Ixodes granulatus*, are recognised for their medical importance. The *Haemaphysalis* sp. tick is a significant vector, transmitting various pathogens, including *Babesia* sp., *Tularemia* sp., *Rickettsia* sp., and arboviruses. Their bites can cause stress and blood loss in both animal and human hosts. Similarly, *I. granulatus* serves as the primary vector of the Langkat Virus (Tijjani *et al.*, 2019).

Lastly, lice are known to harbor and transmit plague bacilli and transmit tularemia and bartonellosis to humans. Their bites in humans can lead to a condition called pediculosis. Furthermore, *Polyplax spinulosa* can transmit *Trypanosoma lewisi* and *Rickettsia typhi* (Chuluun *et al.*, 2005).

### 2.2.2 Endoparasitic infection of wild rats

Endoparasites are classified into two major categories: protozoans and helminths. The term helminth encompasses a diverse group of parasitic metazoans from the phyla Platyhelminthes (flatworms), Acanthocephala (thorny-headed worms), and Nematoda (roundworms). The flatworms (Platyhelminthes) are then further classified into Cestode (tapeworm), Trematode (flukes), and Tubercularia. The Tubercularia possesses a ciliated body and is a free-living aquatic organism, whereas the rest are exclusively parasitic (Bowman *et al.*, 2002).

Cestode or tapeworm is known for its ribbon-shaped body, multisegmented proglottids, and scolex. The body consists of three parts: the head (scolex), which is the anterior part of the holdfast organ; a neck (germinative segment); and a body divided into segments. The body segment known as a proglottid is equipped with both female and male reproductive organs, as tapeworms exhibit hermaphroditic characteristics (Cobo, 2014). The *Hymenolepis nana* and *Hymenolepis diminuta* are commonly found infecting wild rats, which act as the definitive host (Tijjani, 2020).

In addition to the Platyhelminthes phyla, trematode or flukes typically exhibit a flattened, leaf-shaped structure in a dorso-ventral orientation. The external surface of their bodies is covered with a layer known as the tegument, often equipped with small-scale-like spines. These organisms possess a pair of suckers: one located at the oral end and the other on the ventral side. The oral sucker leads to the oesophagus, which connects to a small muscular organ called the pharynx. Interestingly, their digestive

system lacks a distal opening. Instead, it branches into two ceca (Waikagul *et al.*, 2015).

The flatworm is divided into two groups, Monogenea and Digenea. The difference between these two groups is that the Monogenea exhibit external parasitism, commonly found on aquatic vertebrates' excretory bladder or gills, requiring only a single host to fulfil their life cycle. Conversely, the Digenea follow a complex life cycle that involves two or more host species (Bhatia *et al.*, 2010). Trematode in rodents is very rare with only a few species documented. The first report on the presence of a medically significant fluke was by Lie and Virik (1963) who reported the presence of *Echinostoma malayanum* in *Suncus murinus*, *Rattus argentiventer*, *Rattus rattus diardii*, and *Rattus tiomanicus*. Subsequently, *Prosthodendrium* sp. was detected infecting house rats in Kuala Lumpur (Singh *et al.*, 1997). Additionally, *Schistosoma* sp. fluke was also discovered in *Bandicota indica*, the giant paddy field rats (Singh *et al.*, 1997).

Meanwhile, the nematodes, also known as roundworms, generally exhibit a cylindrical body form without segments, gradually narrowing towards both the front and back. These organisms are enveloped by a resilient, transparent cuticle that displays delicate horizontal stripes on its outer surface. They exist in distinct male and female sexes, with variations in size and contrasting characteristics evident primarily at their posterior end (Soulsby, 1968). In Malaysia, nematodes in wild rats are well documented. The earliest reported nematodes in wild rats from the Muridae family by Alessandrini (1905) and Adams (1933) were *Heterakis* sp., *Syphacia obvelata* and *Cyclodontostomum purivisi* which were found in the large

intestine of rats captured in the Raub, Pahang. Later, comprehensive descriptions of the *Heterakis spumosa*, *Angiostrongylus malaysiensis*, *Gongylonema neoplasticum*, *Trichuris muris*, *Subulura Anderson*, *Strongyloides ratti*, *Physaloptera* sp., and *Syphacia muris* was done (Paramasvaran, 2012).

On the other hand, the acanthocephala commonly referred to as spiny-headed worms exhibit a cylindrical unsegmented body configuration that is covered with thick cuticles. The body structure is composed of two distinct sections: a posterior trunk and an anterior presoma. The anterior presoma comprises a distinctive spiny proboscis alongside an unspined neck, forming a unique anatomical arrangement (Soulsby, 1968). The males of the species exhibit a notably smaller physical size when compared to their female counterparts. They entail a complex life cycle that prominently involves intermediary hosts. These intermediary hosts are small crustaceans or arthropods (Soulsby, 1968). Acanthocephala is relatively rare among rodents with only one documented occurrence of *Moniliformis moniliformis* by Sandosham (1957). Later, Lim *et al.* (1974), Khairul (1977), and Leong *et al.*, (1979) reported and documented the same species of acanthocephalan, *Moniliformis moniliformis* in house rats in Malaysia. Previous studies have shown that *Moniliformis moniliformis* has only been observed in house rats in Malaysia.

On the other hand, in contract with helminth, a protozoan is a single-celled organism characterised by the presence of at least one nucleus, and it is classified into five categories: sporozoans, ciliates, flagellates, amebae, and pseudopods (Soulsby, 1968; Deplazes *et al.*, 2016). The most prevalent