UTILISATION OF DNA BARCODING IN ASSESSING THE DIVERSITY OF BATS BASED ON TAXONOMIC RECORDS AND IDENTIFYING THEIR PLANT-BASED DIET IN PENINSULAR MALAYSIA

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FACULTY OF SCIENCE
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KUALA LUMPUR

2018

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THESIS SUBMITTED IN FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE OF DOCTOR OF PHILOSOPHY

INSTITUTE OF BIOLOGICAL SCIENCES
FACULTY OF SCIENCE
UNIVERSITY OF MALAYA
KUALA LUMPUR

2018

UNIVERSITY OF MALAYA

ORIGINAL LITERARY WORK DECLARATION

Name of Candidate: **LIM VOON CHING**

Matric No: **SHC150038**

Name of Degree: **DOCTOR OF PHILOSOPHY**

Title of Thesis:

UTILISATION OF DNA BARCODING IN ASSESSING THE DIVERSITY OF BATS BASED ON TAXONOMIC RECORDS AND IDENTIFYING THEIR PLANT-BASED DIET IN PENINSULAR MALAYSIA

Field of Study:

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UTILISATION OF DNA BARCODING IN ASSESSING THE DIVERSITY OF BATS BASED ON TAXONOMIC RECORDS AND IDENTIFYING THEIR PLANT-BASED DIET IN PENINSULAR MALAYSIA

ABSTRACT

In Peninsular Malaysia, the diversity of bats was previously assessed through morphological identification of captured bats while the diet of plant-visiting bats were examined through morphological identification of seeds and pollen grains collected from bats. Yet morphological identification is often of limited service when applied to identification of morphologically similar bat and plant species. The objective of this research is to use a molecular approach, DNA barcoding to review the diversity of bats and their plant-based diet in Peninsular Malaysia. Through literature review and Neighbour-Joining analyses of DNA barcodes available from bats sampled in Peninsular Malaysia, at least 110 bat species have been documented in the region and eighteen of them are species complex which deserve further investigation. The diet of frugivorous bat, *Cynopterus brachyotis*, at secondary forest, oil palm plantation and urban area were compared by identifying pulps and seeds found in the bats' faeces using DNA barcoding. Native and introduced plants were detected from bat faeces at all sampling sites, suggesting the dual role of *C. brachyotis* in dispersing (i) native plants which aid in forest regeneration, and (ii) introduced plants which potentially facilitate their invasion. The diet of nectarivorous bat, *Eonycteris spelaea* at urban area was examined by identifying the plant material present in the bat faeces using DNA metabarcoding. Many plant species which were detected from the bat faeces have not been reported in previous dietary studies of *E. spelaea* including ferns and figs, consequently suggesting that *E. spelaea* may not be specialised nectarivore. Therefore, the use of DNA barcoding has highlighted the taxonomic uncertainties in bats and provided new insights into diet of plant-visiting bats.

Keywords: DNA barcoding, Chiroptera, taxonomy, diet, species' interaction

PENGGUNAAN "DNA BARCODING" DALAM MENGKAJI KEPELBAGAIAN KELAWAR BERDASARKAN REKOD TAKSONOMI DAN MENGENAL PASTI DIETNYA YANG BERASASKAN TUMBUHAN DI SEMENANJUNG MALAYSIA

ABSTRAK

Kepelbagaian kelawar di Semenanjung Malaysia sering dikaji melalui pengenalpastian morfologi kelawar yang ditangkap manakala diet kelawar yang berasaskan tumbuhan sering dikaji melalui pengenalpastian morfologi biji dan debunga yang didapati dari kelawar. Namun begitu, ciri-ciri morfologi kurang membantu bagi pengenalpastian spesies kelawar dan tumbuhan yang mempunyai morfologi yang serupa. Objektif kajian ini adalah menggunakan kaedah molekul, *DNA barcoding* untuk mengkaji kepelbagaian kelawar dan dietnya yang berasaskan tumbuhan di Semenanjung Malaysia. Sorotan kajian dan analisis kod bar DNA kelawar menunjukkan bahawa sebanyak 110 spesies kelawar telah direkodkan di Semenanjung Malaysia dan lapan belas daripadanya adalah spesies kompleks. Diet kelawar frugivor, *Cynopterus brachyotis* di hutan sekunder, ladang kelapa sawit dan kawasan bandar dibanding melalui pengenalpastian pulpa dan biji tumbuhan dalam najis kelawar menggunakan *DNA barcoding*. Tumbuhan asli dan eksotik dikesan dalam najis kelawar mencadangkan bahawa *C. brachyotis* menyebarkan (i) tumbuhan asli lalu membantu pemulihan hutan, dan (ii) tumbuhan eksotik lalu memudahkan proses pencerobohannya. Diet kelawar nektarivor, *Eonycteris spelaea* di kawasan bandar dikaji melalui pengenalpastian bahagian tumbuhan dalam najisnya menggunakan *DNA metabarcoding*. Kebanyakan spesies tumbuhan yang dikesan dari najis tersebut belum pernah dilaporkan oleh kajian terdahulu termasuk paku pakis dan pokok ara justeru menunjukkan bahawa *E. spelaea* bukan kelawar nektarivor yang khusus. Penggunaan DNA barcoding berjaya merungkai ketidakpastian taksonomi kelawar dan memberi maklumat baru mengenai diet kelawar yang berasaskan tumbuhan.

Kata kunci: DNA barcoding, Chiroptera, taksonomi, diet, interaksi spesies

ACKNOWLEDGEMENTS

I would like to express my deepest gratitude to everyone who have contributed to the completion of this research particularly my supervisors, Dr. John James Wilson (who never stop nagging yet encouraging me), Assoc. Prof. Dr. Rosli Ramli and Assoc. Prof. Dr. Subha Bhassu for their constant guidance, advice and constructive criticism.

This research would never have happened without the financial support from Ministry of Higher Education, Malaysia (MyPhD), National Geographic Society (Asia 59-16), Malaysian Nature Society (YERG16-12) and University of Malaya (PG060-2016A). Thank you for believing in me and acknowledging the potential of my research.

Special gratitude to my colleagues: Lee Ping Shin, Brandon Mong, Jisming Shi and Sing Kong Wah for their assistance and guidance in my fieldwork and laboratory work. I thank my collaborators: Dr. Elizabeth Clare and Dr. Joanne Littlefair for their technical and financial support. I would like to thank the UM's Rimba Project for providing the support and laughter during fieldwork. I am grateful to Dr. Sugumaran Manickam for providing the invaluable advice on plant species identification. I thank Peninsular Malaysia's Department of Wildlife and National Parks and Dark Cave Management for providing the specimens and assistance during fieldwork.

Last but not least, I would like to thank the following people (in alphabetical order) who have contributed to this research in their own way: Afiqah Zainuri, Benjamin Ong, Dr. Charles Francis, Chua Li Pei, Dr. Christine Fletcher, Jo Leen Yap, Keane Lai Soen Liong, Lim Aik Hean, Lim Tze Tshen, Nurul Fitrah Marican, Dr. Puan Chong Leong, Dr. Sheema Abdul Aziz, Prof. Stephen Rossiter, Tan Kai Ren, Tan Yi Wen, Vanessa Ting, and Prof. Emeritus Yong Hoi Sen. Thank you!

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LIST OF SYMBOLS AND ABBREVIATIONS

~	:	Approximate
>	:	More than
≥	:	Greater than or equal to
%	:	Percentage
BIN	:	Barcode Index Number
BOLD	:	Barcode of Life Datasystems
CCDB	:	Canadian Centre for DNA barcoding
COI	:	Cytochrome *c* oxidase I
DNA	:	Deoxyribonucleic acid
IUCN	:	International Union for Conservation of Nature
ITS2	:	Internal transcribed spacer 2
km^2	:	Square kilometres
mtDNA	:	Mitochondrial deoxyribonucleic acid
NADH	:	Nicotinamide adenine dinucleotide hydride
NCBI	:	National Center for Biotechnology Information
ND2	:	Mitochondrially encoded NADH dehydrogenase 2
NGS	:	Next-generation sequencing
PCR	:	Polymerase chain reaction
NJ	:	Neighbour-Joining
RAG1	:	Recombination activating gene 1
rbcL	:	Ribulose bisphosphate carboxylase gene
sp.	:	Species (singular)
spp.	:	Species (plural)

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CHAPTER 1: INTRODUCTION

1.1 Bats (order: Chiroptera)

Over 25% of the world's bat species occur in Southeast Asia yet they are threatened by the rapid deforestation and land-use changes across the region including Peninsular Malaysia (Kingston, 2013). Knowledge of bat diversity of Peninsular Malaysia remains limited due to the absence of a comprehensive checklist of bats specifically for the region. Bat surveys in Peninsular Malaysia are generally based on morphological identification of captured bats (Jayaraj *et al.*, 2012a; 2013a) which often requires high level of taxonomic expertise. However, researchers with limited expertise in taxonomy of bats may face difficulties in identifying newly encountered species and distinguishing morphologically similar and sympatric species, which consequently may provide limited information for understanding the diversity of bats among geographical regions (Francis *et al.*, 2010; Wilson *et al.*, 2014).

Knowing what species occur in the region is imperative for developing conservation plans for the bats which provide important ecosystem services through their feeding behaviour. Generally, bats in Peninsular Malaysia feed mainly on insects with only few species from family Pteropodidae feed mainly on plants (Medway, 1969; Kingston *et al.*, 2006). Studies from Peninsular Malaysia have found that insectivorous bats fed predominantly on agricultural insect pests, suggesting the role of the bats as biological pest controller (Zubaid, 1988a; 1988b). Several studies have demonstrated how frugivorous bats aid in forest regeneration by feeding on fruits of pioneer plants and thus dispersing the seeds away from mother trees (Tan *et al.*, 2000; Hodgkison *et al.*, 2003). Others have supported the significant role of nectarivorous bats in pollination of food crops and mangrove plants through their feeding on the nectar and pollen (Start & Marshall, 1976; Nor Zalipah *et al.*, 2016). Therefore, understanding the diet of bats are necessary for fully understanding the ecological and economic roles of the bats.

Previous dietary studies of plant-visiting bats (family: Pteropodidae) in Peninsular Malaysia relied on the morphological-identification of seeds and pollen grains which are physically identifiable (Start & Marshall, 1976; Tan *et al.*, 1998; 2000; Hodgkison *et al.*, 2003; 2004; Fletcher *et al.*, 2012). Plant material which were ingested in liquid form (e.g., nectar) and digested into fragments (e.g., pulp) were disregarded by the previous studies due to the difficulties in identifying them. In addition, seeds and pollen grains of certain plant taxa lack distinctive morphological characteristics which consequently limited the identification of plants consumed by the bats (Pompanon *et al.*, 2012; Bell *et al.*, 2016).

1.2 DNA barcoding: prospects in conservation

In recent years, DNA barcoding has emerged as a novel tool for species identification. A short fragment of DNA at a specific region which is unique among species (e.g., *COI* for animals, Hebert *et al.*, 2003; *rbcL* for plants, CBOL, 2009) can be extracted from unknown specimen and matched to taxonomically verified DNA reference sequences for identification (Hebert *et al.*, 2003; Kress *et al.*, 2015). This technique can distinguish morphologically similar species which occur in sympatry and has minimal adverse impact on study species (Francis *et al.*, 2010; Sing *et al.*, 2013; Wilson *et al.*, 2014). Several studies from Peninsular Malaysia have demonstrated the capability of DNA barcoding to detect cryptic species in bats (Sing *et al.*, 2013), butterflies (Wilson *et al.*, 2013; Jisming-See *et al.*, 2016) and filaria (Uni *et* al., 2017).

The development of high-throughput sequencing platforms has introduced DNA metabarcoding which has been applied in a mammal survey by identifying DNA barcodes of mammal obtained from blowflies sampled in particular locations (Lee *et al.*, 2016). DNA metabarcoding has also been used to assess the foraging preference of honey bees by identifying the plant material present in honey (Hawkins *et al.*, 2015). Both DNA barcoding and metabarcoding have been used to study the diet of insectivorous (Clare *et al.*, 2009; 2014) and frugivorous bats (Hayward, 2013; Aziz *et al.*, 2017a) by identifying

the remains of consumed species in bat faeces. A global effort to build a comprehensive DNA barcode reference library for the species identification has generated large public databases such as Barcode of Life Datasystems – BOLD (Ratnasingham & Hebert, 2007) and GenBank (NCBI, 2016), providing a feasible means for species identification.

1.3 Objectives and research questions

The primary aim of this thesis is to use DNA barcoding (in general) to review the diversity of bats based on taxonomic records and the diet of two plant-visiting bat species in Peninsular Malaysia based on the following research questions:

1.3.1 What is the taxonomic status of the bats of Peninsular Malaysia based on the analyses of DNA barcodes which are publicly available on BOLD?

The diversity of bats in Peninsular Malaysia was reviewed in this study. The objectives were: (1) to review the taxonomic status of the bat species in the checklist based on analyses of DNA barcodes which are publicly available on the DNA barcode reference library, BOLD, (2) to chart the progress towards a comprehensive DNA barcode reference library (i.e., BOLD) for the bats of this region, and (3) to create a checklist of bat species reported from Peninsular Malaysia. This project has been published as Lim *et al.* (2017). A checklist of the bats of Peninsular Malaysia and progress towards a DNA barcode reference library. *PLoS ONE*, *12*(7), e0179555.

1.3.2 What is the diet of frugivorous bat, *Cynopterus brachyotis* based on the identification of pulps and seeds found in the bat faeces using DNA barcoding?

The diet of frugivorous bat, *C. brachyotis* at secondary forest, oil palm plantation and urban area in Peninsular Malaysia were compared in this study. The objectives were: (1) to examine the diet of *C. brachyotis* by identifying the pulps and seeds present in bat faeces using DNA barcoding which utilises Sanger sequencing, and (2) to investigate (i)

whether *C. brachyotis* can adapt to changing landscapes by exploiting cultivated and introduced plants as novel food resource and thus potentially dispersing these plants, or (ii) whether *C. brachyotis* feed on native plants, hence may aid in forest regeneration. This project has been published as Lim *et al.* (2018). Impact of urbanisation and agriculture on the diet of fruit bats. *Urban Ecosystems, 21*(1), 61-70.

1.3.3 What is the diet of nectarivorous bat, *Eonycteris spelaea* based on the identification of plant material present in the bat faeces using DNA metabarcoding?

The diet of nectarivorous bat, *E. spelaea* roosting in an urban cave in Peninsular Malaysia was examined in this study. The objectives were: (1) to examine the diet of *E. spelaea* by identifying the plant material present in bat faeces using DNA metabarcoding which utilises high-throughput next-generation sequencing, and (2) to investigate whether *E. spelaea* in an urban environment, (i) exploit introduced plants as food resources, thus potentially pollinating them and impacting the reproductive success of native plants, or (ii) feed primarily on native plants and hence remain as crucial pollinators of native plants in a highly disturbed habitat. This project has been published as Lim *et al.* (2018). Pollination implications of the diverse diet of tropical nectar-feeding bats roosting in an urban cave. *PeerJ, 6*, e4572.

CHAPTER 2: LITERATURE REVIEW

2.1 Land cover changes in Peninsular Malaysia

Between the year 2000 and 2010, the urban land in East-Southeast Asia has expanded by more than 22% (Schneider *et al.*, 2015). Similar trends are observed in Peninsular Malaysia where the forested area has shrunk by 14% between year 2000 and 2012 (Butler, 2013a) while the urban land and oil palm plantation are expanding 1.5% and 7% annually (Butler, 2013b; Schneider *et al.*, 2015). Such rapid land-cover changes are mainly driven by urbanisation and agriculture which are associated with the growing human population. In Peninsular Malaysia, the human population was estimated to be 18 million in year 2000 but has since increased to 25 million in year 2016 (DOSM, 2017).

Changes in land use are often associated with alterations to biogeochemical cycles, climate and biodiversity (Grim *et al.*, 2008; Fitzherbert *et al.*, 2008). For example, the introduction of non-native species in human-dominated areas (Grim *et al.*, 2008; Fitzherbert *et al.*, 2008) may compete with and extirpate native species (Faeth *et al.*, 2005; McConkey *et al.*, 2012). Despite the loss of biodiversity, important ecological processes still take place in urban and agricultural habitats. For example, intense landscaping often increases the species richness and homogeneity of plants in urban areas, where there are an increasing number of same non-native plants planted for urban beautification (Grimm *et al.*, 2008; Kowarik, 2011). These plants support a diverse assemblage of bee, birds and bats (Corlett, 2005; Aida *et al.*, 2016; Sing *et al.*, 2016), which in turn provides seed dispersal and pollination services, and consequently aid in maintaining green spaces in urban areas (Tan *et al.*, 2000; Corlett, 2005; Sheherazade *et al.*, 2017).

Understanding how ecosystem services in human modified environments are maintained, albeit often involving introduced species and novel interactions (Corlett, 2005), is a serious and growing challenge. The preference for planting particular plant species in urban areas, especially ornamental introduced plants, may create competition

between native and introduced plants for seed dispersal and pollination services which could affect the reproductive success and survival of native plants (Faeth *et al.*, 2005). Therefore, it is important to understand how a population uses plant resources in human modified environments for assessing how planting schemes will impact biodiversity and associated ecosystem services.

2.2 Bats of Peninsular Malaysia

Rapid deforestion and habitat degradation (driven by agriculture and urbanisation) have resulted in climatic and vegetation changes across Southeast Asia which consequently threatened the bats of the region (Hughes *et al.*, 2012; Kingston, 2013). About 25% of more than 1300 bat species in the world occur in Southeast Asia (Kingston, 2013; Voigt & Kingston, 2016). Of the 323 species in Southeast Asia assessed by IUCN, about 20% are considered to be threatened or near threatened while another 20% are categorised as “Data deficient"; the population trends for 24% are decreasing, 57% are unknown, 18% are stable while only 1% (representing *Cynopterus sphinx*) is thought to be increasing (Kingston, 2013).

Knowing (i) what bat species are present in Peninsular Malaysia, (ii) their distributions across the region, and (iii) their taxonomic status are crucial for developing suitable conservation plans (Francis *et al.*, 2010; Kingston, 2010; Tsang *et al.*, 2016). Several published checklists of bats have covered Peninsular Malaysia as part of a broader region, for example, “Walker’s bats of the world” (Nowak, 1994), “Horseshoe bats of the world” (Csorba *et al.*, 2003), and/or in combination with other mammal groups, for example, “A handlist of Malaysian mammals” (Chasen, 1940), “The mammals of the Indomalayan region: a systematic review” (Corbet & Hill, 1992), “Checklist of mammals from Malaysia” (Davison & Zubaid, 2007), and “Red list of mammals for Peninsular Malaysia” (DWNP, 2010). Other researchers have produced comprehensive checklists for particular localities: Krau Wildlife Reserve (Kingston *et al.*, 2006) and Ulu Gombak

(Sing *et al.*, 2013). Yet a comprehensive checklist of bats specifically for the entire geopolitical region of Peninsular Malaysia has never been published.

