

**OESTROUS BEHAVIOUR OF WILD-CAUGHT FEMALE
MALAYAN TIGERS (*PANTHERA TIGRIS JACKSONI*) IN
CAPTIVITY**

NUR QAMARINA BINTI SHAHARIN

DOCTOR OF VETERINARY MEDICINE

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MALAYAN TIGERS (*PANTHERA TIGRIS JACKSONI*) IN
CAPTIVITY**

By

NUR QAMARINA BINTI SHAHARIN

(D20A0073)

A THESIS SUBMITTED IN FULFILLMENT OF THE
REQUIREMENTS OF DOCTOR OF VETERINARY MEDICINE

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2024

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DATE: 8 February 2025

OESTRUS BEHAVIOUR OF WILD-CAUGHT FEMALE MALAYAN TIGERS (*PANTHERA TIGRIS JACKSONI*) IN CAPTIVITY

ABSTRACT

The Malayan tiger (*Panthera tigris jacksoni*) found in the Malay peninsula, a critically endangered species, is facing a rapidly declining population due to habitat loss, poaching and human-wildlife conflict. This research focuses on understanding the oestrus behaviour of wild-caught female Malayan tigers to aid in effective population and conservation strategies. The first objective was to identify specific behaviours related to the different phases of the oestrous cycle in these tigers. By observing their behaviours, including prustenings, and body movements, the study aimed to identify the oestrus behaviour in order to correlate with the oestrus phase. The second objective is to assess the influence of environmental factors on the oestrous cycle in wild-caught captive female Malayan tigers. Wild-caught female tigers were observed in enriched environments to see if it influenced their reproductive behaviours after being transferred to a captive and had to adapt with the new environment. This study was conducted at the National Wildlife Rescue Centre (NWRC) in Sungkai, Perak. There were two subjects involved in this study, which were wild-caught female Malayan tigers, Mek Santong and Yong Tawai. Their behaviour was observed to identify the timing of oestrous behaviour shown in order to get the overview pattern of oestrous cycle. This research study was conducted by visual observation through video surveillance without interrupting their behaviour with the presence of humans near their area for a total of 28 days in 252 hours. Based on this study, both Mek Santong and Yong Tawai had not been mixed with male in one night stall and only Mek Santong showed partial oestrous behaviour whereas Yong Tawai did not express any oestrous behaviour throughout the study. However, even though Mek Santong showed only partial oestrous behaviour, it is sufficient to indicate that she had experienced oestrus during the observation period. Therefore, these preliminary results can be used to identify oestrus behaviour in wild-caught Malayan tigress like Mek Santong. This would enhance captive breeding programs and support the conservation of Malayan tiger. While for Yong Tawai, she requires more time and observation, allowing her to adapt and adjust to the captive environment with presence of human for her to express natural behaviours.

TINGKAH LAKU ESTRUS HARIMAU MALAYA (*PANTHERA TIGRIS JACKSONI*)

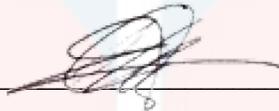
BETINA YANG DITANGKAP LIAR DALAM KURUNGAN

ABSTRAK

Harimau Malaya (*Panthera tigris jacksoni*) yang ditemui di Semenanjung Malaysia merupakan spesies yang sangat terancam dengan populasi yang semakin berkurang akibat kehilangan habitat, pemburuan haram, dan konflik antara manusia dan hidupan liar. Kajian ini bertujuan untuk memahami tingkah laku estrus pada harimau Malaya betina yang ditangkap dari alam liar dalam usaha strategi pemuliharaan yang lebih berkesan. Objektif pertama adalah untuk mengenal pasti tingkah laku tertentu berhubung dengan fasa-fasa dalam kitaran estrus harimau ini. Pemerhatian tingkah laku, ngauman, dan pergerakan badan mereka adalah bertujuan untuk mengenal pasti hubungan tingkah laku estrus dan kitaran estrus. Untuk menilai pengaruh faktor persekitaran terhadap kitaran estrus pada harimau Malaya betina yang ditangkap liar dalam kurungan. Kajian ini dijalankan di Pusat Menyelamat Hidupan Liar Kebangsaan (NWRC) di Sungkai, Perak. Dua subjek yang terlibat dalam kajian ini adalah harimau betina Malaya yang ditangkap dari alam liar, Mek Santong dan Yong Tawai. Tingkah laku mereka telah diperhatikan untuk mengenal pasti masa dan pola tingkah laku estrus yang berlaku bagi mendapatkan gambaran keseluruhan kitaran estrus. Kajian ini dijalankan melalui pemerhatian visual dengan menggunakan pemantauan video tanpa mengganggu tingkah laku mereka dengan kehadiran manusia di sekitar kawasan mereka, selama 28 hari bersamaan 252 jam. Berdasarkan kajian ini, kedua-dua Mek Santong dan Yong Tawai tidak dicampur bersama harimau jantan dalam kandang malam yang sama dan hanya Mek Santong menunjukkan beberapa tingkah laku estrus, manakala Yong Tawai tidak menunjukkan sebarang tingkah laku estrus sepanjang kajian dijalankan. Walaupun Mek Santong hanya menunjukkan sebahagian tingkah laku estrus, ini sudah cukup untuk menunjukkan bahawa dia telah mengalami estrus sepanjang tempoh pemerhatian. Manakala Yong Tawai mungkin mengalami fasa anestrus di mana dia tidak menunjukkan tanda-tanda estrus dan tidak bersedia untuk mengawan. Oleh itu, hasil awal ini boleh digunakan untuk mengenal pasti tingkah laku estrus pada harimau betina Malaya seperti Mek Santong yang ditangkap dari alam liar. Ini akan membantu mempertingkatkan program pembiakan dalam kurungan dan menyokong usaha pemuliharaan harimau Malaya. Manakala bagi Yong Tawai, ia perlu diberikan lebih masa dan pemerhatian untuk memastikan dia mulai menyesuaikan diri pada persekitaran dalam kurungan dan kehadiran manusia di sekeliling, dan boleh menunjukkan tingkah laku semulajadinya.

CERTIFICATIONS

This is to certify that we have read this research paper entitled **Oestrous Behaviour Of Wild-Caught Female Malayan Tigers (*Panthera tigris jacksoni*) In Captivity**’ by Nur Qamarina **Binti Shaharin**, and in our opinion, it is satisfactory in terms of scope, quality, and presentation as partial fulfillment of the requirements for the course DVT 55204 – Research Project.



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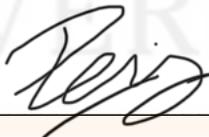
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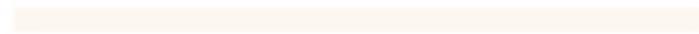
Finally, I would also like to dedicate this project to my classmates, Neofelis (DVM XII), and also my housemate, Pakistan Harapan, who stood by me offering both emotional and intellectual support as I completed this study. To all of you, I am truly grateful, and this work would not have been possible without your love, guidance, and endless support.

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LIST OF ABBREVIATIONS

- NWRC - National Wildlife Rescue Center
- DNA - Deoxyribonucleic acid
- IUCN - International Union for Conservation of Nature
- EN - Endangered
- TRAFFIC - The Wildlife Trade Monitoring Network
- DWNP - Department Wildlife National Parks
- MTCC - Malayan Tiger Conservation Centre
- MyBIT - Malayan Tiger Breeding In Captive Program
- NGO - Non-governmental Organizations
- WWF - World Wide Fund for Nature
- CL - Corpus Luteum
- GnRH - Gonadotropin - releasing Hormone
- LH - Luteinizing Hormone
- ID - Identification

CHAPTER 1

INTRODUCTION

1.1 Research Background

Panthera tigris jacksoni commonly known as Malayan tiger, one of the smallest tiger subspecies, lives in the southern and central portions of the Malay Peninsula, as well as southern Thailand. This majestic feline, Malaysia's national symbol, was only recognised as a unique subspecies in 2004, after previously being misidentified as the Indochinese tiger. The Malayan tiger, with its brilliant orange coat and tiny black stripes, is a master of camouflage, allowing it to hunt prey discreetly or withdraw into deep forest for protection. The Malayan tiger, like all tigers, is a solitary animal, though social dynamics change during mating season, which lasts from November to March (Malaysian Wildlife, 2021).

