

Occurance of Ectoparasite-Small Mammals in Hutan Lagenda and Waterfall of Gunung Ledang

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Abstract

The study of interaction between ectoparasites and small mammals were conducted at Gunung Ledang Johor National Park, Johor. The objectives of this study were to assess the distribution of small mammals in Gunung Ledang National Park and to identify the ectoparasites found on small mammals in Gunung Ledang National Park. Both hosts species and ectoparasites were identified by laboratory examination using 24 cage traps and 2 mist nets during a 12-day sampling period. The baits that were used in this study were cut ripe banana, dried salted fish and mango. In this study, the results showed a total of 31 volant and non- volant small mammals had been captured and a total of 32 ectoparasites had been collected from the infested small mammals. 14 out of 31 small mammals had infested by ectoparasites which represented 13 of volant species and only 1 of the non-volant species. Among 32 ectoparasites, there were 5 species had examined represented 4 species from bat flies and 1 species from ticks. Results also showed that the data analysis on both small mammals and ectoparasites, and ecological factors such as habitat type, elevation and microclimate affect ectoparasite diversity and prevalence. This study effectively assessed the distribution of small mammals in Gunung Ledang, finding ectoparasites linked to these small mammals at both Taman Lagenda and Waterfall Gunung Ledang. This has showed the biological richness of these sites with a notable variety of small mammal species and their ectoparasites. Ectoparasite prevalence and abundance among host species provided important information about host-parasite interactions, highlighting the role of the host behaviour.

1. Introduction

Malaysia, as a biodiversity hotspot in Southeast Asia's tropical regions, boasts a rich diversity of wildlife, particularly small mammals. These species play a vital role in maintaining ecological balance with over 70% of the global vertebrate mammal diversity classified as small mammals ranging from 2g to 5kg, including bats, shrews, and rodents [1] [2]. Cage trapping, which uses various kind of bait to capture small mammals, are commonly used in rodent pest control and small mammals' research. It is crucial to have an efficient trapping system due to this technique

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aids researchers in not only estimating the population density and community structure of small mammals, but also to estimate the structure, composition and population size of small mammals and understanding the ecological health of the habitats' ecosystem [3][4][5].

Ectoparasites are microorganisms that infest an animals' exterior, including its skin and can result in apparent sores and health issues. These parasites such as lice and fleas are classified in to two major groups, which are Arachnida which includes ticks and mites, and Insecta, which includes lice and fleas. Ectoparasite infestation are a major problem for animal production since they can cause discomfort, weight loss and even higher mortality among them. Researchers can look at the various ectoparasites that are present in various mammal species in habitat such as Gunung Ledang in order to investigate the association between ectoparasites and small mammals.

Mount Ledang Taman Lagenda and Gunung Ledang's Waterfall region are two separate but related ecosystems. Because of its dense vegetation and cooler, shaded environments, Taman Lagenda, a lush forested area, provides a variety of habitats for small mammals and ectoparasites. On the other hand, species that are adapted to such conditions are drawn to the waterfall area because of its more dynamic environment, which includes rushing water, increased humidity and open spaces. Although the biodiversity of both regions are great, differences in their microhabitats lead to differences in the distribution of the species.

Malaysia's Gunung Ledang National Park is known for its abundant biodiversity, which includes a wide range of ectoparasites and small mammals and also diverse range of habitats through different vegetation types such as lowland dipterocarp, hill dipterocarp, upper dipterocarp and montane-oak forests were an example that consist rich vertebrate fauna assemblage that has yet to be explored. Hence, a rapid study aimed at assessing distribution of small mammals and their interaction with ectoparasites of Gunung Ledang was carried out over a 12 days sampling period by Universiti Tun Hussein Onn Malaysia. However, limited research studies are known about the interactions between these ectoparasites and small mammals, particularly in the areas such as Taman Lagenda and Waterfall Gunung Ledang. Hence, implementing sustainable conservation initiatives and maintaining updated on the ecology become more difficult by this lack of information. Hence, this study aimed to assess the distribution of small mammals in Gunung Ledang National Park. This study also aimed to identify, assess the diversity, distribution and prevalence of ectoparasites on small mammals within both sub-habitats in Gunung Ledang through field sampling and laboratory identification.