While Davison and Zubaid (2007) have reported 106 bat species for Peninsular Malaysia, the number is increasing with discoveries of new species. For example, *Kerivoula krauensis* (Francis *et al.*, 2007) and *Rhinolophus luctoides* (Volleth *et al.*, 2015) were recently recognised on the basis of divergences in mitochondrial DNA sequences and subtle distinctive morphological characteristics. Francis *et al.* (2010) had suggested that the species richness of bats across Southeast Asia may be underestimated by 50%, while Sing *et al.* (2013) had demonstrated how further intensive surveys may increase the species richness of bats in Peninsular Malaysia.

2.2.1 Morphological-based identification

In Peninsular Malaysia, bat species are traditionally recognised on the basis of the morphological characteristics of bats. For example, *Rhinolophus convexus* was described on the basis of its distinct noseleaf shape, and external and cranial measurements (Csorba, 1997). The congeneric *R. chiewkweeae* was once considered to be conspecific with *R. pearsoni* but is now recognised as a distinct species on the basis of external, cranial and dental measurements, consequently eliminated the occurrence of the latter species in Peninsular Malaysia (Yoshiyuki & Lim, 2005).

Examination of morphological characters may be of limited service when applied to identification of sympatric and morphologically similar species (Francis *et al.*, 2010; Wilson *et al.*, 2014). For example, *Hipposideros bicolor* sensu lato is a widespread species complex that comprises two sympatric species, *H. bicolor* and *H. atrox*, which are morphologically similar and overlap in forearm length but are acoustically and genetically distinct (Kingston *et al.*, 2001; Douangboubpha *et al.*, 2010a). Many recent reports from Peninsular Malaysia used *H. bicolor* to represent both species (Joann *et al.*,

2011; Hasan *et al.*, 2012; Jayaraj *et al.*, 2012a; 2013a) due to the difficulties in distinguishing the two species based on morphological characters.

2.2.2 Echolocation-based identification

For certain bat species, particularly those that feed on insects, the distinctiveness in their echolocation calls can aid in species identification. The *H. bicolor* sensu lato comprises two morphologically similar species that are acoustically distinct: *H. bicolor* which echolocate at 131 kHz and *H. atrox* which echolocate at 142 kHz (Kingston *et al.*, 2001; Douangboubpha *et al.*, 2010a). Another example is the *Kerivoula intermedia* and *K. minuta* which are morphologically similar but are generally distinguishable based on the forearm length (*K. intermedia*= >27 mm; *K. minuta*= ≤27 mm), body mass (*K. intermedia*= >25 mm; *K. minuta*= ≤2.5 mm) and echolocation frequency (*K. intermedia*= start frequency is 173±8 kHz and end frequency is 77±5 kHz; *K. minuta*= start frequency is 175±7 kHz and end frequency is 85±8 kHz) (Kingston *et al.*, 1999). However, for certain taxa, echolocation frequency may not be sufficiently distinct and may vary due to several factors including age, habitat and geographic locations (Kingston *et al.*, 1999; Hayes *et al.*, 2009).

2.2.3 Microsatellite analysis

This approach has been used to examine the genetic structure of populations of *Cynopterus* (Campbell *et al.*, 2006) and *Rhinolophus* bats (Lim, 2012) in Peninsular Malaysia. Microsatellites are simple tandemly repeated DNA sequences that occur throughout the genome (Rossiter, 2009). As microsatellites exhibit high degree of variation among individuals and within a population, analysis of multiple microsatellite loci can provide individuals with unique DNA profiles (Hillis *et al.*, 1996; Piggott & Taylor, 2003). Besides being non-lethal, microsatellite analysis can provide genetic data with small amount of sample (e.g., wing punch samples) through PCR amplification (Palsbøll, 1999; Lim, 2012). The mutation rate of microsatellites is also higher than

allozymes, with longer microsatellites generally exhibiting greater numbers of alleles (Rossiter, 2009). However, microsatellites cannot be targeted with universal markers as they occur in non-coding regions characterised by high rates of substitution (Rossiter, 2009). Therefore, this approach requires the development of specific microsatellite primers for closely-related species, normally within the same family (Hillis *et al.,* 1996; Burland & Worthington Wilmer, 2001; Piggott & Taylor, 2003). Microsatellite analysis is also highly prone to error when the quality and quantity of DNA is low (Piggott & Taylor, 2003), besides being laborious (i.e., lab procedures) and expensive (i.e., primers, reagents and sequencing) (Rossiter, 2009). Although microsatellite analysis can potentially be used to identify the species of an individual, this approach remains costly and laborious due to the need for developing large number of specific microsatellite primers (Tuler *et al.,* 2015).

2.2.4 Allozyme electrophoresis

Allozymes are variants of polypeptides produced by different alleles at the same gene locus (Buth, 1984; Hillis *et al.,* 1996). Allozyme electrophoresis utilises these allelic variations of allozymes as genetic markers to (i) analyse the population structure of a species, (ii) delineate species boundaries, (iii) trace the evolutionary relationships of more than two taxa, and (iv) identify the genetic similarities/differences between taxa (Hillis *et al.*, 1996; Richardson *et al.,* 2012). To date, there are no studies from Peninsular Malaysia which used this approach to examine the genetic structure of bats. Nevertheless, early genetic studies of bats from elsewhere were based on the single-locus screening and utilized allozyme electrophoresis (Rossiter, 2009). However, allozymes may not be sufficiently variable in some taxa (Hillis *et al.*, 1996). For example, Cooper *et al.* (1998) examined 45 loci of two morphologically distinct *Rhinolophus megaphyllus* and *R. philippinensis* in Australia using allozyme electrophoresis, and discovered low allozyme divergence among the two species which suggested that the two species are monophyletic

and recently diverged, contradicting their analysis of control region mtDNA which suggested that two species are polyphyletic. Allozyme electrophoresis also involves lethal tissue collection and requires immediate cryogenic storage of tissue samples which is difficult in tropical and isolated sampling sites, and therefore is rarely used now (Burland & Worthington Wilmer, 2001).

2.2.5 Chromosomal analysis

Generally, chromosome identification involved the banding of chromosomes for identifying the homologs. Once the homologs are identified, chromosomes are arranged as karyotype by cutting out photographic prints of chromosomes and pasting the homologs in paris on white cardboard. The chromosomes are measured (with either a rule of digitizer map) to obtain relative lengths and centromere indices, providing quantitative data for classifying each chromosome's morphology (Sessions, 1996). This approach has been widely used to examine the variations in chromosomes and hence the genetic diversity of bats in Malaysia (Heller & Volleth, 1984; Volleth *et al.*, 2015). One example is the case of *Kerivoula lenis* and *K. papillosa* which are grouped closely by Corbet and Hill (1992) but are recognised to be distinct by Khan *et al.* (2008) on the basis of karyotypic characters: *K. papillosa* has a diploid number of chromosomes=38 and fundamental number=54 whereas *K. lenis* has a diploid number of chromosomes=38 and fundamental number=52. However, the reliability of chromosome identification relies on the banding patterns or chromosome-specific markers, in addition to the limitations posed by the techniques used in preparing the samples (e.g., hybridization using radioactive probes and accessibility of chromosomal target DNA to the reagents) (Sessions, 1996).

2.3 Diet of plant-visiting bats in Peninsular Malaysia

In Peninsular Malaysia, the diet of plant-visiting bats (family: Pteropodidae), particularly the most common frugivorous bat, *Cynopterus brachyotis* sensu lato and the nectarivorous bat, *Eonycteris spelaea,* have been well-studied.

The lesser dog-faced fruit bat, *Cynopterus brachyotis* sensu lato is a species complex, often reported as *C. brachyotis* (Campbell *et al.*, 2004; Wilson *et al.*, 2014) and is the most common species of bat in Peninsular Malaysia, often recorded at primary and secondary forests, agricultural land, and urban areas (Campbell *et al.*, 2004; Jayaraj *et al.*, 2012a). Because of its ubiquitous presence, *C. brachyotis* sensu lato is an excellent model of ecological flexibility with a potentially important role in seed dispersal. *C. brachyotis* sensu lato has been reported feeding on sixteen plant species in primary forest (Hodgkison *et al.*, 2004), 66 plant species in secondary forests (Tan *et al.*, 1998) and 38 species in urban areas (Tan *et al.*, 2000). While *C. brachyotis* sensu lato in urban areas demonstrated distinct food preferences during fruiting seasons (Tan *et al.*, 2000), *C. brachyotis* sensu lato in primary forest exploited both "steady state" and "big bang" plants and did not show variation in capture rate over time during the bat survey (Hodgkison *et al.*, 2004). The apparent flexibility of *C. brachyotis* sensu lato in diet suggests a significant capability to adapt to changing environments. However, the flexible use of modified habitats may also bring the fruit bats into conflict with farmers in agricultural areas where bats may be perceived as foraging for food in cultivated commercial crops and consequently targeted as crop pests (Fujita & Tuttle, 1991).

The cave nectar bat, *Eonycteris spelaea*, is generally categorised as specialised nectarivorous bat (Fleming *et al.*, 2009; Stewart & Dudash, 2017) that feeds on nectar and pollen, and consequently provides pollination services (Srithongchuay *et al.*, 2008; Bumrungsri *et al.*, 2009; Acharya *et al.*, 2015a; Nor Zalipah *et al.*, 2016). *E. spelaea* is one of three nectarivorous bats present in Peninsular Malaysia and is often recorded in urban and agricultural areas (Lim *et al.*, 2017). The capability of *E. spelaea* to travel long distances for food and visit night-blooming plants with high frequency likely contributes to an important role as a pollinator (Start & Marshall 1976; Stewart & Dudash, 2017). The diet of *E. spelaea* in Southeast Asia was previously assessed through morphological

identification of pollen grains (found in faeces and on the body of bats) examined microscopically. Start and Marshall (1976) observed 31 distinct types of pollen in faeces of *E. spelaea* collected under two roosts at Batu Caves and Gua Sanding in Peninsular Malaysia but could only identify the pollen grains of 17 plant species. Bumrungsri *et al.* (2013) collected eleven types of pollen from captured individuals of *E. spelaea* at Khao Kao Cave in Thailand but could only identify the pollen grains of four plant species. Similarly, Thavry *et al.* (2017) recorded thirteen types of pollen in faeces of a roosting colony at Bat Khteas Cave in Cambodia but could only identify the pollen grains of four plant species.

2.3.1 Morphological-based identification of plant material

Previous dietary studies of frugivorous (Phua & Corlett, 1989; Tan *et al.*, 1998; 2000; Hodgkison *et al.*, 2004; Fletcher *et al.*, 2012) and nectarivorous bats (Start & Marshall, 1976; Bumrungsri *et al.*, 2013; Thavry *et al.*, 2017) mainly relied on the morphological identification of seeds and pollen grains found in the faeces of bats, on the bodies of captured bats and under the roosts of bats. However, seeds and pollen grains of certain plant taxa lack distinctive morphological characteristics (e.g., genera *Artocarpus* and *Ficus*) which consequently limited the identification of plants consumed by the bats (Pompanon *et al.*, 2012; Bell *et al.*, 2016). Seeds that could not be morphologically identified were germinated for morphological identification of the seedlings (Hodgkison *et al.*, 2003; 2004) but this approach is laborious and time-consuming. Such morphological identification also relies heavily on the availability of botanical reference specimens with diagnostic pollen grain, seed, flower and fruit, yet these botanical reference specimens are often incomplete (Aziz *et al.*, 2017a; Kress, 2017).

In addition, these particular studies prioritised solid plant material such as seeds and pollen grains which are physically identifiable in faeces and on bodies of bats, and by necessity disregarded other types of plant material defecated by the bats (i.e., nectar and

leaf fragments). As a result, these particular studies may have overestimated the importance of less digestible plant material (i.e., seeds and pollen grains) as food source for the bats (Voigt *et al.*, 2009) and overlooked the exact ecological role of the bats (Pompanon *et al.*, 2012). Identifying the fragmented and liquid plant material remains necessary for fully understanding the ecological role of the bats and determining whether the interactions between the bats and plants are mutualistic or antagonistic (Kress, 2017).

2.3.2 Direct observation of bat's feeding behaviour

The foraging preference of *C. brachyotis* (Tan *et al.*, 2000; Fletcher *et al.*, 2012) and *E. spelaea* (Gould, 1978) have been directly observed as part of behavioural studies of the bats but were often difficult due to the low light condition at night. A recent study from Peninsular Malaysia has used camera traps to observe the feeding behaviour of island flying fox, *Pteropus hypomelanus* (Aziz *et al.*, 2017b) but this method is expensive due to the cost of camera traps and thus limits the number and angle of observation points.

2.3.3 Stable isotope analysis

This technique can provide long-term and quantitative information on diet of plant-visiting bats and their foraging range, by considering the fact that composition of isotopes in the diet of the animal can be explained by the ratios of stable carbon and nitrogen isotopes in the animal tissues (Voigt *et al.*, 2009). Stable isotopes of carbon occur at varying ratios due to the particular enzymatic route of CO_2 fixation in plants and in plant-visiting animals based on their diet (DeNiro & Epstein, 1978) while nitrogen isotopes may be unequally distributed in an ecosystem due to the presence of nitrogen fixing plants (e.g., Fabaceaea) and usage of chemical fertilizer (DeNiro & Epstein, 1981). Stable isotope analysis has been used to identify the turnover rate of stable isotopes in tissues and blood of bats to determine the relative importance of fruits and insects as food sources for bats (Herrera *et al.*, 2001) and understand how specific diet impact the metabolic rate of bats (Voigt *et al.*, 2003). However, the ability of stable isotope analysis to determine

the relative importance of particular plants as food source for the bats depends on the assumptions and model adopted, and therefore, this technique is subject to biases in estimating the dietary preference of bats, especially those that feed on various food items (Herrera *et al.*, 2001; Voigt *et al.*, 2009). Moreover, this technique is only effective for providing information on generalized trophic levels and could not identify the plant remains in faeces and ejecta specifically to species (Herrera *et al.*, 2001).

2.4 DNA barcoding and metabarcoding for assessing species diversity and diet of bats

One potential tool to examine the diversity and diet of bats is the molecular method, DNA barcoding (Hebert *et al.*, 2003). To date, there are only few related studies from Peninsular Malaysia and therefore, the potential of DNA barcoding to assess the diversity of bats and their plant-based diet in the region remains to be explored.

2.4.1 DNA barcoding

DNA barcoding, which utilises Sanger sequencing, focuses on the variation in the amplified short, standardised region of the genome for identification of closely related taxa and unknown specimens (Hebert *et al.*, 2003; Hajibabaei *et al.*, 2007; Kress *et al.*, 2015). These short DNA fragments (also known as DNA barcodes) are represented by unique arrangement of nucleotide codes (i.e., A, C, G and T) – such variation occurs among and within species and therefore is particularly useful in drawing the species boundary. In a general workflow of DNA barcoding, DNA is extracted from specimens, PCR amplified at a specific standardised region e.g., *COI* for animals (Hebert *et al.*, 2003); *rbcL* and *ITS2* for plants (CBOL, 2009; Chen *et al.*, 2010) with universal group-specific PCR primers, and Sanger sequenced for DNA barcode which is later matched to taxonomically verified DNA sequences for species identification.

DNA barcoding can provide informative genetic data for resolving problems in taxonomy of certain taxa of bats albeit with some limitations (Francis *et al.*, 2010). The

bat diversity of Peninsular Malaysia was previously estimated to be 106 species (Davison & Zubaid, 2007) but the number is increasing, particularly with the recent recognition of cryptic species as distinct species based on DNA barcoding at COI mtDNA (e.g., Francis *et al.*, 2007). Cryptic species (Bickford *et al.*, 2007) is first detected when their supposedly conspecific DNA barcodes fail to match closely and display high divergence with reference sequences from taxonomically verified specimens, consequently demonstrating the potential of DNA barcoding as a species discovery tool (Francis *et al.*, 2007; Sing *et al.*, 2013). Furthermore, DNA can be extracted from hair, tail membrane and wing punch samples; the collection of which has minimal adverse impacts on live bats (Faure *et al.*, 2009; AMNH, 2012). Therefore, DNA barcoding can assist in estimating the phylogenetic diversity of bats of Peninsular Malaysia with implications for conservation approaches for bats and their habitats in the region which are in dire need for protection.

DNA barcoding can also aid in identification of the remains of consumed species in faeces of insectivorous (Clare *et al.*, 2009) and frugivorous bats (Hayward, 2013) even without the high level of taxonomic expertise which is required for morphological-based identification (Pompanon *et al.*, 2012). However, this targeted approach requires the isolation of physical remains of consumed species (i.e., insect legs and plant pulp) from the faeces which consequently limits the amount of physical remains for analysis and recovery of DNA from many consumed species present in the faeces (Pompanon *et al.*, 2012; Shokralla *et al.*, 2012). Nevertheless, DNA barcoding remains a feasible approach for identifying the plant material in faeces of frugivorous bats, of which to date has been demonstrated by only one study (Hayward, 2013). On the other hand, traditional DNA barcoding may not be suitable for dietary study of nectarivorous bats which tend to ingest and defecate plant material in liquid form (e.g., nectar).

2.4.2 DNA metabarcoding

The recent advances in high-throughput sequencing platforms have introduced DNA metabarcoding which utilises next-generation sequencing (NGS) (Brandon-Mong *et al.*, 2015; Kress *et al.*, 2015). DNA metabarcoding involves simultaneous DNA sequencing of multiple templates in complex samples (e.g., faeces) and allows detection of multiple species at once (Pompanon *et al.*, 2012; Brandon-Mong *et al.*, 2015; Lee *et al.*, 2016). This technique has been used to identify the digested material in faeces of insectivorous (Clare *et al.*, 2014) and frugivorous bats (Aziz *et al.*, 2017a), providing insights into diet and ecological role of the bats.

To date, DNA metabarcoding has not been used to examine the diet of nectarivorous bats but has been used to identify the plant material present in honey (a complex sample in liquid form), consequently provided information regarding the sources of nectar collected by the honey bees (Hawkins *et al.*, 2015; de Vere *et al.*, 2017). Previous dietary studies of nectarivorous bats in Southeast Asia (Start & Marshall, 1976; Bumrungsri *et al.*, 2013; Thavry *et al.*, 2017) identified only pollen grains which are physically identifiable in faeces and on bodies of bats, and by necessity disregarded the plant material ingested and defecated in liquid form such as nectar. As nectarivorous bats feed mainly on nectar and pollen (Start & Marshall, 1976; Fleming *et al.*, 2009; Stewart & Dudash, 2017), it is necessary to identify the nectar in order to fully understand the ecological role of the bats. Therefore, the utility of DNA metabarcoding to examine the plant material present in faeces of nectarivorous bats remains to be explored.

CHAPTER 3: METHODOLOGY

3.1 Bat diversity of Peninsular Malaysia

3.1.1 Literature search

A preliminary checklist for Peninsular Malaysia was compiled from published checklists (Medway, 1969; Corbet & Hill, 1992; Kingston *et al.*, 2006; Davison & Zubaid, 2007; DWNP, 2010; Sing *et al.*, 2013). A search for additional published records of bat species reported from Peninsular Malaysia was conducted through Google Scholar (https://scholar.google.com), Web of Science (https://www.webofknowledge.com), PubMed (http://www.ncbi.nlm.nih.gov/pubmed), Cab Direct (http://www.cabdirect.org) and Biodiversity Heritage Library (http://www.biodiversitylibrary.org) using keywords "Chiroptera", "bats", "bat species", "Peninsular Malaysia", and "DNA barcoding". Data from bat surveys conducted in Peninsular Malaysia were also requested directly from government agencies (Department of Wildlife and National Parks and Forest Research Institute Malaysia) and researchers known to be active in this region (Dr. Charles M. Francis and Prof. Dr. Zubaid Akbar Mukhtar Ahmad).