With only a few hundred Malayan tigers remaining in the wild, Malaysia is on the verge of losing its unique tiger subspecies. The world faces the awful reality of losing yet another subspecies, after three have already gone extinct: the Bali, Caspian, and Javan tigers. The greatest risks are habitat loss and fragmentation, which cause prey shortages and a significant drop in tiger populations. Because these wonderful animals require broad home ranges, destruction and urbanization are forcing them into increasingly isolated areas of forest. As a result, conflict between humans and tigers has increased in recent years. Furthermore, unlawful poaching and hunting for their body parts endanger the remaining Malayan tiger population, emphasising the critical need for conservation measures to safeguard these iconic beasts. (Malaysian Wildlife, 2021)

The Malayan tiger (*Panthera tigris jacksoni*) is recognized as a distinct subspecies with unique DNA (Kawanishi, 2015) which make both of it rare and irreplaceable. Within nearly seven decades of persecution and prime habitat degradation (Sharma, 2006; Shevade, 2017), the population plummeted drastically from 3000 individuals in the 1950s to less than 200 animals in 2019. The Malayan tiger population was predicted to be 500 animals in 1990 (Topani, 1990), but had dropped

to 250-340 by 2013 (Kawanishi, 2015). Unfortunately, the dramatic tendency has continued to worsen to the point of a near-population crash. The estimated wild Malayan tiger population in 2018 was less than 200 (Harimau.my, 2024). As a result, the wild Malayan tiger population has declined by almost 60% over the last three decades, dating back to 1990, prompting the International Union for Conservation of Nature (IUCN) to classify it as critically endangered (Stoner *et al.*, 2016). However, in 2022, this species was classified as endangered (EN) based on the IUCN Red List of Threatened Species, Peninsular Malaysia (version 3.1) (MyBIS, 2024).

Malaysia was not previously recognized as a key player in the trade of tiger products; nevertheless, recent TRAFFIC surveys and confiscations by the Department of Wildlife and National Parks (DWNP) show that poaching and trafficking have resurfaced as important challenges to tiger conservation efforts. Both poachers and traffickers have reported a drop in tiger populations due to overhunting, prompting greater efforts to locate them. Human-tiger conflicts worsen the Malayan tiger's problems (Kawanishi *et al.*, 2009).

The Department of Wildlife and National Parks of Peninsular Malaysia (DWNP) had implemented a Malayan Tiger breeding programs called as Malayan Tiger Breeding In Captive Program (MyBIT) with the aim to increase Malayan tiger's population which this program involved 7 facilities for the placement of Malayan tigers. There are five zoos consisting of Zoo Negara, A'Famosa Safari Wonderland, Zoo Melaka, Zoo Taiping & Night Safari as well as Zoo Kemaman and including another two facilities, which are National Wildlife Rescue Centre (NWRC) and Malayan Tiger Conservation Centre (MTCC).

1.2 Problem Statement

Further decline in Malayan tiger wild population with a total population from an estimated 3000 individuals to fewer than 150 individuals in the fragmented forest of Malaysia, mainly due to the threat of hunting pressure for illegal trade and habitat loss as a result of urbanization (Abdullah, 2022; Ten *et al.*, 2021; Alberry *et al.*, 2024) despite the ongoing conservation efforts by various domestic and international organizations (World Wide Fund for Nature Malaysia, n.d.) with zoos and conservation center play a very important role in the managed care and rehabilitation of the big cat. The relentless poaching, illegal trade, habitat loss and dwindling prey availability (Kawanishi, 2015) primarily drive this sharp decrease in the Malayan tiger population. These disturbing trends underline the critical need for governments, non-governmental organizations (NGOs) and policymakers at all levels to work together to safeguard Malayan tigers and their habitats. However, successful captive breeding is not recorded despite several attempts and this could be due to the gap in literature about estrous behavior of captive wild Malayan tiger and or other factors, thus results from this study is important toward providing and understanding reproductive behavior of the captive female tiger for successful breeding program.

1.3 Research Question

1. What are the oestrus behaviours of wild caught female Malayan tigers (*Panthera tigris jacksoni*) in captivity.
2. Which specific behavioural indications are reliable for detecting oestrus phases in wild-caught female Malayan tigers (*Panthera tigris jacksoni*)?
3. How does the oestrous behaviour of wild-caught Malayan tigers differ from that of captive-bred females?

1.4 Research Hypothesis

1. Behavioural indicators in captive wild-caught female Malayan tigers (*Panthera tigris jacksoni*) will be significantly correlated with estrus.
2. Specific estrous behaviour will be reliable to detect oestrus phases in captive wild-caught Malayan tigers.

3. Wild-caught and captive-bred Malayan tigers display oestrous behaviours differently.

1.5 Research Objectives

1. To identify and describe behavioural indicators of oestrus in captive wild-caught female Malayan tigers (*Panthera tigris jacksoni*).
2. To assess if specific behavioural indicators would be more reliable to determine oestrus in wild-caught female Malayan tigers.
3. To compare oestrous behaviours of wild-caught and captive-bred Malayan tigers.



CHAPTER 2

LITERATURE REVIEW

2.1 Animal Behaviour

Animal behaviour is the study of how animals move in their environment; how they interact socially, learn about their surroundings, and how an animal might develop cognitive knowledge of its surroundings (Breed & Moore, 2015). Species-specific behaviours, for example captive big felines exhibit abnormal repetitive behaviour, pacing, which this behaviour can be a sign of frustration and stress, particularly in situations where animals feel they lack control over their surroundings (Bashaw *et al.*, 2007). Visual contact without the option to engage in appropriate behaviour has been linked to increased pacing (Meulendijks *et al.*, 2024). Behaviours are often driven by strong internal motivations and are rewarding for the animal to express. Frustration and tension can arise when an animal is unable to accomplish the desired behaviour. To promote mental health, animals should be able to engage in their full range of behaviours.

2.2 Oestrous cycle

Tiger reaches sexual maturity at three to four years in females and four to five years in males (Mazak, 1979). Tiger mating can occur throughout the year, but is most common between November and April (Sankhala, 1978). Female oestrous cycles can be highly variable, ranging from 18 to 40 days, with the oestrus phase lasting approximately a week (Seal *et al.*, 1985; Wildt *et al.*, 1998; Graham *et al.*, 2006). According to Mazak (1967), female tigers have a receptive period of three to six days and oestrus every three to nine weeks. Female tigers are known as induced ovulators, requiring mating causes the release of an egg for fertilization to stimulate ovulation. This intriguing process frequently involves several days of mating activities to

adequately promote ovulation and ensure egg fertilization (SeaWorld Parks & Entertainment, Inc., 2024).

Female cats' oestrous cycle consists of four separate phases: proestrus, oestrus, dioestrus, and anoestrus (Wildt *et al.*, 1998). Proestrus normally lasts less than a day and is distinguished by the presence of growing ovarian follicles and increased oestrogen levels, which frequently attract male attention without culminating in copulation. In proestrus the female showed heightened interest in males by more frequent chuffing and head rubbing, which corresponds with several behavioural studies on felid cyclicity (Brown 2010, Kutzler 2007, Schaller 1972). Oestrus, which is characterized by advanced follicular growth and peak oestradiol levels, is followed by mating behaviours including prustening, lordosis, rolling, rubbing and tail lifting, as well as male mounting. During the dioestrus phase, one or more corpora lutea (CL) release progesterone, which remains elevated for different periods regardless of conception. The anoestrus phase, on the other hand, occurs between waves of follicular growth and is distinguished by low circulating oestrogen levels (Brown, 2010).

Tigers usually will reach their puberty at the age of 3 - 4 years old. Female reproductive cycles in many species even among felids including tigers can be highly variable in terms of both timing and duration of the cycles. The reproductive cycles of a tiger typically span over a period of 18 - 40 days and the variability can be influenced by other factors such as environmental condition, health, age, and individual differences in the tiger. In reproductive cycles, there were four phases of the oestrous cycle, proestrus, oestrus, dioestrus and anoestrus. Proestrus typically lasts less than a day and is characterized by the presence of ovarian follicles, an increase in circulating oestrogens and occasionally male interest while copulation does not occur during this phase. During proestrus, common behaviour that may be shown by tiger would be restlessness, aggression, low appetite, and urine spraying.

