Salt-lick use by mammals in tropical rainforests of

Peninsula Malaysia

Yuko Tawa-Doi 2023

Abstract

Salt-licks are known as gathering places for various animals to eat soils or drink seeping water in order to supplement minerals or alleviate gastrointestinal problems etc. Salt-licks may relieve mammals from physiological stress and increase their fitness, and it may increase the carrying capacity of areas with salt-licks. Because the salt-licks are used by various animals including endangered species, the conservation of the area around the salt-licks has been proposed at several regions. In this study, I studied the use of salt-licks by mammals in the rainforests of Peninsular Malaysia, a biodiversity hotspot but with limited reports on salt-licks, and discussed the role and necessity of salt-licks in the ecosystems of the region. This study was conducted in the Belum-Temengor Forest Complex, State of Perak, Malaysia. I placed the infrared sensor cameras around the four wet-type salt-licks, where the animals drink water with low turbidity.

First, the food habits and behavior of mammals that visited salt-licks were analyzed to understand the purpose of their salt-lick visits. The results on analyses of camera-trapping data indicated that herbivores visited salt-licks and drinking water much more frequently than other food-habit species. These suggest that the studied saltlicks were mainly used by herbivores to meet physiological needs such as supplement minerals and/or alleviate gastrointestinal problems by drinking salt-lick water.

Second, in order to examine the possible mineral supplementing function of salt-licks of this area, I compared mineral concentration between the water from salt-licks and nearby streams as the reference sites. Only the concentrations of sodium and calcium were significantly higher in water from the salt-licks than those of nearby streams, indicating that mammals could supplement these minerals by drinking salt-lick water. I also analyzed relationship between mineral concentration and salt-lick visit by mammals. Herbivores most frequently visited the salt-lick with the sodium concentration significantly higher than other salt-licks, indicating that sodium supplementation is an important purpose of salt-lick visit by them. However, not all the herbivore species showed such tendency, suggesting that the factors other than sodium concentration might also influence the selection of salt-licks by each species.

Third, I focused on the salt-lick use and behavior around salt-licks of wild Malayan tapirs, a threatened mammal which visits there frequently, to reveal different aspects of the ecology. Only one or two tapirs (male-female combination) were recorded at the same time; no same-sex individuals were recorded at the same time. Multiple identified males and females were recorded at the same salt lick, suggesting the overlap of their home ranges. Analyses of one-night tapir recordings suggested that one particular individual or one male-female pair occupied a salt lick for an entire night, indicating their exclusive behavior particularly between same-sex individuals. In addition to frequent water-drinking behavior, communication behaviors were also observed around the salt-licks, suggesting the importance of salt-licks for their survival and social interactions.

This study revealed that the salt-licks in Peninsular Malaysia also have physiological function such as mineral supplementation and secondary function as a communication site, and therefore the salt-licks are important for survival of mammals in this region. Detailed analysis of the camera-trapping records at salt-licks will lead to some new clues to the living conditions of poorly studied species, such as Malayan tapir, and be useful for planning of ecosystem conservation in this region.

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Note on Archiving

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Chapter 1:

General Introduction

1.1 Importance of salt-licks in conservation

In forest ecosystems including tropical rainforests, there are specific sites which are known to local people as gathering places for animals. Many terrestrial herbivores and even some predominantly arboreal primates have been recorded eating soil and/or drinking seeping water at these sites (Corlett 2009). These sites are called "salt licks" (Molina et al. 2014), "mineral licks" (Moe 1993), "natural licks" (Matsubayashi et al. 2007a), or "mineral spring" (Bechtold 1996), etc. because it is assumed that these sites can provide animals minerals, especially sodium, in many cases (described in detail in <u>Chapter 1.2</u>). I, hereafter, refer to the sites where animals show eating soil or drinking seeping water as "salt-licks".

Salt-licks may relieve mammals, especially in rainforest habitats, from physiological stress and increase their fitness, and it may increase the carrying capacity of areas with salt-licks compared to areas where they are absent (Klaus et al. 1998). The conservation of the area around the salt-licks has been proposed for conservation of forest animals, as the salt-licks are used by various animals including endangered species (in northern British Columbia, Canada: Bechtold 1996, in Borneo:

Matsubayashi et al. 2011, in Columbian Amazon foothills: Molina et al. 2014). In Southeastern Asia, Matsubayashi et al. (2007a) reported that various mammals in tropical rain forests of Borneo, including endangered species such as orangutan (*Pongo pygmaeus*), drank the salt-lick water with much minerals such as sodium and calcium, suggesting the mineral supplementing function of the salt-licks and importance of the salt-licks in wildlife conservation. In order to develop specific plans for the conservation of forest ecosystems, it is necessary to understand the salt-lick use by various animals and the functions of salt-licks in each region.

Although there are many natural salt-licks in the tropical rainforest of Peninsular Malaysia, one of the biodiversity hotspots of the world, we still have limited reports on these salt-licks (Chong et al. 2005, Magintan et al. 2015). Especially, information needed to understand the functions of these salt-licks is very limited. In this region, the Department of Wildlife and National Parks (DWNP) introduced habitat enrichment program, involving maintenance of grazing field, forest replanting, clearance of the forest for new shoots and maintenance of artificial salt-licks (Ali et al. 2021). The artificial salt-licks provide various minerals such as sodium, phosphorous, and calcium in the protected areas under jurisdiction of DWNP (Magintan et al. 2015), supposing the mineral supplement function of the salt-licks. At these artificial salt-licks, some ungulate species such as wild boars (*Sus scrofa*), red muntjacs (*Muntiacus muntjak*), and Malayan tapirs (*Tapirus indicus*) were reported to drink water frequently (Simpson et al. 2020). We still do not understand well, however, why animals visit natural salt-licks. It is necessary to gain a better understanding of the purposes of visits to natural salt-licks by animals and the function of salt-licks in the rainforest ecosystem for conservation of various fauna in Peninsular Malaysia, where there is little information on the use of natural salt-licks.

1.2 Physiological function of salt-licks

Salt-licks can be divided into several types. At the study site in the rainforest of Peninsular Malaysia (mentioned later in <u>Chapter 2</u>), I visually identified two types of salt-licks: the soil-eating type and water-drinking type. At the soil-eating type salt-licks, animals ingest soil particles (geophagy), and at water-drinking type salt-licks, animals ingest seeping water. Number of previous studies have conducted the chemical analyses of the soil or water of salt-licks, for example the measurements of mineral and minor elements concentration, bicarbonate, pH, conductivity, particle size of soil, clay minerals, cation exchange capacity. Besides, some studies have reported on the salt-lick use by mammals or birds in each region and the correlation with chemical characteristics as described below.

(1) The mineral supplementing function was proposed in a number of studies, in particular sodium supplementation (Tankersley 1981, Clayton and MacDonald 1999, Holdø 2002, Montenegro 2004, Ayotte et al. 2006, Matsubayashi et al. 2007a, Matsubayashi et al. 2011, Owen et al. 2014). The herbivores need to supplement sodium because plants do not contain enough sodium for animal physiological functioning (Corlett 2009). For example, Moe (1993) showed that sodium was the main element sought at the salt-licks, based on concentration differences between salt-licks and controls and the significant positive correlation between axis deer (*Axis axis*) use and the sodium content of the salt-lick. Some salt-licks were also reported to have the function for supplementation of calcium, magnesium, phosphorus, iron, manganese, cobalt, and molybdenum (Dormaar and Walker 1996, Klaus et al. 1998, Clayton and MacDonald 1999, Montenegro 2004, Mills and Milewski 2007).

(2) The function of detoxification was proposed in several studies on soileating type salt-licks, because eating soils with higher contents of clay that adsorb the plant secondary metabolites could be effective for detoxification (Oates 1978, Klaus et al. 1998, Gilardi et al. 1999, Houston et al. 2001, Wakibara et al. 2001). The clay contents in soils also have a role for gastrointestinal mucosal cytoprotection (Gilardi et al. 1999).

(3) It has been also proposed that ingested soils or water can alleviate gastrointestinal problems. The clay minerals such as kaolinite in the ingested soils absorb excess water in the feces and alleviate diarrhea (Wakibara et al. 2001, Molina et al. 2014). The ingested soils or water also contribute to the buffering capacity in the gut. Highly herbivorous species are prone to gastrointestinal acidosis in the season when highly digestive foods, such as abundant seeds and fruits, or young leaves and shoots, are more available, and thus the frequency of salt-lick use was reported to increase during such season (Davies and Baillie 1988, Ayotte et al. 2006). Bicarbonates that buffer the pH in the gastrointestinal tract (Bechtold 1996, Ayotte et al. 2006), and clay minerals with cation exchange sites that adsorb potassium and short-chain fatty acids, could prevent gastrointestinal acidosis (Davies and Baillie 1988).

Several hypotheses on the functions of salt-licks have been proposed, however, the reasons of salt-lick visit might vary depending on the animal species, the type of salt-lick, and the region. And this makes it difficult to understand fully why animals gather around the salt-licks. In order to clarify the function of the salt-licks, it is needed to study them separately by types. In this study, I conducted a camera-trapping survey around the wet-type salt-licks, where the animals drink water with low turbidity. At such salt-licks, the possibility of the ingestion of clay particles can be eliminated, which is needed for detoxification.

First, I focused on the visit frequency to salt-licks by mammals and the behaviors (Chapter 3). The following hypotheses are tested: if the purposes of the visit to salt-licks are mineral supplementation or alleviation of gastrointestinal acidosis, (i) the salt-licks would be visited mainly by herbivores and (ii) water-drinking behavior would also be shown by herbivores much more frequently than omnivores and carnivores. Second, I focused on mineral concentration in the water of salt-licks (Chapter 4). The following hypotheses are tested: the herbivores would visit the salt-licks with higher mineral concentration more frequently if the mineral supplementation is a major reason for their salt-lick visits.

1.3 Salt-lick use by mammals in Peninsular Malaysia

Several previous studies showed the list of mammal species observed at primary and secondary forest in Peninsular Malaysia by camera trapping or line transects, etc (<u>Table 1-1</u>). Some ungulate species such as wild boar (*Sus scrofa*), red muntjac (*Muntiacus muntjac*) and Malayan tapir (*Tapirus indicus*) were most frequently recorded (Kawanishi and Sunquist 2004, Azlan 2006). The lists also include the endangered mammal species such as tiger (*Panthera tigris*), Asian elephant (*Elephas maximus*), white-handed gibbon (*Hylobates lar*), and Sunda pangolin (*Manis javanica*), etc. At the artificial salt-licks at three wildlife reserves (Krau Wildlife Reserve, Sungai Dusun Wildlife Reserve, and Sungkai Wildlife Reserve), wild boar, Malayan tapir, mouse deer (*Tragulus* sp.), red muntjac, and Malayan porcupine (*Hystrix brachyura*) were most frequently recorded (Simpson et al. 2020).

Actually, Malayan tapir is categorized as endangered (EN) species in IUCN red list (Traeholt et al. 2016), and it is crucial to understand their ecology and behavior for conservation purposes. However, only a very limited number of studies have examined ecology and behavior of wild Malayan tapirs because of the difficulty in direct observation of this solitary and nocturnal species. For the moment, several previous studies present fragmentary evidence of frequent visit to the natural salt-licks in the rainforests of Southeastern Asia (Holden et al. 2003, Novarino 2005, Khadijah-Ghani 2010). Therefore, I conducted detailed survey of salt-lick use by this threatened mammal which visits there frequently to reveal different aspects of the ecology.

In Chapter 5, I focused on the salt-lick use and behavior around salt-licks of wild Malayan tapirs. The salt-licks will offer a good opportunity to observe this

otherwise very elusive mammal for which we know very little, and to get new insights for further studies. The detailed observation of behavior and social interaction among individuals will lead to deeper understanding on the significance of salt-lick use, and contribute to plan for conservation of rainforest ecosystem. At the last of the thesis, I discussed the importance of salt-licks for conservation of mammals in the rainforest of Peninsular Malaysia and future research directions.

Tables and Figures

Table 1-1. List of medium and large-sized mammals in the forest area of Peninsular Malaysia recorded in the three previous studies.

"Previous study 1, 2, 3" in column headings indicate the reports by Kawanishi and Sunquist (2004), Azlan (2006), and Simpson et al. (2020), respectively. X indicates species recorded.

	Scientific name	Common name	Previous Study 1	Previous Study 2	Previous Study 3
Proboscidea	Elephas maximus	Asian elephant	Х	Х	
Perrisodactyla	Tapirus indicus	Malayan tapir	Х	Х	Х
	Dicerorhinus sumatrensis	Sumatran rhinoceros	Х		
Artiodactyla	Tragulus sp.*1	mouse deer	Х	Х	Х
	Muntiacus muntjak	Red muntjac	Х	Х	Х
	Rusa unicolor	Sambar	Х		Х
	Sus scrofa	Wild boar	Х	Х	Х
	Capricornis sumatraensis	Southern serow	Х	Х	
	Bos gaurus	Gaur	Х		
Carnivora	Prionodon linsang	Banded linsang	Х	Х	Х
	Hemigalus derbyanus	Banded civet	Х		Х
	Paradoxurus hermaphroditus	Common palm civet	Х	Х	Х
	Paguma larvata	Masked palm civet	Х	Х	Х
	Viverra zibetha	Large Indian civet	Х		Х
	Viverra tangalunga	Malayan civet	Х	Х	Х
	Arctictis binturong	Binturong	Х	Х	
	Arctogalidia trivirgata	Small-toothed palm civet			
	Herpestes urva	Crab-eating mongoose		X^{*2}	
	Herpestes brachyurus	Short-tailed mongoose		X^{*2}	Х
	Martes flavigula	Yellow-throated marten	Х	Х	Х
	Mastera nudipes	Malay weasel	Х		
	Aonyx cinerea	Oriental small-clawed otter			Х
	Lutra spp.	Otter ^{*3}	Х		
	Helarctos malayanus	Sun bear	Х	Х	Х
	Cuon alpinus	 Dhole	Х	Х	
	Prionailurus bengalensis	Leopard cat	Х	Х	
	Pardofelis marmorata	Marbled cat	Х	Х	Х
	Catopuma temminckii	Asian golden cat	Х		Х
	Neofelis nebulosa	Clouded leopard	Х	Х	
	Panthera pardus	Leopard	Х	Х	Х

	Panthera tigris jacksoni	Tiger	Х	Х	
Primates	Macaca fascicularis	Long-tailed macaque	Х	Х	Х
	Macaca nemestrina	Pig-tailed macaque	Х	Х	Х
	Presbytis siamensis	White-thighed langur			Х
	Trachypithecus obscurus	Dusky leaf monkey	Х		
	Presbytis femoralis	Banded langur	X^{*4}		
	Hylobates lar	White-handed gibbon	X^{*4}		
	Hylobates syndactylus	Siamang	X^{*4}		
Pholidota	Manis javanica	Sunda pangolin			Х
Rodentia ^{*5}	Echinosorex gymnurus	Moonrat			Х
	Atherurus macrourus	Brush-tailed porcupine	Х	Х	Х
	Trichys fasciculata	Long-tailed porcupine		Х	Х
	Hystrix brachyura	Malayan porcupine	Х	Х	Х

*1 Both species of mouse deers were counted as *Tragulus* sp.

*2 Four species of mongoose were pooled in Previous Study 2.

*3 Both species of *L. lutra* and *L. sumatrana* were counted as *Lutra* spp.

^{*4} The presence was confirmed by sighting or vocalization in Previous Study 1.

*5 Excluding small-sized rats, squirrels, treeshrews.