2. Methodology

2.1 Sampling Site

The study was conducted at Gunung Ledang National Park (2.367°N, 102.600°E), where located at Tangkak District of Johor, Malaysia. Two primary forest trail entrances, Taman Lagenda (2.22°N, 102.36°E) (Fig. 2a), an old-growth forest with diverse plants and many small mammals, and Waterfall Gunung Ledang (2°21.29'N, 102°37.93'E) (Fig. 2b), located in dipterocarp forests for plants adapt to montane and hilly conditions, were selected as sampling site.



Fig. 1 Map of Gunung Ledang in Southernmost Part of Peninsular Malaysia



Fig. 2 (a) Trail of Gunung Taman Lagenda; (b) Trail of Waterfall Gunung Ledang

2.2 Sampling Method

The sampling method used in this study aimed to thoroughly capture volant and non-volant small mammal species through cage traps and mist nets and their associated ectoparasites in Taman Lagenda and Waterfall Gunung Ledang. Two 600-meter line transects with 300-meter of each were set up at each sampling site to provide thorough spatial coverage. 24 cages trap and 2 mist nets were placed thoughtfully along these two lines transect with 12 cage traps and 1 mist nets for each at both sampling sites. The study area was conducted for a total of twelve days with four days a week for three weeks at both study site, Taman Lagenda and waterfall Gunung Ledang respectively. All of the cage traps were baited with ripe banana, mango and burned dried salted fish to attract non-volant small mammals. The traps were left open all day and checked three times between 8:30 a.m. and 5:00 p.m. at intervals of 2-3 hours and replaced damaged baits regularly. Mist nets were set up at 5:30 p.m. and checked the next morning at 9:00 a.m., then closed to prevent harm to volant species or other animals. Captured small mammals were then placed in cloth bags to prevent ectoparasites transfer before identification [6].

2.3 Data Collection

2.3.1 Identification of Small Mammals

For identification, the captured volant and non-volant small mammals were quickly taken to field laboratory. Key features such as sexual status, ear size, and tail-to-body length were compared with standard taxonomic guides [7]. Identification was confirmed with photographic records and references to existing databases for small mammal's native to Malaysia [8] [9]. All the small mammals were handled ethically, following the University Tun Hussein Onn Malaysia's guidelines to minimize stress and harm. The identified species were then marked and released back into their natural habitat.

2.3.2 Collection of Ectoparasites

Essential tools for collecting ectoparasites include gloves, data sheets, labels, disinfection supplies, toothbrush, flat painting brush, forceps and collection tube with 70% alcohol. All instruments were sanitized before to use in order to ensure data accuracy. Ectoparasites were collected by brushing the fur of small mammals by three to five times with a toothbrush, flat painting brush and forceps on a white A4 paper. In order to reach the skin, the brushing was done slowly and deliberately by concentrating on their head, ears, tail and other body parts. The small mammals were then released back at capture site after being collected and having their toes clipped for identification. Ectoparasites were transferred with forceps to alcohol containers. A last check was done to make sure no ectoparasites are overlooked and brushes that were clogged with fur were cleansed.

2.3.3 Identification of Ectoparasites

For identification, the preserved ectoparasite specimens were placed on object glass slides and examined by using a Leica stereomicroscope equipped with 40x magnification in the laboratories at Universiti Tun Hussein Onn Malaysia,

Pagoh Campus. The ectoparasites were identified via the genus and species group using morphological characteristics and taxonomic keys for lice, tick, mites, fleas and bat flies [10] [11] [12]. Each ectoparasite's total number obtained from distinct hosts were noted in a notebook.

2.3.4 Data Analysis

For this study, quantitative information on ectoparasites and small mammals was required, such as species count, abundance, diversity including species evenness and species richness, and species prevalence. Relative abundance, evenness and species richness will be determined using the Shannon-Weiner Index (H') and Simpson Diversity Index (D). D was used for evaluating species evenness whereas H' represents species diversity. The prevalence will be shown as a proportion of the ectoparasite-infested population. The average ectoparasite abundance across the data also were determined by using measures of central tendency, particularly the mean. All of the quantitative data were calculated by using PAST software [13].