The oestrous cycle phase in which a period of females is sexually receptive and are capable of mating generally lasts about a week (Seal *et al.*, 1985; Wildt *et al.*, 1998; Graham *et al.*, 2006). During this period, hormonal changes in the female's body triggers the oestrus behaviour that signal her readiness to mate. Elevated oestrogen levels are correlated with the behavioural signs

shown by females such as prusten with male, followed by rubbing, rolling and lordosis (Seal et al., 1987). However, the length and intensity of oestrus can vary among individuals as we can see from this study both of the wild-caught tigers showed different behaviour. During dioestrus, corpus luteum (CL) secrete progesterone which remains elevated for varying durations regardless of whether conception takes place. Whereas anoestrus refers to the phase between follicular development cycles, when circulating oestrogen levels are at their lowest (Brown, 2010b). During dioestrus and anoestrus, the obvious signs shown would be resting most of the time.

2.3 Induced ovulators

One of the key reproductive strategies in female tigers is an induced ovulation, in which mating will trigger the release of eggs from the ovaries. Unlike spontaneous ovulators, who ovulate on a regular cycle, female tigers require copulation to initiate the process. Most felids require multiple mating interactions over several days to stimulate the release of gonadotropin-releasing hormone (GnRH) from the medial basal hypothalamus, which leads to surges of luteinizing hormone (LH) from the anterior pituitary gland (Shille *et al.*, 1983; Johnson and Gay, 1981). This complex hormonal cycle is necessary for the final development of follicles and oocytes, which leads to ovulation after mating. This process enables female tigers to synchronize ovulation and mating, increasing their chances of successful reproduction.

2.4 Oestrus behaviour

The behaviour from the female towards other individuals is a very important tool for determining the cycle stage. During oestrus, a female tiger will often present herself to the male, ready to mate; a behaviour that is not observed in other phases of the cycle. When not in oestrus, the female showed less interest in and less positive interactions toward males. Male and female tigers typically engage in mating interactions that involve increased vocalization, purring, chuffing, and moaning. Key female indicators of oestrus include increased scent spraying and frequent water consumption. Malayan tigers rely significantly on vocalization throughout the mating phase. Female tigers in estrus frequently make a characteristic, low frequency sound called "prusten," which involves exhaling air through their parted lips and nose. This sound is used as a greeting signal when a

female approaches a male in a pre-mating situation, and the male swiftly reciprocates, showing his eagerness to mate. Both the female and the male make this vocalization before participating in rubbing behaviours and the male follows the female during the mating phase (Palita, 1996).

Furthermore, behavioural cues like rolling is an important indicator of oestrus in female Malayan tigers. During this time, rolling behaviour helps spread pheromones, which improves scent marking and attracts males (Umopathy *et al.*, 2006). During oestrus, behaviours like vocalization, rolling and rubbing are more frequently displayed (Brown 2010).

The lordosis reflex is an important behaviour in female mammals that happens during mating. It involves the female arching her back in response to a male's mounting, providing for optimal copulation placement (Beach, 1976). This position is aided by hormonal changes, particularly oestrogen, which prepare the female body for mating. By expressing this reaction, the female expresses her desire to mate and her sexual receptivity to the male. This behaviour not only improves the chances of successful mating, but it also helps with reproduction by guaranteeing effective communication between mates and boosting the likelihood of conception (Guarraci & Frohardt, 2019).

2.5 Ethology

Ethology is defined as the scientific exploration of animal behaviour, with an emphasis on behaviour in natural environments and the idea of behaviour as an evolutionary adaptive attribute. Ethology, a branch of zoology that involves systematic and unbiased study of animal behaviour, often emphasizing behaviours observed in natural environments and viewing behaviour as an evolutionary adaptive trait. It offers a scientific lens through which to understand various aspects of animal behaviour including influenced by other factors (Rathod, 2019).

CHAPTER 3

RESEARCH METHODOLOGY

3.1 Study Area

The study focused on two wild-caught female Malayan Tigers and was conducted at National Wildlife Rescue Centre (NWRC), Sungkai, Perak under the Department of Wildlife National Parks (DWNP), Peninsular Malaysia, which is situated on the western side of Perak in the Batang Padang district. The NWRC was established within Sungkai Wildlife Reserve, Perak in 2018. The wildlife reserve was founded in 1928 with a total area of 1805 hectares and was expanded to 3966 hectares in 1940. In 1957, the land was gazetted, with 428 hectares removed for rubber plantation and other purposes, leaving only 2,468 hectares remaining (Department of Wildlife and National Parks [DWNP], n.d.). This wildlife rescue centre serves as an ex-situ conservation hub with the aim to protect endangered species by relocating them from their natural habitats to safer environments. The primary goal is to conserve wildlife, but it also seeks the sustainability of breeding programs for species reintroduction, promote in wildlife research and education, raise public awareness on conservation and contribute to eco-tourism through both ex-situ and in-situ species management. At NWRC, in addition to Malayan tiger, several species are conserved including Sun Bear, different kinds of aviary species, Malayan tapir and gibbon. Malayan tiger unit consists of five blocks, A, B, C, D and E, with each of the blocks having four night stalls and two exercise yards.

In the year 2022, there were 19 tigers in the rescue centre but few were distributed to Zoo's and other centres thus currently left with a total of 15 tigers. All the tigers were kept in night stalls and only a few will be released to the exercise yard every morning until evening and will come back into stalls for feeding time. The daily cleaning routine is at around 9:30 am and tigers will be transferred to the next stall or keeper walk hall temporarily while cleaning their stall. The feeding time is scheduled at 3:30 pm daily, except on Saturdays. The tigers are provided with either chicken or meat with the total amount of feed given calculated based on 10% of their body weight. Each

night stalls have been provided with a few pieces of furniture such as hanging logs and a table with logs laid horizontally next to it with additional enrichment tire toys.

3.2 Study Design

Study design for the Malayan tiger in NWRC is to record the oestrous behaviour of the wild-caught female tiger through video surveillance for a period of 28 days in order to identify at least one cycle of oestrous behaviour as the tiger would experience oestrus at intervals in three to nine weeks every twenty-five days. Observational study will be performed from 8 am to 5 pm in order to allow recording of the oestrous behaviour during that particular time in which the behaviour will be recorded using ethogram and frequency will be calculated using tally numeration system. The main behaviours that correlate with the oestrous cycle such as prustening, rubbing, rolling, tail lifting and lordosis will be recorded for analysis.

3.3 Study Population

The study population involved two female wild-caught tigers that were reported in human-wildlife conflict cases at residential areas. Both tigers were selected to be observed in the enclosure to determine their oestrous cycle as they are young individuals with good breeding potentials, and would bring new genetic diversity to the captive population in NWRC. This study will require information regarding the animal of interest such as its species, identification (name), chip number, date of acquisition, age, sex, weight, and reproduction status, feeding management and the time of observation from the beginning until the end will be recorded in the data sheet. There will be two female wild-caught tigers involved in this study.

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Table 3.1: Information of both study population involved in this study

ID Tiger	Age	Chip Number	Date of acquisition	Origin	Unit of night stall	Pairing with male	Daily routines
Mek Santong	7 years old	458098500-301004	30/3/2023	Bukit Santong, Kerteh, Terengganu	C	Not paired	Daily stalls cleaning is every morning at 9:30 am and Mek Santong will be transferred to the next stall temporarily. Feeding time was scheduled at 3:30 pm daily, except fasting on Saturday.
Yong Tawai	5 years old	458098500-301272	29/2/2024	Felda Tawai, Gerik, Perak	D	Not paired	Daily stalls cleaning is every morning at 9:30 am and Yong Tawai will be transferred to the next stall temporarily. Feeding time was scheduled at 3:30 pm daily, except fasting on Saturday.