Chapter 2:

General methods

2.1 Study area

This study was conducted in the Belum-Temengor Forest Complex (BTFC) located in the State of Perak in northern Peninsular Malaysia (5°30' N, 101°20' E) (Fig. 2-1), which is mainly covered by upland and lowland dipterocarp forests (Misni et al. 2017). The BTFC consists of two sections, the Royal Belum State Park (RBSP) in the north and Temengor Forest Reserve (TFR) in the south, separated by the East-West Highway (Gerik-Jeli Highway). The RBSP covers an area of 1,175 km² (260–1,533 m a.s.l.) and consists of primary forests. By contrast, TFR (1,489 km², 260–2,160 m a.s.l.) is classified as a production forest where selective logging has been ongoing since the 1970s (Ching and Leong 2011). According to the data of rainfall at Kg. Lalang (about 30 km west of the study site) provided by Water Resources Management and Hydrology Division, the rainy season around BTFC was from April to December with the break from June to August during the study period (Fig. 2-2).

2.2 Locations of studied salt licks

There are at least 12 salt-licks known by local people in the BTFC and some of them are shown in the sightseeing maps of this area. Automated infrared sensor cameras were established around four salt-licks easily accessible for humans. All the four saltlicks are of the "wet" type; i.e., salt-licks where water that may contain minerals is seeping or flowing from the ground. These salt-licks are relatively open areas surrounded by forest, lacking large trees and where the underbrush is not very dense. One of the four studied salt-licks, called the Tersau salt-lick (5°19' N, 101°22' E), is located 16 m from the shoreline of Temengor Lake in TFR. This area is mainly covered by upland dipterocarp forest (Misni et al. 2017). The other three salt-licks are located along the Tiang River in RBSP. These salt-licks are called (A) Sira Kuak (5°42' N, 101°27' E), (B) Sira Batu (5°43' N, 101°27' E), and (C) Sira Tanah or Sira Dinding (5°43' N, 101°27' E) by the local people; however, in this study, I hereafter refer to them as "Tiang A," "Tiang B," and "Tiang C," respectively. This area is covered by mixture of upland and lowland dipterocarp forest (Misni et al. 2017). Fig. 2-3 shows the detail information on the studied salt-licks. The distance between Tiang A and Tiang B, Tiang A and Tiang C, and Tiang B and Tiang C is about 2.4 km, 2.2 km and 0.44 km, respectively (Fig. 2-4). Odden and Wegge (2007) estimated that the home range size of

red muntjacs was 0.70 km² in males, and 0.59 km² in females (95% Fixed Kernel Density) on average in Nepal. Chatterjee et al. (2014) estimated that the home range size of sambars was 7.5 km² in males, and 4.3 km² in females (50% Fixed Kernel Density) on average in Western India. Thus, the medium-large mammals in this area might easily move between Tiang B and C salt-licks. Besides, large ungulates such as sambar might access all of the three salt-licks in the Tiang area. In contrast, it is more than 40 km between the salt-licks in the Tersau area and those in the Tiang area which are also separated by a highway as shown in Fig. 2-1. Thus, Tersau salt-lick was probably visited by mammal populations different from those that visited the studied salt-licks in the Tiang area.

2.3 Camera trapping

I placed the motion-sensitive infrared-triggered digital cameras at and around the four salt-licks between 25 February 2014 and 24 February 2016 (730 days). In <u>Chapter 5</u>, only the data recorded between 26 February 2014 to 9 April 2015 (408 days) was analyzed. I set the cameras not only at each salt-lick but also along the animal trails leading to each salt-lick (within 20 m of the salt-lick) which seemed to be used as gateways to and from the salt-lick by many animals (hereafter referred to as "gateway"). The data recorded at the gateways was analyzed only at <u>Chapter 5</u> to gain a deeper understanding of the salt-lick use by Malayan tapirs.

The models of the cameras used in this study were Trophy Cam HD 2013 (Bushnell Outdoor Products, Kansas, USA), Stealth Cam Sniper Shadow STC-SNX1 (Stealth Cam LLC, Texas, USA) and TREL 20J (GISupply Co. Ltd., Hokkaido, Japan). I used these cameras in video mode or photograph mode. The records by photograph mode were analyzed only at <u>Chapter 5</u> mainly for individual identification of Malayan tapirs. The cameras in video mode collected 1-min-long videos per trigger event. The triggering interval was a minimum of 20 seconds in video mode and 5 seconds in photograph mode. The photographs recorded within 1 min were counted as one picture to avoid repetitive count of many images by continuous shooting. And then the videos and pictures recorded continuously at intervals of 30 minutes or less were considered to be not independent and counted as "one capture" to avoid repetitive count of many images by continuous shooting during a single visiting event by same animals.

All camera units were mounted on trees at approximately 1 m above the ground to record medium- to large-sized mammals. Two or three cameras were placed at each salt-lick to cover a large portion of the open area; however, some parts of each salt-lick were often not adequately covered due to issues with the faulty cameras, SD memory cards, or batteries. In each Chapter, I mentioned the number of active cameras and days with active camera (total days when at least one camera was active at each studied saltlick).

Tables and Figures

Fig. 2-1. Location of Belum-Temengor Forest Complex (BTFC). The area of BTFC is about 320,000 ha which includes 17,200 ha Temengor Lake. The stars show the location of the Tersau salt-lick and three salt-licks in the Tiang area, respectively. The location of the three salt-licks in the Tiang area is shown by one star in this figure because they are closely located to each other.

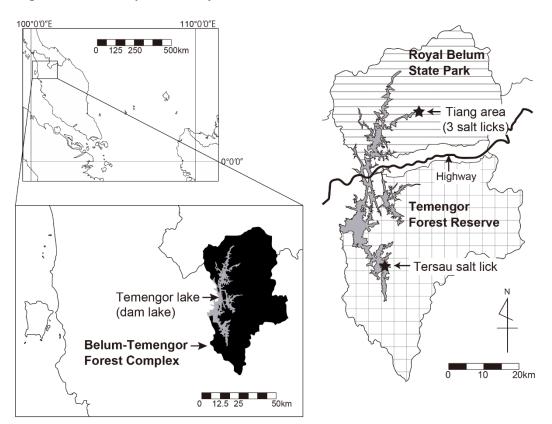
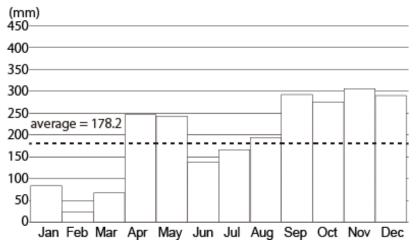


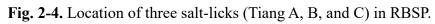
Fig. 2-2. The average monthly rainfall at Kg. Lalang during two years of Feb 2014 – Feb 2016.

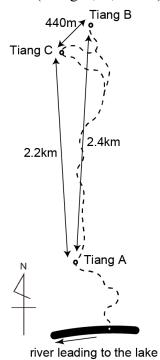
Kg. Lalang is about 30 km west of the Belum-Temengor Forest Complex, Perak, Malaysia. The data was provided by Water Resources Management and Hydrology Division.



	Tersau	Tiang A (Sira Kuak)	Tiang B (Sira Batu)	Tiang C (Sira Tanah or Dinding)
Picture of the salt-lick (size of water drinking sites)	(23 m x 17m)	(10 m x 10m)	(1.5m x 1.5m) + (1.5m x 1.5m)	(1.7 m x 5m)
Simplified illustration of salt lick area	animal trails leading to a salt lick open area drink salt-lick water (23m × 17m) 16m away from the lakeshore lake wide trail wide trail (1m × 1m puddle)	open, mud area where animals drink salt-lick water (10m × 10m) animal trails leading to a salt lick small stream	two puddles where animals drink salt-lick water (1.5m × 1.5m each) upslope stream open area with some rocks wide trail	upsiope where animals wide water (1.7m × 5m) open, large plains small stream animal trails leading to a sait lick
Characteristics of water drinking sites and vegetation in the salt-lick	Water was springing from two spots (bubbles came up constantly) forming two puddles. The spring water flew into a small stream flowing across the salt-lick. Large, flat, open area with grasses.	Animals drunk water from one relatively large mud area. Thick, dense underbrush and vines were all around the area where animals drink water.	Animals drunk water from Two adjacent small puddles. Relatively few underbrush, and presence of subcanopy trees (about 5m) around the open area.	Animals drunk water from one relatively sma mud area. Large open area with some underbrush and a few large trees (10m~) on one side.
Surroundings	The small stream flew across the salt-lick into the Temengor Lake. Dense underbrush and large trees (15m~) were surrounding the open area of the salt-lick.	A small stream flew about 5m away from the mud area. Large trees (15m~) were also around the salt-lick area.	A stream flew adjacent to the puddles. Both sides of the stream were rocky. Subcanopy trees and large trees (10m~) were along the stream and around the salt-lick area.	A small stream flew adjacent to the mud area. A steep upslope on one side of the mud area, and large plains with some subcanopy trees and large trees (10m~) on the other sides.
Vegetation around the salt-lick*	Mostly upland dipterocarp forest	Mixture of upland and lowland dipterocarp	Mixture of upland and lowland dipterocarp	Mixture of upland and lowland dipterocarp forest

Fig. 2-3. The detail information of the four studied salt-licks in BTFC, Perak, Malaysia.





Chapter 3:

Salt-lick use in Malaysian tropical rainforests reveals behavioral differences by food habit in medium and large-sized mammals

Summary

The food habits and behavior of medium and large sized mammals that visited natural salt-licks in a tropical rainforest of Belum-Temengor Forest Complex, Peninsular Malaysia, were analyzed to understand the purpose of their salt-lick visits. The study was conducted at the wet-type salt-licks, where the animals drink clear water, thus the ingestion of clay particles needed for detoxification of plant secondary compounds, one of the possible purposes of salt-lick visits, seemed very low. Ten herbivorous, seven omnivorous, and four carnivorous species were recorded at the salt-licks. 95.3% of all video capture records of animals at the salt-licks were of herbivores, while the records of omnivores and carnivores were only 3.5% and 1.2%, respectively. These results indicated that herbivores visited studied salt-licks much more frequently than other food-habit species. The herbivores also tended to stay longer at the salt-licks than omnivores and carnivores. In addition, water-drinking behaviors were recorded significantly more frequently in herbivores (73% of video captures) than in omnivores

(28%) and carnivores (0%). The carnivores showed only resting, passing, and hunting behaviors. The results suggest that the studied salt-licks were mainly used by herbivores and some omnivores, possibly to supplement minerals and/or alleviate gastrointestinal problems by drinking salt-lick water.

3.1 Introduction

There are generally two types of natural salt-licks found at forest reserve in Peninsular Malaysia (Chong et al. 2005). The minerals from water-drinking type saltlicks are ingested by animals when they drink the mineral-soaked water, and mammals and other wildlife take the minerals from the soil-eating type salt-licks by ingesting the soils. As described in Chapter 1, the study of natural salt-licks is lacking in this region (Chong et al. 2005, Magintan et al. 2015). At protected rainforests in this region, the Department of Wildlife and National Parks (DWNP) embarked on the Habitat Enrichment Programme through artificial development of salt-licks with a total of 30 sites developed throughout 2011-2012 (Magintan et al. 2015), and some previous studies assessed the use of artificial salt-licks by wild animals. Simpson et al. (2020) evaluated the effectiveness of artificial salt-licks by not only species richness but also through additional indicators which may assess comparative frequencies, staying time and behaviors observed at salt-licks.

In this chapter, I conducted a camera-trapping survey in Belum-Temengor Forest Complex to gain a better understanding of the purposes of visits to natural saltlicks by mammals of Peninsular Malaysia. Among the possible purposes of salt-lick visits by mammals (mineral supplementation, detoxification, alleviation of gastrointestinal problems) which have been proposed in Chapter 1, I examined the possibility of mineral supplementation by analyzing food habit and behavior of animals visiting the salt-licks. This study was conducted at the wet-type salt-licks, where the animals drink clear water with low turbidity, and the ingestion of clay particles needed for detoxification seemed very low.

The omnivores and the carnivores are thought to have less need for mineral supplementation and/or alleviation of gastrointestinal acidosis than the herbivores. Therefore, I hypothesized that if the purposes of the visit to these salt-licks are mineral supplementation or alleviation of gastrointestinal acidosis, (i) the salt-licks would be visited mainly by herbivores and (ii) water-drinking behavior would also be shown by herbivores much more frequently than omnivores and carnivores. I analyzed the video records at the salt-licks to assess these hypotheses and discuss the purposes and theories of salt-lick visits by the medium-large sized mammals of the Belum-Temengor area.

3.2 Methods

3.2.1 Camera trapping and data analysis

I placed motion-sensitive infrared-triggered digital cameras at the four saltlicks between 25 February 2014 and 24 February 2016 (730 days) as mentioned in Chapter 2. At each of Tersau, Tiang A, and B salt-lick, three cameras were deployed and at Tiang C salt-lick, two cameras were deployed towards the water-drinking point. The videos recorded continuously at intervals of 30 minutes or less were considered to be not independent and counted as "one capture". Even if multiple cameras at the same salt-lick recorded an animal walking into the salt-lick, the video records of this event were counted as "one capture".

The capture frequencies (the number of captures per day with active camera) of each studied salt-lick and each species were calculated. Besides, I calculated the time difference between the first and last record of each capture as an index of stay length around the salt-lick (staying time). The staying time was recorded as 1 minute for the captures including only one video. The staying times of each capture were compared among species with enough records (more than 5 captures).

The monthly capture frequencies were calculated for three herbivores, red muntjac, sambar (*Rusa unicolor*) and Malayan tapir, to examine monthly changes in

their visit frequency to the salt-licks. For this analysis, the data of three salt-licks at the Tiang area was used because the sambar was not distributed in Temengor Forest Reserve.

3.2.2 Analysis of the animal behaviors

I analyzed the behavior of mammals recorded in videos at salt-licks, especially focusing on "water-drinking behavior". Water-drinking behavior was defined as a behavior in which the animal is staying at a water site in the salt-lick touching its mouth to the water for more than 1 second. For Asian elephant, however, it was defined as a behavior in which the animal is staying at a water site touching the tip of its trunk to the water more than 1 second. When it was clear that the animal was not drinking water even if it fulfills the definitions mentioned above, for example, when a wild boar was digging soils around the water site, it was not included in water-drinking behavior. In counting the number of "capture with water-drinking behavior", when multiple individuals were recorded in one capture, it was counted as a "capture with waterdrinking behavior" if at least one of them was drinking water.

The number of individuals recorded in each capture and the number of individuals that showed water-drinking behavior among them was counted for four herbivores, red muntjac, sambar, Malayan tapir and white-thighed langur (*Presbytis siamensis*), which were recorded at the salt-licks most frequently and wild boar, the only omnivorous species in which water-drinking behavior was repeatedly observed. In this assessment, the maximum number of individuals seen in a video was recorded for that capture event. The percentage of individuals that showed water-drinking behavior in the total recorded individuals in each capture was calculated.

3.2.3 Statistical analysis

The statistical tests were computed in R version 4.2.0. Because the results of Shapiro-Wilk test showed that the staying time of each capture was not normally distributed either in each species or in each food-habit (P < 0.001), Wilcoxon rank-sum tests were used to compare the staying time of each capture between each herbivore and other food-habit species. Similarly, the number of captures in a day was not normally distributed in three herbivores (red muntjac, sambar, and Malayan tapir; Shapiro-Wilk test, P < 0.001). Therefore, Wilcoxon rank-sum tests were used to compare the number of captures in a day of three herbivores between each month and entire survey period. Pearson's chi-squared test was used to compare the rate of water-drinking individuals among four herbivores (red muntjac, sambar, Malayan tapir, white-thighed langur) and

one omnivore (wild boar). For all tests, a p-value of < 0.05 was taken to indicate statistical significance. Data are presented as the mean \pm standard deviation (*SD*).