Museum collection numbers of type specimens were obtained from literature. The following abbreviations were used for museum collections: Natural History Museum, London, UK, (BM(NH)); Centre for Thai National Reference Collections, Bangkok, THAILAND (TNRC); National Museum of Malaysia, Kuala Lumpur, MALAYSIA (MNM); National Museum of Natural History, Washington D.C., USA (USNM); Forschungsinstitut und Natur-Museum Senckenberg, Frankfurt am Main, GERMANY (SMF); Hungarian Natural History Museum, Budapest, HUNGARY (HNHM); National Science Museum, Tokyo, JAPAN (NSMT); Museum National d'Histoire Naturelle, Paris, FRANCE (MNHN), Museum für Naturkunde, Berlin, GERMANY (MNB), National Museum of Natural History Naturalis, Leiden, NETHERLANDS (NMNL), Field Museum of Natural History, Chicago, Illinois, USA (FMNH), and Department of

Wildlife and National Parks, MALAYSIA (DWNP). Scientific names were checked against usage in the Mammals of the World list maintained by Dr. Nancy Simmons of the American Museum of Natural History whereas common English (vernacular) names followed the "Field Guide to the Mammals of Southeast Asia" (Francis, 2008). The current conservation status for each species were obtained from IUCN (2016).

3.1.2 Progress of DNA barcoding

Based on the checklist obtained as above, the BOLD Taxonomy Browser (Ratnasingham & Hebert, 2007) was searched for the availability of DNA barcodes (the standard COI mtDNA region for animals) on BOLD representing each species. The localities and associated Barcode Index Numbers (BINs) (Ratnasingham & Hebert, 2013) of all public DNA barcodes for the listed species were recorded. A BIN is a molecular operational taxonomic unit with high correspondence to "traditional" species boundaries and also a unique alphanumeric code associated with the DNA barcodes (>500bp) it comprises on BOLD. In several cases detailed below, DNA barcodes are likely to represent certain species based on their placement on taxon identification (taxon ID) trees produced by BOLD v.4 (Ratnasingham & Hebert, 2007) but are not presently recorded as those species (i.e., unnamed or recorded under different names). For certain taxa, MEGA 7 (Kumar *et al.*, 2016) was used to construct Neighbour-Joining (NJ) trees of the public DNA barcodes using the Kimura 2-parameter model (Kimura, 1980) and bootstrapping with 500 replicates (Soltis & Soltis, 2003).

3.2 Diet of frugivorous bat, *C. brachyotis* in Peninsular Malaysia

3.2.1 Ethics

Faecal collection and bat sampling were conducted with authorisation from Department of Wildlife and National Parks, Peninsular Malaysia (JPHLandTN(IP)100-

34/1.24 Jld. 4(34)) using protocol approved by Institutional Animal Care and Use Committee, University of Malaya (ISB/10/06/2016/LVC (R)).

3.2.2 Study sites and bat species

Faecal sampling was conducted at three sites with either urban, agricultural or secondary forest land use (Figure 3.1). The urban site was an abandoned residential area located between University of Malaya and MAHSA University in Kuala Lumpur city in close proximity to a busy hospital and occupied residences. The agricultural site was located within a 2940 ha oil palm plantation (*Elaies guineensis* x *Elaies oleifera*) at Bemban, Melaka. The secondary forest site was located at the University of Malaya Field Studies Centre which is situated within 120 hectares of a secondary forest selectively logged from 1956 to 1958 (Medway 1966; Sing *et al.*, 2013).

Fresh faeces were collected from individual bats (identified as *C.* cf. *brachyotis* SUNDA following Jayaraj *et al*. (2012b) but referred as *C. brachyotis* in this study) captured using mist nets at the urban site for eleven days from 10 June to 18 December 2015 and at the agricultural site for four days from 12 January to 15 January 2016. Most of the bats defecated immediately when captured, but those that did not were kept in individual cloth bags for one hour to produce faeces and were then released. The faeces collected from one individual was considered as a single independent sample.

A roosting colony (identified as *C.* cf. *brachyotis* SUNDA by capturing and measuring four individuals from the colony following Jayaraj *et al.* (2012b) but referred as *C. brachyotis* in this study) was located at the secondary forest site. The floor below the roost was cleaned daily and fresh faeces from the colony were collected from the floor non-invasively between 10 July and 25 September 2015. Each faecal sample (i.e., collected into an individual Eppendorf tube) was treated as an independent sample.

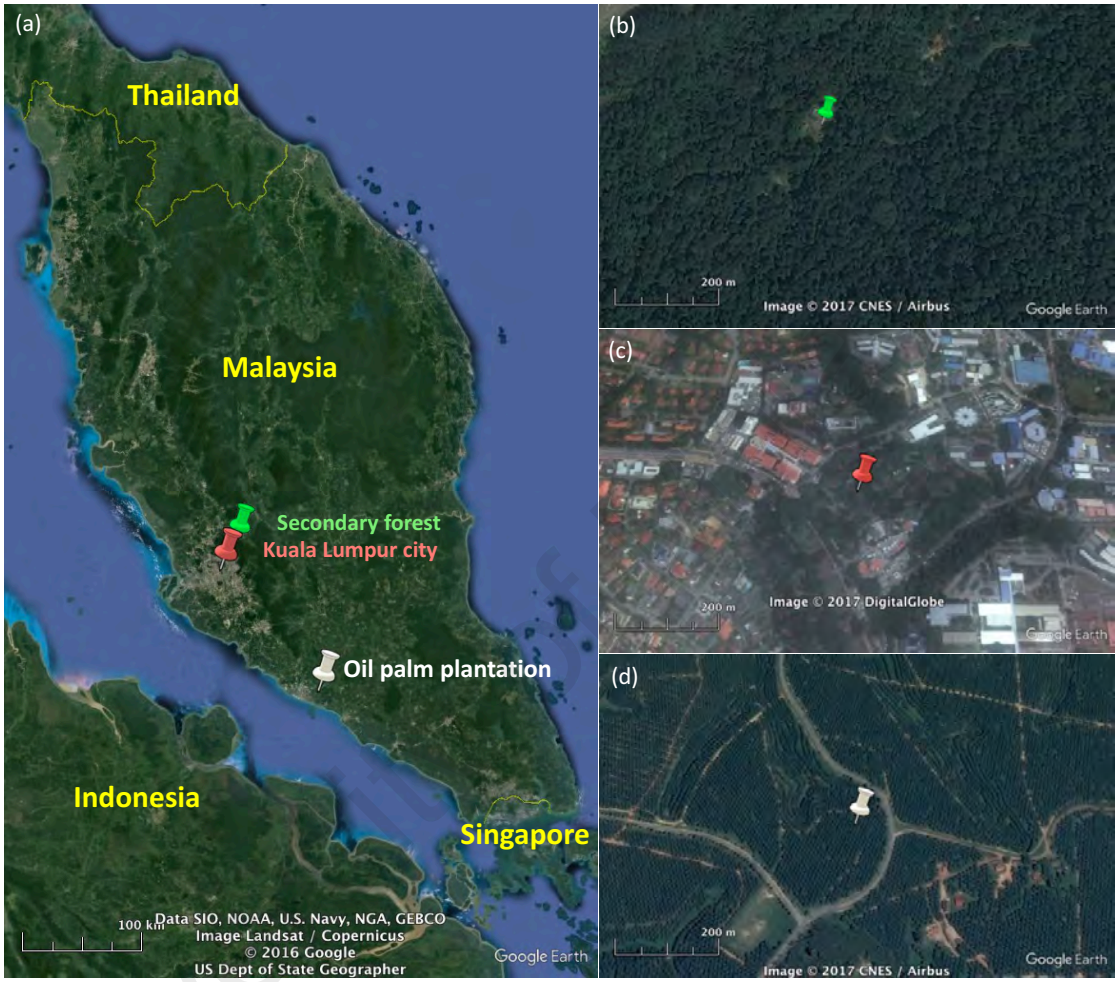

Figure 3.1: The sampling locations in Peninsular Malaysia. (a) The map of Peninsular Malaysia. (b) The sampling location at secondary forest. (c) The sampling location at urban area. (d) The sampling location at oil palm plantation.

The faeces were kept in 1.5 ml Eppendorf tubes filled with 99.8% ethanol and stored at -20°C prior to analysis. Ethanol is not normally used to preserve plant material, but is recommended to prevent fungal and bacterial growth in bat faeces. The ethanol was evaporated from samples prior to extraction. Due to the limit of the plant box which allows 96 samples for each analysis, a total of 95 faecal samples were selected for plant DNA barcoding incorporating approximately equal number of samples from each site (i.e., 32 samples from the urban site, 32 samples from the agricultural site and 31 samples from the secondary forest site) and one positive sample.

3.2.3 DNA extraction, amplification and sequencing

Seeds were prioritised over pulps to ensure the amplification of DNA. In cases where seeds were not found in the faecal samples, the pulps were used. The seeds and pulps were isolated from the faecal samples and sent to the Canadian Centre for DNA barcoding (CCDB) for DNA extraction, PCR amplification, and Sanger sequencing of two gene regions (*rbcL*: ~550 bp and *ITS2*: ~350 bp), following the standard plant protocols of the CCDB (Ivanova & Grainger, 2008; Ivanova *et al.,* 2011; Kuzmina & Ivanova, 2011a; 2011b).

3.2.4 Plant species identification

The resultant DNA barcodes of *rbcL* and *ITS2* regions were BLAST-ed (searched) (Boratyn *et al.,* 2013) against GenBank (NCBI, 2016) to assign taxonomic names to the barcodes. The results of *ITS2* searches were prioritised over *rbcL* due to the greater taxonomic resolution of this gene fragment (Chen *et al.,* 2010; Kuzmina *et al.,* 2012). Species names were assigned based on *ITS2* and *rbcL* matches using a customised set of criteria (Figure 3.2). See Appendix A for details of the assignment criterion.

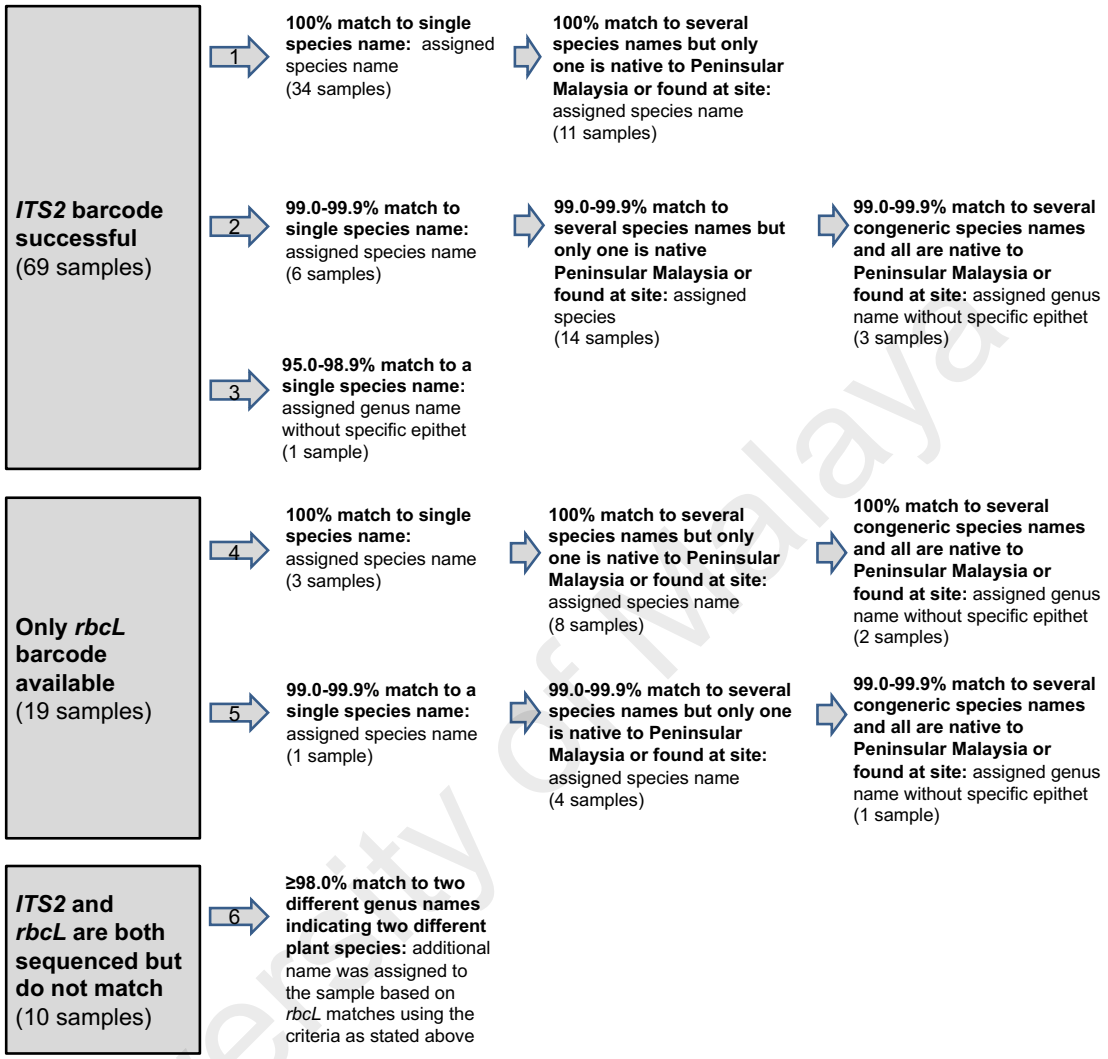

Figure 3.2: Criteria used to assign taxonomic names to the plant DNA barcodes based on matches returned by BLAST searches on Genbank, NCBI database.

The DNA barcodes and the sample metadata were uploaded onto the BOLD v4. (Ratnasingham & Hebert, 2007) under project code VCCBD. The barcodes are available in GenBank under accessions KY080541 to KY080613 and KY080617 to KY080686.

3.2.5 Species richness and sampling completeness ratio

The following analyses were performed using R version 3.3.1. (R Core Team, 2017). The occurrence of identified plants in faeces was used to estimate species richness and sampling completeness ratio of the faecal sampling using SpadeR package (Chao & Shen, 2010). Chao1, which is good for datasets skewed towards low abundance class, is used to estimate the plant species richness in the faecal samples based on the observed frequency of each plant species in each sampling site (Chao & Chiu, 2016). Several Chao1 models were used to assess consistency of estimates provided by each model. A homogeneous model was also included under the assumption that all plant species have the same detection probabilities, but usually severely underestimates the true species richness if heterogeneity exists (Chao & Chiu, 2016).

3.2.6 Dietary resource overlap

An interaction figure between the bats and detected plants was created to compare the food resource use of *C. brachyotis* at three sampling sites with different land use using the bipartite package (Dormann *et al.*, 2008). The occurrence of identified plants in faeces was used to quantify the dietary resource overlap between the fruit bats in three sampling sites using Pianka's measure of niche overlap (Pianka, 1973; Equation 3.1) in EcoSimR with niche null model with RA3 algorithm (Gotelli & Ellison, 2013). The following was considered based on Rödder and Engler (2011): (1) there is no overlap or limited overlap in dietary resource between the fruit bats if the value is in the range of 0 to 0.2, (2) there is low overlap if the value is in the range of 0.2 to 0.4, (3) there is moderate overlap if the value is in the range of 0.4 to 0.6, (4) there is high overlap if the value is in the range of 0.6 to 0.8, and (5) there is a very high overlap if the value is in the range of 0.8 to 1.

Equation 3.1:

$$O_{jk} = \frac{\sum_i^n p_{ij} p_{ik}}{\sqrt{\sum_i^n p_{ij}^2 \sum_i^n p_{ik}^2}}$$

where p_{ij} is the proportion that resource i is of the total resources used by species j; $p_{ik}$is the proportion that resource i is of the total resources used by species k; and n is the total number of resource states.

3.3 Diet of nectarivorous bat, *E. spelaea* in Peninsular Malaysia

3.3.1 Ethics

Faecal collection was conducted at Dark Cave, Batu Caves with authorization from the Department of Wildlife and National Parks, Peninsular Malaysia (Ref: JPHL&TN(IP)100-34/1.24 Jld. 4(34)), the Malaysian Nature Society and Majlis Perbandaran Selayang (Ref: Bil(35)dlm.MPS 3/3-117/153 JL) using a protocol approved by the Institutional Animal Care and Use Committee, University of Malaya (Ref: ISB/10/06/2016/LVC (R)).

3.3.2 Study site and bat species

Batu Caves constitute an extensive karst cave system developed within an isolated 329 m high limestone massif located in Gombak District, part of the Klang Valley conurbation in Selangor state adjacent to Kuala Lumpur Federal Territory (Moseley *et al.*, 2012; Grismer *et al.*, 2014). Batu Caves is surrounded by industrial parks and residential areas (Grismer *et al.*, 2014) and includes a Hindu temple that has become a major tourist attraction (Kasim, 2011). The cave complex includes the Dark Cave, a protected cavern with >2000 m of passages (Price, 2002) managed by the Cave Management Group under the Malaysian Nature Society (http://www.darkcavemalaysia.com/). Dark Cave is an ecologically diverse karst cave

system which supports a large number of animals (Moseley, 2009; Moseley *et al.*, 2012) including a colony of *E. spelaea*. Start and Marshall (1976) estimated that the colony comprised >10,000 individuals whereas Beck and Lim (1972) and Gould (1988) estimated >4000 individuals. For this study, faecal samples were collected under the *E. spelaea* roost at Dark Cave (Figure 3.3).

3.3.3 Faecal collection

Approximately 10 ml of fresh faecal samples were collected non-invasively under the roost of *E. spelaea* once every week from 31 December 2015 to 4 March 2016 (i.e., 10 days over 10 weeks). Overall, a total of ~100 ml of fresh faecal material was collected and used for the study. As the Cave Management Group cleans the floor below the roost daily to prevent the accumulation of bat faeces (which is unappealing to tourists), faeces below the roost were assumed to be deposited the previous night. The faeces were kept in 1.5 mL tubes filled with 99.8% ethanol and stored at -20°C prior to analysis.

3.3.4 Preparation of faecal samples

The faeces were centrifuged to form pellets and the supernatant were discarded. The pellets were incubated at 56ºC for 2 hours to evaporate moisture (i.e., ethanol), pooled according to collection week and homogenised using a TissueLyser II (Qiagen, Germany) with 3 mm tungsten carbide beads (Qiagen, Germany) for 4 minutes at 30 1/s.

3.3.5 Plant DNA extraction, PCR amplification, clean-up and sequencing

DNA extraction was performed twice using the QIAamp DNA stool mini kit (QIAGEN, Germany) following the manufacturer's protocol which resulted in two DNA replicates for each weekly sample. The purity and concentration of the DNA was examined with NanoDrop 2000c UV-Vis Spectrophotometer (Thermo Fisher Scientific). DNA extracts with a purity range from 1.8 to 1.9 and concentrations $\geqslant$50 ng/ µl were used for PCR amplification.