3.4 Sampling Method and Procedure

3.4.1 Sampling Technique

In this study, a behavioural sampling technique was used to document and analyze the oestrous behaviour displayed by two wild-caught female Malayan tigers (*Panthera tigris jacksoni*). This method provided an approach to observing and recording behaviours associated with the reproductive cycle. The focal sampling method was applied, where each tiger was observed individually over a 28-day period to identify patterns of oestrous behaviour and determine their reproductive phases. The use of this sampling technique allowed for detailed and

accurate data collection while minimizing external disturbances that could influence natural behaviours. The frequency data will be collected in the ethogram and frequency tables and were analysed to identify the reproductive phase of the female wild-caught Malayan tigers.

3.4.2 Sampling Procedure

A 28-day observational study was conducted to document oestrous behaviour patterns in these tigers. The study followed a focal animal sampling method utilizing an ethogram, allowing for systematic documentation of reproductive behaviours. Observations were conducted daily from 08:00 to 17:00 hrs using live video surveillance through CCTV cameras to observe and record the behaviour and their frequencies at the same time to minimize observer interference, ensuring their natural behaviour was not influenced by human presence for detailed analysis of the oestrous pattern shown.

A standardized ethogram was used to document and categorize oestrous-related behaviours, which included prustenning, rubbing, rolling, tail lifting, and lordosis. The frequency of these behaviours was recorded in a focal sampling worksheet to capture their occurrence across different observation periods. Environmental variables, such as the presence of construction's worker or vehicle's sound, were also noted to determine any external influences on oestrous behaviour. For data analysis, the behavioural frequency data were processed using descriptive statistics, and graphical interpretations such as bar charts and frequency distribution graphs were used to visualize the patterns of oestrous behaviour over the 28-day period. The occurrence of behaviours was examined to identify different oestrous phases, including proestrus, oestrus, and post-oestrus, based on the intensity and recurrence of key reproductive behaviours. The sampling procedure was designed to ensure systematic, controlled, and non-invasive monitoring of oestrous behaviour in wild-caught captive Malayan tigers.

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Results

In this project, raw observational data of the estrous behaviour shown by both Mek Santong and Yong Tawai obtained from the ethogram and frequency chart that have been collected for a total of 28 days from 8 am until 5 pm.

Results for Mek Santong were plotted in the form of bar chart and line graph to get the overview of pattern oestrous behaviour.

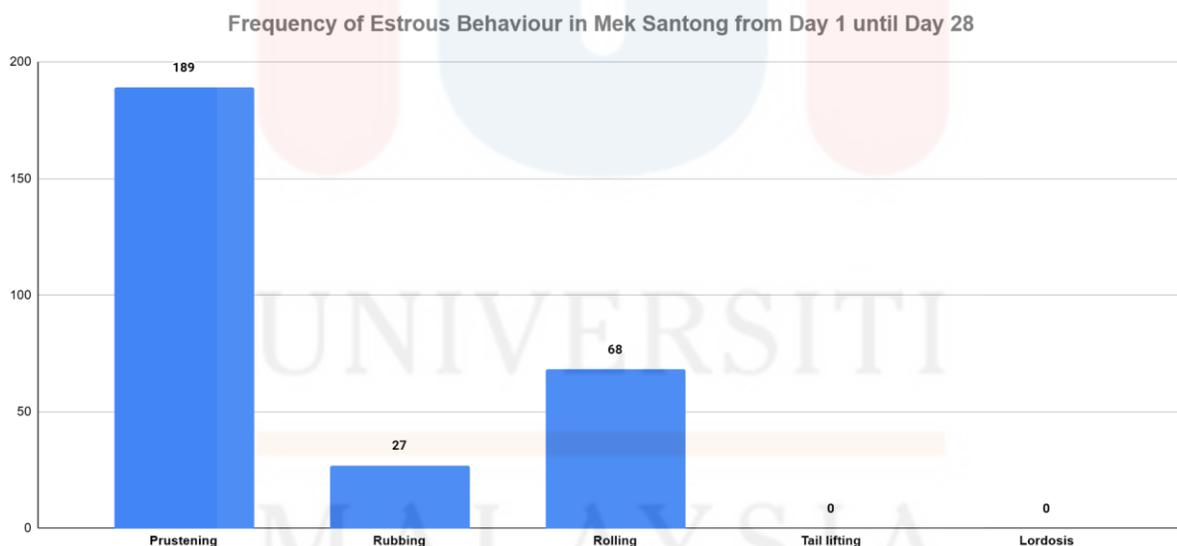


Figure 4.1: The graph chart representing frequency of estrous behaviour exhibited by Mek Santong

The most expressed behaviour shown by Mek Santong throughout 28 days would be prustening, with a total of 189 times, directed at the male beside her night stall. The next most expressed behaviour was rolling while facing male night stall and the occurrences were concurrent with

rubbing behaviour to show her interest towards the male. Total frequency of rolling shown by Mek Santong was 68 times and she would roll at the exact location in front of connecting doors of male stall and was mostly seen after her expressing restlessness. Rubbing towards the wall, furniture, and doors by Mek Santong only had a total of 27 times. However, Tail lifting and lordosis were not displayed at all throughout the observation for 28 days.

Prustening Behaviour from Day 1 until Day 28

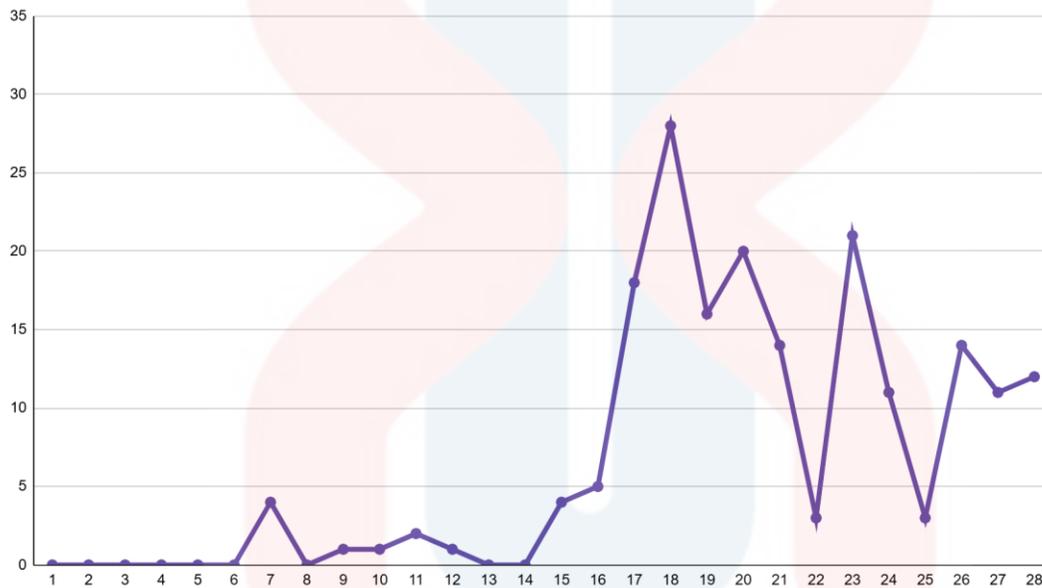


Figure 4.2: Line graph for prustening behaviour of Mek Santong

Line graph illustrated on figure 4.2 showed that from Day 1 to Day 6, the prustening behaviour remains at a low frequency and stable level close to zero occurrences. Starting on Day 7, there is a small peak suggesting a slight increase of the behaviour. The behaviour began to rise on Day 15 and reached a peak of 28 occurrences on Day 18. However, after the peak, prustening behaviour becomes more variable as it drops significantly with only displayed three times by Mek Santong, then fluctuated with smaller peaks around Day 20 and 23.