3.3 Results

3.3.1 Recorded animals

Medium-large mammals were recorded in 2,679 captures over 1,763 days with active cameras at the four salt-licks during the study period (1.52 captures / day, Table 3-1). The capture frequency was highest at Tiang C among the studied salt-licks. 10 herbivore species, seven omnivore species and four carnivore species were recorded during the study period. Table 3-2 shows the list of medium-large mammals recorded at salt-licks, capture frequency and recorded group size of each species. The herbivores were recorded in 2,553 captures (95.3% of all captures), the omnivores in 93 captures (3.5%), and the carnivores in 33 captures (1.2%). Red muntjac was the most frequently recorded species (in 1,116 captures, 0.633 captures / day), followed by sambar (in 558 captures, 0.317 captures / day) and Malayan tapir (in 405 captures, 0.230 captures / day). Relatively small carnivorous species, which have been reported from this area (Rayan et al. 2013, Table 3-3), such as mongooses and leopard cats, were not recorded in this study. This can be because the camera units installed to record medium-large

sized mammals could not record small sized animals, or more likely, these species were not present at the salt-licks.

Table 3-4 shows the average and maximum staying time at the salt-licks of each species which was recorded in more than 5 captures. The minimum staying time was 1 minute in all recorded species except gaur (Bos gaurus) that stayed for 19 minutes at minimum. The average staying time of herbivore species ranged from 2.56 to 56.44 minutes (18.16 ± 18.63 minutes on average, n = 9), while those of omnivores and carnivores were 2.56–4.64 minutes (3.60 minutes on average, n = 2) and 1–3.30 minutes (1.93 minutes on average, n = 3), respectively. The average of average staying times of herbivore species was significantly longer than that of other food-habit species (2.60 \pm 1.30 minutes on average, n = 5) (Wilcoxon rank sum test, Z = 2.4667, P = 0.01199). The maximum staying time of herbivore species ranged from 24 to 580 minutes (160 \pm 166.43 minutes on average, n = 9), while those of omnivores and carnivores ranged 15– 37 minutes (26 minutes on average, n = 2) and 1–24 minutes (11 minutes on average, n = 3), respectively. The average of maximum staying times of herbivore species was also significantly longer than that of other food-habit species $(17 \pm 12.57 \text{ minutes on})$ average, n = 5) (Wilcoxon rank sum test, Z = 2.5361, P = 0.007493).

<u>Table 3-5</u> shows the results of comparison of the staying times between each herbivore species and other food-habit species which was recorded in more than 5 captures. In five of nine analyzed herbivore species, the staying times were significantly longer than those of all analyzed omnivore and carnivore species. In contrast, the staying time of herbivore species was significantly shorter than that of other food-habit species, only in two species (mouse deer *Tragulus* sp. and Malayan porcupine *Hystrix brachyura*) when compared with the wild boar (<u>Table 3-5</u>). These results suggest that the herbivores tended to stay longer at the salt-licks than omnivores and carnivores.

The monthly changes in the capture frequency at the Tiang area were examined in the three herbivores, red muntjac, sambar, and Malayan tapir, the three most frequent visitors to the salt-licks (Fig. 3-1). Significant monthly variation was observed in all three species; the red muntjac showed higher capture frequency in April (0.85 captures / day; Wilcoxon rank sum test, W = 99958, P = 0.008024) and November (1.78 captures / day; W = 64142, P < 0.001) and lower in March (0.41 captures / day; W = 78140, P =0.008478) than annual capture frequency (0.69). The sambar showed higher capture frequency in April (0.95 captures / day; W = 102807, P < 0.001) and lower in July (0.27 captures / day; W = 75788, P = 0.04796) and December (0.32 captures / day; W = 59100, P = 0.02217) than annual capture frequency (0.46). The Malayan tapir showed higher capture frequency in January (0.29 captures / day; W = 42257, P = 0.007362), September (0.43 captures / day; W = 42257, P = 0.007362) and December (0.37 captures / day; W = 73014, P < 0.001) and lower in May (0.015 captures / day; W = 73178, P < 0.001) and June (0.057 captures / day; W = 70108, P = 0.01519) than annual capture frequency (0.18). However, the months with capture frequencies significantly higher or lower than the annual capture frequency were different among these species and remarkable common seasonal change was not observed (Fig. 3-1).

3.3.2 Behaviors at the salt-licks

Water-drinking behaviors were often recorded in various species (1,877 captures in total), while no soil eating behavior were observed in the recorded mammals. Water-drinking behaviors were recorded in 73% of the captures recording herbivores (n = 2,553), and in 28% of the captures recording omnivores (n = 93). In the captures recording carnivores (n = 33), no water-drinking behavior was observed (Fig. 3-2). The percentage of captures recording water-drinking behavior was significantly higher in the herbivores than in the omnivores (Pearson's chi-squared test, $X^2 = 18.616$,

P < 0.001) or in the carnivores ($X^2 = 22.1, P < 0.001$).

Nine of 10 recorded herbivorous species showed water-drinking behavior. The southern serow was the only herbivore species that showed no water-drinking behavior, however, it was recorded only once. In contrast, among the seven omnivorous species, only the wild boars repeatedly showed water-drinking behaviors in 24 of the 76 captures (32%). They were also observed to dig the ground, probably searching for food, in 22 captures (29%). Such ground digging behavior was not observed in any other species. The rate of water-drinking individuals of the four herbivore species (red muntjac: 72% of 1,271 individuals in 1,116 captures; sambar: 76% of 700 individuals in 558 captures; Malayan tapir: 78% of 511 individuals in 405 captures; white-thighed langurs: 89% of 482 individuals in 145 captures) was significantly higher than that of an omnivore species (wild boar: 23% of 192 individuals in 76 captures) (Pearson's chi-squared test, p-values are shown in Table 3-6).

In contrast, the recorded carnivores did not show any water-drinking behavior even though they were recorded several times at the salt-licks. The tigers, the most frequently recorded carnivorous species (14 captures in 11 days) showed only resting and/or traveling behavior, but no water-drinking or hunting behavior. A tiger was once recorded at Tiang B salt-lick very close (about 3 m) to Malayan tapir which was drinking water there (at 10:18 PM, 12th Jan 2015)^{*1}. However, the tiger was only shown lying prone on the ground and did not attack the Malayan tapir. Whether this encounter led to a hunting event is uncertain. In Tersau salt-lick, a group of dholes (*Cuon alpinus*) (at least 3 individuals) was recorded to chase a wild boar (at 0:27 PM, 8th May 2014)^{*2}. This was the only hunting behavior observed in the four carnivores recorded at the saltlicks (33 captures).

*1,2 The online video clips are available at <u>https://doi.org/10.1007/s10344-022-01600-y</u>.

3.4 Discussion

Not only herbivores, but also omnivores and carnivores were recorded at the studied salt-licks, although most of the records (95.3%) were those of herbivores. Rayan et al. (2013) conducted a camera-trapping survey on mammals in the whole area of BTFC (not only at the salt-licks as this study) and reported 31 medium-large mammal species (11 herbivores, eight omnivores, and 12 carnivores) (Table 3-3). In this study, almost all (10 of 11) of the herbivores reported by Rayan et al. (2013) in the landscape, except brush-tailed porcupine (*Atherurus macrourus*), and most (6 of 8) of omnivores were also recorded at the salt-licks. On the other hand, few (4 of 12) carnivores were

recorded at the salt-lick sites in this study. These results suggest that the studied saltlicks were mostly visited by available herbivores in the landscape. It is possible, however, that the records of herbivores were much higher than omnivores and carnivores because it reflects their abundance in the mammal population in this area. Previous studies estimated the rates of herbivores, omnivores and carnivores by camera trapping in other tropical rainforests of Peninsular Malaysia (Kawanishi and Sunquist 2004, Azlan 2006) and Malaysian Borneo (Bernard et al. 2013). In addition, visits to artificial salt-licks by animals in Peninsular Malaysia (Simpson et al. 2020) is also comparable with this study.

Table 3-7 compares my results with those of previous studies. The table shows that the percentages of herbivore records in the whole forest area (27.9%–62.0 %) estimated by previous studies (Kawanishi and Sunquist 2004, Azlan 2006, Bernard et al. 2013), indicating the relative abundance of herbivores, were lower than that at the salt-licks (95.3%) in this study. This suggests that the very high percentage of herbivore records at the studied salt-licks was not due to the high rate of herbivores in the mammal population of the study area. Thus, the very high percentage of herbivore records at the salt-licks rather shows that the herbivores visited studied salt-licks much more frequently.

Simpson et al. (2020), who studied the use of artificial salt-licks by mammals in Peninsula Malaysia, reported that the red muntjacs, Malayan tapirs, and wild boars spent an average of 14.1 (\pm 21.1), 16.9 (\pm 18.3), and 7.4 (\pm 13.2) minutes per visit at the artificial salt-lick, respectively. They also reported that these species spent longer time there than at control sites inside the forest area $(1.5 \pm 2.2, 1, \text{ and } 2.6 \pm 5.2 \text{ minutes}, 1.5 \pm 2.2, 1.5 \text{ minutes})$ respectively), deliberately targeting the artificial salt-licks. The staying time at control sites in the forest was not measured in this study, however, the average staying times at the natural salt-licks in this study (red muntjac 7.37 ± 12.83 minutes, Malayan tapir 13.74 ± 19.20 , wild boar 4.64 ± 7.28 , Table 3-4) were longer than those at control sites reported by Simpson et al. (2020), while a few minutes shorter than those at artificial salt-licks. The herbivores tended to stay longer at the salt-licks than omnivores and carnivores, although the wild boar, one of the omnivores, stayed longer than some herbivores (Table 3-5).

These results strongly suggest that the studied wet-type salt-licks were visited mainly by herbivores in Peninsula Malaysia. The wet-type salt-licks in other regions were also visited by a number of ungulates, which feed on leaves or grasses, such as moose (*Alces alces*) (Tankersley 1981, Bechtold 1996, Ayotte et al. 2006), white-tailed deer (*Odocoileus virginianus*) (Bechtold 1996, Atwood and Weeks 2003, Owen et al. 2014), and lowland tapir (*Tapirus terrestris*) (Blake et al. 2011). The frugivorous primate, white-bellied spider monkey (*Ateles belzebuth*) (Izawa 1993, Link et al. 2011), and some omnivores which eat mainly plants, such as babirusa (*Babyrousa babyrussa*) (Clayton and MacDonald 1999) and the orangutans (*Pongo pygmaeus*) (Matsubayashi et al. 2011), also visit wet-type salt-lick repeatedly. This coincides with my results that the wild boar, an omnivorous species, is one of the most frequent visitors to the studied salt-licks.

To clarify the purpose of salt-lick visit by those mammals, I needed to analyze their behavior at the salt-licks in addition to capture frequency and staying time. As I hypothesized, the water-drinking behavior at the salt-licks was also observed in herbivores more frequently than omnivores and not observed in the carnivores. Although nine out of 10 herbivorous species recorded at the salt-licks showed waterdrinking behavior, only one out of seven omnivorous species, the wild boar, showed the water-drinking behavior repeatedly. In addition, the frequently recorded herbivores, red muntjac, sambar, Malayan tapir and white-thighed langur showed water-drinking behavior in more than 70% of the observed individuals, while the wild boars showed the behavior in 23% of the observed individuals (<u>Table 3-6</u>). Since the omnivores and the carnivores are thought to have less need for mineral supplementation and/or alleviation of gastrointestinal acidosis than the herbivores, the results suggest that the wet type saltlicks of this area were mainly used by the herbivores for mineral supplementation and/or alleviation of gastrointestinal acidosis. The possibility that mammals rehydrate in the puddles of salt-licks cannot be ruled out, however, this alone does not explain the reason for frequent water-drinking behavior by only herbivores.

In tropical rainforest of Malaysian Borneo, Matsubayashi et al. (2007a) showed that the wet type salt-licks with greater concentrations of minerals in seepage soil water were significantly preferred by not only the herbivorous sambar, but also the omnivorous bearded pig, than those with lower concentrations of minerals. Therefore, it is possible that the wild boars in the study area also use the salt-licks for mineral supplementation. At the artificial salt-licks in wildlife reserves of Peninsular Malaysia, Simpson et al. (2020) reported that wild boars visit more frequently and spend longer time there than at control sites inside the forest, although in more than half of the records they were simply passing by or spending short time (less than 2 min) at the saltlicks. In addition, Simpson et al. (2020) who described the behavior of wild boars at the artificial salt-licks, reported that they were often wallowing or rooting up the ground. In this study, the wild boars also showed ground digging behavior repeatedly, probably searching for food. It is possible that the wild boars are attracted to the salt-licks not only for mineral supplementation or alleviation of gastrointestinal acidosis but for food or to wallow.

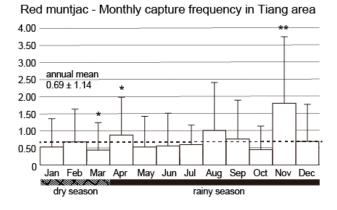
Several previous studies reported hunting behavior observed in salt-licks. Montenegro (2004) reported that carnivores such as jaguars (*Panthera onca*) and ocelots (*Leopardus pardalis*) were also recorded at the salt-licks in Peruvian Amazon. Matsuda and Izawa (2008) observed the killing of an adult male spider monkey by a jaguar and a predation attempt by a puma (*Felis concolor*) on an adult female spider monkey at a salt-lick in La Macarena, Colombia. The hunting behavior by the dholes was observed also in this study, however, this was the only record of hunting (the online video clips are available at <u>https://doi.org/10.1007/s10344-022-01600-y</u>). Further study is needed to clarify whether the hunting frequency is higher at salt-lick areas than in other places in the forests.

This study showed that most of the medium-large mammals that visited the wet-type salt-licks in the study area were herbivores and they often drink water there, while omnivores and carnivores drink water less frequently. These results suggest that the wet-type salt-licks in the study area were mainly used by herbivores, probably for mineral supplementation and/or alleviation of gastrointestinal acidosis. In addition, the salt-licks are suggested to be possible hunting sites for some carnivores. Future research should compare the capture frequencies of mammals between at salt-licks and at control sites (ex: streams used for their usual water-intake) to better clarify the importance of the salt-licks for mammals.

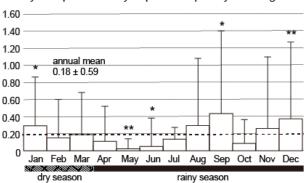
Tables and Figures

Fig. 3-1. The capture frequency of the red muntjac, the sambar, and the Malayan tapir in each month at the Tiang area.

According to the data of rainfall at Kg. Lalang, about 30 km west of the study site provided by Water Resources Management and Hydrology Division, the rainy season around BTFC was from April to December with the break from June to August during the study period. The asterisk (*) indicates the significant difference in the number of captures in a day between the month and entire year at P < 0.05; the double asterisk (**) at P < 0.001 (Wilcoxon rank-sum tests).



Sambar - Monthly capture frequency in Tiang area 3.00 ** 2.50 annual mean 0.46 ± 1.04 2.00 1.50 1.00 * 0.50 0 Jan Feb Mar Apr May Jun Jul Aug Sep Oct Nov Dec rainy season dry season



Malayan tapir - Monthly capture frequency in Tiang area

Fig. 3-2. The number of captures with / without water-drinking behavior by each food-habit mammal recorded at salt-licks.