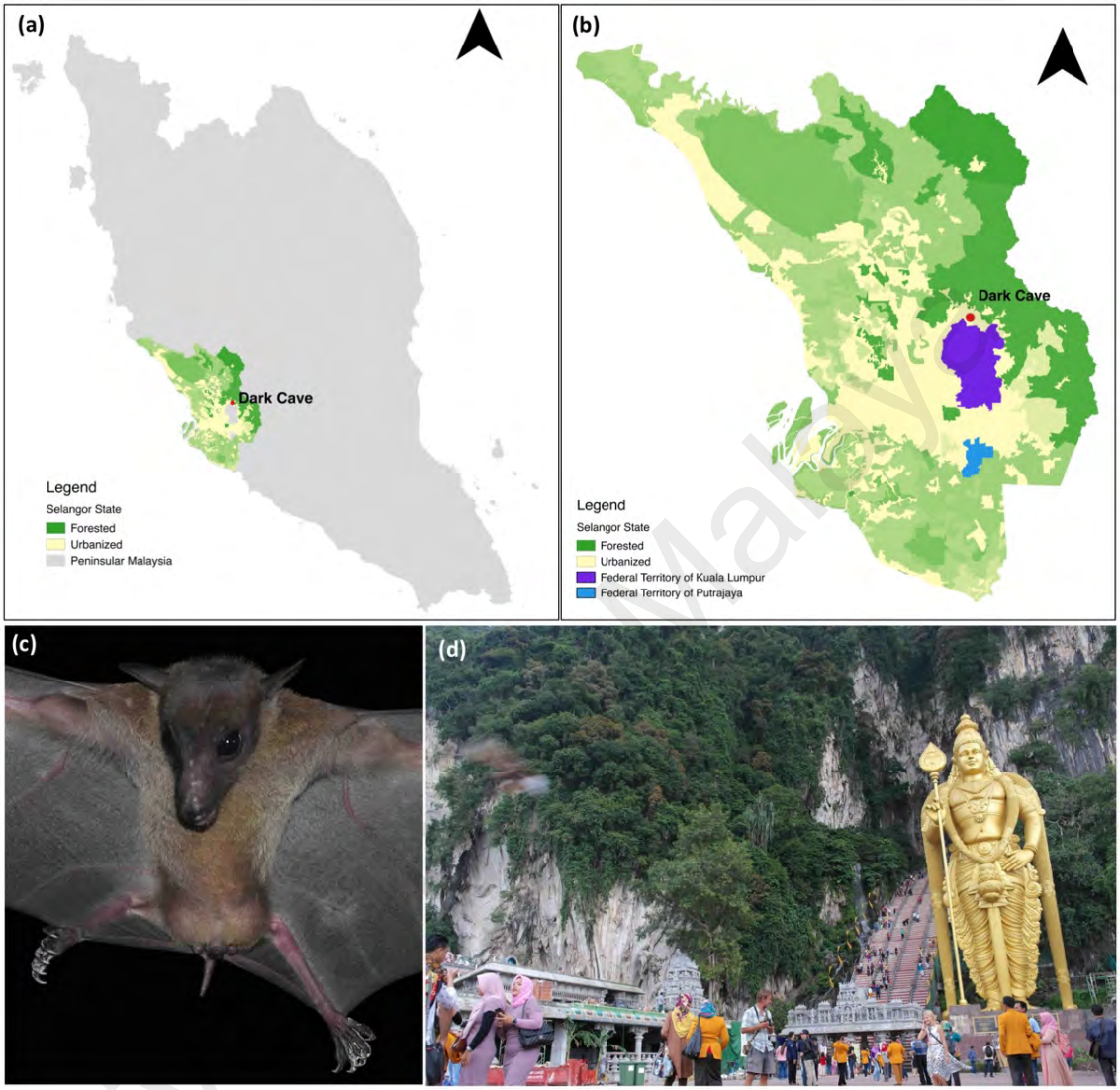

Figure 3.3: A permanent roosting colony of *Eonycteris spelaea* was located at Dark Cave Conservation Site, one of the caves in Batu Caves. (a) The location of Dark Cave Conservation Site in Peninsular Malaysia (b) Land cover of Selangor state where Dark Cave is located (source: www. http://www.globalforestwatch.org/) (c) Close-up of *E. spelaea* (d) Batu Caves serves as temple for Hindu prayers and tourist attraction for its cultural and natural heritage.

Two DNA barcode markers were selected for this study: *rbcL* due to its relative universality (i.e., universal primers; CBOL, 2009) and *ITS2* due its higher taxonomic resolution (Chen *et al.*, 2010). Both markers have a large number of taxonomically verified DNA reference sequences available in NCBI GenBank (http://www.ncbi.nlm.nih.gov/) (*rbcl*= 155,634; *ITS2*= 243,155; Bell *et al.*, 2016) and have been used successfully to examine the diet of rolled-leaf beetles in a tropical rainforest in Costa Rica (García-Robledo *et al.*, 2013) and the plant sources of honey (Prosser & Hebert, 2017).

Fragment of *rbcL* and *ITS2* were amplified using universal primers with Illumina adaptors (Table 3.1:). Five PCR amplifications were performed for each DNA extract replicate together with one positive (*Musa* sp.) and one negative (ddH_20) control. Each PCR amplification was performed in a total volume of 25 µL consisting of 12.5 µL EconoTaq PLUS GREEN 2X Master Mix (Lucigen, USA), 0.25 µL of each forward and reverse primer (100 µM), 7 – 8 µL of ddH_2O, and 4 – 5 µL of DNA. The thermocycling profile for *rbcL* was: initial denaturation at 95 ºC for 2 minutes, denaturation, annealing and extension at 95 ºC for 30 seconds, 55 ºC for 30 seconds, 72 ºC for 10 seconds (35 cycles), and a final extension at 72 ºC for 6 minutes. The thermocycling profile for *ITS2* was: initial denaturation at 94 ºC for 2 minutes, denaturation, annealing and extension of 94 ºC at 30 seconds, 55 ºC at 30 seconds, 72 ºC at 20 seconds (35 cycles), and a final extension at 72 ºC for 10 minutes.

PCR products were checked on 2% agarose gels and extracted and purified using a NucleoSpin Gel and PCR Clean-up kit (Macherey-Nagel, Germany) following the manufacturer's instructions. The purified products were assessed with a NanoDrop 2000c UV-Vis Spectrophotometer (Thermo Fisher Scientific). Products with purity ranging from 1.8 to 1.9 and concentration $\geqslant$50 ng/ µl were used for a second round of PCR to

generate amplicons containing dual-index multiplex identifier (MID) tags and sequencing on an Illumina Miseq Sequencer (Illumina, USA) with 2 × 300 bp paired-end read setting.

Paired-end reads were sorted into datasets (i.e., weeks) by MID and merged (for *ITS2*). *RbcL* reads could not be merged due to the lack of overlapping sequence between paired-end reads. Therefore, *rbcL* reads containing only the forward primer were used in subsequent steps as these sets of reads were longer and more abundant.

3.3.6 Filtering pipeline

Using the Galaxy web server (https://usegalaxy.org/, Giardine *et al.*, 2005), files were converted to Illumina1.8+ format using "FASTQ Groomer" (Blankenberg *et al.*, 2010). Primers were removed using "Clip" (http://hannonlab.cshl.edu/fastx_toolkit/). Short (rbcL<100 bp; ITS2<320 bp) and low quality (QV<20) reads were discarded using "Filter FASTQ" (Blankenberg *et al.*, 2010). Remaining reads were de-replicated with 100% identity using "VSearch dereplication" (Rognes *et al.*, 2015). Duplicates and possible chimeras were then removed using "cd-hit-dup" (Fu *et al.*, 2012). Remaining reads were clustered into operational taxonomic units (OTU) with 98% identity using "VSearch clustering" (Rognes *et al.*, 2015).

3.3.7 Assignation of taxonomic names

OTU were BLAST-ed (searched) against NCBI GenBank (Boratyn *et al.*, 2013; https://blast.ncbi.nlm.nih.gov/Blast.cgi) with the following Megablast parameters: Identity=100%, Minimum score=300, and Maximum Expected Value=0.01. Taxonomic names were assigned to OTU using the following criteria: (i) when the OTU matched to records from one species only, the species name was assigned; (ii) when the OTU matched to records from multiple species from one genus only, the genus name was assigned; (iii) when the OTU matched to records from multiple genera belonging to one family only, the family name was assigned. Taxonomic names were checked against

Corner (1997) and Boo *et al.* (2014) for the local uses of the species (e.g., food, medicinal and aesthetic), and against local botanical records from the Herbarium, University of Malaya for information regarding the flowering phenology. The status of the species as native or introduced was checked with the Catalogue of Life (www.catalogueoflife.org). See Appendix B for further details on each OTU. Raw sequence data related to this study are available in Sequence Read Archive (SRA) at Genbank, NCBI under accessions SAMN07956186 to SAMN07956205.

Table 3.1: Primers used in PCR amplification of plant DNA extracted from faecal samples of *Eonycteris spelaea*. Illumina adaptors are underlined whereas primer sequence are shown in regular font.

Target Amplicon	**Direction**	**Illumina adaptor + Primer sequence (5'-3')**	**References for Primers**
ITS2 (350bp)	Forward	<u>TCGTCGGCAGCGTCAGATGTGTATAAGAGACAG</u>ATGCGATACTTGGTGTGAAT	Chen *et al.* (2010)
ITS2 (350bp)	Reverse	<u>GTCTCGTGGGCTCGGAGATGTGTATAAGAGACAG</u>TCCTCCGCTTATTGATATGC	White *et al.* (1990)
rbcL (600bp)	Forward	<u>TCGTCGGCAGCGTCAGATGTGTATAAGAGACAG</u>ATGTCACCACAAACAGAGACTAAAGC	Kress and Erickson (2007)
rbcL (600bp)	Reverse	<u>GTCTCGTGGGCTCGGAGATGTGTATAAGAGACAG</u>AGTCCACCGCGTAGACATTCAT	de Vere *et al.* (2012)

3.3.8 Species richness and sampling completeness ratio

All analyses were performed using R version 3.3.1. (R Core Team, 2017). The detection of plant species in faecal samples of *E. spelaea* was recorded as absent or present following Prosser and Hebert (2017). Currently, DNA metabarcoding cannot be considered quantitative due to biological (e.g. varying copy numbers of plastid and nuclear DNA in pollen among and within species) (Bell *et al.*, 2016) and methodological (e.g. PCR amplification bias caused by universal primers) (Prosser & Hebert, 2017) factors. The species richness and the sampling completeness ratio were estimated using the SpadeR package (Chao & Shen, 2010). Chao2 is more suitable for the incidence-type data collected in this study as it estimates the species richness based on the incidence of each species (i.e., presence or absence) recorded in each sampling unit (Chao & Chiu, 2016). Several Chao2 models were used to assess consistency of estimates provided by each model. A homogeneous model was also included under the assumption that all plant species have the same detection probabilities, but usually severely underestimates the true species richness if heterogeneity exists (Chao & Chiu, 2016). Rarefaction and extrapolation sampling curves of estimated species richness and the sampling completeness ratio were created using the iNEXT package (Hsieh *et al.*, 2016) with Chao2 and a 95% confidence interval.

3.3.9 Relative detection rate of each plant species in faeces of *E. spelaea*

To apply a consistent terminology, if a plant species was detected in (i) ≥8 of the 10 weekly samples, it was considered "frequently" detected, (ii) >3 but <8 of the 10 weekly samples, it was considered "moderately" detected, and (iii) ≤3 of the weekly samples, it was considered "infrequently" detected.

CHAPTER 4: RESULTS

4.1 Bats of Peninsular Malaysia and their DNA barcode reference library

At least 110 bat species have been reported from Peninsular Malaysia. A checklist was created and available in the Checklist section of BOLD as "A checklist of bats of Peninsular Malaysia and progress towards a DNA barcode reference library" (CL-PMBAT). The search of BOLD revealed that 86 of the 110 species have public records, of which 48 species have DNA barcodes collected from bats sampled in Peninsular Malaysia. Based on NJ analyses and allocation of DNA barcodes to BINs by BOLD (as discussed below), several DNA barcodes recorded under the same species name showed variations in COI mtDNA, which may or may not represent distinct taxa pending on further analysis. Of the eight families included in this checklist, Vespertilionidae has the highest number of recorded species (n=44, 40% of the total species for Peninsular Malaysia), followed by Hipposideridae (n=20, 18%), and Pteropodidae (n=18, 16%). Nycteridae has the lowest number of recorded species with only one species (0.9%).

4.1.1 Family: Pteropodidae

4.1.1.1 *Aethalops alecto* [Thomas, 1923a]

Aethalodes alecto Thomas, 1923a: 251. Indrapura Peak, Sumatra, INDONESIA (Collector unknown; BM(NH) 1923.1.2.1).

Aethalops alecto Thomas, 1923b.

Common English name: Grey Fruit Bat

Barcode Index Number: DNA barcodes recorded as *A. alecto* are associated with the BIN, BOLD:AAB6984, but there are no DNA barcodes from Peninsular Malaysia.

Remarks: Jayaraj *et al.* (2016a) commented that "unpublished genetic data suggests that the Javan and Borneon forms are distinct". The relationship between these two forms and

the bats in Peninsular Malaysia could not be evaluated in this study due to the lack of DNA barcodes from Peninsular Malaysia and Java.

IUCN status: Least Concern

Recorded at: **Perak**: Maxwell Hill (Medway, 1969); **Pahang**: Gunong Benom and Cameron Highlands (Medway, 1969).

A. alecto is not common and confined to hill and montane forests, normally above 1000 m (Medway, 1969; Francis, 2008).

4.1.1.2 *Balionycteris seimundi* Kloss, 1921

Balionycteris maculata seimundi Kloss, 1921: 229. Junction of Tahan and Teku rivers, foot of Gunung Tahan, Pahang, MALAYSIA (E. Seimund, collector; MNM 1/21).

Balionycteris maculata Chasen, 1940.

Common English name: Spotted-winged Fruit Bat

Barcode Index Number: BOLD:AAB7907 (14 DNA barcodes from Peninsular Malaysia; Figure 4.1).

Remarks: Originally described as a subspecies of *B. maculata* (Corbet & Hill, 1992). Khan *et al.* (2008) reported a high genetic divergence (12%) in cytochrome *b* mtDNA between populations of *B. maculata* sensu lato in Peninsular Malaysia and Borneo, and consequently raised *B. seimundi* as a distinct species. The same pattern was also observed in COI mtDNA (Figure 4.1; see Figure 2 in Francis *et al.*, 2010). Following Khan *et al.* (2008), the name of the taxon in Peninsular Malaysia should be updated to *B. seimundi.*

IUCN status: Least Concern

Recorded at: As *B. maculata*: **Pahang**: Gunung Tahan (Kloss, 1921), Merapoh (Ratnam *et al.*, 1989), Krau Wildlife Reserve (Zubaid, 1993; Anan *et al.*, 1998; Kingston *et al.*, 2006), Tasik Chini (Lim & Ratnam, 1999), Kuala Atok National Park (Tingga *et al.*, 2012), Lata Bujang Forest Reserve (Mohd-Hanif *et al.*, 2015); **Selangor**: Ulu Gombak (Medway, 1966; Heller & Volleth, 1984; 1989), Bukit Lanjan (Ratnam *et al.*, 1989),

Bangi Forest Reserve (Zubaid, 1993), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a), Air Hitam Forest Reserve (Azlan *et al.*, 2000), Sungai Dusun Forest Reserve (Mohd-Hanif *et al.*, 2015); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990); **Perak**: Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013), Royal Belum State Park (Tamrin *et al.*, 2010), Bayor River-Rantau Panjang (Shafie *et al.*, 2011); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Perlis**: Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Johor:** Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al.*, 2015).

B. seimundi roosts in small harem groups in sites with bell-shaped cavities and smooth surfaces. Individuals have been found roosting in crowns of palms, clumps of epiphytic ferns, arboreal ant nests, hollowed arboreal termite nests and hollowed detached large branches (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.1.3 *Chironax melanocephalus* [Temminck, 1825]

Pteropus melanocephalus Temminck, 1825: 190; Gunung Karang, Bantam, west Java, INDONESIA (Collector unknown; Type unknown).

Chironax melanocephalus Corber & Hill, 1992.

Common English name: Black-capped Fruit Bat

Barcode Index Number: BOLD:ACG2580 (1 DNA barcode from Peninsular Malaysia; Figure 4.1)

Remarks: Sing *et al.* (2013) first reported that a DNA barcode collected at Ulu Gombak, shared 95.8% similarity with DNA barcodes of *Chironax melanocephalus* from Java, Indonesia (Figure 4.1). The DNA barcodes from Java are likely to represent *C. melanocephalus* sensu stricto as they were collected from type locality and are assigned to a different BIN (BOLD:AAE9045). Whether several forms of *Chironax* occur in Peninsular Malaysia remains to be determined. Two distinct morphotypes of *C. melanocephalus* sensu lato were recently described from Sumatra, Indonesia, neither matching with the currently recognised subspecies: *C. m. melanocephalus* and *C. m.*

tumulus (Huang *et al.*, 2014). No DNA barcodes were provided for these specimens but it remains possible that the taxon in Peninsular Malaysia is one of these putative species.

IUCN status: Least Concern

Recorded at: Selangor: Ulu Gombak (Medway, 1966; Heller & Volleth, 1989; Sing *et al.*, 2013), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a), Sungai Dusun Forest Reserve (Mohd-Hanif *et al.*, 2015); **Pahang**: Cameron Highland (Medway, 1969; Shahfiz *et al.*, 2008a), Krau Wildlife Reserve (Kingston *et al.*, 2006), Fraser Hill Forest Reserve (Mohd-Hanif *et al.*, 2015); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Perak**: Royal Belum State Park (Tamrin *et al.*, 2010), Bayor River-Rantau Panjang (Shafie *et al.*, 2011); **Kelantan**: Air Panas-Gua Musang (Hasan *et al.*, 2012), Gunung Chamah (Jayaraj *et al.*, 2016b); **Johor:** Endau Kluang Forest Reserve (Mohd-Hanif *et al.*, 2015).

C. melanocephalus is common in lowland, hill and montane forests where the species roosts in large colonies in caves and rock shelters but in smaller groups in tree ferns (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.1.4 *Cynopterus* cf. *brachyotis* SUNDA

Pachysoma brachyotis Müller, 1838: 146. Dewei River, central Kalimantan, INDONESIA (Collector unknown; Type unknown).

Cynopterus brachyotis Medway, 1969.

Cynopterus cf. *brachyotis* SUNDA Campbell *et al.*, 2004.

Common English name: Sunda Short-nosed Fruit Bat

Barcode Index Number: BOLD:AAA9800 (20 DNA barcodes from Peninsular Malaysia; Figure 4.1)

Remarks: Campbell *et al.* (2004) reported two distinct species under *C. brachyotis* sensu lato with a mean divergence of 8.3% in mtDNA (combined control region and cytochrome *b*) between them. The two species are commonly annotated as *C.* cf. *brachyotis* SUNDA and *C.* cf. *brachyotis* FOREST (Figure 4.1). The SUNDA species is

larger than the FOREST species with a longer forearm (>64 mm) and is abundant in highly disturbed habitat (e.g., agricultural and suburban areas) but is absent in mature forests (Kingston *et al.*, 2006; Francis, 2008; Campbell *et al.*, 2004; 2007).