Rubbing behaviour from Day 1 until Day 28

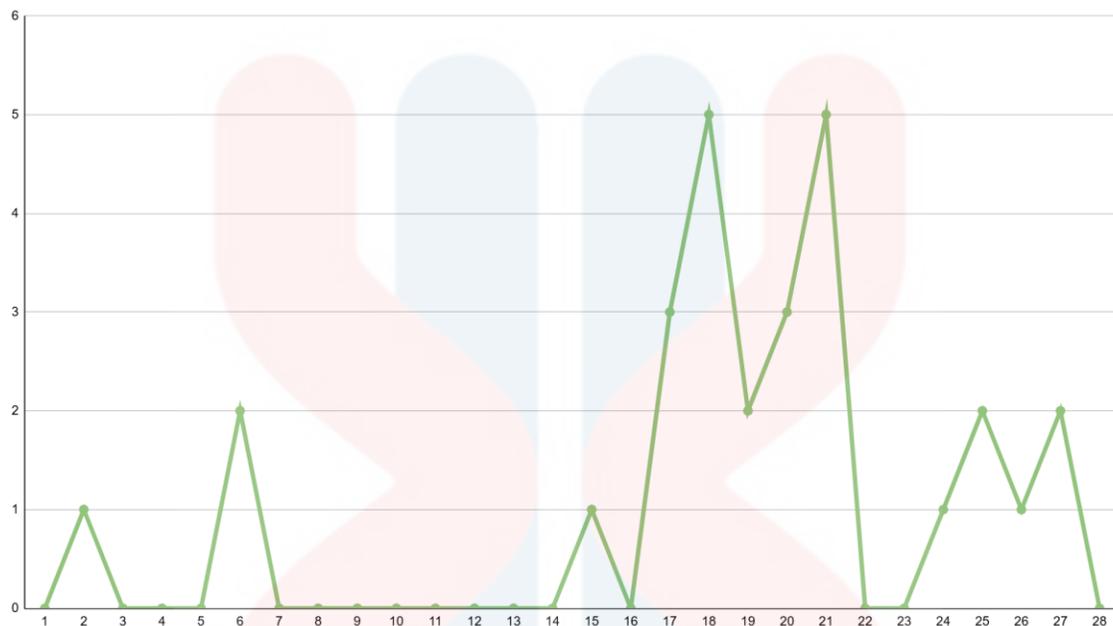


Figure 4.3: Line graph for rubbing behaviour of Mek Santong

Based on the figure 4.3, rubbing behaviour was at a low frequency level with minimal to no activity recorded during Day 1 to Day 5. There was an initial increase in the behaviour peaking at Day 6 with two times exhibited and followed by a drop to zero by Day 7. On Day 16, rubbing behaviour increased significantly reaching its highest occurrences of five times displayed at Day 17 and 21, which shows the most consistent and elevated rubbing behaviour across the 28 days total observation. After Day 21, the behaviour showed decline in frequency but remained variable with smaller peaks on Day 24 to Day 27 before plummeting to zero on Day 28.

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Rolling Behaviour from Day 1 until Day 28

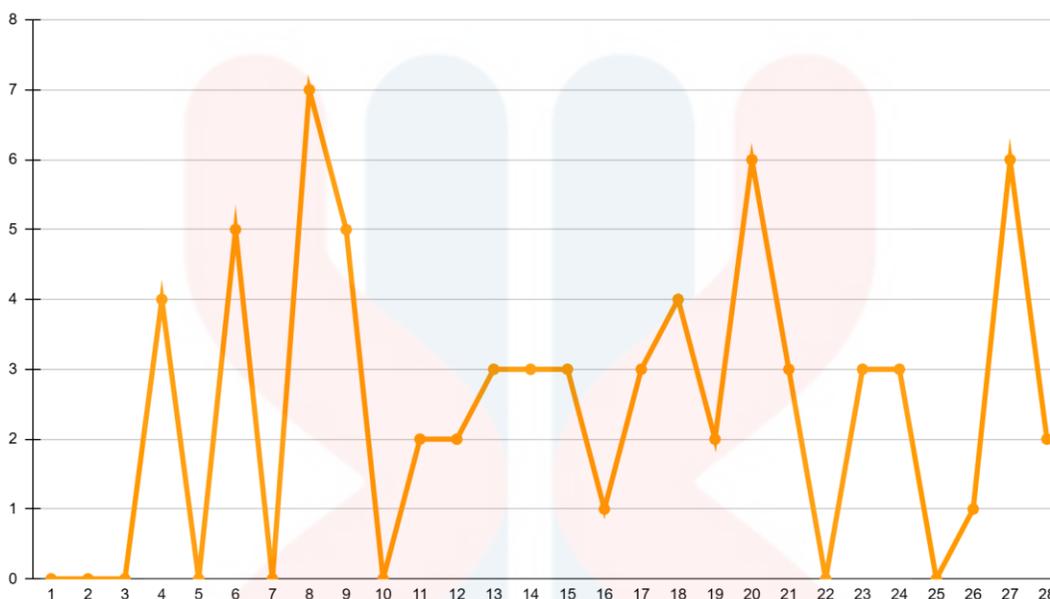


Figure 4.4: Line graph for rolling behaviour of Mek Santong

Based on the figure 4.4, from Day 1 until Day 3, there is no rolling behaviour shown by Mek Santong. It began to increase on Day 4 reaching a peak at Day 7 with a value of seven followed by a sharp decline by Day 9. Rolling behaviour remains relatively low with some fluctuations at a lower range on Day 10 to Day 15. Another peak in rolling behaviour occurs on Day 17, reaching a similar high level as seen in the initial peak. Fluctuations continue, but the behaviour generally remains more active than in the earlier low period. Around Day 26 and Day 28, the behaviour remains variable with smaller peaks and Day 27 had a value of six occurrences and dropped to just two on the last day of observation.

Another tigress, Yong Tawai, was also collected at the same period using ethogram and frequency tables but she showed zero reproductive behaviour patterns suggesting the absence of an oestrous cycle. During the observation time, Yong Tawai demonstrated a variety of behaviours, including restlessness, resting, and aggression. Restlessness was characterized by frequent pacing and movement within the night stall, whereas resting behaviour was defined by extended periods of inactivity or lying down. Aggressive behaviours were seen, such as vocalizations, posturing, or

physical confrontations towards the keeper and visitor, but they were not consistently associated with any phase of the oestrous cycle.

From the data collected throughout the duration of 28 days, there were no distinct patterns indicative of oestrous behaviour in Yong Tawai. The behaviours observed did not align with oestrus-related activities listed in this study such as prustening, rubbing, rolling, tail lifting and lordosis. The behaviours displayed by Yong Tawai did not demonstrate a clear correlation with the tiger's reproductive cycle.

4.2 Discussion

Based on the observational study conducted on two wild-caught Malayan tigers, Mek Santong and Yong Tawai, only the former exhibited partial oestrus behaviour. One of the reasons for the tigresses not fully expressing oestrous behaviour could be that both of them were not mixed with male throughout the 28-days duration. In contrast, Yong Tawai did not display any sign of oestrus behaviour, which may be attributed to her recent capture in February 2024 compared to Mek Santong who was captured in March 2023. It is possible that Yong Tawai had not fully adapted to her new environment in captivity, which would have contributed to her to display stereotypical behaviours instead of oestrous behaviours. The behaviours exhibited by Mek Santong were tabulated and represented in a line graph to provide a clear overview of the oestrous cycle pattern. These graphical representations highlight the frequency of each behaviour observed daily, allowing for a better understanding of the changes in her behaviour throughout the observation period.