The number in brackets shows the percentage of captures with water-drinking behavior. The black bar shows the capture with water-drinking behavior, and the white bar shows without such behavior.

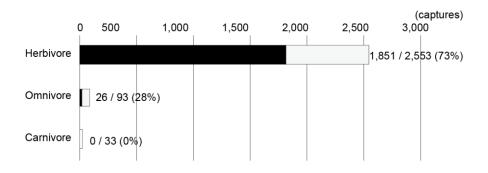


Table 3-1. The number of active cameras, captures with mammal records, days with active camera, and capture frequency.

	Number of active cameras	Number of captures with mammal records	Number of days with active cameras ^a	Capture frequency ^b		
Tersau	1–3	614	537	1.14		
Tiang A	1–3	781	507	1.54		
Taing B	1–3	685	498	1.38		
Tiang C	1–2	599	221	2.71		
Total	1-11	2,679	1,763	1.52		

Cameras were deployed between February 2014 and February 2016.

^aTotal days when at least one camera was active at each studied salt-lick

^bThe number of captures per day with active camera

Food habit	Scientific name	Common name	Number of captures	Capture frequency	Recorded group size
Herbivore	Muntiacus muntjak	Red muntjac	1,116	0.633	1–3
	Rusa unicolor	Sambar	558	0.317	1–4
	Tapirus indicus	Malayan tapir	405	0.230	1–2
	Presbytis siamensis	White-thighed langur	145	0.082	1–11
	Tragulus sp.	Mouse deer	118	0.067	1–2
	Elephas maximus	Asian elephant	105	0.060	1–10<
	Hystrix brachyura	Malayan porcupine	80	0.045	1–4
	Trachypithecus obscurus	Dusky leaf monkey	16	0.009	1–3
	Bos gaurus	Gaur	9	0.005	1–3
	Capricornis sumatraensis	Southern serow	1	0.001	1
Omnivore	Sus scrofa	Wild boar	76	0.043	1–14
	Macaca nemestrina	Pig-tailed macaque	9	0.005	1–3
	Arctogalidia trivirgata	Small-toothed palm civet	3	0.002	1
	Paradoxurus hermaphroditus	Common palm civet	2	0.001	1
	Hemigalus derbyanus	Banded civet	1	0.001	1
	Paguma larvata	Masked palm civet	1	0.001	1
	Helarctos malayanus	Sun bear	1	0.001	1
Carnivore	Panthera tigris jacksoni	Tiger	14	0.008	1
	Viverra zibetha	Large Indian civet	10	0.006	1
	Prionodon linsang	Banded linsang	5	0.003	1
	Cuon alpinus	Dhole	4	0.002	3–10

Table 3-2. List of mammals recorded at salt-licks, the number of captures, frequency, the average staying time, and the recorded group size.

Herbivore			Omnivore			Carnivore			
	WWF	this study		WWF	this study		WWF	this study	
Asian elephant	Х	Х	Wild boar	Х	Х	Banded linsang	Х	Х	
Malayan tapir	Х	Х	Banded civet	Х	Х	Large Indian civet	Х	Х	
Mouse deer	Х	Х	Common palm civet	Х	Х	Crab-eating mongoose	Х		
Red muntjac	Х	Х	Masked palm civet	Х	Х	Yellow-throated marten	Х		
Sambar	Х	Х	Binturong	Х		Oriental small-clawed otter	Х		
Southern serow	Х	Х	Small-toothed palm civet		Х	Dhole	Х	Х	
Gaur	Х	Х	Sun bear	Х	Х	Leopard cat	Х		
White-thighed langur	Х	Х	Long-tailed macaque	Х		Marbled cat	Х		
Dusky leaf monkey	Х	Х	Pig-tailed macaque	Х	Х	Asian golden cat	Х		
Brush-tailed porcupine	Х					Clouded leopard	Х		
Malayan porcupine	Х	Х				Leopard	Х		
						Tiger	Х	Х	

Table 3-3. The list of mammals recorded in Belum-Temengor Forest Complex; comparison between the prior WWF study and this study."WWF" in column headings indicates the WWF-Malaysia report by Rayan et al. (2013). X indicates species present.

Food habit	Species	Average staying time	Maximum staying time (minutes)		
Food haon	(recorded in more than 5 captures)	$(\pm SD)$ (minutes)			
Herbivore	Red muntjac ($n = 1,116$)	7.37 (± 12.83)	117		
(9 species)	Sambar ($n = 558$)	14.08 (± 21.28)	260		
	Malayan tapir (n = 405)	13.74 (± 19.20)	193		
	White-thighed langur $(n = 145)$	12.46 (± 15.37)	77		
	Mouse deer $(n = 118)$	2.80 (± 4.94)	30		
	Asian elephant ($n = 105$)	47.46 (± 78.70)	580		
	Malayan porcupine $(n = 80)$	2.56 (± 5.35)	31		
	Dusky leaf monkey $(n = 16)$	6.50 (± 7.77)	24		
	Gaur $(n = 9)$	56.44 (± 38.98)	128		
	Average of herbivores	10.16 (+ 10.62)	160 (± 166.43)		
	(n = 9 species)	18.16 (± 18.63)			
Omnivore	Wild boar $(n = 76)$	4.64 (± 7.28)	37		
(2 species)	Pig-tailed macaque $(n = 9)$	2.56 (± 4.40)	15		
	Average of omnivore species	2 (0	26		
	(n = 2 species)	3.60			
Carnivore	Tiger $(n = 14)$	1.50 (± 1.80)	8		
(3 species)	Large Indian civet (n = 10)	3.30 (± 6.90)	24		
	Banded linsang $(n = 5)$	1	1 11		
	Average of carnivore species	1.02			
	(n = 3 species)	1.93			

Table 3-4. The average and maximum staying time (minutes) of each species which was recorded in more than 5 captures.

Table 3-5. Comparison of the staying times between each herbivore species and other food-habit species which was recorded in more than 5 captures.

The inequality sign ">" indicates that the staying time of the herbivore species was significantly longer than the other food-habit species, the sign "<" indicates that significantly shorter (Wilcoxon rank sum test, P < 0.05). "NS" indicates that no significant difference (Wilcoxon rank sum test, P > 0.05).

	Omnivores		Carnivores	arnivores			
Herbivores	Wild boar $(n = 76)$	Pig-tailed macaque (n = 9) Tiger (n = 14)		Large Indian civet (n = 10)	Banded linsang $(n = 5)$		
Red muntjac ($n = 1,116$)	>	>	>	>	NS		
Sambar ($n = 558$)	>	>	>	>	>		
Malayan tapir (n = 405)	>	>	>	>	>		
White-thighed langur $(n = 145)$	>	>	>	>	>		
Mouse deer $(n = 118)$	<	NS	NS	NS	NS		
Asian elephant ($n = 105$)	>	>	>	>	>		
Malayan porcupine (n = 80)	<	NS	NS	NS	NS		
Dusky leaf monkey $(n = 16)$	NS	NS	>	>	NS		
Gaur $(n = 9)$	>	>	>	>	>		

	Average of group size	Water-drinking individuals / all recorded individuals in captures	Pearson's chi-squared test results
Red muntjac	1.14	913 / 1,271 (72%)**	$X^2 = 45.598, P < 0.001$
Sambar	1.25	535 / 700 (76%)**	$X^2 = 48.293, P < 0.001$
Malayan tapir	1.26	400 / 511 (78%)**	$X^2 = 48.16, P < 0.001$
White-thighed langur	3.32	431 / 482 (89%)**	$X^2 = 60.609, P < 0.001$
Wild boar	2.53	45 / 192 (23%)	_

Table 3-6. The rate of water-drinking individuals in four herbivores and one omnivore. The double asterisks (**) indicate significantly higher rate of water-drinking species than that of the wild boar at P < 0.001 (Pearson's chi-squared test).

			Camera days	Total species	Total captures				
Study area	Forest type	Camera	(No. of cameras	of medium-	(or photos) with	Herbivore	e Omnivore	Carnivore	Insectivore
Study area		installation site	* installation	large	medium-large	neibivoie			
			days)	mammals	mammals				
Taman Negara National						2038	870	374	
Park, Peninsular Malaysia	Primary forest	whole forest (over	14054	31 ^a	3288 ^b	(62.0%, 10	(26.5%, 9	(11.3%, 11	6 (0.2%, 1
(Kawanishi and Sunquist,	Timary lorest	200km ²)	14034	51	5288	(02.076, 10 species)	(20.576, 9 species)	species)	species)
2004)						species)	species)	species)	
Jerangau Forest Reserve,		whole forest (over				592	1100	429	
Peninsular Malaysia (Azlan	Secondary forest	170 km^2) and the	5972	33	2121	(27.9%, 8		, 8 (20.2%, 11	0 (0.0%, 0 species)
2006)	Secondary lorest	plantation-forest	3972				(51.9%, 8		
2000)		fringe				species)	species)	species)	
Imbak Canyon		whole forest (13				270	220		
Conservation Area,	Primary forest (surrounded by secondary	plots; each plot is	1426	27	5()	279	229	53 (9.4%,	3 (0.5%, 1
Malaysian Borneo (Bernard	forest)	approximately	1436	27	564	(49.5%, 9		9 species)	species)
et al. 2013)		3.5km in radius)				species)	species)		
	Krau WR: primary forest, Sungai Dusun								
Krau WR, Sungai Dusun	WR and Sungkai WR: secondary forest. All					670	724	0- <i>(1</i> 00 <i>/</i>	0 (0 00 (0
WR, and Sungkai WR,	the areas share borders with forested areas,	At 15 artificial	846°	21°	1421°	(47.1%, 7	(51.0%, 8	27 (1.9%,	0 (0.0%, 0
Peninsular Malaysia	local communities and large agricultural	salt-licks				species)	species)	6 species)	species)
(Simpson et al. 2020)	developments of oil palm and rubber.								
Belum-Temengor Forest		only around four salt-licks	2340			2553			
Complex, Peninsular	Primary forest			21	2679	(95.3%, 10	93 (3.5%,	33 (1.2%,	0 (0.0%, 0
Malaysia (this study)						species)	7 species)	4 species)	species)

Table 3-7. Comparison of camera-trapping results between previous studies and this study for forest and salt-lick areas in Malaysia

Chapter 4:

Mineral contents of salt-lick water and mammal visitation to salt-lick in tropical rainforests of Peninsula Malaysia

Summary

Salt-licks are considered to be important places for conservation of animals, however, functions of salt-licks were still not well studied especially in Peninsular Malaysia. In order to examine the possible mineral-supplementing function of salt-licks of this area, I compared mineral concentration between the water from salt-licks mainly visited by herbivorous mammals to drink water and nearby streams, possible water sources for mammals at the area. I also analyzed relationship between mineral concentration and salt-lick visit by mammals. Among analyzed minerals, only the concentrations of sodium and calcium were significantly higher in water from all studied salt-licks than those of nearby streams in both dry and rainy seasons, indicating that mammals could supplement these minerals by drinking salt-lick water. Herbivores most frequently visited the salt-lick with the sodium concentration significantly higher than other saltlicks. In contrast, omnivores and carnivores didn't show such tendency. Red muntjac (Muntiacus muntjak) and sambar (Rusa unicolor), the first and second most-frequent

visitors of salt-licks (41.7% and 20.8% of the visitation record by mammals, respectively), most frequently visited the salt-lick with highest sodium concentration. On the other hand, Malayan tapir (*Tapirus indicus*) and white-thighed langur (*Presbytis siamensis*), the third and fourth most-frequent visitors (15.1% and 5.4%, respectively), frequently visited the salt-lick with lower sodium concentration. The results are consistent with hypotheses that sodium supplementation is an important purpose of saltlick visit by herbivores, and indicated that other factors which would vary by species, such as social interaction, also affect selection of salt-licks to visit by herbivores.

4.1 Introduction

In Chapter 3, I analyzed the behavior of medium-large mammals recorded at wet-type natural salt-licks in the tropical rainforests of Belum-Temengor Forest Complex, Peninsular Malaysia. I showed that 95.3% of all records of animals at the saltlicks were herbivores, while the records of omnivores and carnivores were only 3.5% and 1.2%, respectively. In addition, I reported that water-drinking behaviors were recorded significantly more frequently in herbivores (73% of video captures) than in omnivores (28%) and carnivores (0%) at the salt-licks. These results suggested that animals, especially herbivores, visited the salt-licks to meet physiological needs such as mineral supplementation which are deficient in plant-based diets and/or alleviation of gastrointestinal acidosis related to plant-based diets by drinking the salt-lick water. It is still not clear, however, whether the water of these salt-licks contains a higher concentration of minerals and/or bicarbonate which have been reported to alleviate gastrointestinal acidosis by neutralizing increased rumen acidity (Ayotte et al. 2008; Davies and Baillie 1988) than the water of other places.

At the artificial salt-licks developed by DWNP from 2011-2012 in protected rainforests of Peninsular Malaysia, common course-grain salt (NaCl) and mineral blocks (enriched in sodium, phosphorous, calcium, magnesium, iron, cobalt, iodine, manganese, selenium and zinc) were added to newly dug pits together. The salt and mineral blocks were dissolved by rainfall and small pools of salty water were formed. Some ungulate species such as wild boars, Malayan tapirs, and red muntjacs were reported to drink water frequently from there, indicating that artificially added minerals attracted these animals (Simpson et al. 2020). There were still limited studies on the function of natural salt-licks of this area, however, I expected that some components such as minerals in the wet-type natural salt-licks can also attract herbivorous animals in this area, the same as these artificial salt-licks.

In this Chapter, to clarify whether the salt-licks in BTFC actually have the

mineral supplementing function for herbivorous mammals of this area and which minerals are important for their mineral supplementation, I compared the mineral concentrations of the water from the salt-licks and the reference sites. I also analyzed the relationship between mineral concentration and frequency of salt-lick visit by animals, supposing that herbivores would visit the salt-licks with higher concentration of more preferred minerals more frequently if mineral supplementation is a major reason for their salt-lick visits. In this study, I did not discuss the possible function of the salt-lick water to alleviate gastrointestinal acidosis of herbivores because I did not analyze bicarbonate concentration in the water.

4.2 Methods

4.2.1 Mineral analyses in the water of salt-licks

Mineral contents were compared between the water of the salt-licks and the nearby streams where mammals of those areas were likely to use for their usual waterintake, and examine the possibility that water of the salt-licks could provide minerals which were not available from the stream water. The method of sample collection followed Matsubayashi et al (2007a). The water samples were collected at four saltlicks and nearby streams in 4–5 February (dry season) and 18–19 November (rainy season) 2015. At the Tersau salt-lick, the water samples were collected right near the water-spring vents on the bottom of a puddle (about 1m x 1m size, 5cm deep) from where the bubbles were rising up. At three salt-licks in the Tiang area, the water-spring vent was not detected, and thus the water samples were collected randomly from the puddle. The four streams selected as the reference sites were at a distance of 10–20 m away from each salt-lick. Water samples of the streams were collected from the points on the same day as the sampling at the salt-lick, at a point where no surface inflow from the salt-lick was found upstream. Nine or 10 samples were collected using a syringe (Nipro syringe 20 ml 08753, Nipro corporation, Osaka, Japan) at each salt-lick and stream. All the samples were filtered through a syringe filter of 0.20 µm pore size (Sartorius, Minisart 17597, Sartorius AG, Goettingen, Germany) to remove debris and particulates. The concentration of sodium, potassium, magnesium, and calcium in the water samples were measured using Atomic Absorption Spectrometer (AAnalyst 800, Perkin Elmer, Shelton, USA). The water samples were collected and analyzed independently, without blending.