It is unclear which species represents *C. brachyotis* sensu stricto despite the cryptic taxa being widely acknowledged (N. Simmons, personal communication, March 31, 2017). Medway (1969) recognised three subspecies of *C. brachyotis* in Peninsular Malaysia: (i) *C. b. brachyotis* found in lowlands and islands in the northern part of Peninsular Malaysia, including Perak with a forearm length: 57 – 68 mm and an ear length: 14.5 – 18.5 mm; (ii) *C. b. angulatus* which intergrades with the nominal subspecies at the northern range and has a forearm length: 68 – 72 mm and an ear length: 18 – 22 mm; and (iii) *C. b. altitudinus* found in the central highlands above 3, 000 ft from Gunung Brinchang, Pahang to Gunung Bunga Buah, Selangor with a forearm length: 60 – 68 mm and an ear length: 18 – 21 mm. A thorough examination of all relevant types in this genus is required in order to correctly attribute currently existing Linnaean names.

IUCN status: As *C. brachyotis*: Least Concern

Recorded at: These records refer to *C. brachyotis* sensu lato, so may represent "SUNDA" or "FOREST". **Pahang**: Gunung Brinchang (Medway, 1966), Pulau Tioman (Medway, 1969; Campbell *et al.*, 2004), Merapoh (Ratnam *et al.*, 1989), Tasik Chini (Lim & Ratnam, 1999), Krau Wildlife Reserve (Zubaid, 1993; Anan *et al.*, 1998; Kingston *et al.*, 2006), Kuala Lipis and Cherating (Campbell *et al.*, 2004), Cameron Highland (Shahfiz *et al.*, 2008a), Kuala Atok National Park (Tingga *et al.*, 2012), Lata Bujang Forest Reserve and Fraser Hill Forest Reserve (Mohd-Hanif *et al.*, 2015); **Kedah**: Pulau Langkawi (Medway, 1966), Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Pulau Pinang**: Pulau Pinang (Medway, 1966); **Perak**: Pulau Pangkor (Medway, 1969; Campbell *et al.*, 2004), Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013), Taping (Campbell *et al.*, 2004), Royal Belum State Park (Shahfiz *et al.*, 2008b; Tamrin *et al.*, 2010), Bayor River-

Rantau Panjang and Selama (Shafie *et al.*, 2011); **Terengganu**: Pulau Redang (Medway, 1969), Pulau Perhentian (Campbell *et al.*, 2004); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990); **Kelantan:** Air Panas-Gua Musang (Hasan *et al.*, 2012), Gunung Stong State Park (Jayaraj *et al.*, 2012a), Gunung Reng, Gua Musang, and Lojing Highlands (Jayaraj *et al.*, 2016b); **Selangor**: Ulu Gombak (Medway, 1966; Heller & Volleth, 1984; 1989; Ratnam *et al.*, 1989; Sing *et al.*, 2013), Gunung Bunga Buah (Medway, 1966), Bukit Kemandul and Bukit Lanjan (Ratnam *et al.*, 1989), Bangi Forest Reserve (Zubaid, 1993), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a); Air Hitam Forest Reserve (Azlan *et al.*, 2000), Sungai Dusun (Campbell *et al.*, 2004); **Melaka:** Melaka town (Campbell *et al.*, 2004); **Perlis**: Perlis State Park and Kangar (Campbell *et al.*, 2004); Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Johor:** Endau-Kluang Forest Reserve and Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al*, 2015).

4.1.1.5 *Cynopterus* cf. *brachyotis* FOREST

Cynopterus brachyotis FOREST Campbell *et al.*, 2004.

Cynopterus JLE sp. A Francis *et al.* 2010 (and as in BOLD).

Common English name: Forest Short-nosed Fruit Bat

Barcode Index Number: BOLD:AAA9308 (19 DNA barcodes from Peninsular Malaysia; Figure 4.1)

IUCN status: As *C. brachyotis*: Least Concern

Remarks: The FOREST form is smaller than the SUNDA form (average forearm length is <63 mm) and is confined to primary and mature secondary forests (Kingston *et al.*, 2006; Francis, 2008; Campbell *et al.*, 2004). See remarks on *C.* cf. *brachyotis* SUNDA.

Recorded at: Confirmed records of "FOREST": **Pahang**: Krau Wildlife Reserve (Campbell *et al.*, 2004); **Johor**: Endau Rompin (Campbell *et al.*, 2004); **Perlis**: Perlis State Park and Kuala Perlis (Campbell *et al.*, 2004); **Kelantan**: Gua Musang (Campbell *et al.*, 2004), **Perak**: Taiping (Campbell *et al.*, 2004), Gunung Stong State Park (Jayaraj

et al., 2012a); **Melaka:** Unspecified (Shahfiz *et al.*, 2009); **Selangor**: Ulu Gombak (Sing *et al.*, 2013); **Terengganu**: Tasik Kenyir and Temenggor Lake (Syaripuddin *et al.*, 2014).

C. cf. *brachyotis* FOREST is generally restricted to primary and mature secondary forests (Kingston *et al.*, 2006; Francis, 2008; Campbell *et al.*, 2004) and has not been reported from disturbed habitats (Campbell *et al.*, 2007).

4.1.1.6 *Cynopterus horsfieldii* Gray, 1843

Cynopterus horsfieldii Gray, 1843: 38; Java, INDONESIA (Collector and type unknown).

Common English name: Horsfield's Fruit Bat

Barcode Index Number: BOLD:AAD1477 (3 DNA barcodes from Peninsular Malaysia; Figure 4.1)

IUCN status: Least Concern

Recorded at: Selangor: Ulu Gombak (Medway, 1966; Heller & Volleth, 1984; 1989); Bukit Lanjan (Ratnam *et al.*, 1989), Bangi Forest Reserve (Zubaid, 1993), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a); **Pulau Pinang**: (Medway, 1969); **Pahang**: Merapoh (Ratnam *et al.*, 1989), Krau Wildlife Reserve (Zubaid, 1993; Kingston *et al.*, 2006), Tasik Chini (Lim & Ratnam, 1999), Cameron Highland (Campbell *et al.*, 2004; Shahfiz *et al.*, 2008a), Cherating (Campbell *et al.*, 2004), Kuala Atok National Park (Tingga *et al.*, 2012), Fraser Hill Forest Reserve (Mohd-Hanif *et al.*, 2015); **Perak**: Temengor Forest Reserve (Francis, 1995), Taiping (Campbell *et al.*, 2004), Bayor River-Rantau Panjang & Selama (Shafie *et al.*, 2011), Temenggor Lake (Syaripuddin *et al.*, 2014); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Kelantan**: Gua Musang (Campbell *et al.*, 2004; Hasan *et al.*, 2012), Gunung Stong State Park (Jayaraj *et al.*, 2012a), Gunung Reng (Jayaraj *et al.*, 2016b); **Perlis:** State Park (Campbell *et al.*, 2004), Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Terengganu**: Tasik Kenyir (Syaripuddin *et al.*, 2014); **Johor:** Endau Kluang Forest Reserve (Mohd-Hanif *et al.*, 2015).

C. horsfieldii has a wide range of habitats (e.g., lowland, hill, and montane forests, mangroves, orchards and plantations) (Kingston *et al.*, 2006) and has been reported roosting gregariously in caves, cavities in limestone caves and rock shelters (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.1.7 *Cynopterus sphinx* [Vahl, 1797]

Vespertilio sphinx Vahl, 1797: 123; Tranquebar, Madras, INDIA (Collector unknown; Type unknown).

Cynopterus sphinx Cuvier, 1824.

Common English name: Greater Short-nosed Fruit Bat

Barcode Index Number: DNA barcodes recorded as *C. sphinx* are associated with BIN, BOLD:AAA3386, but there are no DNA barcodes from Peninsular Malaysia. Another BIN (BOLD:AAD9139) contains a single DNA barcode of *C. sphinx* from India and DNA barcodes recorded as *Pteropus vampyrus*, *P. lylei*, and *Rousettus leschenaultii*, which may represent erroneous records or contamination.

Remarks: *C. sphinx* resembles *C.* cf. *brachyotis* closely in morphology with overlapping forearm length (Francis, 2008). Examination of specimens from Peninsular Malaysia identified as *C. sphinx*, *C.* cf. *brachyotis* SUNDA and *C.* cf. *brachyotis* FOREST revealed that *C. sphinx* is 8.9% divergent from *C.* cf. *brachyotis* SUNDA and 7.5% divergent from *C.* cf. *brachyotis* FOREST in mtDNA (combined control region and cytochrome *b*) (Campbell *et al.*, 2004). In this study, DWNP specimens labelled as *C. sphinx* and *C.* cf. *brachyotis* from Peninsular Malaysia were examined which revealed that the two species are distinct at lower last molar. Specimens of *C. sphinx* have lower teeth which are almost uniform in size whereas specimens of *C.* cf. *brachyotis* have non-uniformed lower teeth with extremely small lower last molars (Boonsong & McNeely, 1977).

IUCN status: Least Concern

Recorded at: Pahang: Cameron Highland (Campbell *et al.*, 2004); **Perak**: Taiping (Campbell *et al.*, 2004), Selama (Shafie *et al.*, 2011); **Perlis**: Kuala Perlis, Perlis State Park and Kangar (Campbell *et al.*, 2004), Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Kelantan**: Gunung Stong State Park (Jayaraj *et al.*, 2012a), Gunung Reng and Lojing Highlands (Jayaraj *et al.*, 2016b).

C. sphinx is commonly found in disturbed habitats and ecotones but not in the forest interior (Francis, 2008; Campbell *et al.*, 2007).

4.1.1.8 *Dyacopterus spadiceus* [Thomas, 1890]

Cynopterus spadiceus Thomas, 1890: 235; Baram, Sarawak, MALAYSIA (Charles Hose, collector; BM(NH) 1890.1.28.4).

Dyacopterus spadiceus Andersen, 1912a.

Common English name: Dayak Fruit Bat

Barcode Index Number: There are two public DNA barcodes of *D. spadiceus* on BOLD, but neither are associated with any BIN due to their short sequence length (<500 bp). One DNA barcode (BM447-04) is from Peninsular Malaysia (Francis *et al.*, 2010). NJ analysis revealed that this DNA barcode exhibited little divergence with the DNA barcode from Kalimantan, Indonesia (BM265-04) (Figure 4.1).

IUCN status: Near Threatened

Recorded at: Perak: Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013); **Pahang**: Krau Wildlife Reserve (Anan *et al.*, 1998; Kingston *et al.*, 2006); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Selangor**: Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a), Sungai Dusun Forest Reserve (Mohd-Hanif *et al.*, 2015).

D. spadiceus roosts in tree cavities and ferns, and has been recorded in lowland, hill and montane forests, and nearby limestone caves (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

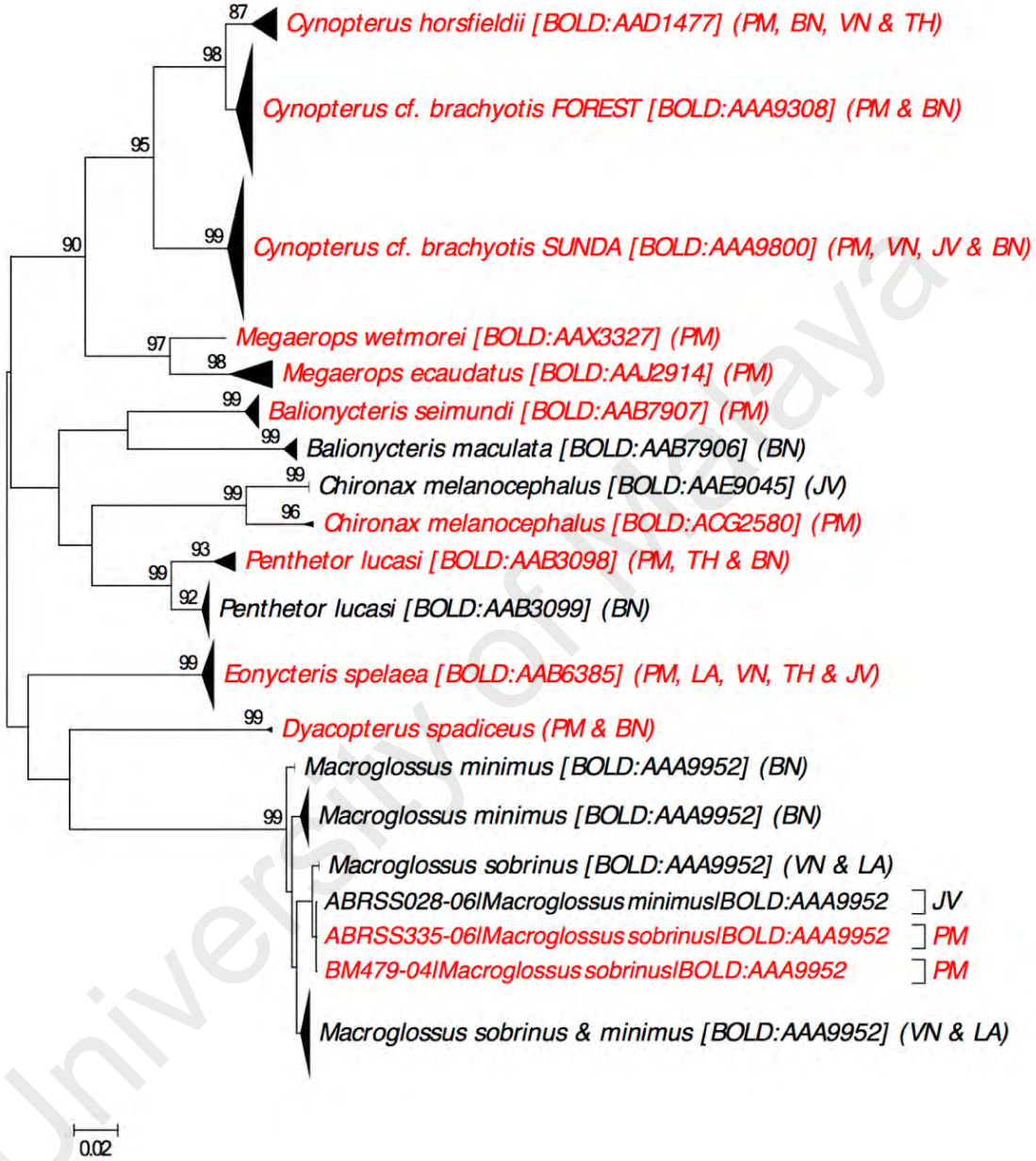

Figure 4.1: Neighbour-joining tree showing all available DNA barcodes for species in family Pteropodidae reported from Peninsular Malaysia. The percentage of pseudoreplicate trees ($\geq$70%) in which the DNA barcodes clustered together in the bootstrap test (500 pseudoreplicates) are shown above the branches. Abbreviation as follows: PM=Peninsular Malaysia, VN=Vietnam, JV=Java, Indonesia, BN=Borneo (including Sabah, Sarawak, Brunei and Kalimantan), TH=Thailand, LA=Laos.

4.1.1.9 *Eonycteris spelaea* [Dobson, 1871]

Macroglossus spelaeus Dobson, 1871: 105, 106; Farm Caves, Moulmein, Tenasserim, MYANMAR (Collector unknown; Type unknown).

Eonycteris spelaea Dobson, 1873a.

Common English name: Cave Nectar Bat

Barcode Index Number: BOLD:AAB6385 (1 DNA barcode from Peninsular Malaysia; Figure 4.1)

IUCN status: Least Concern

Recorded at: Pahang: Krau Wildlife Reserve (Zubaid, 1993; Kingston *et al.*, 2006), Pulau Tioman (Medway, 1969; Csorba *et al.*, 1997), Tasik Chini (Lim & Ratnam, 1999); **Selangor**: Batu Caves (Medway, 1969), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a), Ulu Gombak (Medway, 1966; Heller & Volleth, 1984; 1989); **Perak**: Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013), Bayor River-Rantau Panjang and Selama (Shafie *et al.*, 2011); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Kelantan:** Air Panas-Gua Musang (Hasan *et al.*, 2012), Gunung Reng, Gua Musang and Lojing Highlands (Jayaraj *et al.*, 2016b), Gunung Stong State Park (Jayaraj *et al.*, 2012a); **Melaka:** Unspecified (Shahfiz *et al.*, 2009).

E. spelaea is a cave dweller and roosts in large colonies with thousands of individuals (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.1.10 *Macroglossus minimus* [Geoffroy, 1810a]

Pteropus minimus Geoffroy, 1810a: 97; Java, INDONESIA (Leschnault de la Tour, collector; Type unknown).

Macroglossus minimus Cuvier, 1824.

Common English name: Lesser Long-tongued Nectar Bat

Barcode Index Number: DNA barcodes recorded as *M. minimus* are associated with BIN, BOLD:AAA9952, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Remarks: *M. minimus* is morphologically similar to *M. sobrinus* but has a deep median groove on the upper lip (which is absent in *M. sobrinus*) and shorter rostrum (26 – 28mm) and muzzle (Boonsong & McNeely, 1977; Francis, 2008). In this study, DWNP specimens labelled as *M. minimus* and *M. sobrinus* from Peninsular Malaysia were examined and they fit the description of *M. minimus* and *M. sobrinus* respectively, cosequently support the presence of both taxa in Peninsular Malaysia. However, the taxa showed very shallow divergence in COI mtDNA in NJ analysis with DNA barcodes from both type localities (i.e., Java and Peninsular Malaysia) being grouped together (Figure 4.1; see Figure 6 in Francis *et al.*, 2010). It remains unclear whether *M. minimus* and *M. sobrinus* are actually the same species or whether they represent two taxa that diverged recently. Further analysis of nuclear DNA would be required to determine this. The taxa are tentatively retained as distinct species in this checklist.

Recorded at: Pahang: Pulau Tioman (Medway, 1969), Krau Wildlife Reserve (Zubaid, 1993); **Selangor**: Kuala Selangor (Ratnam *et al.*, 1989), Bangi Forest Reserve (Zubaid, 1993), Ulu Gombak (Medway, 1966); **Perak**: Bayor River-Rantau Panjang (Shafie *et al.*, 2011), Temengor Forest Reserve (Shahfiz *et al.*, 2013); **Kelantan**: Air Panas-Gua Musang (Hasan *et al.*, 2012), Gunung Stong State Park (Jayaraj *et al.*, 2012a), Gunung Chamah, Gunung Reng, Gua Musang and Lojing Highlands (Jayaraj *et al.*, 2016b).

M. minimus has been recorded in mangroves, coastal areas and disturbed areas (Medway, 1969; Francis, 2008).

4.1.1.11 *Macroglossus sobrinus* Andersen, 1911

Macroglossus minimus sobrinus Andersen, 1911: 641, 642; Mount Igari, Perak, MALAYSIA (A.L. Butler, presenter; BM(NH) 1898.11.29.1).

Macroglossus sobrinus Chasen, 1940.

Common English name: Greater Long-tongued Nectar Bat

Barcode Index Number: BOLD:AAA9952 (2 DNA barcodes from Peninsular Malaysia; Figure 4.1).