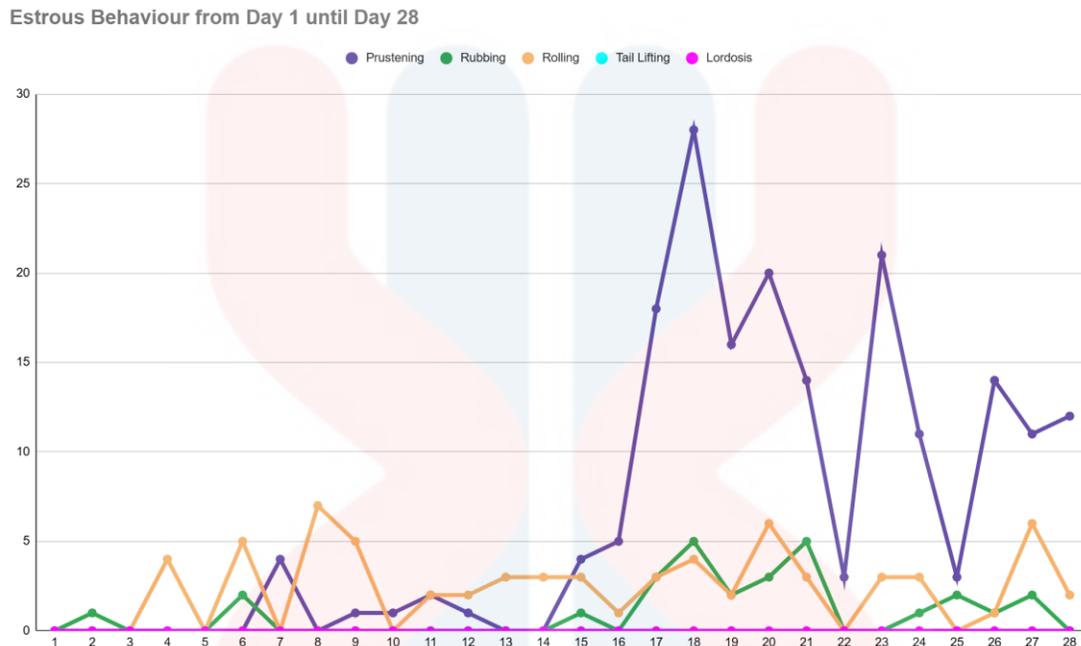


Figure 5.1: Line graph of estrous behaviour in Mek Santong for 28 days

Based on the line graph above in Figure 5.1, all the data showed a clear periodicity of oestrous behaviour in Mek Santong with peak frequency at around mid-cycle on Day 15 to 21 which is likely the most fertile period. Prustening behaviour being the highest behaviour shown may serve as a reliable indicator of oestrus in Mek Santong. Behaviour of rubbing and rolling further aligned with this time frame, reinforcing her roles in courtship and ready to mate. Even though hallmark behaviours of oestrus, which are lordosis and tail lifting, appeared to be less prominent in Mek Santong, this could be attributed to individual variation or could be influenced by other external factors that prevent the expression of the behaviour. Additionally, lordosis and tail lifting may not be shown by Mek Santong due to the absence of male in the same night stall. This is supported by another study on the breeding behaviour of captive-bred female Malayan tiger where one of the subjects expressed all the behaviour, including tail lifting against the wall and presenting herself in lordosis before her mate in the same night stall. This demonstrates that there is individual variability in expression of oestrus signs and not all individuals will exhibit all the behaviours as an indicator of oestrus (Logington, 2022).

In the early stage of the observation, Mek Santong showed low frequency of rubbing, rolling and no signs of prusten towards male beside her night stall. Prusten is commonly associated with social and courtship behaviour in tigers where during this study, Mek Santong showed minimal prusten activity from Day 1 to Day 13 and mostly expressed signs of restlessness circling the night stall repetitively and resting. Rubbing behaviour is likely a means of scent marking or to stimulate sensory cues during oestrus that was observed sporadically with little to no activity from Day 1 to 13. This suggests that these days correspond to early proestrus when the tiger may not be sexually receptive and had no interest with male by showing minimal signs of the oestrous behaviour. Rubbing behaviour showed less frequency on Day 6 preceding the rise of prusten on Day 14, which suggests rubbing may serve for the onset of the full oestrus phase before showing other behaviour. The earlier onset of rubbing may reflect the initial stages of pheromone release to attract the male in which during this phase, Mek Santong was seen to display her rubbing behaviour concurrently with prusten to express her interest to her potential mates.

Based on the line graph above, rolling behaviour shows periodic activity throughout the 28-day observation period with notable peaks on Day 5 to 7, Day 18 to 19 and on Day 26 thus these peaks align with the prusten and rubbing behaviour in the mid-cycle. In this study, Mek Santong frequently exhibited rolling behaviour in the same location, directly in front of the connecting door to the male's night stall. The behaviour of Mek Santong usually started with circling repetitively in front of the connecting door followed by rubbing against the wall or furniture, which then displayed rolling a few times at a time with a duration of 10 seconds.

A significant peak occurs between Day 15 to 20 with the highest frequency prusten observed on Day 16 that aligns with the oestrus phase suggesting that prusten is one of the indicator oestrus onset or peak receptivity. With prusten, female tigers actively seek male attention and exhibit reproductive readiness and during this phase, other behaviour like rubbing and rolling follows a similar pattern with significant peaks between Day 18 to 21. All the behaviours that overlap between Day 18 to Day 20 are closely linked to the estrus phase where the alignment of peak prusten, rubbing and rolling behaviour during mid-cycle may highlight the synergistic role in expressing oestrus signs.

Throughout the 28 days observation, variability of behaviour shown by Mek Santong may have been influenced by the external factors, such as construction of new pool in block C for night stalls C3 and C4 starting on Day 13 and prolonged until the end of study. During this period, her behaviour was characterized by an increase in restlessness, including circling the night stall, aggression and resting, indicating potential stress from the noise and the constant presence of humans in the area throughout the daytime. She exhibited aggressive behaviour by attempting to attack people walking in front of the night stall but even though with the distraction, she still displayed the oestrus behaviour but with low frequency. After a few days, she tried to adapt to the noise and humans, Mek Santong eventually started displaying behaviours indicative of interest towards the male with peak prustening followed by rolling on Day 23.

After Day 24, the behaviours had become less consistent with showing inconsistent increase and drop. On the day with less behaviour displayed from Day 24 - 26, Mek Santong spends most of her time resting and becomes active mainly around feeding time. This decline is related with the reduction of hormonal stimulation where progesterone levels rise while oestrogen levels decline. This variability may indicate the transition out of oestrus into dioestrus phase. It is a reliable indicator of the end of oestrus and the start of the dioestrus phase.

As a recent wild-caught Malayan tiger, Yong Tawai's behavioural rhythms cannot be seen which might be due to external influences like the captive breeding conditions and it is believed she still has not yet adapted to the new captive environment. This may be due to the stress of transitioning six months ago from the wild to captivity. The adjustment period might be challenging since she was recently caught and may struggle with changes in her surroundings, unfamiliar sounds, sight and also new daily routines. Such environmental changes can lead to anxiety and discomfort which affect her natural behaviour including oestrous cycle. From the observation, Yong Tawai only showed resting behaviour, and restlessness plus aggression when there was presence of keepers near her night stall during daily cleaning. The resting behaviour indicates that Yong Tawai experienced an anoestrus phase where her reproductive activity was low, and there is no sexual behaviour or mating readiness towards a male beside her night stall during the observational study.

Moreover, when the tiger has stress due to other external factors, this can alter the balance of reproductive hormones that are essential for the oestrous cycle in females. When they are stressed, the level of cortisol will rise which can interfere with the production of the oestrogen and progesterone which thus resulting in inhibiting the normal onset of oestrus in females leading to the anoestrus phase that was experienced by Yong Tawai. The elevated cortisol levels can suppress the GnRH which has a function to regulate the release of sex hormones in the reproductive system which cause no expression of any oestrus behaviour indicating no oestrus experiencing at the moment (Dobson *et al.*, 2000; Einarsson *et al.*, 2008).

The discussion above explained how the transition from wild to captive settings affects the oestrous cycles of tigers that have been captured from the wild. Outcomes from the present study can be compared to the work by Logington (2022) that focused on oestrous behaviour in captive-bred Malayan tigers. Based on findings from both of this study, it can be concluded that there are slight differences in the frequency and pattern of oestrous behaviour between wild-caught and captive-bred tigers. The captive-bred tigers typically exhibit more consistent oestrous cycles, in particular Tiger A demonstrated all common oestrous behaviours such as prustenng, rubbing, rolling, lordosis, and tail lifting for two cycles within 28 days. Whereas wild-caught tigers in this study showed more variability in oestrous cycles where Mek Santong displayed inconsistent oestrous behaviour without expressing all behaviours, while Yong Tawai experienced an anoestrus phase, not showing any sign of oestrous activity. These differences can largely be due to the stress of adapting to captivity, which can disrupt the oestrous cycle. In contrast, captive-bred tigers that were raised in controlled environments are less likely to experience these interruptions as compared to the wild-caught tigers.