2.4 Equation

2.4.1 Species Richness

Species richness quantifies the variety of species found in a given region or environment.

$$\text{Species Richness } (S) = N \quad (1)$$

2.4.2 Shannon-Weiner Index (H')

The Shannon-Weiner Index measures the diversity of a community by considering both the number of species, species richness and how evenly the species are distributed, species evenness.

$$\text{Shannon-Weiner Index } (H') = - \sum_{i=1}^S \pi_i \ln \pi_i \quad (2)$$

2.4.3 Simpson Diversity Index (D)

The Simpson Diversity Index is used to measure the diversity of a community by considering both the number of species, species richness and the relative abundance of each species.

$$\text{Simpson Diversity Index } (D) = 1 - \frac{\sum n(n-1)}{N(N-1)} \quad (3)$$

2.4.4 Prevalence

Prevalence quantifies the frequency of a characteristic or condition in a population at a certain point in time. It describes the proportion of individuals in a community that are infected with a specific ectoparasite species in ecology and parasitology.

$$\text{Prevalence} = \frac{\text{Total number of infested small mammals}}{\text{Total number of small mammals captured}} \times 100\% \quad (4)$$

2.4.5 Mean Abundance

Mean abundance used to calculate the average number of individuals of a species or group within a given sample or population.

$$\text{Mean Abundance} = \frac{\text{Number of ectoparasites}}{\text{Number of infested small mammals}} \quad (5)$$

3. Results and Discussion

3.1 Host Species

Taman Lagenda and Waterfall Gunung Ledang were the two study sites where a total of 31 small mammals, 23 volant and 8 non-volant species were captured. Numerous families of both volant and non-volant species were found in Taman Lagenda (Table 1). Among the volant species were bats belonged to the Rhinolophidae, Pteropodidae, and Hipposideridae families and a collared scops owl from the Strigidae family. Rats from the Muridae family, treeshrews from the Tupaiidae family and squirrel from Sciuridae family were examples of non-volant species. 5 species were captured at Taman Lagenda and 26 species were found at Waterfall Gunung Ledang. 14 out of 23 volant species, *Cynopterus brachyotis* (Fig. 3a) was the highest number captured at Waterfall whereas 2 out of 8 non-volant species were referred to *Tupaia glis* (Fig. 3b). Reducing human activities during the suspension of tourism activities is thought to have contributed to the increasing number of species at Waterfall Gunung Ledang by causing less disruption and attracting more small mammals.

Table 1 List of Host Species Caught at Taman Lagenda and Waterfall Gunung Ledang

	Family	Host Species	Gender		Total caught	Habitat Types *	
			Male	Female			
Volant	Rhinolophidae	<i>Rhinolophus refulgens</i>	1	-	1	tlgl	
		<i>Rhinolophus creaghi</i>	-	1	1	tlgl	
		Total	-	-	2		
	Pteropodidae	<i>Cynopterus brachyotis</i>	9	5	14	wfgl	
		<i>Cynopterus sphinx</i>	-	1	1	wfgl	
		<i>Balionycteris seimundi</i>	1	1	2	wfgl	
		<i>Aethalops alecto</i>	1	-	1	wfgl	
		Total	-	-	18		
		Hipposideridae	<i>Hipposideros armiger</i>	1	1	2	wfgl
		Total	-	-	2		
Strigidae	<i>Otus lettia</i>	-	1	1	tlgl		
	Total	-	-	1			
Non-Volant		<i>Callosciurus notatus</i>	-	-		tlgl	
	Sciuridae	<i>Sundasciurus tenuis</i>	-	1	1	wfgl	
		Total	-	-	2		
	Muridae	<i>Rattus tiomanicus</i>	-	1	1	tlgl	
		<i>Leopoldamys sabanus</i>	1	-	1	wfgl	
		<i>Maxomys rajah</i>	-	1	1	wfgl	
		<i>Rattus norvegicus</i>	-	-	1	wfgl	
		Total	-	-	4		
	Tupaiidae	<i>Tupaia glis</i>	1	1	2	wfgl	
		Total	-	-	2		
Grand Total			15	14	31		

tlgl=Taman Lagenda Gunung Ledang, wfgl=Waterfall Gunung Ledang



Fig. 3 (a) Volant species, *Cynopterus brachyotis*; (b) Non-Volant species, *Tupaia glis*