IUCN status: Least Concern

Remarks: *Macroglossus sobrinus* was first described as a subspecies of *M. minimus* but was later considered as a distinct species (Boonsong & McNeely, 1977; Kingston *et al.*, 2006; Francis, 2008). See remarks on *M. minimus.*

Recorded at: Selangor: Ulu Gombak (Heller & Volleth, 1984; 1989), Bangi Forest Reserve (Zubaid, 1993), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a); **Pahang**: Krau Wildlife Reserve (Zubaid, 1993; Kingston *et al.*, 2006), Tasik Chini (Lim & Ratnam, 1999), Cameron Highland (Shahfiz *et al.*, 2008a); **Kedah:** Ulu Muda Forest Reserve (Norsham *et al.*, 1999), Gunung Jerai (Jayaraj *et al.*, 2013a); **Perak**: Bayor River-Rantau Panjang (Shafie *et al.*, 2011); **Kelantan:** Air Panas-Gua Musang (Hasan *et al.*, 2012), Gunung Stong State Park (Jayaraj *et al.*, 2012a), Gunung Chamah, Gunung Reng, Gua Musang and Lojing Highlands (Jayaraj *et al.*, 2016b); **Perlis:** Wang Kelian State Park (Jayaraj *et al.*, 2013a).

M. sobrinus has been recorded in dipterocarp and montane forests, and disturbed habitat (Francis, 2008), and has been reported roosting in rolled young banana leaves and pollinating wild banana plants (Kingston *et al.*, 2006).

4.1.1.12 *Megaerops ecaudatus* [Temminck, 1837]

Pachysoma ecaudatum Temminck, 1837: 94; Padang, West Sumatra, INDONESIA (Collector unknown; Type unknown).

Megaerops ecaudatus Temminck, 1841.

Common English name: Sunda Tailless Fruit Bat

Barcode Index Number: BOLD:AAJ2914 (7 DNA barcodes from Peninsular Malaysia; Figure 4.1)

IUCN status: Least Concern

Recorded at: Pahang: Fraser Hill, Gunung Brinchang, and Cameron Highland (Medway, 1969), Krau Wildlife Reserve (Anan *et al.*, 1998; Kingston *et al.*, 2006), Tasik Chini (Lim & Ratnam, 1999), Lata Bujang Forest Reserve (Mohd-Hanif *et al.*, 2015); **Perak**: Bidor (Andersen, 1912a), Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Selangor**: Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a); **Perlis:** Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Johor:** Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al.*, 2015); **Kelantan:** Gua Musang (Jayaraj *et al.*, 2016b).

M. ecaudatus predominantly inhabits pristine forest but has been recorded in disturbed forests (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.1.13 *Megaerops wetmorei* Taylor, 1934

Megaerops wetmorei Taylor, 1934: 191; near Tatayan, Cotobato, Mindanao Island, PHILIPPINES (E. H. Taylor, collector; Described based on specimen No. 770 in E.H. Taylor's collection with unknown current location).

Common English name: White-collared Fruit Bat

Barcode Index Number: BOLD:AAX3327 (1 DNA barcode from Peninsular Malaysia; Figure 4.1)

IUCN status: Vulnerable

Remarks: The species was first recorded in Peninsular Malaysia as a new subspecies, *M. w. albicollis* in Pasoh Forest Reserve (Francis, 1989) with distinctive white tufts on the shoulders and neck (Francis, 1989; Kingston *et al.*, 2006). The type specimen of *M. wetmorei* lacked the white neck tufts (which was followed in the description by Corbet & Hill, 1992) and has a short tail of 1.5 mm (Taylor, 1934). Specimens of *M. w. albicollis* from Pasoh Forest Reserve have a short tail of ~4 mm (Francis, 1989) whereas specimens from Krau Wildlife Reserve (Kingston *et al.*, 2006) are tailless. Further analysis, and more

DNA barcordes, would be required to determine whether *M. w. albicollis* deserves to be recognised as a species distinct from *M. w. wetmorei.*

Recorded at: Pahang: Krau Wildlife Reserve (Kingston *et al.*, 2006); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1989;1990). *M. wetmorei* has only been recorded in mature forests (Kingston *et al.*, 2006; Francis, 2008).

4.1.1.14 *Penthetor lucasi* [Dobson, 1880]

Cynopterus (Ptenochirus) lucasi Dobson, 1880: 163; Sarawak, MALAYSIA (Frederic A. Lucas, presenter; Described based on a male specimen from collection of Ward's Museum, Rochester, New York with unknown current location).

Penthetor lucasi Andersen, 1912a.

Common English name: Dusky Fruit Bat

Barcode Index Number: BOLD:AAB3098 (1 DNA barcode from Peninsular Malaysia; Figure 4.1)

Remarks: High divergence in cytochrome *b* mtDNA were reported within a population of *P. lucasi* in Miri, Sarawak, Borneo (4.9%) and within a population in Kuching, Sarawak (4.7%) (Mohd & Abdullah, 2012). This is congruent with Khan *et al.* (2008) who reported "~5%" divergence in cytochrome *b* mtDNA among specimens from Sarawak. Khan *et al.* (2008) did not include specimens from Peninsular Malaysia whereas Mohd Ridwan and Abdullah (2012) included specimens from Kelantan, Peninsular Malaysia. The DNA sequences from Kelantan were clustered with sequences from Kuching, Miri and Sri Aman (Borneo) and exhibited 3.88% divergence in cytochrome *b* mtDNA from another cluster from Borneo which consists of DNA sequences from Miri and Kuching.

DNA barcodes recorded as *P. lucasi* are associated with two BINs, BOLD:AAB3098 and BOLD:AAB3099 (Figure 4.1). Currently, no subspecies has been described for *P. lucasi* but considering two DNA clusters could occur within a population

(Mohd & Abdullah, 2012), further analyses including nuclear DNA, morphology and specimens from several localities are required for a taxonomic revision.

IUCN status: Least Concern

Recorded at: Pahang: Gunung Brinchang (Medway, 1969; Kingston *et al.*, 2006), Cameron Highlands (Medway, 1969), National Park (Yatim *et al.*, 1985), Krau Wildlife Reserve (Zubaid, 1993; Kingston *et al.*, 2006), Tasik Chini (Lim & Ratnam, 1999), Fraser Hill Forest Reserve (Mohd-Hanif *et al.*, 2015); **Selangor**: Ulu Gombak (Medway, 1966; Heller & Volleth, 1989), Ulu Langat Forest Reserve and Sungai Dusun Game Reserve (Yatim, 1983), Bangi Forest Reserve (Zubaid, 1993), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a); **Terengganu**: Kenyir Dam (Yatim *et al.*, 1985); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Kelantan:** Air Panas-Gua Musang (Hasan *et al.*, 2012), Gunung Stong State Park (Jayaraj *et al.*, 2012a); **Perak**: Temengor Forest Reserve (Shahfiz *et al.*, 2013).

P. lucasi roosts gregariously in caves, rock shelters and crevices, and occasionally under palm trees in forests (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.1.15 *Pteropus hypomelanus* Temminck, 1853

Pteropus hypomelanus Temminck, 1853: 61; Ternate Island, North Molucca islands, INDONESIA (Collector unknown; Type unknown).

Common English name: Island Flying-Fox

Barcode Index Number: DNA barcodes recorded as *P. hypomelanus* are associated with the BIN, BOLD:AAZ4957, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Recorded at: Johor: Pulau Pemanggil (Medway, 1969); **Terengganu**: Pulau Redang (Medway, 1969), Pulau Perhentian (Medway, 1969); **Kedah**: Pulau Paya (Medway, 1969); **Pahang**: Pulau Tioman (Dobson, 1873a; Medway, 1969; Aziz *et al.*, 2017a).

P. hypomelanus roosts close to shores on islands, under the fronds of coconut palms and branches of trees, and flies to mainland to feed (Medway, 1969; Francis, 2008).

4.1.1.16 *Pteropus vampyrus* [Linnaeus, 1758]

Vespertilio vampyrus Linnaeus, 1758: 31; Java, INDONESIA (Collector unknown; Type unknown).

Pteropus vampyrus Chasen, 1940.

Common English name: Large Flying-Fox

Barcode Index Number: A DNA barcode recorded as *P. vampyrus* is associated with the controversial BIN, BOLD: AAD9139 (see remarks on *C. sphinx*) but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Near Threatened

Recorded at: Pahang: Gunung Tahan (Bonhote, 1908), Sungai Tembeling (Medway, 1969), Taman Negara (Yatim *et al.*, 1985), Tanjung Agas (Epstein *et al.*, 2009); **Selangor**: Ulu Gombak (Medway, 1966); Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a); **Terengganu**: Kenyir Dam (Yatim *et al.*, 1985), Kampung Gong Tengah, Permaisuri and Kampung Kepah (Epstein *et al.*, 2009); **Johor:** Benut (Epstein *et al.*, 2009); **Perak**: Lenggong,Teluk Memali and Tambun (Epstein *et al.*, 2009), Temengor Forest Reserve (Shahfiz *et al.*, 2013).

P. vampyrus travels a long distance to feed, and often roosts in mangroves, on nipah palms and on open branches of trees (Medway, 1969; Francis, 2008).

4.1.1.17 *Rousettus amplexicaudatus* [Geoffroy, 1810a]

Pteropus amplexicaudatus Geoffroy, 1810a: 96, pl. 4; Timor Island, Lesser Sunda Islands, INDONESIA (Collector unknown; Type unknown).

Rousettus amplexicaudatus Chasen, 1940.

Common English name: Geoffroy's Rousette

Barcode Index Number: DNA barcodes recorded as *R. amplexicaudatus* are associated with a BIN, BOLD:AAC4982, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Recorded at: Pahang: Gunung Brinchang, (Medway, 1969), Krau Wildlife Reserve (Kingston *et al.*, 2006); **Selangor**: Batu Caves (Medway, 1969), Ulu Gombak (Heller & Volleth, 1984; 1989), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a): **Kedah**: Pulau Langkawi (Medway, 1969), Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Melaka**: Unspecified (Shahfiz *et al.*, 2009); **Perak**: Selama (Shafie *et al.*, 2011), Temengor Forest Reserve (Shahfiz *et al.*, 2013).

R. amplexicaudatus is a cave dweller and sometimes roosts in crevices of large rock boulders in complete darkness (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.1.18 *Rousettus leschenaultii* [Desmarest, 1820]

Pteropus leschenaultii Desmarest, 1820: 110; Pondicherry, INDIA (Collector unknown; Type unknown).

Rousettus leschenaultii Chasen, 1940.

Common English name: Leschenault's Rousette

Barcode Index Number: DNA barcodes recorded as *R. leschenaultii* are associated with the BIN, BOLD:AAB5823, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Recorded at: **Selangor**: Batu Caves based on skeletal remains (Kock *et al.*, 2000); **Perlis**: Wang Kelian State Park (Jayaraj *et al.*, 2013a). *R. leschenaultii* roosts primarily in caves and sometimes in wells, mines and cave-like structures (Francis, 2008).

4.1.2 Family: Emballonuridae

4.1.2.1 *Emballonura monticola* Temminck, 1838

Emballonura monticola Temminck, 1838: 25, pl. 2; Mountain Munara, Java, INDONESIA (Collector unknown; Type unknown).

Common English name: Lesser Sheath-tailed Bat

Barcode Index Number: BOLD:AAX76 (2 DNA barcodes from Peninsular Malaysia; Figure 4.2)

IUCN status: Least Concern

Recorded at: Pahang: Pulau Tioman (Medway, 1969), Krau Wildlife Reserve (Zubaid, 1993; Kingston *et al.*, 2006), Tasik Chini (Lim & Ratnam, 1999); **Terengganu**: Pulau Redang (Medway, 1969); **Johor**: Pulau Aur (Medway, 1969), Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al.*, 2015); **Kedah**: Pulau Langkawi (Medway, 1969), Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Selangor**: Ulu Gombak (Medway, 1966; Heller & Volleth, 1984; 1989), Bukit Lanjan (Ratnam *et al.*, 1989), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990); **Perak**: Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013); **Kelantan:** Gua Musang (Jayaraj *et al.*, 2016b).

E. monticola is confined to forests and roosts in small groups of two to 20 individuals, often in rock crevices, shallow caves, buttresses of fallen trunks, hollowed logs, and overhanging earth banks (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.2.2 *Taphozous longimanus* Hardwicke, 1825

Taphozous longimanus Hardwicke, 1825: 525; Calcutta, Bengal, INDIA (Collector unknown; Type unknown).

Common English name: Long-winged Tomb Bat

Barcode Index Number: DNA barcodes recorded as *T. longimanus* are associated with the BIN, BOLD:AAH9837, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Recorded at: Unspecified locations in **Selangor**, **Perak**, and **Pahang** (Medway, 1969); **Johor**: Endau-Rompin (Medway, 1969); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Pahang**: Krau Wildlife Reserve (2 DWNP specimens caught in year 2017).

T. longimanus roosts in buildings, caves, tree hollows, crowns of palm trees, and among rocks (Medway, 1969; Francis, 2008). The latest DWNP specimens were caught in crowns of coconut tree, approximately 3 meters tall (VC Lim, personal observation).

4.1.2.3 *Taphozous melanopogon* Temminck, 1841

Taphozous melanopogon Temminck, 1841: 287; Bantam, West Java, INDONESIA (Collector unknown; Type unknown) (Temminck, 1841).

Common English name: Black-bearded Tomb Bat

Barcode Index Number: DNA barcodes recorded as *T. melanopogon* are associated with the BIN, BOLD:AAD2120, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Recorded at: Pahang: Krau Wildlife Reserve (Zubaid, 1993; Kingston *et al.*, 2006); **Johor**: Pulau Pisang (Medway, 1969); **Pulau Pinang**: island (Medway, 1969); **Selangor**: Pulau Angsa, Ulu Gombak and Batu Caves (Medway, 1969), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a); **Kedah**: Pulau Langkawi (Medway, 1969), Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Perak**: Bukit Jerneh Cave and Tumang Lembing Cave (Douangboubpha *et al.*, 2010b); **Terengganu**: Bukit Dendong (Yeap, 2003).

T. melanopogon is primarily a cave dweller but has been recorded in lowland and hill forests, plantations and buildings. Individuals have been reported roosting at the entrance of caves, in rock crevices and hollowed dead trees (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

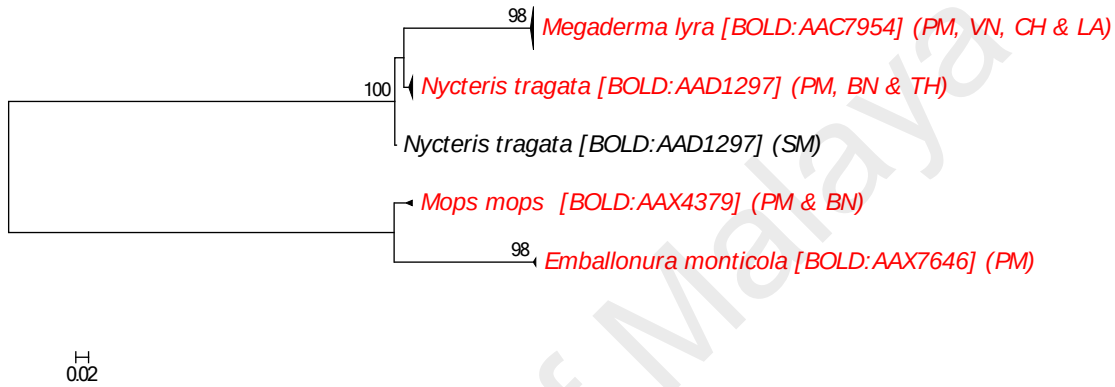

Figure 4.2: Neighbour-joining tree showing all available DNA barcodes for species in families Emballonuridae, Megadermatidae, Molossidae and Nycteridae reported from Peninsular Malaysia. The percentage of pseudoreplicate trees (⩾70%) in which the DNA barcodes clustered together in the bootstrap test (500 pseudoreplicates) are shown above the branches. Abbreviation as follows: PM=Peninsular Malaysia, VN=Vietnam, BN=Borneo (including Sabah & Sarawak of East Malaysia, Brunei and Kalimantan Indonesia), TH=Thailand, LA=Laos, SM=Sumatera Indonesia, CH=China.

4.1.2.4 *Saccolaimus saccolaimus* [Temminck 1838]

Taphozous saccolaimus Temminck, 1838: 14; Java, INDONESIA (Collector unknown; Syntype: BM(NH) 1874.10.26.2).

Saccolaimus saccolaimus Simmons, 2005.

Common English name: Pouched Tomb Bat

Barcode Index Number: The only DNA barcode recorded as *S. saccolaimus* is from Vietnam and is not associated with any BIN due to its short sequence length (<500bp).

IUCN status: Least Concern

Recorded at: Pulau Pinang: Pulau Pinang (Medway, 1969); **Melaka**: Masjid Tanah (Medway, 1969); **Selangor**: Ulu Gombak (Heller & Volleth, 1989).

S. saccolaimus has been reported roosting in the eaves of buildings, hollowed trees and rock crevices (Medway, 1969) with colony size varying from a few to hundreds of individuals (Francis, 2008).

4.1.3 Family: Megadermatidae

4.1.3.1 *Megaderma lyra* Geoffroy, 1810b

Megaderma lyra Geoffroy, 1810b: 190; INDIA (Collector unknown; Type unknown).

Common English name: Greater False-Vampire

Barcode Index Number: DNA barcodes recorded as *M. lyra* are associated with the BIN, BOLD:AAC7954. Two DNA barcodes from Peninsular Malaysia (RONP005-14 and RONP020-14) are not associated with any BIN due to their short sequence length (<500bp) but showed little divergence with other *M. lyra* DNA barcodes (Figure 4.2).

IUCN status: Least Concern

Recorded at: Selangor: Ulu Gombak (Medway, 1969), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a); **Perak**: Selama (Shafie *et al.*, 2011). *M. lyra* has been reported roosting in shallow caves, buildings and tunnels (Medway, 1969; Francis, 2008).

4.1.3.2 *Megaderma spasma* [Linnaeus, 1758]

Vespertilio spasma Linnaeus, 1758: 32; Ternate Island, Moluccas, INDONESIA (Collector unknown; Type unknown).

Megarderma spasma Chasen, 1940.

Common English name: Lesser False-Vampire

Barcode Index Number: DNA barcodes recorded as *M. spasma* are associated with the BIN, BOLD:AAC8422, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Recorded at: Pulau Pinang: Unspecified (Medway, 1969); **Johor**: Pulau Pisang and Pulau Aur (Medway, 1969); **Kedah**: Pulau Langkawi (Medway, 1969), Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Selangor**: Ulu Gombak (Medway, 1966; Heller & Volleth, 1989), Sungai Dusun Game Reserve (Yatim, 1983), Bangi Forest Reserve (Zubaid, 1993), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a); **Pahang:** National Park (Yatim *et al.*, 1985), Krau Wildlife Reserve (Ratnam *et al.*, 1989; Kingston *et al.*, 2006), Merapoh (Ratnam *et al.*, 1989), Pulau Tioman (Csorba *et al.*, 1997), Tasik Chini (Lim & Ratnam, 1999), Kemasul (Lim *et al.*, 2014); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990), Berembun Forest Reserve (Joann *et al.*, 2013); **Perak**: Temengor Forest Reserve (Shahfiz *et al.*, 2013), Bukit Jerneh Cave and Tumang Lembing Cave (Douangboubpha *et al.*, 2010b); **Perlis**: Wang Kelian State Park (Jayaraj *et al.*, 2013a).