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CHAPTER 5

CONCLUSIONS AND RECOMMENDATION

5.1 Conclusion

In conclusion, this study successfully answers the research questions, supports the hypotheses, and achieves the objectives that provide important findings into the oestrous behaviour of wild-caught female Malayan tigers (*Panthera tigris jacksoni*) in captivity. Behaviours such as prustenning, rubbing, and rolling were identified as key indicators of oestrus in Mek Santong, confirming the hypothesis that specific behaviours are reliable for detecting the oestrous phase. However, behaviours like tail lifting and lordosis were not observed, likely due to external factors such as the absence of a male in the same night stall or stress from ongoing construction near the tiger unit. In contrast, Yong Tawai, who was recently captured in February 2024, did not show any oestrous behaviour, which could be due to her difficulty adapting to the new captive environment. Her stress may have caused an imbalance in reproductive hormones, leading to an anoestrus phase throughout the study. These findings compared with other studies may highlight the differences between wild-caught and captive-bred tigers, as the latter tend to have more consistent oestrous cycles and are less affected by stress. This study achieved its goals by identifying reliable oestrous behaviours, assessing their use in wild-caught tigers, and comparing the behaviour of wild-caught and captive-bred individuals. These results are essential for improving breeding programs and ensuring the conservation of Malayan tigers.

5.2 Recommendations

Further recommendation for future research is to include a longer observation period, ideally extending to 24 hours to gather more data on the oestrous behaviour frequency. This would help ensure sufficient data collection while minimizing the influence of external factors particularly during daytime hours. For a recent wild-caught Malayan tiger, it would be recommended for her to adapt with the captive for at least one year in order for her to be able to express natural behaviour, including oestrous behaviour because she may require more time to adjust and the transition to captivity can be quite shocking and may involve individual variation. In addition to that, hormonal analysis could also strengthen the understanding of how these behaviours correlate with the oestrous cycle in wild-caught Malayan tigers. This can be done by performing faecal progesterone analysis to determine the oestrous cycle. This test could be more accurate in identifying the oestrus phase of the Malayan tigress, although costly due to the use of reagents and laboratory services.

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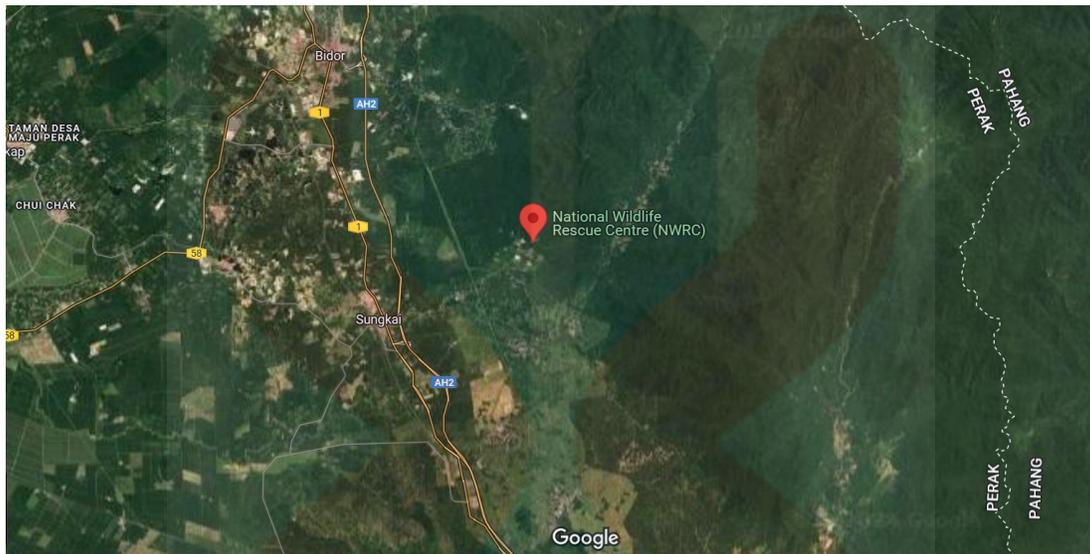
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Appendix A



Appendix A1: Satellite view of location National Wildlife Rescue Centre (NWRC)



Appendix A2: Main gate of National Wildlife Rescue Centre (NWRC)



Appendix A3: Video surveillance room



Appendix A4: Television for video surveillance observation



Appendix A5: Mek Santong showing aggression and anxiety with human presence.

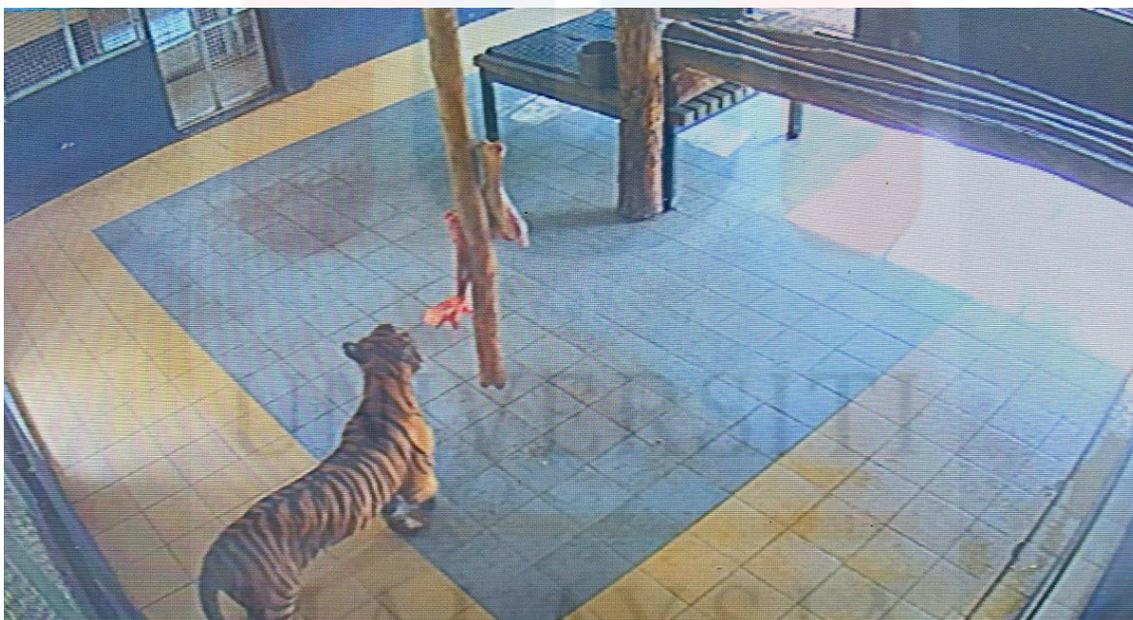


Appendix A6: First view video surveillance of Mek Santong's night stall (C2)

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Appendix A7: Second view video surveillance of Mek Santong's night stall (C2)



Appendix A8: Feeding time Mek Santong by applying Food Hanging Enrichment method

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Appendix A9: Mek Santong seen resting on raised platform



Appendix A10: Mek Santong displayed rolling behaviour in front of connecting door with male