3.2 Ectoparasite Species

A total of 32 ectoparasite species were collected from 31 small mammal species at both study sites. These ectoparasites included five types of bat fly parasites from three families and one tick species from Ixodidae family shown in Table 2. The most common ectoparasite was *Nycteribia stylidiopsis* (Fig. 4), found on four volant host species, 3 *Cynopterus brachyotis* and 1 *Balionycteris seimundi*. Among volant hosts, *Cynopterus brachyotis* carried to ectoparasites types, which were *Nycteribia stylidiopsis* and *Eucampsipoda madagascarensis* (Fig. 5a). Only one of the non-volant species, *Tupaia glis*, was infested, hosting *Ixodes granulatus* (Fig. 5b). Volant species showed higher infestation rates due to their larger movement ranges, increasing trap encounters while non-volant species, being arboreal or ground-dwelling, likely avoided traps.

Table 2 List of Ectoparasites Species and Number of Ectoparasite Collected from Particular Host Species

Ectoparasites Species	Total	Family	Genus	Number of Ectoparasites Individual	Host Species	Total Caught	Number of Host Infested
Bat Flies							
<i>Brachytarsina sp.</i>	6	Streblidae	<i>Brachytarsina</i>	5	<i>Rhinolophus refulgens</i>	1	1
				1	<i>Rhinolophus creaghi</i>	1	1
<i>Nycteribia stylidiopsis</i>	9	Hippoboscidae	<i>Nycteribia</i>	8	<i>Cynopterus brachyotis</i>	14	3
				1	<i>Balionycteris seimundi</i>	2	1
<i>Eucampsipoda madagascarensis</i>	8	Nycteribiidae	<i>Eucampsipoda</i>	1	<i>Cynopterus sphinx</i>	1	1
				5	<i>Cynopterus brachyotis</i>	14	3
				1	<i>Balionycteris seimundi</i>	2	1
				1	<i>Aethalops alecto</i>	1	1

<i>Raymondia alulata</i>	7	Hippoboscidae	<i>Raymondia</i>	7	<i>Hipposideros armiger</i>	2	1
Ticks							
<i>Ixodes granulatus</i>	2	Ixodidae	<i>Ixodes</i>	2	<i>Tupaia glis</i>	2	1
Total	32	-	-	32	-	-	14

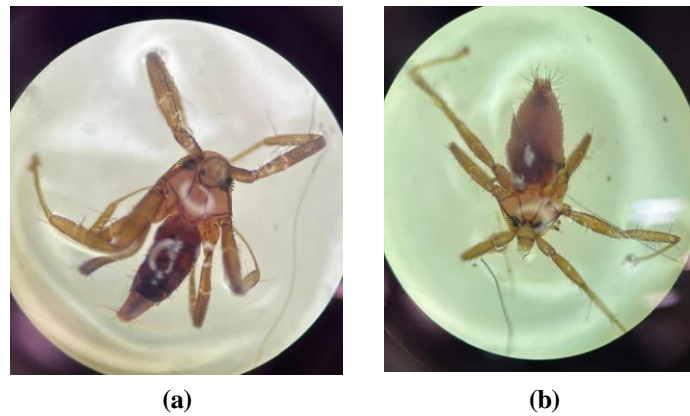


Fig. 4 (a), (b) Bat Flies, *Nycteribia stylidiopsis*

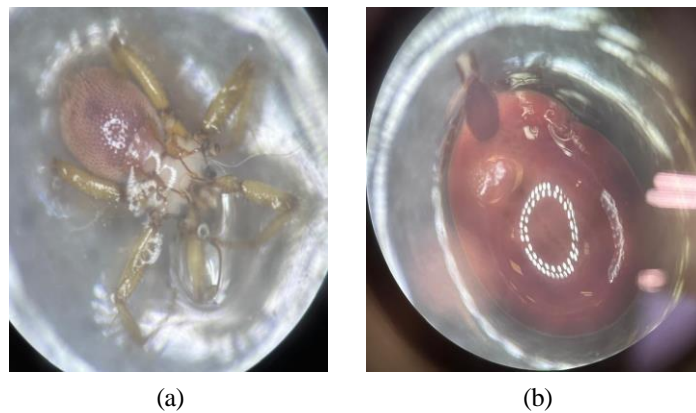


Fig. 5 (a) Bat Flies, *Eucampsipoda madagascarensis*; (b) Ticks, *Ixodes granulatus*

3.3 Species Richness, Shannon-Weiner Index (H') and Simpson Diversity Index (D) of Small Mammals at Two Study Sites Respectively

At Taman Lagenda and Waterfall Gunung Ledang, the species richness, H' and D for small mammals were examined. The results are summarized in Table 3.