M. spasma has been found roosting in caves, tunnels, culverts, large tree hollows, rock crevices and abandoned buildings (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.4 Family: Molossidae

4.1.4.1 *Cheiromeles torquatus* Horsfield, 1824

Cheiromeles torquatus Horsfield, 1824: pt 8; Pulau Pinang, MALAYSIA (John Crawfurd, Esq., collector; Type unknown).

Common English name: Naked Bat

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

IUCN status: Least Concern

Recorded at: Pulau Pinang: Unspecified (Horsfield, 1824); **Selangor**: Ulu Gombak (Medway, 1966), Batu Cave (Medway, 1969), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a); **Pahang**: Pulau Tioman (Corbet & Hill, 1992), Krau Wildlife Reserve (Zubaid, 1993; Kingston *et al.*, 2006); **Perak**: Temengor Forest Reserve (Shahfiz *et al.*, 2013).

C. torquatus has been reported roosting in caves, tree hollows and abandoned buildings, often with *Mops mops* (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.4.2 *Chaerephon johorensis* [Dobson, 1873b]

Molossus (Nyctinomus) johorensis Dobson, 1873b: 22; Johor, MALAYSIA (Collector unknown; Type unknown).

Chaerephon johorensis Chasen, 1940.

Common English name: Johore Wrinkle-lipped Bat

Barcode Index Number: There is no DNA barcode recorded under this name on BOLD.

IUCN status: Vulnerable

Recorded at: Unspecified locations at **Johor (**Dobson, 1873b) and **Selangor** (Medway, 1969); **Pahang**: Krau Wildlife Reserve (Zubaid, 1993; Kingston *et al.*, 2006); **Kedah**: Gunung Jerai (Jayaraj *et al.*, 2013b); **Terengganu:** Belukar Bukit (Roslan *et al.*, 2016).

C. johorensis has been reported foraging high over the canopy and large rivers in forest (Kingston *et al.*, 2006; Francis, 2008).

4.1.4.3 *Chaerephon plicatus* [Buchannan, 1800]

Vespertilio plicatus Buchannan, 1800: 261, pl. 13; Bengal, INDIA (Collector unknown; Type unknown).

Tadarida plicata Medway, 1969.

Chaerephon plicata Nowak, 1994.

Chaerephon plicatus Simmons, 2005.

Common English name: Asian Wrinkle-lipped Bat

Barcode Index Number: DNA barcodes recorded as *C. plicatus* are associated with the BIN, BOLD:AAK0536, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Remarks: *C. plicatus* is considered to be widespread across Peninsular Malaysia (Medway, 1969; Csorba *et al.*, 2014) despite only a few records from the region, though there are many records from Thailand and Myanmar (Csorba *et al.*, 2014). There are no specimens deposited in DWNP collection. Specimens labelled as *C. plicatus* deposited in Institute of Medical Research, Malaysia, could not be identified due to the damaged band above head which distinguishes *C. plicatus* from *C. johorensis*. *C. plicatus* resembles *Mops mops* closely but is distinguishable by having five teeth in each upper jaw including extra small anterior upper premolars whereas *M. mops* has only four teeth in upper jaw (Medway, 1969; Boonsong & McNeely, 1977; Francis, 2008). Such subtle differences are difficult to be used as identification characteristics in field for live specimens, which may explain the lack of recent records for *C. plicatus* in Peninsular Malaysia.

Recorded at: Kedah: Ulu Muda Forest Reserve (Norsham *et al.*, 1999).

C. plicatus roosts in large, densely packed colonies and has been reported roosting in buildings (Medway, 1969; Francis, 2008).

4.1.4.4 *Mops mops* [Blainville, 1840]

Dysopes mops Blainville, 1840: 101; Sumatra, INDONESIA (Collector unknown; Type unknown).

Mops mops Chasen, 1940.

Common English name: Sunda Free-tailed Bat

Barcode Index Number: BOLD:AAX4379 (1 DNA barcode from Peninsular Malaysia; Figure 4.2)

IUCN status: Near Threatened

Recorded at: Pahang: Krau Wildlife Reserve (Zubaid, 1993; Kingston *et al.*, 2006; Francis *et al.*, 2010); **Selangor**: Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999).

M. mops is a forest inhabitant and roosts in dead or hollowed trees, often with *Cheiromeles torquatus* (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.5 Family: Nycteridae

4.1.5.1 *Nycteris tragata* [Andersen, 1912b]

Petalia tragata Andersen, 1912b: 546; Bidi Caves, Sarawak, Borneo, MALAYSIA (Cecil J. Brooks, Esq., presenter; BM(NH) 1903.3.31.1).

Nycteris tragata Chasen, 1940.

Common English name: Malayan Slit-faced Bat

Barcode Index Number: BOLD:AAD1297 (5 DNA barcodes from Peninsular Malaysia; Figure 4.2)

Remarks: Two species names of genus *Nycteris*: *N. javanica* and *N. tragata* have been used in Peninsular Malaysia. All records of *N. javanica* from the region are from old reports dated before year 2000 (Medway, 1969; Francis, 1990; Zubaid, 1993; Anan *et al.*, 1998; Lim *et al.*, 1999a; Norsham *et al.*, 1999). *N. tragata* was once considered as a synonym of *N. javanica* (Medway, 1969). The taxa were later considered to be distinct with *N. javanica* being confined to Java and some of the surrounding islands whereas *N. tragata* confined to Peninsular Malaysia and Borneo (Van Cakenberghe & De Vree. 1993; Kingston *et al.*, 2006). In this checklist, previous reports of *N. javanica* are treated as reports of *N. tragata* and only one species, *N. tragata*, is present in Peninsular Malaysia.

IUCN status: Near Threatened

Recorded at: Pahang: Krau Wildlife Reserve (Kingston *et al.*, 2006), Kuala Atok, National Park (Tingga *et al.*, 2012), Jengka (Lim *et al.*, 2014), Lata Bujang Forest Reserve (Mohd-Hanif *et al.*, 2015); **Melaka**: Unspecified (Shahfiz *et al.*, 2009); **Perak**: Temengor Forest Reserve (Joann *et al.*, 2011); **Kelantan**: Air Panas-Gua Musang (Hasan *et al.*, 2012); **Negeri Sembilan**: Gunung Angsi Forest Reserve (Joann *et al.*, 2013; Lim *et al.*, 2014); **Selangor**: Semangkok Forest Reserve (Joann *et al.*, 2013); **Perlis**: Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Kedah**: Bukit Hijau (Lim *et al.*, 2014); **Johor:** Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al.*, 2015).

N. tragata is confined to mature primary forests and roosts in small groups in hollowed trees, caves, crevices of large boulders and man-made hollows such as culverts (Kingston *et al.*, 2006; Francis, 2008).

4.1.6 Family: Hipposideridae

4.1.6.1 *Aselliscus stoliczkanus* [Dobson, 1871]

Asellia stoliczkana Dobson, 1871: 106; Pulau Pinang, MALAYSIA (Dr. Stoliczka; Type unknown).

Aselliscus stoliczkanus Medway, 1969.

Common English name: Trident Roundleaf Bat

Barcode Index Number: DNA barcodes recorded as *A. stolickanus* are associated with ten BINs, BOLD:AAA6446, BOLD:AAA6447, BOLD:AAA6448, BOLD:AAA6449, BOLD:AAA6450, BOLD:AAA6451, BOLD:ABY9671, BOLD:ABY9672, BOLD:ACF3013, and BOLD:ACF3014. All the DNA barcodes are from Vietnam, Laos, China and Myanmar (Appendix C). Whether any of these DNA barcodes represent the valid *A. stolickanus* remains to be determined as none of them are from bats caught near the type locality. There are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Recorded at: Pulau Pinang (Dobson, 1871); **Pahang**: Pulau Tioman (Zubaid, 1988c).

Both records from Peninsular Malaysia are from islands in northern Peninsular Malaysia. *A. stolickanus* roosts in limestone caves and forages in forested and disturbed areas (Boonsong & McNeely, 1977; Francis, 2008). In Thailand, *A. stolickanus* is uncommon but widespread (Boonsong & McNeely, 1977). Its rarity in bat survey could be due to its ability to detect and hence avoid mist nets (Zubaid, 1988c).

4.1.6.2 *Coelops frithii* Blyth, 1848

Coelops frithi Blyth, 1848: 251; Sunderbans, BANGLADESH (R. W. G. Frith, Esq., presenter; Type unknown).

Common English name: Asian Tailless Roundleaf Bat

Barcode Index Number: DNA barcodes recorded as *C. frithii* are associated with two BINs, BOLD:AAF3920 and BOLD:AAF3921 (Appendix D). One DNA barcode (ABBM313-05) is not associated with any BIN due to its short sequence length (<500bp). There are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Recorded at: Selangor: Ulu Gombak (Medway, 1969), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999).

C. frithii has been reported foraging in forests and roosting in small groups in caves and hollowed trees (Francis, 2008).

4.1.6.3 *Coelops robinsoni* Bonhote, 1908

Coelops robinsoni Bonhote, 1908: 4; foot of Mountain Tahan, Pahang, MALAYSIA (Mr Robinson, collector; BM(NH) 1906.10.4.9).

Common English name: Malaysian Tailless Roundleaf Bat

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

IUCN status: Vulnerable

Recorded at: Pahang: Gunung Tahan (Bonhote, 1908; Medway, 1969), Krau Wildlife Reserve (Kingston *et al.*, 2006); **Selangor**: Port Swettenham=Port Klang (Medway, 1969).

The type specimen was caught in a young, rolled-up leaf of a wild banana plant (Bonhote, 1908). Individuals have been reported roosting in hollowed buttresses of large trees and in caves in primary lowland forest (Kingston *et al.*, 2006; Francis, 2008).

4.1.6.4 *Hipposideros armiger* [Hodgson, 1835]

Rhinolophus armiger Hodgson, 1835: 699; NEPAL (Collector unknown; Type unknown).

Hipposideros armiger Chasen, 1940.

Common English name: Greater Roundleaf Bat

Barcode Index Number: BOLD:AAA8161 (2 DNA barcodes are from Peninsular Malaysia; Figure 4.3).

Remarks: DNA barcodes recorded as *H. armiger* are associated with four BINs, BOLD:ABX5993, BOLD:AAA8161, BOLD:AAA8163, and BOLD:AAA8164. The BIN, BOLD:AAA8161 contains DNA barcodes from across Southeast Asia including Peninsular Malaysia (ABBSI001-04 and ABBSI002-04). The remaining BINs appear to be more geographically restricted (Figure 4.3). Four subspecies were recognised by Simmons (2005): *H. a. armiger* (type locality: Nepal), *H. a. tranninhensis* (type locality: Vietnam), *H. a. terasensis* (type locality: Taiwan), and *H. a. fujianensis* (type locality: China). Whether each BIN represents a subspecies or a distinct species and whether BOLD:AAA8161 represents *H. armiger* sensu stricto remains to be determined.

IUCN status: Least Concern

Recorded at: Kedah: Pulau Langkawi (Medway, 1969), Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990); **Pahang**: Tasik Chini (Lim & Ratnam, 1999), Krau Wildlife Reserve (Kingston *et al.*,

2006), Kenong (Lim *et al.*, 2014); **Perak**: Bukit Jerneh Cave and Tumang Lembing Cave (Douangboubpha *et al.*, 2010b), Bayor River-Rantau Panjang (Shafie *et al.*, 2011); **Perlis**: Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Pulau Pinang**: Bukit Panchor (Lim *et al.*, 2014); **Kelantan:** Gunung Reng (Jayaraj *et al.*, 2016b).

H. armiger has been reported roosting in large chambers in caves, sometimes in mixed colonies with other species, and roosting solitarily on *bertam* plants and in crevices of large boulders in forest (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.6.5 *Hipposideros halophyllus* Hill & Yenbutra, 1984

Hipposideros halophyllus Hill & Yenbutra, 1984: 77; Khao Sa Moa Khon (=Khao Sa Moa Khon), Tha Woong (=Ta Woong), Lop Buri, THAILAND (Kitti Thonglongya, collector; TNRC 54-3694).

Common English name: Thai Roundleaf Bat

Barcode Index Number: BOLD:AAX1220 (1 DNA barcode from Peninsular Malaysia; Figure 4.3)

Remarks: The BIN also contains a DNA barcode recorded as *H. ater* from India which was originally from Genbank. The DNA barcode of *"H. ater"* is likely to be a case of misidentification as the two species are morphologically distinct. *H. halophyllus* has a kidney-shaped internarial septum whereas *H. ater* has a slightly inflated and triangular internarial septum (Douangboubpha *et al.*, 2010b). Although Peninsular Malaysia is included in the distribution range of *H. ater* in some literature (Corbet & Hill, 1992; Davison & Zubaid, 2007), it is unlikely that *H. ater* occurs in the region due to the absence of any records. The DNA barcode of *"H. ater"* is excluded from the NJ analysis.

IUCN status: Vulnerable

Recorded at: Perak: Bukit Jerneh Cave and Tumang Lembing Cave (Douangboubpha *et al.*, 2010b).

H. halophyllus has been recorded in and near limestone caves in Peninsular Malaysia and Thailand (Francis, 2008; Douangboubpha *et al.*, 2010b). It is unknown whether *H. halophyllus* is strictly confined to limestone areas or this association is an effect of limited sampling but it is likely that the species requires specialised roosting habitat (Douangboubpha *et al.*, 2010b).

***Hipposideros bicolor* species complex**

H. bicolor sensu lato is a species complex which comprises two phonic types with individuals echolocate either at 131 kHz (=*H.* bicolor131) or 142 kHz (=*H.* bicolor142) (Kingston *et al.*, 2001). The two phonic types are 6.5 – 6.8% divergent in cytochrome *b* mtDNA but are morphologically similar with overlapping forearm length (Kingston *et al.*, 2001). Although the two phonic types are widely recognised as two distinct species, many reports still use *H. bicolor* to represent both species (Joann *et al.*, 2011; Hasan *et al.*, 2012; Jayaraj *et al.*, 2012a; 2013a), causing ambiguity regarding the distribution of the two species in Peninsular Malaysia. The two species were recently formalised under Latin names: *H. bicolor* (=bicolor131) and *H. atrox* (=bicolor142) (Douangboubpha *et al.*, 2010a). Yet the search for DNA barcodes on BOLD coupled with NJ analysis suggest that this species complex is even more complicated (Figure 4.3). Whether *H. nequam* represents any of the phonic types remains to be determined (see remarks on *H. nequam*).

4.1.6.6 *Hipposideros bicolor* [Temminck, 1834]

Rhinolophus bicolor Temminck, 1834: 19. pl. 1; Anjer Coast, Northwestern Java, INDONESIA (Collector unknown; Type unknown).

Hipposideros bicolor Chasen, 1940.

Hipposideros bicolor131 Kingston *et al.*, 2001.

Common English name: Bicolored Roundleaf Bat

Barcode Index Number: BOLD:AAC0447 (2 DNA barcodes from Peninsular Malaysia) and BOLD:AAD3329 (6 DNA barcodes from Peninsular Malaysia). The two BINs showed 4.2% of divergence in COI mtDNA (Figure 4.3).

IUCN status: Least concern

Recorded at: As *H.* bicolor131: **Pahang**: Krau Wildlife Reserve (Kingston *et al.*, 2006), Bukit Ibam, Kemasul, Jengka, Klau Besar, Kenong and Gunung Aais (Lim *et al.*, 2014); **Perak**: Royal Belum State Park (Shahfiz *et al.*, 2008b), Kledang Saiong Forest Reserve (Joann *et al.*, 2013; Lim *et al.*, 2014); **Melaka:** Unspecified (Shahfiz *et al.*, 2009); **Selangor**: Semangkok Forest Reserve (Joann *et al.*, 2013), Ulu Gombak (Sing *et al.*, 2013; Lim *et al.*, 2014); **Terengganu**: Gunung Tebu Forest Reserve (Joann *et al.*, 2013), Pasir Raja, Dungun (Wilson *et al.*, 2014); **Negeri Sembilan**: Pasoh Forest Reserve (BM454-04 and BM455-04, Francis *et al.*, 2010), Berembun Forest Reserve (Joann *et al.*, 2013), Gunung Angsi Forest Reserve, (Joann *et al.*, 2013; Lim *et al.*, 2014); **Johor**: Endau Rompin National Park (ABRSS332-06, ABRSS333-06, ABRSS379-06, and BM423-04, Francis *et al.*, 2010), Gunung Panti and Labis Forest Reserve (Lim *et al.*, 2014); **Kelantan**: Gunung Stong State Park (Lim *et al.*, 2014); **Pulau Pinang**: Bukit Panchor (Lim *et al.*, 2014); **Kedah**: Bukit Hijau and Ulu Muda Forest Reserve (Lim *et al.*, 2014).

As *H. bicolor* (could be either *H. bicolor* or *H. atrox*): **Perak**: Temengor Forest Reserve (Joann *et al.*, 2011); **Kelantan**: Air Panas-Gua Musang (Hasan *et al.*, 2012), Lojing Highlands (Jayaraj *et al.*, 2016b); **Perlis**: Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Pahang:** Lata Bujang Forest Reserve and Fraser Hill Forest Reserve (Mohd-Hanif *et al.*, 2015); **Johor:** Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al.*, 2015).

H. bicolor species complex has been recorded in wide range of habitats: primary and secondary lowland forests, cultivated areas including rubber plantations, and near limestone areas (Kingston *et al.*, 2006; Francis, 2008, Douangboubpha *et al.*, 2010a).

Individuals have been reported roosting in caves, tunnels and rock crevices with other *Hipposideros* species (Medway, 1969; Douangboubpha *et al.*, 2010a).

4.1.6.7 *Hipposideros atrox* Andersen, 1918

Hipposideros gentilis atrox Andersen, 1918: 381; Semangko Gap, Selangor, MALAYSIA, 2800 ft (A. L. Butler, Esq., presenter; BM(NH) 1901.3.9.4).

Hipposideros bicolor atrox Hill *et al.*, 1986.

Hipposideros bicolor142 Kingston *et al*., 2001.

Hipposideros atrox Douangboubpha *et al*., 2010a.

Common English name: Lesser Bicoloured Roundleaf Bat

Barcode Index Number: BOLD:ACE5015 (2 DNA barcodes from Peninsular Malaysia) and BOLD:ACE6229 (11 DNA barcodes from Peninsular Malaysia). The two BINs showed 2.12% of divergence in COI mtDNA (Figure 4.3).

IUCN status: Not Evaluated but Least Concern as *H. bicolor*

Recorded at: As *H.* bicolor142: **Pahang**: Krau Wildlife Reserve (Kingston *et al*., 2006), Bukit Ibam, Jengka, Klau Besar, and Kenong (Lim *et al.*, 2014); **Selangor**: Semangkok Forest Reserve (Joann *et al*., 2013), Ulu Gombak (Sing *et al*., 2013; Joann *et al*., 2013); **Terengganu**: Gunung Tebu Forest Reserve (Joann *et al*., 2013), Pasir Raja, Dungun (Wilson *et al*., 2014), Tasik Kenyir (Syaripuddin *et al.*, 2014); **Negeri Sembilan**: Berembun Forest Reserve (Joann *et al.*, 2013), Gunung Angsi Forest Reserve (Joann *et al*., 2013; Lim *et al*., 2014); **Perak**: Temenggor Lake (Joann *et al*., 2013), Kledang Saiong Forest Reserve (Lim *et al*., 2014); **Kelantan**: Gunung Stong State Park (Lim *et al*., 2014); **Pulau Pinang**: Bukit Panchor (Lim *et al.*, 2014); **Kedah**: Bukit Hijau (Lim *et al*., 2014).