4.2.2 Camera trapping and data analysis

As mentioned in Chapter 2, I placed motion-sensitive infrared-triggered digital

cameras at the four salt-licks between 25 February 2014 and 24 February 2016 (730 days). At each of Tersau, Tiang A, and B salt-lick, three cameras were deployed. At Tiang C salt-lick, two cameras were deployed. I used these cameras in video mode and collected 1-min-long videos per trigger event. The videos recorded continuously at intervals of 30 minutes or less were considered to be not independent and counted as "one capture". Even if multiple cameras at the same salt-lick recorded an animal and/or group of animals walking into the salt-lick, the video records of this event were counted as "one capture", regardless of the group size. The capture frequencies (the number of captures per day with active camera) of each studied salt-lick and each species were calculated.

To analyze the relationship between mineral concentrations and frequency of salt-lick visit by medium-large mammals, I compared the mineral concentration in the salt-lick water and the capture frequencies of animals among the three salt-licks closely located to each other in the Tiang area. The salt-lick in the Tersau area was excluded from the analysis because it is separated by a high way from other studied salt-licks.

4.2.3 Statistical analysis

Wilcoxon rank sum test was used to compare the mineral concentration

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between the water from salt-licks and nearby streams. Steel-Dwass test was used to compare the mineral concentration among the three salt-licks at the Tiang area. Fisher's exact test was used to compare the record frequency of mammals among three salt-licks at the Tiang area. These tests were computed in R version 4.2.0. For all tests, a *p*-value of < 0.05 was taken to indicate statistical significance. Data are presented as the mean \pm standard deviation (*SD*).

4.3 Results

4.3.1 Mineral concentration of the salt-lick water and stream water

Sodium concentrations were significantly higher at all four salt-licks studied than the average of stream water both in dry and rainy season (Wilcoxon rank sum test, $P \le 0.004$, <u>Table 4-1</u>). Calcium concentrations were also significantly higher at most salt-licks than the stream water in both seasons ($P \le 0.001$, <u>Table 4-1</u>), except at Tiang A in rainy season. On the other hand, potassium and magnesium concentrations were not always significantly higher at the salt-licks than the stream water. Thus, I focused on the concentrations of sodium and calcium in the rest of my analyses.

4.3.2 Relationship between mineral concentration and mammal records

Capture frequencies of mammals at each salt-lick ranged from 1.14 to 2.71 captures / day, as shown in Chapter 3 (Table 3-1). The mammal species most frequently recorded at the studied salt-licks was red muntjac (41.7% of all animal captures, 0.633 captures / day), followed by sambar (20.8%, 0.317 captures / day), Malayan tapir (15.1%, 0.230 captures / day), and white-thighed langur (5.4%, 0.082 captures / day), as reported in Chapter 3. The red muntjacs and the Malayan tapirs were recorded at all the four studied salt-licks, while the sambar and white-thighed langur were distributed only in RBSP and recorded at three salt-licks in the Tiang area. The percentage of red muntjacs, sambars, Malayan tapirs and white-thighed langur in the total animal captures at the three studied salt-licks in the Tiang area was 41.0%, 27.0%, 10.6%, and 7.0%, respectively.

Compared among three salt-licks in the Tiang area, Tiang C salt-lick showed significantly higher sodium concentration than Tiang A and B salt-licks (Steel-Dwass test, Tiang C-Tiang B: t = 3.69, P = 0.00130; Tiang C-Tiang A: t = 5.41, P < 0.001, Fig. 4-1). Tiang C salt-lick also showed the highest calcium concentration among the three salt-licks, however, the difference was only significant between Tiang C and Tiang A

(Steel-Dwass test, t = 4.71, P < 0.001).

The record frequency of mammals with each food-habit was compared among the three salt-licks in the Tiang area (Fig. 4-2). The herbivores were recorded significantly more frequently at Tiang C salt-lick (590 captures, 2.67 captures / day) with the highest sodium concentration than at Tiang A and B salt-licks (Fisher's exact test, P < 0.001 for both pairs of Tiang C-Tiang B and Tiang C-Tiang A), while omnivores and carnivores showed no such tendency. The omnivores were recorded significantly more frequently at Tiang A salt-lick (39 captures, 0.077 captures / day) than Tiang B and C. The carnivores were recorded most frequently at Tiang B salt-lick (16 captures, 0.032 captures / day), though there was no significant difference among these salt-licks in the Tiang area (Fig. 4-2).

The red muntjacs and the sambars were recorded significantly more frequently at Tiang C salt-lick (352 captures, 1.59 captures / day, and 157 captures, 0.71 captures / day, respectively) than at Tiang A and B salt-licks (Fisher's exact test, P < 0.001 for both pairs of Tiang C-Tiang B and Tiang C-Tiang A), in accordance to the difference in sodium concentration (Fig. 4-2). The Malayan tapirs, however, were recorded at Tiang B salt-lick significantly more frequently (163 captures, 0.327 captures / day) than at Tiang A and C salt-licks (Fisher's exact test, P < 0.001 and P = 0.00655, respectively). In addition, the white-thighed langurs were also significantly less recorded at Tiang A salt-lick than at Tiang B and C salt-licks (Fisher's exact test, P < 0.001 for both pairs) and there was no significant difference in the capture frequency between at Tiang B and Tiang C (Fisher's exact test, P = 0.130).

4.4 Discussion

Comparison of mineral concentration between salt-lick water and stream water in this study suggested that the herbivorous mammals could supplement sodium and/or calcium by drinking water at the studied salt-licks; the concentration of sodium in saltlick water was higher than that of stream water regardless of seasons or sites, and that of calcium was also higher in most cases. On the other hand, those of potassium and magnesium in salt-lick water were not necessarily higher than the stream water, suggesting that animals could not always supplement these minerals much more by drinking the salt-lick water than by stream water (Table 4-1).

Some previous studies on wet-type salt-licks also reported the higher mineral concentration (including sodium and calcium) in the salt-lick water than the reference-site water in various regions (<u>Table 4-2</u>). For example, Clayton and MacDonald (1999)

who studied wet-type salt-licks visited by babirusa in Sulawesi Island, Indonesia reported higher concentration of sodium and calcium in the salt-lick water than in reference-site water. Matsubayashi et al. (2007a) who studied wet-type salt-licks in Borneo Island, Malaysia also reported higher mineral concentration (sodium, potassium, magnesium, and calcium) of the salt-lick water. In addition, these authors compared animal visits between two adjacent salt-licks at a distance of only 16 m and reported that the sambars and bearded pigs (*Sus barbatus*) visited the salt-lick with higher concentration of all four minerals more frequently than the other.

In this study, the herbivores most frequently visited the Tiang C salt-lick with the highest sodium concentration among the three salt-licks in the Tiang area, while the omnivores and carnivores did not show such tendency. Since omnivores and carnivores are thought to have less need for mineral supplementation than the herbivores, the result also suggests that the herbivores visited the salt-licks for supplementation of minerals, especially sodium.

Some previous studies also suggested that herbivores visited salt-licks mainly for sodium supplementation. Couturier and Barrette (1988) who studied salt-lick use by moose in Quebec, Canada reported that among the four wet-type salt-licks where they

observed moose, the one that had the most visits also had the highest sodium content. Moe (1993) who analyzed soils of dry-type salt-licks in southeastern Nepal showed that sodium concentration was significantly higher in soil from salt-licks than in reference soils, while other mineral concentrations were not. He also showed positive correlation between the sodium concentration and visit frequency by axis deer (Axis axis) using the data from 12 dry-type salt-licks in the study area. Holdø et al. (2002) who studied African elephants (Loxodonta africana) in a Kalahari-sand habitat, Zimbabwe analyzed the mineral concentration of the food plants, drinking water, and soils of dry-type saltlicks the elephants ate. They estimated that the elephants could not ingest enough amount of sodium for their requirements from the food plants and drinking water, while they could take enough amount of calcium and magnesium. They also reported that the salt-lick soils contained higher concentration of sodium than other soils, while the calcium and magnesium concentrations of salt-lick soils were lower than the food plants and drinking water, indicating that elephants supplement sodium, not calcium and magnesium, from salt-lick soils.

On the other hand, it is also reported that the salt-licks have an important role of supplementing not only sodium but also other minerals such as calcium for animals. Atwood and Weeks (2003), who studied the salt-lick use by white-tailed deer in Indiana, USA, reported that females visited salt-licks with various mineral contents more frequently than salt-licks with only sodium, pointing out that females during lactation might need minerals such as calcium and phosphorus in addition to sodium.

The calcium concentration in salt-lick water $(3.24 \pm 1.54 \text{ ppm}, \text{ range } 0.51 -$ 12.79, n = 79) analyzed in this study, however, was much lower in absolute values compared to those that reported by these previous studies (Clayton and MacDonald 1999: 90 ppm; Matsubayashi et al. 2007a: 83.4 ± 50.0 ppm, Table 4-2). Therefore, the supplementation of sodium seems more important than that of calcium for animals that visited the salt-licks in this study. In addition, the herbivores (especially the red muntjacs and sambars, which were the main users of the salt-licks) most frequently visited the Tiang C salt-lick with the highest sodium concentration among the three saltlicks in the Tiang area, while the calcium concentration at Tiang C was not always higher than other salt-licks. This suggests that sodium is likely more important than calcium in mineral supplementation of herbivores at the salt-licks of this area. I cannot eliminate, however, the possibility that the animals supplement calcium at the salt-licks, because calcium concentration in the salt-lick water was higher than that in the stream water in most cases.

It also cannot be denied that the animals supplement some minerals other than

sodium and calcium which were not measured in this study but critical for herbivore nutrition. Especially, phosphorous concentration and its relationship with salt-lick use by animals should be examined in future studies in this area because the artificial saltlicks containing phosphorous in addition to sodium and calcium succeeded to attract mammals in protected forest in Peninsular Malaysia (Magintan et al. 2015).

In contrast to the red muntjacs and sambars that visited Tiang C with the highest sodium concentration most frequently, the Malayan tapirs and the white-thighed langurs, the third and fourth most-frequent herbivorous visitors to the studied salt-licks in the Tiang area, did not visit Tiang C most frequently. The Malayan tapirs and the white-thighed langur visited Tiang B more frequently than Tiang C while sodium concentration at Tiang B was significantly lower than at Tiang C, though the difference in the white-thighed langur was not significant (Fig. 4-2). These results indicate that not all the herbivore species visited the salt-lick with higher sodium concentration more frequently. Thus, other factors which may vary with species could influence the frequency of salt-lick visit by animals in addition to the sodium concentration.

The micro-habitat preferences of each species were also considered to influence the visit frequency to the salt-licks by animals in this area. Razali et al. (2020)

analyzed the relationships between the physical environments around the salt-licks and the visit frequency by animals in BTFC. And they suggested that ungulates preferred the salt-lick with dense understory foliage that could be their foraging site, and arboreal primates and birds preferred the salt-lick with closed canopy cover that could provide perching site for them.

Lazarus et al. (2019) conducted camera trapping surveys at the same three saltlicks in the Tiang area as this study. Their results on the record frequency of red muntjac were not consistent with this study, while the results on sambar and Malayan tapir were consistent. They reported that the red muntjacs were recorded significantly more frequently at Tiang A than other two salt-licks based on the research of six months (the exact survey dates were not described), while in this study the red muntjacs were observed to visit Tiang C most frequently, followed by Tiang A and Tiang B based on the research of two years. Although they did not conduct mineral analysis of salt-lick water, they discussed that the red muntjacs might preferred Tiang A because of the thick, dense underbrush and vines around Tiang A where they could quickly hide from predators. They also speculated that the sambars were observed significantly more frequently at Tiang C because they can visit there in group due to the large open area in front of the water source in Tiang C.

Although it is still not clear why their results on red muntjacs are different from ours, the difference in survey season and studied period, however, could cause the difference because the red muntjacs could seasonally change the visit frequency to the salt-licks. In addition, the physiological demand for minerals could also seasonally change by pregnancy and lactation for example. Holdø et al. (2002) reported that the female African elephants spent more time than males for soil-eating at the dry-type licks. In Borneo, Matsubayashi et al. (2007b) reported that female sambars visited the wet-type salt-lick more frequently in rainy season than dry season while visit frequency of males did not seasonally change. Couturier and Barrette (1988) showed that the various age-sex (yearling or adult, male or female) moose had different seasonal patterns of salt-lick use, suggesting possible differences in mineral needs related to molting, antler grows, calf growth, lactation, or estrus.

Mineral analyses of the salt-lick water by this study showed that the wet-type salt-licks studied in BTFC could provide the sodium and calcium supplementation for the herbivorous mammals. Especially, the high concentration of sodium was consistent with the hypothesis that the herbivorous mammals visit the salt-licks mainly for sodium supplementation. However, the results on relationship between the mineral concentration of salt-lick water and visit frequency by mammals suggest that the factors other than sodium concentration might also influence the selection of salt-licks by each species.

Tables and Figures

Fig. 4-1. Comparison of mean (±SD) sodium and calcium concentrations in the water of three salt-licks in the Tiang area in RBSP.

The three salt-licks are closely located to each other and large-sized mammals in this area might easily access all of them. The graphs are shown in order of the average concentration. The asterisks (*) indicate significant differences at P < 0.05; double asterisks (**) at P < 0.001 (Steel-Dwass test).

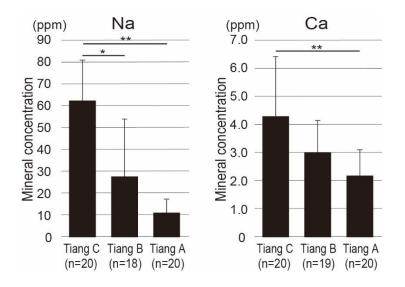


Fig. 4-2. Comparison of record frequency of mammals among three salt-licks at the Tiang area in RBSP.

The graphs are shown in order of the average sodium concentration. The asterisks (*) indicate significant differences at P < 0.05; double asterisks (**) at P < 0.001 (Fisher's exact test).

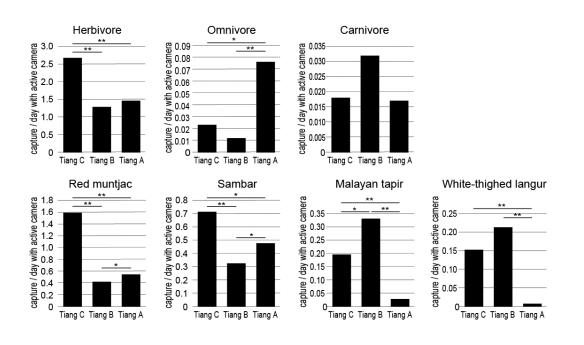


Table 4-1. Mineral concentration of each salt-lick water and those of nearby stream water in each season. The average concentration among four sites was calculated for streams. The asterisks (*) indicate that the mineral concentration of the salt-lick was higher than that of all stream sites at P < 0.05; double asterisks (**) at P < 0.001 (Wilcoxon rank sum test).