Table 3 Species Richness, Shannon Wiener Diversity Index and Simpson Diversity Index of Small Mammals at Two Study Sites

Study Site	Species Richness	Shannon-Weiner Index, H'	Simpson Diversity Index, D
Taman Lagenda	5	1.61	0.8
Waterfall	10	1.68	0.68

According to the results, Taman Lagenda (5) had a lower species richness than Waterfall Gunung Ledang (10). A higher number of small mammal species may be supported by a variety of microhabitats, the availability of food supplies and the absence of human interference. According to the H', the areas' levels of diversity are comparable, with Waterfall having slightly higher diversity (1.68). It indicates that although a range of species can be found on both locations, the distribution of individuals among species was marginally more even at the Waterfall regions. On the other hand, Taman Lagenda had greater Simpson Diversity Index (D=0.8) than Waterfall (D=0.68), indicating that a small number of species were more prevalent there. This could indicate that some species were more frequently found at Taman Lagenda, whereas the species composition of the Waterfall site were more balanced. A more varied community was indicated by Waterfall Gunung Ledang's slightly higher H' and greater species richness than Taman Lagenda. However, a lower D assessing on D showed less evenness in the distribution of species, which indicates a greater dominance of some species in Taman Lagenda.

3.4 Species Richness, Shannon-Weiner Diversity Index and Simpson Diversity Index of Ectoparasites

Five ectoparasite species were identified at both study sites, including 4 at the Waterfall and only 1 at Taman Lagenda Gunung Ledang. In Table 4, it showed that only bat flies, named *Brachytarsina sp.* were observed at Taman Lagenda, where they infested *Rhinolophus refulgens* and *Rhinolophus creaghi*. In comparison, the Waterfall area featured 4 ectoparasite species. The results showed that ectoparasite richness was influenced by host diversity. There are more ectoparasite species because the Waterfall Gunung Ledang supported a greater diversity of hosts. Taman Lagenda exhibits decreased ectoparasite richness, might due to fewer or less diverse hosts.

Table 4 Species Richness of Ectoparasites Infestation at Two Study Sites

Study Sites	Ectoparasite Species	Host Species	Total Species Richness
Taman Lagenda	<i>Brachytarsina sp.</i>	<i>Rhinolophus refulgens</i> , <i>R. creaghi</i>	1
Waterfall Gunung Ledang	<i>Nycteribia stylidiopsis</i> , <i>Eucampsipoda madagascarensis</i> , <i>Raymondia alulata</i> , <i>Ixodes granulatus</i>	<i>Cynopterus brachyotis</i> , <i>Balionycteris seimundi</i> , <i>Cynopterus sphinx</i> , <i>Balionycteris seimundi</i> , <i>Aethalops alecto</i> , <i>Hipposideros armiger</i> , <i>Tupaia glis</i>	4

Taman Lagenda and Waterfall Gunung Ledang's diversity indices for ectoparasite infestation on small mammals were shown in Table 5. Taman Lagenda had a lower H' (0.69), compared to Waterfall Gunung Ledang (1.47). As well, Waterfall Gunung Ledang had a higher D (0.69) than Taman Lagenda (0.50). increased diversity at Waterfall Gunung Ledang was associated with species richness and host abundance, which support more stable and diverse ectoparasite populations. Conversely, Taman Lagenda showed lower diversity indices, most likely as a result of a decreased species richness of ectoparasites and limited small mammal hosts populations.

Table 5 Shannon Wiener Diversity Index and Simpson Diversity Index of Ectoparasites Infestation at Two Study Sites

Study Sites	Total Caught	No. of Hosts Infested	Shannon-Weiner Index, H'	Simpson Diversity Index, D
Taman Lagenda	5	2	0.69	0.50
Waterfall Gunung Ledang	26	13	1.47	0.69

3.5 Prevalence of Ectoparasites

According to Table 8, the overall prevalence of ectoparasites among captured small mammals were 43.8%. Out of 31 small mammals captured, 13 volant (56.5%) individuals and only 1 non-volant (12.5%) had ectoparasite infestations. Bat flies were the ectoparasites found on volant species listed in Table 8 and *Ixodes granulatus*, a tick, was the only ectoparasite found on non-volant species.