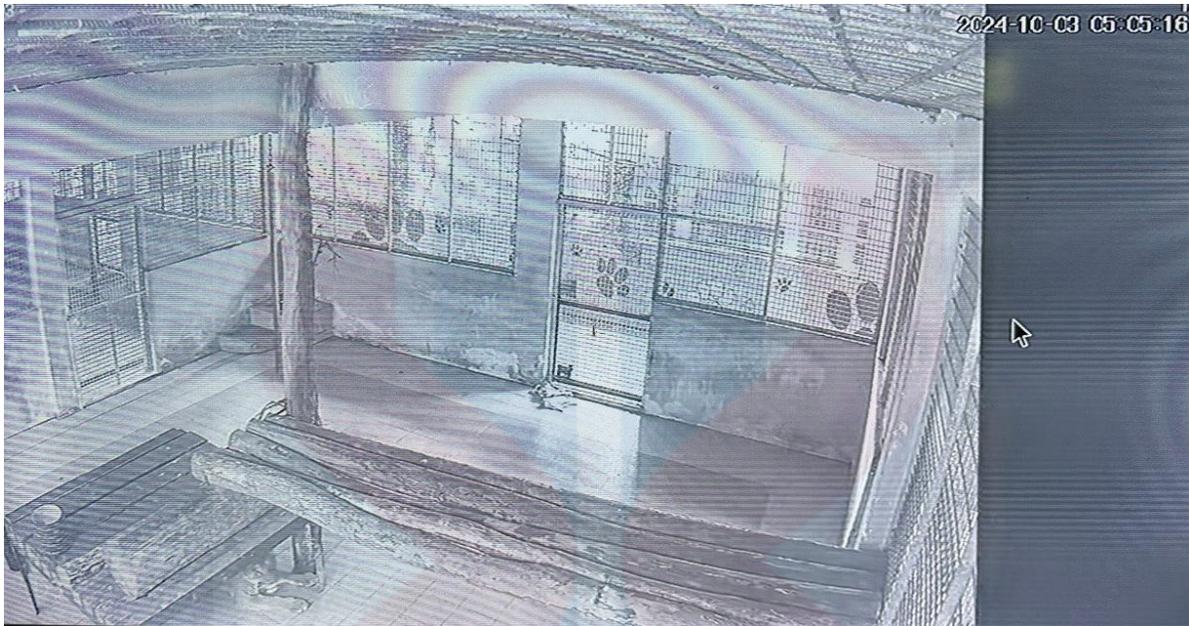
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Appendix A11: Mek Santong showing interest towards male beside her night stall



Appendix A12: Yong Tawai resting under the raised platform



Appendix A13: Second view video surveillance of Yong Tawai's night stall (D3)



Appendix A14: Daily cleaning routine of night stall

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Ethogram of estrous behaviours in female Malayan tigers (*Panthera tigris jacksoni*)

Behaviour acts	Description	Duration
Scent marking	Urinating or spraying in strategic locations	
Prustening	A low-frequency sound called "prusten," which involves exhaling air through their parted lips and nose	
Rubbing	Rub bodies against trees, rocks, or other surfaces around enclosure	
Rolling	Rolling on the ground, ying on the back and turning over	
Tail lifting	Upward or erect position of tail to exposes the genital region	
Lordosis	Lowering the forelimbs while hindlimbs are extended, raised hips, ventral arching of the spine and raising or sideward displacement of the tail	
Other		

Appendix A16: Ethogram of estrous behaviour

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Observer : Animal ID : Sex : Date : Start to end time :

Frequency chart of estrous behaviour female Malayan tiger (*Panthera tigris jacksoni*).

Estrous behaviour	Prustening	Rubbing	Rolling	Tail lifting	Scent marking	Lordosis	Others
Code	P	Rb	Ro	Tl	Sm	L	O

Non - breeding behaviour	Grooming	Eating	Drinking	Yawning	Defecating	Resting
Code	G	E	Dr	Y	De	Re

Activity :	Tally marks recorded of estrous behaviours		Frequency
	P		
Rb			
Ro			
Tl			
Sm			
L			
O			
Note:			

Appendix A17: Frequency chart of estrous behaviour



IBU PEJABAT
JABATAN PERLINDUNGAN HIDUPAN LIAR DAN
TAMAN NEGARA (PERHILITAN) SEMENANJUNG MALAYSIA
HEADQUARTERS
DEPARTMENT OF WILDLIFE AND NATIONAL PARKS (DWNP)
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Rujukan : JPHLTN.600-6/1/4 JLD 5(85)

Tarikh : 28 November 2024

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No. 33 Lorong Kp1
Taman Kota Permai,
34700 Simpang
PERAK

Puan,

**KEPUTUSAN PERMOHONAN PERUBAHAN/PINDAAN BUTIRAN PERMIT
PENYELIDIKAN BERTAJUK ESTROUS BEHAVIOUR MONITORING IN WILD
CAUGHT FEMALE MALAYAN TIGERS (*Panthera tigris jacksoni*)**

Dengan hormatnya saya merujuk kepada perkara di atas dan keputusan Mesyuarat Jawatankuasa Penyelidikan Bil 10/2024 adalah berkaitan.

2. Sukacita dimaklumkan bahawa Jabatan PERHILITAN meluluskan perubahan/pindaan butiran **Penyelidikan Nur Qamarina Binti Shaharin (Universiti Malaysia Kelantan)** seperti butiran di **Lampiran** bermula **September 2024 hingga Oktober 2025 (1 tahun)**.

- a. Menghubungi rakan saing dan *Co-author* yang dilantik iaitu **Dr Zubaidah binti Kamarudin, Pegawai Veterinar (NWRC)** bagi memantau dan membantu perjalanan penyelidikan ini di lapangan;
- b. Berkongsi hasil penyelidikan seperti laporan/penerbitan kertas saintifik/tesis dengan Jabatan melalui rakan saing yang telah dilantik;
- c. Mengemukakan satu laporan hasil penyelidikan yang lengkap kepada Jabatan dalam tempoh dua (2) bulan selepas tamat penyelidikan; dan
- d. Penyelidikan hendaklah diselesaikan dalam tempoh yang dinyatakan dalam permit.

'HIDUPAN LIAR UNTUK GENERASI AKAN DATANG'

SELAMATKAN HARIMAU MALAYA

www.harimau.my

Appendix A18: Letter approval from DWNP



JABATAN PERLINDUNGAN HIDUPAN LIAR DAN TAMAN NEGARA
DEPARTMENT OF WILDLIFE AND NATIONAL PARKS



D-01013-15-24

PERMIT PENYELIDIKAN ATAU KAJIAN KE ATAS HIDUPAN LIAR-AKADEMIK(DILINDUNGI)

Sekyen 10(1)(a)

AKTA PEMULIHARAAN HIDUPAN LIAR 2010 (AKTA 716)

BUTIRAN PEMILIK

NAMA PEMILIK : NUR QAMARINA BINTI SHAHARIN
NO MYKAD / PASPORT : 010419080708
PEKERJAAN : LAIN-LAIN

NAMA PREMIS : UNIVERSITI MALAYSIA KELANTAN
NO PENDAFTARAN PREMIS :
ALAMAT PREMIS : KAMPUS KOTA
16100-KOTA BAHRU, KELANTAN



TEMPOH SAH
10/12/2024 - 31/10/2025

Syarat-syarat Permit Penyelidikan Atau Kajian Ke Atas Hidupan Liar-Akademik(Dilindungi)

1. Permit ini tidak boleh dipindah milik.
2. Permit ini sah untuk tempoh yang telah dinyatakan pada permit.
3. Permit ini hanya sah bagi kajian spesies yang dinyatakan sahaja.
4. Ketua Pengarah boleh menggantung atau membatalkan permit jika pemegang permit melanggar syarat-syarat yang telah dinyatakan.
5. Pemegang tertakluk kepada peruntukan Akta Pemuliharaan Hidupan Liar 2010 (Akta 716) dan apa-apa perintah yang pada masa itu berkuatkuasa.
6. Pemegang permit perlu membuat permohonan permit yang baharu dalam tempoh empat belas (14) hari dari tarikh tamat tempoh sah permit.

Syarat-syarat tambahan

1. Menghubungi rakan saing dan Co-author yang dilantik iaitu Dr Zubaidah binti Kamarudin, Pegawai Veterinar (NWRC) bagi memantau dan membantu perjalanan penyelidikan ini di lapangan;
2. Berkongsi hasil penyelidikan seperti laporan/penerbitan kertas saintifik/tesis dengan Jabatan melalui rakan saing yang telah dilantik;
3. Mengemukakan satu laporan hasil penyelidikan yang lengkap kepada Jabatan dalam tempoh dua (2) bulan selepas tamat penyelidikan; dan
4. Penyelidikan bertajuk Estrous Behaviour Monitoring In Wild Caught Female Malayan Tigers (Panthera Tigris Jacksoni) hendaklah diselesaikan dalam tempoh yang dinyatakan dalam permit September 2024 hingga Oktober 2025 (1 tahun)

Ini adalah cetakan komputer dan tidak perlu ditandatangani

Nama Pegawai : CHE MUHANDI BIN CHE ISMAIL
No Lesen : D-01013-15-24

Appendix A19: Permit approval from DWNP