		Area	Salt lick (ppm)	(Range, N)	Ave. of streams (ppm)	(Range, N)	Test results
Sodium	Dry	Tersau	$38.30 \pm 7.97^{\ast\ast}$	(24.36–45.55, n=10)			W = 390, P < 0.001
	season	Tiang A	$11.00 \pm 1.22^{\ast\ast}$	(10.17–14.05, n=10)	5.06 + 2.11	(2.81–10.57,	W = 384, P < 0.001
		Tiang B	$41.02\pm 31.90^{\ast\ast}$	(11.22–79.01, n=9)	5.96 ± 2.11	n=39)	W = 351, P < 0.001
		Tiang C	$79.19 \pm 3.87^{\ast\ast}$	(73.28–84.85, n=10)			W = 390, P < 0.001
	Rainy	Tersau	$36.53 \pm 7.75^{**}$	(26.38–44.47, n=10)			W = 400, P < 0.001
	season	Tiang A	$10.66\pm9.18^{\ast}$	(4.69–27.75, n=10)	5 05 1 5 70	(2.25–24.95,	W = 320, P = 0.00373
		Tiang B	$13.19 \pm 12.82^{\ast}$	(5.74–46.38, n=9)	5.85 ± 5.72	n=40)	W = 300, P = 0.00202
		Tiang C	$45.00 \pm 12.01^{\ast\ast}$	(31.49–71.28, n=10)			W = 400, P < 0.001
Potassium	Dry	Tersau	2.13 ± 0.07	(2.01–2.20, n=10)			W = 102.5, P = 0.0224
	season	Tiang A	$3.13 \pm 0.09^{\ast \ast}$	(2.99–3.28, n=10)	0.04 + 0.05	(1.90–2.67,	W = 390, P < 0.001
		Tiang B	8.86 ± 19.14	(0.93–59.69, n=9)	2.34 ± 0.25	n=39)	W = 125.5, P = 0.191
		Tiang C	$6.20 \pm 1.54^{**}$	(3.95-8.42, n=10)			W = 390, P < 0.001
	Rainy	Tersau	1.80 ± 0.08	(1.68–1.90, n=9)			W = 90, P = 0.0208
	season	Tiang A	2.66 ± 1.05	(1.54–4.18, n=10)	2.02 + 0.25	(1.45–2.64,	W = 260, P = 0.149
		Tiang B	9.19 ± 24.00	(0.51–77.45, n=10)	2.03 ± 0.35	n=40)	W = 140, P = 0.149
		Tiang C	$41.16\pm 38.17^{**}$	(4.06–77.48, n=10)			W = 400, P < 0.001
Magnesium	Dry	Tersau	0.21 ± 0.04	(0.16–0.29, n=10)			W = 44.5, P < 0.001
	season	Tiang A	$2.52 \pm 0.52^{\ast \ast}$	(1.99–3.10, n=10)	0.04 + 0.64	(0.17–2.04,	W = 384, P < 0.001
		Tiang B	$1.80\pm0.94^{\ast}$	(0.25–3.02, n=9)	0.94 ± 0.64	n=39)	W = 274, P = 0.00962
		Tiang C	1.25 ± 0.27	(0.86–1.63, n=10)			W = 246, P = 0.21
	Rainy	Tersau	0.26 ± 0.11	(0.16–0.44, n=10)			W = 41, P < 0.001
	season	Tiang A	1.18 ± 0.60	(0.55–2.13, n=10)		(0.26–2.59,	W = 146.5, P = 0.199
		Tiang B	$2.40\pm1.09^{\ast}$	(0.79–3.62, n=10)	1.50 ± 0.77	n=40)	W = 305, P = 0.0112
		Tiang C	2.03 ± 2.55	(0.45-8.70, n=10)			W = 206, P = 0.894
Calcium	Dry	Tersau	$3.69 \pm 0.46^{**}$	(2.69–4.29, n=10)			W = 390, P < 0.001
	season	Tiang A	$2.78 \pm 0.14^{\ast \ast}$	(2.60–2.95, n=10)	1.00 + 0.74	(0.22–2.05,	W = 390, P < 0.001
		Tiang B	$2.61 \pm 0.95^{**}$	(1.19–3.81, n=9)	1.20 ± 0.74	n=39)	W = 309, P < 0.001
		Tiang C	$3.75 \pm 0.53^{\ast \ast}$	(3.16–4.51, n=10)			W = 390, P < 0.001
	Rainy	Tersau	$3.33 \pm 0.51^{**}$	(2.65–3.86, n=10)			W = 390, P < 0.001
	season	Tiang A	1.54 ± 1.02	(0.51–3.15, n=10)		(0.23–3.01,	W = 200.5, P = 1
		Tiang B	$3.36\pm1.27^{\ast}$	(1.44–5.72, n=10)	1.72 ± 0.90	n=40)	W = 333.5, P = 0.0012
		Tiang C	$4.78 \pm 3.10^{**}$	(2.70–12.79, n=10)			W = 388.5, P < 0.001

Table 4-2. Concentration of minerals in water-drinking type salt-licks.

The unit of mineral concentration is ppm. The asterisks (*) indicate that the mineral concentration of the salt-lick was higher than that of reference sites at P < 0.05 (¹t-test, ²Mann-Whitney U test, ³Wilcoxon rank-sum test).

			Na	(range)	K	(range)	Mg	(range)	Ca	(range)
Matsubayashi et al. (2007a), Borneo,	salt-lick	(n=59)	801.8 ± 1173.5*	(38.7–2710.2)	$14.4 \pm 12.6^{*}$	(6.8–29.8)	$21.4\pm9.8^*$	(13.6–35.1)	$83.4\pm50.0^{\ast}$	(41.7–155.9)
Malaysia ¹	Referenc e	(n=18)	6.9 ± 2.4	(4.6–8.7)	1.6 ± 0.6	(1.5–1.8)	2.7 ± 1.0	(2.3–3.1)	13.8 ± 8.5	(5.6–20.4)
Clayton and MacDonald (1999),	salt-lick	(n=1)	259	-	1.0	-	0.1	-	90	-
Sulawesi, Indonesia	Referenc e	(n=1)	7.1	-	< 0.1	-	6.1	-	25	-
Owen et al. (2014), Maine, USA	salt-lick	(n=2)	12.6	(9.4–15.7)	1.6	(1.0–2.3)	1.1	(0.6–1.6)	4.5	(3.9–5.0)
mane, corr	Referenc e	(n=2)	1.4	(0.9–1.9)	0.3	(0.2–0.3)	0.7	(0.5–0.9)	4.1	(3.0–5.1)
Couturier and Barrette (1988),	salt-lick	(n=17)	$172.5 \pm 97.1^{*}$	(73–472)	-	-	-	-	-	-
Quebec, Canada ²	Referenc e	(n=10)	3.6 ± 2.6	-	-	-	-	-	-	-
Bechtold (1996), British Columbia,	salt-lick	(n=20)	$107.8 \pm 119.7^{*}$	-	$2.7 \pm 3.3^{*}$	-	$10.0\pm12.0^{\ast}$	-	29.3 ± 32.2	-
Canada ¹	reference	(n=20)	2.1 ± 2.1	-	0.5 ± 0.4	-	4.4 ± 3.7	-	17.4 ± 12.2	-
this study ³	salt-lick	(n=79)	$34.5 \pm 25.2^{*}$	(4.7–84.9)	$9.5\pm20.7^{\ast}$	(0.5–77.5)	1.5 ± 1.3	(0.2–8.7)	$3.2 \pm 1.5^{*}$	(0.5–12.8)
	reference	(n=79)	5.9 ± 4.3	(2.2–25.0)	2.2 ± 0.3	(1.5–2.7)	1.2 ± 0.8	(0.2–2.6)	1.5 ± 0.9	(0.2–3.0)

Chapter 5:

Salt-lick use by wild Malayan tapirs (*Tapirus indicus*): behavior and social interactions at salt-licks

Summary

Salt-lick use and behavior around salt licks of wild Malayan tapirs were studied using infrared sensor cameras in the Belum-Temengor Forest Complex, Malaysia to observe different aspects of the ecology and behavior of this otherwise very elusive mammal for which we know little. All tapirs recorded at the salt-licks were adult-sized individuals, and they were observed at night. Water-drinking behavior was observed in 73% of the captures at the salt-licks, suggesting that they visited the salt-licks for physiological needs, e.g., to supplement plant-based diet by drinking water containing minerals. Only one or two tapirs (male-female combination) were recorded at the same time; no samesex individuals were recorded at the same time though both tapirs could be confidently sexed in 48% of the 88 captures containing records with two tapirs. Seven males and six females were identified. Each identified individual visited 1-3 salt licks, and multiple identified males and females were recorded at the same salt lick, suggesting the overlap of their home ranges. They were recorded around the studied salt licks once per 36 days

on average, though they could visit other salt licks, too. The frequency varied across individuals (once per 11–204). Analyses of one-night tapir recordings suggested that one particular individual or one male-female pair occupied a salt lick for 3–4 hours, indicating their exclusive behavior particularly between same-sex individuals. The results revealed the frequent salt-lick use by tapirs, suggesting importance of salt licks for their survival and social interactions.

5.1 Introduction

Four Tapiridae species are extant; three of them occur in South and Central America, and only Malayan tapirs inhabit Southeast Asia. Their main habitat is rainforest, wetland, and montane forest. Tapir has a long, flexible proboscis and good sense of smell and hearing. Tapirs utilize smell and voice, and touch each other by their proboscis in their communication. Previous studies on communication behavior in tapirs had mainly focused on those during copulation period in captive conditions. Not so many reports are available on the behavior of wild tapir because it is difficult to observe a nocturnal tapir directly in the dense forest. Especially, tapir is basically solitary, thus few studies have focused on their social interaction in wild.

Although few studies conducted direct observations of wild Malayan tapir,

several studies have suggested that they often visit salt-licks (Holden et al. 2003, Novarino 2005, Khadijah-Ghani 2010) in the rainforests of Peninsular Malaysia and Sumatra. For example, Novarino et al. (2004) reported that 63% of the footprints of Malayan tapirs were recorded at salt-licks in west Sumatra. Kawanishi et al. (2002) also documented that 12.7% of the wild animal images recorded along the trails leading to salt-licks in Taman Negara National Park were those of Malayan tapirs. Therefore, the direct evidence of salt-licks use by Malayan tapirs could provide valuable information concerning their ecology and conservation of them.

Despite these indirect evidences of the frequent salt-lick use of wild Malayan tapirs, very little direct evidence exists of the frequency of use and their behavior at saltlicks. Such data could provide valuable information concerning their ecology and conservation because it is possible that salt-licks are important places for Malayan tapirs where they need to visit frequently for their survival. Thus, in this chapter, I analyzed videos and photographs recorded by infrared sensor cameras established at salt-licks and along the animal trails leading to the salt-licks (gateways) in the Belum-Temengor Forest Complex (BTFC) located in northern Peninsular Malaysia. My objective was to characterize the salt-lick use by Malayan tapirs and their behavior around the salt-licks to understand the purposes of their visits. The salt-licks will offer a good opportunity to observe different aspects of the ecology and behavior of this otherwise very elusive mammal for which we know very little, and to get new insights for further studies. I identified individuals and analyzed their records around the salt-licks to estimate the frequency of their salt-lick visits. I also analyzed the group size and combination of individuals visiting a salt-lick at the same time, duration of stay and observed behavior to understand their social interaction around the salt-licks. I then discuss the purposes of the salt-lick visits and their social interaction, as suggested by the salt-lick use and behavior around there.

5.2 Methods

5.2.1 Camera trapping and data analysis

I placed the motion-sensitive infrared-triggered digital cameras at and around the four salt-licks between February 2014 and February 2016, however, in this Chapter, only the data recorded between 26 February 2014 and 9 April 2015 (408 days) was analyzed. As mentioned in <u>Chapter 2.3</u>, I set the cameras at each salt-lick and gateway (within 20 m of the salt-lick). I assumed that the animals recorded at the gateways had likely visited the corresponding salt-lick and that tapirs visited the salt-lick even in the cases when tapirs were recorded only at the gateway because the sensor cameras could not cover the entire area of each salt-lick. At the Tersau salt-lick, eight cameras were deployed (three at the salt-lick and five at each gateway). At each of the Tiang A, B, and C sites, six cameras were deployed (two at each salt-lick and four at each gateway).

I used the cameras in video mode or photograph mode. The triggering interval was a minimum of 20 s in video mode and 5 s in photograph mode. The photographs recorded within 1 min were counted as one picture, and then videos and pictures recorded continuously at intervals of 30 minutes or less were counted as "one capture". I verified the validity of this assumption through individual identification as described later (<u>Chapter 5.3.4</u>).

5.2.2 Individual identification

The individual Malayan tapirs recorded by the camera traps were identified based on scratches on the body and/or notches on the ears (Holden et al. 2003, Traeholt and Mohamed 2009, Rayan et al. 2012). The sex of each recorded tapir was determined by checking the genital area (Fig. 5-1).

5.2.3 Data analysis and statistical analysis

I calculated the time difference between the first and last record of each

identified tapir in one night as an index of their time of stay around a salt-lick (time-ofstay index). Chi-squared test was used to compare the rate of water drinking images between males and females recorded in video mode. Wilcoxon rank-sum tests were used to compare the record frequency, time-of-stay index between males and females. For both tests, a p-value of < 0.05 was taken to indicate statistical significance. Data are presented as the mean \pm standard deviation (*SD*).

5.3 Results

5.3.1 Tapir records

Tapirs were recorded in 533 captures over 6,326 camera days (1,404 videos over 5,391 camera days and 1,218 pictures over 935 camera days) in total during the study period. They were recorded around the salt-licks for a cumulative total of 265 days (13–128 days at each salt lick), which was an average of 16% of the observation days across all sites or once per 6.1 days (range, 4–31%; once per 3.2–28.2 days) (Table 5-1). Among three salt-licks at the Tiang area, the number of captures per camera day at salt-lick or gateway was highest at Tiang B (0.154), followed by Tiang C (0.092), and lowest at Tiang A (0.011), significantly (Fisher's exact test, P < 0.001 at all pairs).

Most of the tapir images were recorded during the night (Fig. 5-2). All tapirs

were recorded between 1750 h and 0903 h. The peak of their visit was observed between 2300 h and 0500 h.

5.3.2 Recorded tapirs

All recorded tapirs were estimated to be adults or sub-adults judging from body size and body color pattern (Donny et al. 2019). No individuals with the juvenile color pattern nor those with the remarkably smaller body size compared to other recorded tapirs were recorded.

The 391 out of 533 captures (73%) included sex-identifiable records for at least one individual. The 270 captures included male records, the 186 captures included female records and 65 captures included records of both male and female. This value could be biased toward males, as they are more easily identified than are females in terms of sex-determination (Fig. 5-1).

A maximum of two tapirs was recorded at the same time in the same video or photograph. The 88 captures containing records of two tapirs indicated that both remained at the salt-lick at the same time. The sex of both tapirs could be confidently determined in 135 videos and 15 pictures over 42 captures that containing records of two tapirs (48%, n = 88). In all these 42 captures, the sex combination of the two recorded tapirs was one male and one female; no same-sex combinations were observed. This result suggests that most of the two individuals recorded at the same time were adult sized male-female pairs.

In addition to the 88 captures that contained video or photograph recording two tapirs at the same time, 14 captures that contained records of both male and female were also counted as captures with two-tapir records. Two tapirs were recorded in 102 captures and one tapir were recorded in remaining 431 captures; the average number of recorded tapirs in one capture was 1.19 ± 0.39 (n = 533).

5.3.3 Observed behavior

One of the most frequently observed behaviors at the salt-lick was "waterdrinking behavior"; tapirs would often maintain a posture with the head down to the ground, touching their mouth to the salt-lick water for at least 1 second (as mentioned in <u>Chapter 3.2.2</u>). This behavior was observed more than once in 182 captures with the video recorded at the salt-licks (n = 251, 73%). No significant sexual difference was observed in the rate of water drinking captures (males: 87%, 131 of 151 captures, females: 73%, 72 of 99 captures) (chi-squared test, X^2 =0.64764, P = 0.421), though the sex of tapirs in 19% of the captures with video records at the salt-licks (n = 251) were not determined. On the other hand, no clear soil-eating behavior by tapirs nor traces of such behavior was observed at the salt-licks.