Table 6 Prevalence of Ectoparasites on Small Mammals

Host Type	Ectoparasite Species	Prevalence (%)
Volant	<i>Eucampsipoda madagascarensis</i>	26.1
	<i>Nycteribia stylidiopsis</i>	17.4
	<i>Brachytarsina sp.</i>	8.7
	<i>Raymondia alulata</i>	4.3
Non-Volant	<i>Ixodes granulatus</i>	12.5

Eucampsipoda madagascarensis had the greatest ectoparasite prevalence among volant species (26.1%). This might be explained by bats' social nature, as they frequently nap in colonies, which created a stable environment for the reproduction and spread of ectoparasites. *Nycteribia stylidiopsis* had a prevalence of 17.4%, making it the second most common ectoparasite for volant hosts. *Brachytarsina sp.* and *Raymondia alulata* had lower prevalence rates of 8.7% and 4.3% respectively, showing that they were less frequently found on volant hosts. However, only 1 non-volant species had a tick infestation with the prevalence of 12.5% due to the high humidity and dense forest at the study sites might be the reason of the low prevalence in non-volant species. These conditions favored efficient grooming behaviors that aid in getting rid of ectoparasites by these small mammals.

3.6 Mean Abundance of Ectoparasites

Based on the calculation, bat flies had a greater mean abundance (2.30) than ticks (2.0), as showed in Table 9. The anatomy and behaviours of volant species were being considered to be crucial for this gap. Bat flies were able to thrive in greater numbers because bats were less efficient at grooming. On the other hand, tick populations were decreased by non-volant species, which groomed more frequently. Furthermore, ticks usually leave their hosts after feeding in order to reduce their presence on particular hosts, but bat flies remained on their hosts and feed on blood on a regular basis.

Table 7 Mean Abundance of Ectoparasite

Ectoparasites	No. of Examined Volant/Non-Volant Small Mammals	No. of Volant/Non-Volant With Ectoparasites	No. of Ectoparasite/Individual	Mean Abundance
Bat Flies	23/8	13/0	30/0	2.30
Ticks	23/8	0/1	0/2	2.0
Total		14	32	4.30

4. Conclusion

This study concluded by effectively assessing the distribution of small mammals and the ectoparasites that are associated with them at Taman Lagenda and Waterfall Gunung Ledang. Numerous ectoparasites were identified in the laboratory, and some of the parasites had showed host-specific preferences, showing co-evolutionary relationships. The findings highlighted the biodiversity of tropical rainforests and the significance of habitat components of conservation initiatives.

The study had limitations related to sampling techniques and geographic coverage. It was carried out throughout a 12-day period at Taman Lagenda and Waterfall Gunung Ledang. The diversity of ectoparasites found was impacted by the limited instruments, which included 24 cage traps and 2 mist nets. This decreased the possibility of collecting a wide variety of small mammals, especially non-volant species. Furthermore, the use of morphological characteristics

and manual identification may have resulted in observer biases and methodological errors, which could have been minimized with the use of advanced molecular techniques such as DNA barcoding. These limitations emphasized the necessity of more extensive sampling efforts and improved techniques in future studies.

To better understand how ecological elements such as altitude, vegetation and microclimate affect ectoparasite prevalence and host interactions, future studies should examine a greater variety of habitats in Gunung Ledang National Park. The dynamics of ectoparasite life cycles and host availability can also be understood by examining seasonal fluctuations in temperature, humidity and precipitation. Gaining an understanding of these trends will improve our knowledge of biodiversity processes and aid in the development of efficient management and conservation plans for this hotspot for biodiversity.

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Conflict of Interest

Authors declare that there is no conflict of interests regarding the publication of the paper.

Author Contribution

The authors confirm contribution to the paper as follows: **study conception and design:** H'ng Yan Ting; **data collection:** H'ng Yan Ting, Arney Sapaat; **analysis and interpretation of results:** H'ng Yan Ting, Arney Sapaat; **draft manuscript preparation:** H'ng Yan Ting, Arney Sapaat. All authors reviewed the results and approved the final version of the manuscript.

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