Vocalization was recorded in 33 captures with 61 videos over 27 days. Most of the recorded vocal sounds were similar to the sounds previously reported as "squeals" or "hiccups" in captive Malayan tapirs (Naundrup 2012). Although it was difficult to identify the specific individual that vocalized, I was able to observe one case of an interaction between a male and a female with vocalization in a video recorded at the gateway of Tiang B (02:45 AM, 16th July, 2014). In this video, the male chased the female uttering the hiccup-like sound, and the female escaped from the male producing the squeal-like sound (Fig. 5-3). A behavior of spraying urine backward was also observed (32 captures with 37 times in total over 24 days), especially in males (27 captures with 30 times over 20 days).

5.3.4 Identified individuals

In total, 13 individuals (seven males and six females) were identified (Fig. 5-1). The 40% of captures (n = 533) contained records with at least one identified individual; 28% of 1,752 tapir images recorded in videos and 9% of 1,446 tapir images in pictures were identified. While 27 captures contained the records of one identified male and one identified female, no capture contained more than two same-sex identified individuals, suggesting that the assumption of "one capture" in this study was appropriate.

Seven of the nine identified tapirs in the Tiang area where I studied three saltlicks visited multiple (2-3) salt-licks (<u>Table 5-2</u>). At each studied salt-lick, 2–8 identified tapirs including both sexes (1–4 males and 1–4 females) were recorded. At three of the four studied salt-licks (except Tiang A), multiple identified males (2–4) and females (2– 4) were recorded (<u>Fig. 5-4</u>). Each identified individual was recorded with 1–2 oppositesex pair partners at the salt-lick during the study period (<u>Table 5-3</u>). Specific combination of pair partners was not permanent and sometimes changed on a daily basis (<u>Fig. 5-5</u>). In addition, the same pair was sometimes observed at different salt-licks.

The identified tapirs were recorded at the studied salt-licks once per 36 days on average (0.029 ± 0.030 times/day, n = 13), although the frequency varied greatly across individuals (once per 11–204 days, 0.005-0.089 times/day) (<u>Table 5-2</u>). There was no significant difference in the record frequency between males (average 0.030 ± 0.031 times/day, n = 7) and females (average 0.027 ± 0.030 times/day, n = 6) (Wilcoxon ranksum test, W = 19.5, P = 0.8862).

I examined the records to estimate how many tapirs visited a salt-lick in one

night. Over the 133 days with records of identified tapirs, 2.4 ± 1.4 captures (with 11 ± 15 videos and 24 ± 27 pictures of tapirs) were recorded each night on average. These records included averages of 1.7 ± 0.9 and 1.5 ± 0.8 captures (with 4.2 ± 8.6 and 2.5 ± 3.5 videos and 6.4 ± 7.4 and 2.2 ± 2.2 pictures) of specific male and female individuals, respectively, indicating that the same identified individual was repeatedly recorded in one night (Fig. 5-6). In addition, no identified tapirs were recorded with other tapirs of the same-sex in the same night, suggesting that the visitor of the salt-lick in one night was limited to one particular individual or one male-female pair.

The average time-of-stay index, which estimates the time spent around the saltlick, was $204 \pm 156 \text{ min} (n = 71)$. The value did not differ significantly between males $(218 \pm 148 \text{ min}, n = 40)$ and females $(185 \pm 164 \text{ min}, n = 31)$ (Wilcoxon rank-sum test, W = 718.5, P = 0.2559). These results suggest that one particular individual or one male-female pair exclusively occupied the salt-lick for a long period during one night (at least more than 3–4 h on average, likely for the entire night).

5.4 Discussion

5.4.1 Tapirs observed around the salt-licks

All tapirs observed in this study were estimated to be adults. Since middle-

sized mammals such as civets were often recorded, my camera traps could record calfsized tapirs if they visited the salt-licks. However, I cannot deny the possibility that the adult pairs observed in this study included pairs of a mother and her adult-sized son, though the female-female pairs, possible mother-daughter pairs were not observed. The lack of observations of calves may be because no calves were born in this area during the study period and/or because mother tapirs did not bring their calves to salt-licks. The fact that a female adult and a calf-sized individual with the short proboscis and adult color pattern of black and white were recorded by camera trapping around Tersau saltlick in 2010 (Christopher C. T. Wong, WWF-Malaysia, Selangor, Malaysia, personal communication, April 2021), suggests the former possibility. On the other hand, no previous studies on Malayan tapir have reported salt-lick visits by small calves with the juvenile color pattern. In a study on lowland tapir (T. terrestris), Montenegro (1998) reported only one case of salt-lick visit by a mother with her calf with the juvenile color pattern. In this observation, however, he reported that the calf stayed outside the saltlick while the mother was in the salt-lick. This indicates the possibility that calves were not recorded by camera traps because they did not come in the salt-lick area.

Water-drinking behavior was frequently recorded at the salt-licks without sex difference, suggesting that they visited the salt-licks for some physiological needs.

Mountain tapirs (*T. pinchaque*) were also reported to drank water at salt-licks, though no quantitative data was shown (Lizcano and Cavelier 2000). As mentioned in Chapter 1.2, a number of studies has addressed physiological reasons of salt-lick use by herbivorous animals, such as mineral supplementation (Moe 1993, Clayton and MacDonald 1999, Ayotte et al. 2006, Matsubayashi et al. 2007, Molina et al. 2014), detoxification of plant secondary compounds by clay particle ingestion (Klaus et al. 1998, Gilardi et al. 1999, Houston et al. 2001), and alleviation of gastrointestinal problems (Bechtold 1996, Ayotte et al. 2006, Molina et al. 2014). Malayan tapirs are herbivores as other Tapiridae species that consume only plants as their food. They were reported to feed on the leaves, fruits, buds and stems of a wide variety of forest species in the wild (Holden et al. 2003, Khadijah-Ghani 2010, Simpson et al. 2013). Thus, it is possible that they visited the salt-licks for such physiological needs. In this study area, however, it is unlikely that animals visited the salt-licks for detoxification because ingestion of clay particles seemed very low at the studied salt-licks, as mentioned in Chapter 1.2. On the other hand, the water collected at the studied salt-licks contained significantly more sodium than the stream water of this area as shown in Table 4-1 in Chapter 4. Therefore, it is possible that tapirs visited studied salt-licks to supplement their plant-based diet by drinking water containing minerals such as sodium.

If tapirs visited the salt-licks for mineral supplement in this study, it is also possible that small calves with the juvenile color pattern were not recorded at the saltlicks because they still suckled breast milk that contained enough minerals, and were not needed to visit the salt-licks so often as adults for mineral supplement. It is because breast milk contains nutrients including minerals sufficient for the needs of young infants, in general. Although we have no reports on mineral contents of breast milk of tapir species, breast milk of white rhino, an odd-toed ungulate herbivore like tapirs, was reported to contain K (about 300 mg/L), Na (about 300 mg/L), Mg (about 60 mg/L) and Ca (over 500 mg/L) on average (Osthoff et al. 2021).

Two to eight tapir individuals were identified at each salt-lick in this study. Khadijah-Ghani (2010), who conducted camera-trapping survey and individual identification at Krau Wildlife Reserve, Peninsular Malaysia, reported that three to 14 tapirs were identified at each salt-lick or indigenous settlement. Thus, the number of individuals visiting one salt-lick that she reported is not so different from ours.

5.4.2 Estimated frequency of salt-lick visit

In this study, the frequency of record around the salt-licks varied greatly across identified tapirs (once per 11–204 days). The average value of record frequency (at least

once per 36 days) among individuals was probably underestimated as the frequency of their visit to the salt-licks, because it is possible that they visited other unknown salt licks near the study sites in addition to the studied salt-licks.

Although we have no previous reports on the frequency of salt-lick visits by wild Malayan tapirs, we have some reports on lowland tapirs. Tobler (2008) who conducted a GPS telemetry survey of five lowland tapirs reported that they visited the salt-lick once per 9.8 days on average (once per 6.2–14.5 days). In his report, the male that had a home range close to the studied salt-lick visited the salt-lick much more frequently than another male that had a home range far from the salt-lick, although the author did not mention it. These reports suggest that the tapirs visited the salt-licks near the home range more frequently than those far from the home range. If this is the case for Malayan tapirs I studied, their actual frequency of salt-lick visit would close to the values of the most frequent visitors, because individuals with low record frequency at the studied salt-licks might have visited other salt-licks close to their home range more frequently. The values of the most frequent visitors in my study (the male "M1": at least once per 11.3 days, the female "F2": at least once per 11.0 days, Table 5-2) are close to that in Tobler (2008) (once per 9.8 days on average), suggesting that frequency of saltlicks visit of both species is not so different.

At three studied salt-licks, multiple identified males and females were recorded at each salt-lick, though the record frequency varied greatly across individuals. The results suggest that their home ranges overlap each other to some extent (Fig. 5-4). If tapirs tended to visit the salt-licks closer to their home range more frequently, I can speculate spacial relationship among their home ranges from the record frequency of each individual at particular salt-lick. For example, M3 and F2, the most frequently recorded male and female at Tiang B salt-lick, could have home ranges near Tiang B that were close to and/or largely overlap each other. This can be a hypothesis about their home range distribution to be tested in future studies by other methods, e.g., GPS telemetry.

Among three salt-licks in the Tiang area which were closely located each other (Fig. 2-4), the record of tapir visit was most frequent at Tiang B, followed by Tiang C and least frequent at Tiang A. No obvious relationship between the record frequency and characteristics of each salt-lick, such as size of water drinking area, number of animal trails, vegetation and surroundings, were observed (Fig. 2-3). Lazarus et al. (2019) who studied records of various mammal species at these salt-licks also reported that Malayan tapirs were recorded most frequently at Tiang B among these 3 salt-licks

in the Tiang area. They analyzed topographical differences among these salt-licks and pointed out a possibility that Malayan tapirs visited Tiang B most frequently because more subcanopy trees, which were reported as their major diet, were observed around Tiang B than other salt-licks, though they did not conduct precise vegetation survey. Since I also observed more subcanopy trees around Tiang B and Tiang C than around Tiang A, abundance of subcanopy trees edible for them can be a factor affecting their visit to the salt-licks. To understand the difference in the record frequency among these salt-licks, however, I need to examine many other factors, such as seasonal change in the home range of each tapir individual in future studies.

5.4.3 Social interactions observed at the salt-lick

Several previous studies on Malayan tapirs have also shown that they were nocturnal and that the maximum group size at salt-licks was two as I showed in this study (Lynam 1999; Novarino 2005; Khadijah-Ghani 2010). However, no previous studies have provided data on the sex and/or growth stage of the paired individuals observed at the salt-licks at the same time.

My results strongly suggest that the same-sex adult Malayan tapirs avoided to visit a salt-lick at the same time and they remained at the salt-lick alone or with an opposite-sex individual for more than 3–4 hr during the night. The results suggest that same-sex adult individuals strongly avoided each other and that either a single individual or a particular male-female pair dominated a salt-lick for an entire night.

Although no other reports exist regarding salt-lick use by wild Malayan tapirs, several relevant studies have examined salt-lick use by South American tapir species. As for time-of-stay, previous direct observations of lowland tapirs at salt-licks revealed that the duration of each stay at a salt-lick was usually less than 40 min and that tapirs visited salt-licks several times per night (Montenegro 1998). In a study of six lowland tapirs using GPS telemetry, Tobler (2008) reported that they can spend several hours in the area surrounding a salt-lick. For mountain tapirs in the Central Andes, Lizcano and Cavelier (2000) used passive infrared monitors to show that the time tapirs stayed at a salt-lick ranged between 1 and 190 min. Together, these reports suggest that lowland and mountain tapirs also sometimes remain around a salt-lick for long periods of time, similar to the Malayan tapirs in the present study.

The male-female interaction similar to what I observed at the salt-lick (i.e., chasing and escaping with frequent vocalization) has previously been reported as a component of mating behavior in captive Malayan tapirs (Kusuda et al. 2008), lowland tapirs (Mallinson, 1969), and mountain tapirs (Padilla et al., 2010). Mallinson (1969)

reported that female captive lowland tapirs emitted frequent sounds similar to "squeals" when they were chased by males during courtship. Similarly, Padilla et al. (2010) reported that female captive mountain tapirs frequently squealed when they were chased and bitten by a male prior to copulatory behavior. Thus, many of the sounds recorded in the present study could be vocalized as a part of courtship. These results suggest that Malayan tapirs used the salt-licks as a place to meet potential mating partners in addition to drink water there, though no copulation was recorded in this study.

My results revealed the frequent visit to the salt-licks by Malayan tapirs, suggesting importance of salt-licks for their survival and social interactions. Therefore, protection of salt-lick areas is needed for conservation of this species. In addition, my results showed that salt-licks are important study sites where we can observe social interactions of this little-understood endangered mammal.

Tables and Figures

Fig. 5-1. Typical example of the captured images (or pictures) available for sex identification and 13 identified Malayan tapirs

Example of male tapir image



Example of female tapir image





F1 (female) was observed at Tersau over 7 days. It didn' t have auricle of both ears.



F3 (female) was observed at Tiang B over 7 days and Tiang C over 4 days. It had a scar on the top of head.

F2 (female) was observed at Tiang A over 1 day, Tiang B over 23 days and Tiang C over 13 days. It had two head scars and notches on both ears.



F4 (female) was observed at Tiang C over 3 days. It had notches on both ears.



F5 (female) was observed at Tiang B over 3 days and Tiang C over 5 days. It had a notch on left ear.





F6 (female) was observed at Tersau over 5 days. It was different from F1 because it had auricle of both ears. The border line of color around the hip was distinctive.



M1 (male) was observed at Tersau over 36 days. It had a notch on right ear and slits on left ear.



M3 (male) was observed at Tiang B over 25 days and Tiang C over 2 days. It had a large head scar but no notch or slit on ears.



M5 (male) was observed at Tiang B over 4 days and Tiang C over 1 day. It had scratches on left hind leg, but didn' t have any notches or slits on ears.



M7 (male) was observed at Tiang B over 1 day and Tiang C over 5 days. It had scratches on left front and hind legs, but didn't have any notches or slits on ears.



M2 (male) was observed at Tersau over 4 days. It had scratches on the base of right front leg, but didn't have any notches or slits on ears.



M4 (male) was observed at Tiang A over 3 days and Tiang C over 1 day. It had two slits on left ear.



M6 (male) was observed at Tiang B over 2 days. It had a scratch on the left side of hip, but didn't have any notches or slits on ears.



Fig. 5-2. The frequency of salt-lick visits by Malayan tapirs in each hour.

The bar under the graph shows light-dark phase. In the study area, the dark period was between 1900 h and 0700 h.

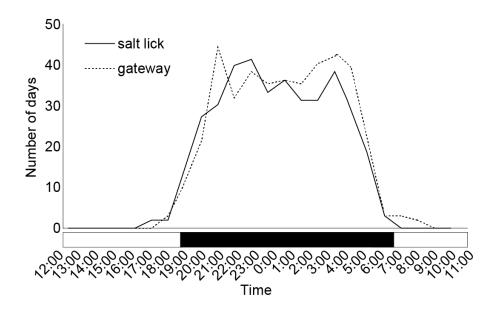


Fig. 5-3. Sound spectrograms of squeal-like sound by a female and hiccup-like sound by a male during a male-female interaction.

(Recorded at Tiang B salt-lick on 16th July 2014)

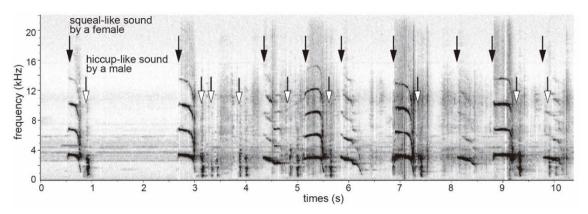


Fig. 5-4. The number of visits by identified individuals to each studied salt-lick. "M1–M7", "F1–F6" indicate the identified individuals.

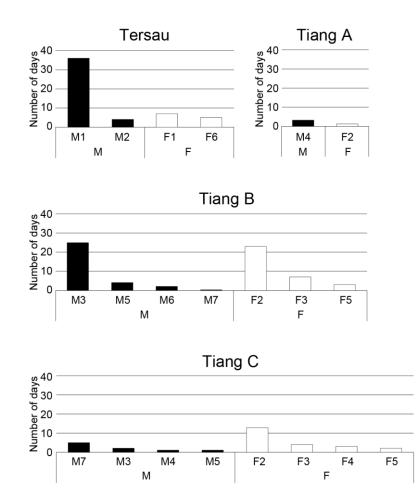


Fig. 5-5. Example of the record of individuals visiting a salt-lick.

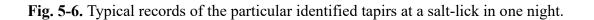
The "unID" means an unidentified tapir. F1 was observed with M1 for 3 days (21st August, 2014 and 15th, 20th November, 2014), and with M2 for 2 days (22nd and 23rd August, 2014) at Tersau in TFR. F2 was recorded with M3 for 5 days (2nd, 9th, 12th and 13th March, 26th April, 4th May, and 23rd September 2014), and with M7 for 1 day (14th December 2014). F2 and M3 were usually recorded at Tiang B salt-lick in RBSP, but recorded at Tiang C only once on 12th March, 2014.

Aug	ust	20	14

<u> </u>						
17	18	19	20	21	22	23
unID	unID,	M2	M2	M1,	M2,	M2,
	F1			F1	F1	F1
24	25	26	27	28	29	30
unID M		M1	unlD	unID M		

November 2014

9	10	11	12	13	14	15
						M1,
						F1
16	17	18	19	20	21	22
unID M	M1	unID M,		M1,		
		F1		F1		



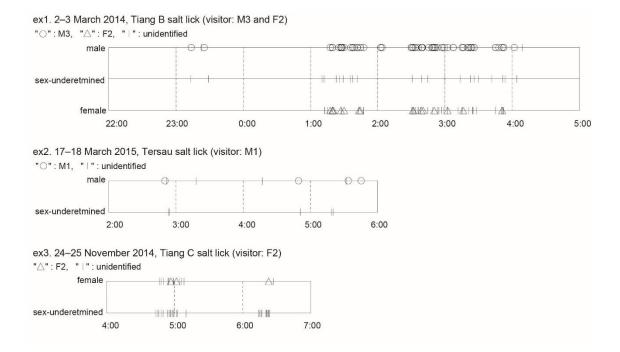


Table 5-1. The number of captures, the number of captures per camera day, and the rate of days with records of tapir visits.

The contents in [] shows the characteristics and the size of open area where animals drink salt-lick water. The number in () shows the days in which tapirs were recorded / the days in which at least one sensor camera was active without any issues.

Salt-lick		The number of	capture / camera day	The rate of days with records of
Salt-lick		captures	capture / camera day	tapir visits
Tersau [puddles and a	salt-lick	86	0.145	9% (32/340)
small stream merged	gateway	64	0.049	12% (51/405)
together, 23m x 17m]	salt-lick or gateway	150	0.079	13% (54/405)
Tiang A [one mud area,	salt-lick	14	0.025	3% (11/351)
10m x 10m]	gateway	3	0.003	1% (4/333)
	salt-lick or gateway	17	0.011	4% (13/366)
Tiang B [Two puddles,	salt-lick	151	0.319	27% (85/322)
1.5m x 1.5m respectively]	gateway	100	0.087	23% (94/408)
	salt-lick or gateway	251	0.154	31% (128/408)
Tiang C [one mud area,	salt-lick	55	0.169	15% (36/246)
1.7m x 5m]	gateway	60	0.065	15% (63/408)
	salt-lick or gateway	115	0.092	17% (70/408)

	Place	No. of video	No. of salt-lick visit	frequency of		Place	No. of video	No. of salt-lick visit	frequency of
	Place	(picture)	days	visit (/day)		Place	(picture)	days	visit (/day)
M1	Tersau	73 (76)	36	0.089	F1	Tersau	48 (9)	7	0.017
M2	Tersau	11 (0)	4	0.010	F2	Tiang A	0(1)	1	0.002
M3	Tiang B	212 (12)	25	0.061		Tiang B	58 (3)	23	0.056
	Tiang C	4 (2)	2	0.005		Tiang C	17 (14)	13	0.032
	total	216 (14)	27	0.066		total	75 (18)	37	0.091
M4	Tiang A	2 (1)	3	0.007	F3	Tiang B	11 (0)	7	0.017
	Tiang C	2 (0)	1	0.002		Tiang C	7 (1)	4	0.010
	total	4 (1)	4	0.010		total	18 (1)	11	0.027
M5	Tiang B	4 (0)	4	0.010	F4	Tiang C	2(1)	3	0.007
	Tiang C	1 (0)	1	0.002	F5	Tiang B	4 (0)	3	0.007
	total	5 (0)	5	0.012		Tiang C	2 (3)	2	0.005
M6	Tiang B	2 (0)	2	0.005		total	6 (3)	5	0.012
M7	Tiang B	1 (0)	1	0.002	F6	Tersau	6(1)	5	0.005
	Tiang C	20 (0)	5	0.012			Avera	ge of identified female	0.027
	total	21 (0)	6	0.015					
		Ave	rage of identified male	0.030					

Table 5-2. List of identified individual and no. of video (picture), frequency of salt-lick visit.No. of video (picture) shows the number of shots available for individual identification.

ID	The number of pair partners	ID of the partners		
M1	2	F1 (3 days), F6 (4 days)		
M2	1	F1 (2 days)		
M3	2	F2 (7 days), F3 (1 day)		
M4	1	unidentified tapir (1 day)		
M5	1	unidentified tapir (1 day)		
M6	1	F3 (1 day)		
M7	1	F2 (1 day)		
F1	2	M1 (3 days), M2 (2 days)		
F2	2	M3 (7 days), M7 (1 day)		
F3	2	M3 (1 day), M6 (1 day)		
F4	0			
F5	1	unidentified tapir (1 day)		
F6	1	M1 (4 days)		

 Table 5-3. The number of pair partners and the combination.

The number in parenthesis shows the total of days when the specific pair was observed.

Chapter 6:

General Discussion

6.1 Physiological function of wet-type salt-licks at BTFC

In this study, I conducted a camera-trapping survey on medium-large mammals in the forest of BTFC, Peninsular Malaysia, which is one of the biodiversity hotspots of the world. Understanding the purposes of visits to salt-licks by animals and the function of salt-licks in the rainforest ecosystem is necessary for conservation of various fauna. In Chapter 3, I analyzed the visit frequency, food habits, and behaviors of medium-large mammals visiting the wet-type salt-licks through 1-min video records collected by camera trapping. In the results, 95.3% of all video capture records of animals at the saltlicks were of herbivores. In addition, water-drinking behaviors were recorded significantly more frequently in herbivores (73% of video captures) than in omnivores (28%) and carnivores (0%). Therefore, the following hypotheses were correct in this area; if the purposes of the visit to these salt-licks are mineral supplementation or alleviation of gastrointestinal acidosis, (i) the salt-licks would be visited mainly by herbivores and (ii) water-drinking behavior would also be shown by herbivores much more frequently than omnivores and carnivores.

And in Chapter 4, I compared mineral concentration between the water from wet-type salt-licks and reference-sites. I also analyzed relationship between mineral concentration and frequency of salt-lick visit by animals. Water collected at the studied salt-licks contained significantly more sodium than the stream water of this area. The fact suggests that the herbivores which showed water-drinking behavior at the studied salt-licks visited there to supplement their plant-based diet by drinking water containing minerals such as sodium. The herbivores most frequently visited the salt-lick with the sodium concentration significantly higher than other salt-licks, while the omnivores and the carnivores didn't show such tendency. Red muntjac and sambar most frequently visited the salt-lick with highest sodium concentration. On the other hand, Malayan tapir and white-thighed langur frequently visited the salt-lick with lower sodium concentration. The following hypotheses were proved to be correct for a part of species: the herbivores would visit the salt-licks with higher mineral concentration more frequently if the mineral supplementation is a major reason for their salt-lick visits. However, the results also indicated that the factors which would vary by species also affect the selection of salt-licks to visit by herbivores.

A number of previous studies that analyzed the water in the wet-type salt-licks

also reported high concentration of minerals such as sodium, supporting the hypothesis that the main purpose of salt-lick use by herbivores is mineral supplementation (Tankersley 1981, Clayton and MacDonald 1999, Ayotte et al. 2006, Matsubayashi et al. 2007a, Owen et al. 2014). Besides, it is also possible that the animals visited the wettype salt-licks to alleviate gastrointestinal acidosis by drinking bicarbonate-rich water. Bechtold (1996) and Ayotte et al. (2006) studied salt-licks in the temperate forests of Northern British Columbia, Canada, and showed that salt-lick water or soil was richer, not only in sodium, but also in bicarbonate than control samples from nearby streams, suggesting that bicarbonate was important for deer species to maintain the proper digestive environment for rumen microbes to function efficiently. In addition, other previous studies indicated that the seasonal changes in visit frequency to salt-licks are also relevant to the needs for alleviation of gastrointestinal problems. Ayotte et al. (2008) who analyzed the records of elks (Cervus elaphus) at these wet-type salt-licks in Northern British Columbia reported that the frequency of both females and males increased in late May following vegetation greening. They discussed that eating more bicarbonate-rich soil at wet salt-licks in May could be important for this deer species to neutralize increased rumen acidity that is associated with the transition of food from fibrous winter forage to lush spring growth (Ayotte et al. 2006).

In <u>Chapter 3</u>, significant monthly variation of the capture frequency at the saltlicks was observed in three herbivorous species, red muntjac, sambar, and Malayan tapir (the most frequent visitors to the salt-licks), though the month with highest capture frequency was different for each species. The red muntjacs and sambars, however, visited the salt-lick significantly more frequently than the annual mean in April, the beginning of rainy season, though no clear seasonal change was observed in their visit frequency. At present, however, I have no precise studies on sprouting and fruiting phenology of this area, and seasonal dietary change of these species. The possibility cannot be eliminated that herbivores visit the salt-licks in the purpose of bicarbonate ingestion to alleviate gastrointestinal problems also in this area because the concentration of bicarbonate in salt-lick water was not measured in this study.

This study did not examine the effects of factors other than some mineral contents of the salt-lick water, however, many other factors, such as unmeasured contents in salt-lick water, could affect the pattern of animal visitation to the salt-licks. In order to understand the functions of salt-licks for the mammals more precisely, it is necessary to examine the effects of these factors on the salt-lick visit by mammals, which may vary by species, in future studies.

6.2 Effects of inter- and intra-species relationships on salt-lick use

Especially herbivorous species visit the salt-licks frequently for physiological reason. Therefore, it is assumed that encounters with other individuals of same/other species often occur. These factors are also possible to affect the visit frequency and behaviors at the salt-licks.

Since some carnivores, tigers and dholes (<u>Table 3-2</u>), and hunting behavior by dholes were recorded at the studied salt-licks as mentioned in <u>Chapter 3.3.2</u>, difference in predation risk among the salt-licks could affect the salt-lick visit by herbivores. Griffiths et al. (2020), who conducted camera trapping survey at 52 salt-licks in the northeastern Peruvian Amazon, showed that three mammals (the paca *Cuniculus paca*, Brazilian porcupine *Coendou prehensilis*, and red brocket deer *Mazama americana*) were less likely to visit salt-licks during nights with bright moon probably due to a heightened risk of predation at the salt-licks when visibility was better for predators. Although it remains to be studied whether the visit frequency of prey mammal is affected by lunar cycles also in this study area, the lunar cycle could not cause the difference in visit frequency among three salt-licks in the Tiang area because the difference was observed during the same study period.

Even the solitary species were sometimes observed staying together with other

individuals at the salt-lick areas. In Sabah state of Malaysia, Matsubayashi et al. (2011) studied the use of salt-licks by orangutans, which are arboreal species. They reported that multiple individuals such as two pairs of females with infants were recorded by sensor cameras in the same frame, nevertheless orangutans are usually solitary. This suggests that the salt-licks have secondary function as a communication site for this species. They strongly recommended that other production forests protect their natural-licks and manage them as monitoring sites for orangutan habitat conservation, and this proposal was actually accepted by the Sabah Forestry Department.

In <u>Chapter 5</u>, I studied the behavior of wild Malayan tapirs around the salt-licks and observed male-female interaction such as chasing and vocalization. These results suggest that they used the salt-licks as a place to meet potential mating partners in addition to drink water there. Thus, such social interaction can affect selection of the salt-licks by tapirs. Conversely, the loss of natural salt-licks could affect the survive and breeding of Malayan tapirs which are living in the vicinity. The evidences of frequent salt-lick use by endangered animals can support the plan for conservation of microhabitat including salt-licks.

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6.3 Conclusion and future research directions

Most of the medium and large mammals visiting the wet-type salt-licks in BTFC were herbivores; they often drank water there, while omnivores and carnivores drank water less frequently. This indicates that wet-type salt-licks are mainly used by herbivores. Mineral analysis of the salt-lick water indicated that the wet-type salt-licks in BTFC could potentially supply sodium and calcium to herbivorous mammals. In particular, the high concentration of sodium was consistent with the hypothesis that herbivores visit the salt-licks primarily for sodium supplementation. However, the relationship between the mineral concentration of salt-lick water and the frequency of visits to salt-licks by mammals suggests that factors other than sodium concentration, for example, other contents in salt-lick water, social interaction, or microhabitat preference might also influence the selection of salt-licks for each species. The results of mineral analyses providing better understandings on the salt-lick functions can contribute to improve the design of artificial salt-licks and the plans for mammal conservation in this area.

Malayan tapirs, in particular, were frequently observed at the wet-type saltlicks despite their endangered status. This suggests that salt-licks are important for their survival and social interactions in BTFC. It was also shown that salt-licks are important study sites, where social interactions of this poorly understood mammal can be observed. In addition, protection of salt-lick areas is needed for conservation of this species.

Combining the results of camera-trapping at the salt-licks with other survey results, such as GPS telemetry, will provide a deeper understanding of the home range, daily lifestyles, space use of each individual, and relationship between individuals. Further survey will clarify the conditions necessary to maintain the population of Malayan tapirs. Moreover, apart from the Malayan tapirs, records of other endangered species such as the gaurs and the dusky leaf monkeys at the salt-licks also provide clues for understanding their status in the region. In particular, the sambars, categorized as vulnerable (VU) in the IUCN red list, were also recorded very frequently at the studied salt-licks and social interaction among sambars can be observed in video records, although not yet analyzed in detail. For example, the male and female sambars were observed staying at the salt-licks, and two males were pitting their horns against each other. Detailed analysis of the camera-trapping records at salt-licks will lead to some new clues to the living conditions of poorly studied species in the area and be useful for planning of ecosystem conservation in this region.

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