As *H.* cf. *bicolor*: **Perlis**: Perlis State Park (ABBSI006-04 and ABBSI007-04, Francis *et al.*, 2010); **Pahang**: Krau Wildlife Reserve (ABBSI011-04 and ABBSI015-04, Francis *et al*., 2010), Kuala Lompat (BM452-04, Francis *et al*., 2010), Kuala Lipis (BM452-04, Francis *et al*., 2010), Bukit Sagu-Kuantan (BM452-04, Francis *et al*., 2010);

Kelantan: Dabong (BM452-04, Francis *et al*., 2010); **Selangor**: Ampang (BM452-04, Francis *et al*., 2010); **Perak**: Gunung Gajah-Ipoh (BM452-04, Francis *et al*., 2010); **Negeri Sembilan**: Pasoh Forest Reserve (BM452-04, Francis *et al*., 2010).

Also see records of *H. bicolor* sensu lato above.

H. atrox has been recorded roosting in varying colony sizes ranging from a few to hundreds of individuals and has been reported roosting with other *Hipposideros* species (Douangboubpha *et al*., 2010a). See remarks on *H. bicolor* sensu lato above for details regarding the habitat of *H. atrox*.

4.1.6.8 *Hipposideros cervinus* [Gould, 1854]

Rhinolophus cervinus Gould, 1854: pl. 34; Cape York and Albany Island, Queensland, AUSTRALIA (Collector unknown, Type unknown).

Phyllorhina labuanensis Tomes, 1859: 537; Labuan Island, Borneo, MALAYSIA (Mr. James Motley, collector; BM(NH) 7.1.1.305) (Tomes, 1858).

Hipposideros schneidersi (misprint = *schneideri*) Thomas, 1904: 722; Upper Langkat, Sumatera, INDONESIA (Collector unknown; BM(NH) 7.1.9.4).

Hipposideros galeritus schneidersi Tate, 1941.

Hipposideros cervinus labuanensis (schneidersi) Jenkins & Hill, 1981.

Common English name: Fawn Roundleaf Bat

Barcode Index Number: BOLD:AAB6249 (19 DNA barcodes from Peninsular Malaysia; Figure 4.3)

Remarks: Jenkins and Hill (1981) described several subspecies under *H. cervinus* based on morphometric analyses. They treated *H. c. schneidersi* as a synonym of *H. c. labuanensis* and concluded that the latter is the only taxon present in Peninsular Malaysia and Borneo. Bates *et al*. (2007) commented that although both have the typical "*cervinus*" noseleaf and rostrum, *H. c. schneidersi* and *H. c. labuanensis* are morphologically distinct with the former having a broader zygomatum, congruent with an earlier taxonomic

treatment of the two as distinct species (Tate, 1941). Murray *et al.* (2012) reported that specimens identified as *H. cervinus* from Peninsular Malaysia and Sabah (Borneo) are 5.5 – 6.1% divergent in NADH dehydrogenase subunit 2 (ND2) mtDNA. DNA barcodes recorded as *H. cervinus* are associated with two BINs which showed 5.17% of divergence in COIT mtDNA (Figure 4.3). Whether specimens from Peninsular Malaysia and Sabah represent the *H. c. labuanensis* or two different species remains to be determined.

IUCN status: Least Concern

Recorded at: Selangor: Ulu Gombak (Heller & Volleth, 1989; Lim *et al.*, 2014), Air Hitam Forest Reserve (Azlan *et al.*, 2000), Semangkok Forest Reserve (Joann *et al*., 2013); **Pahang**: Krau Wildlife Reserve (Zubaid, 1993; Anan *et al*., 1998; Kingston *et al*., 2006), Tasik Chini (Lim & Ratnam, 1999), Kuala Atok National Park (Tingga *et al*., 2012), Bukit Ibam, Kemasul, Jengka, Klau Besar, Kenong and Gunung Aais (Lim *et al*., 2014), Lata Bujang Forest Reserve (Mohd-Hanif *et al*., 2015); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990), Gunung Angsi Forest Reserve (Joann *et al*., 2013; Lim *et al*., 2014), Berembun Forest Reserve (Joann *et al*., 2013); **Kedah:** Ulu Muda Forest Reserve (Norsham *et al*., 1999), Bukit Hijau (Lim *et al*., 2014); **Melaka**: Unspecified (Shahfiz *et al*., 2009); **Perlis:** Wang Kelian State Park (Jayaraj *et al*., 2013a); **Johor**: Gunung Panti and Labis Forest Reserve (Lim *et al*., 2014), Endau-Kluang Forest Reserve and Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al*., 2015); **Kelantan**: Air Panas-Gua Musang, (Hasan *et al.*, 2012); **Perak**: Temenggor Lake (Syaripuddin *et al.*, 2014), Kledang Saiong Forest Reserve, (Lim *et al.*, 2014); **Pulau Pinang**: Bukit Panchor (Lim *et al*., 2014); **Terengganu**: Pasir Raja, Dungun (Wilson *et al*., 2014), Tasik Kenyir (Syaripuddin *et al*., 2014).

H. cervinus forages in understories of forest and roosts in limestone caves and crevices amongst boulders in large colonies of up to 100,000 individuals (Kingston *et al*., 2006; Francis, 2008).

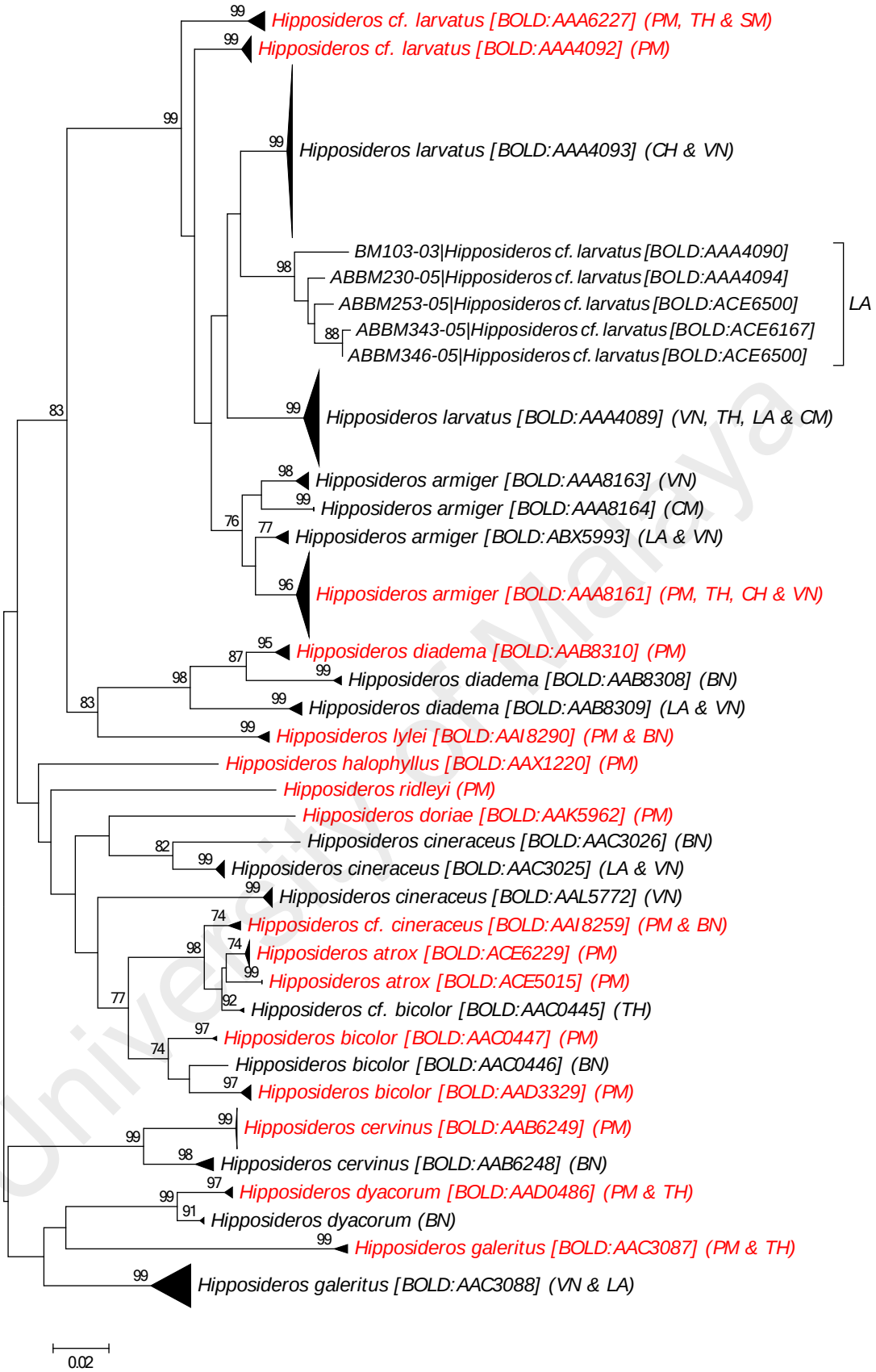

Figure 4.3: Neighbour-joining tree showing all available DNA barcodes for species in family Hipposideridae reported from Peninsular Malaysia. The percentage of pseudoreplicate trees (⩾70%) are shown above the branches. Abbreviation as follows: PM=Peninsular Malaysia, VN=Vietnam, BN=Borneo (including Sabah & Sarawak of East Malaysia, Brunei and Kalimantan Indonesia), TH=Thailand, LA=Laos, SM=Sumatera Indonesia, CH=China, CM=Cambodia.

4.1.6.9 *Hipposideros cineraceus* Blyth, 1853

Hipposideros cineraceus Blyth, 1853: 410; near Pind Dadan Khan, Salt Range, Punjab, PAKISTAN (W. Theobold, Esq., collector; Type unknown).

Common English name: Ashy Roundleaf Bat

Barcode Index Number: A DNA barcode (BM460-04) recorded as *H.* cf. *cineraceus* was collected in Pahang, Peninsular Malaysia and is associated with the BIN, BOLD:AAI8259 (Figure 4.3).

Remarks: Murray *et al.* (2012) reported two forms of *H. cineraceus* from Peninsular Malaysia; a larger specimen from Perak (forearm length = 42.9 mm) which echolocate at 152 kHz and is 9.2 – 15.1% divergent from other specimens in ND2 mtDNA whereas a smaller specimen from Pahang (forearm length = 39.3 mm) which echolocate at 144 kHz and is 10.4 – 12.2% divergent from other specimens in ND2 mtDNA. This finding is congruent with Khan *et al.* (2008) who reported an average divergence of 8.7% in cytochrome *b* mtDNA among specimens of *H. cineraceus* from Krau Wildlife Reserve.

Four BINs are associated with DNA barcodes recorded as *H. cineraceus* on BOLD (Figure 4.3). NJ analysis did not cluster the DNA barcodes from Peninsular Malaysia and Borneo (BOLD:AAI8259) with other DNA barcodes of *H. cineraceus* from Vietnam, Laos and Borneo, but clustered the barcodes closely to *H. atrox* from Peninsular Malaysia (BOLD:ACE5015 and BOLD:ACE6229) (Figure 4.3). *H. cineraceus* resembles *H. bicolor/atrox* closely but is distinguishable by having smaller body size, a slightly raised bump at internarial septum and echolocation frequency=144 kHz, whereas the echolocation frequency of *H. atrox* is 142 kHz (Kingston *et al.*, 2006). Specimens from across the region should be examined to determine the taxonomic status of *H. cineraceus*.

IUCN status: Least Concern

Recorded at: Selangor: Ulu Gombak (Medway, 1966), Ampang (Medway, 1969), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999a); **Johor**: Pulau Pisang (Medway, 1969), Labis

Forest Reserve (Lim *et al.*, 2014); **Kedah**: Pulau Langkawi (Medway, 1969), Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Perak**: Temengor Forest Reserve (Francis, 1995; Tamrin *et al*., 2010; Shahfiz *et al.*, 2013), Royal Belum State Park (Tamrin *et al.*, 2010); **Terengganu**: Bukit Dendong (Yeap, 2003); **Pahang**: Krau Wildlife Reserve (Kingston *et al.*, 2006), Jengka (Lim *et al.*, 2014), Fraser Hill Forest Reserve (Mohd-Hanif *et al*., 2015); **Melaka:** Unspecified (Shahfiz *et al*., 2009); **Kelantan**: Gunung Stong State Park (Lim *et al.*, 2014), Gunung Reng and Gua Musang (Jayaraj *et al.*, 2016b).

H. cineraceus roosts in caves or similar structures such as culverts, often with other *Hipposideros* species (Medway, 1969; Kingston *et al*., 2006; Francis, 2008).

4.1.6.10 *Hipposideros diadema* [Geoffroy, 1813]

Rhinolophus diadema Geoffroy, 1813: 263, pls. 5, 6; Timor Island, INDONESIA (Péron and Lesueur, collector; MNHN 918).

Hipposideros diadema Chasen, 1940.

Common English name: Diadem Roundleaf Bat

Barcode Index Number: BOLD:AAB8310 (7 DNA barcodes from Peninsular Malaysia; Figure 4.3)

Remarks: Murray *et al.* (2012) compared the specimens identified as *H. diadema* from Peninsular Malaysia and *H. pelingensis* from Kabaena Island, Southeast Sulawesi. They reported that the two species have similar body size and are 2.7% divergent in ND2 mtDNA but did not observe *H. diadema*'s distinctive white spots on *H. pelingensis*. They also reported that specimens of *H. diadema* from Peninsular Malaysia and the smaller specimens of *H. diadema* from Sulawesi are 8.5% divergent in ND2 mtDNA.

DNA barcodes recorded as *H. diadema* are associated with three BINs on BOLD which appear to correspond to geographical regions (Figure 4.3), congruent with Murray *et al.* (2012). Kitchener *et al.* (1992) recognised four subspecies under *H. diadema* based on morphological characteristics: *H. d. diadema* (type locality: Timor Island, Indonesia),

H. d. nobilis (type locality: Java, Indonesia), *H. d. griseus* (type locality: Luzon, Phillippine), *H. d. masoni* (type locality: Moulmein, Burma=Myanmar). It is likely that the taxon in Peninsular Malaysia represents either *H. d. nobilis* or *H. d. masoni*. Whether the taxon in Peninsular Malaysia should be recognised as a distinct species remains to be determined through examination of type specimens and specimens from across the region.

IUCN status: Least Concern

Recorded at: Pahang: Merapoh (Ratnam *et al.*, 1989), Krau Wildlife Reserve (Zubaid, 1993; Anan *et al.*, 1998; Kingston *et al.*, 2006), Tasik Chini (Lim & Ratnam, 1999), Kemasul, Jengka, Kenong and Gunung Aais (Lim *et al*., 2014), Lata Bujang Forest Reserve and Fraser Hill Forest Reserve (Mohd-Hanif *et al*., 2015); **Selangor**: Batu Caves (Medway, 1969), Ulu Gombak (Medway, 1966; Heller & Volleth, 1984; 1989), Bangi Forest Reserve (Zubaid, 1993), Bukit Kutu Wildlife Reserve (Lim *et al*., 1999a); **Pulau Pinang**: Bukit Panchor (Medway, 1969; Lim *et al*., 2014); **Kedah**: Pulau Langkawi (Medway, 1969), Ulu Muda Forest Reserve (Norsham *et al*., 1999); Bukit Hijau (*Lim et al.*, 2014); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990); **Perak**: Temengor Forest Reserve (Francis, 1995; Tamrin *et al*., 2010; Shahfiz *et al.*, 2013), Royal Belum State Park (Shahfiz *et al.*, 2008b; Tamrin *et al.*, 2010), Bayor River-Rantau Panjang (Shafie *et al.*, 2011), Temenggor Lake (Syaripuddin *et al*., 2014); **Melaka**: Unspecified location (Shahfiz *et al*., 2009); **Kelantan**: Air Panas-Gua Musang (Hasan *et al*., 2012), Gunung Stong State Park (Lim *et al*., 2014), Gua Musang (Jayaraj *et al*., 2016b); **Perlis**: Wang Kelian State Park (Jayaraj *et al*., 2013a); **Johor**: Gunung Panti and Labis Forest Reserve (Lim *et al*., 2014), Endau-Kluang Forest Reserve (Mohd-Hanif *et al.*, 2015); **Terengganu**: Tasik Kenyir (Syaripuddin *et al.*, 2014).

H. diadema has been reported roosting in limestone caves, in crevices of boulders, tree hollows and solitarily under the fronds of palms, in both primary and secondary forests (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.6.11 *Hipposideros doriae* [Peters, 1871]

Phyllorhina doriae Peters, 1871: 326; Sarawak, Borneo, MALAYSIA (Collector unknown; Type unknown).

Hipposideros sabanus Thomas, 1898a: 243; Lawas, Northeast Sarawak, Borneo, MALAYSIA (A. H. Everett, collector; Type unknown).

Hipposideros doriae Chasen, 1940.

Common English name: Least Roundleaf Bat

Barcode Index Number: BOLD:AAK5962 (1 DNA barcode from Peninsular Malaysia; Figure 4.3)

Remarks: *H. sabanus* is considered as a junior synonym of *H. doriae* (Kingston *et al*., 2006; Khan *et al.*, 2008; Murray *et al.*, 2012).

IUCN status: Near Threatened

Recorded at: Pahang: Krau Wildlife Reserve (Kingston *et al*., 2006; Khan *et al.*, 2008), Genung Benom and Tasik Bera (Khan *et al*., 2008), Kemasul and Gunung Aais (Lim *et al*., 2014); **Perak**: Maxwell Hill (Khan *et al.*, 2008), Kledang Saiong Forest Reserve (Joann *et al*., 2013), Temenggor Lake (Syaripuddin *et al.*, 2014); **Selangor**: Ulu Gombak (Khan *et al*., 2008), Semangkok Forest Reserve (Joann *et al*., 2013); **Kelantan:** Air Panas-Gua Musang (Hasan *et al*., 2012); **Perlis**: Wang Kelian State Park (Jayaraj *et al*., 2013a); **Terengganu**: Tasik Kenyir (Syaripuddin *et al*., 2014); **Johor**: Gunung Panti and Labis Forest Reserve (Lim *et al*., 2014); **Kedah**: Bukit Hijau (Lim *et al*., 2014).

Recorded as *H. sabanus* at: **Perak**: Maxwell Hill (Medway, 1969), Temengor Forest Reserve (Joann *et al.*, 2011); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990); **Pahang**: Krau Wildlife Reserve (Zubaid, 1993); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al*., 1999); **Terengganu:** Bukit Dendong (Yeap, 2003).

H. doriae has been recorded in lowland and submontane forests up to 1500 m (Kingston *et al*., 2006; Francis, 2008).

4.1.6.12 *Hipposideros dyacorum* [Thomas, 1902]

Hipposideros dyacorum Thomas, 1902: 271; Mountain Mulu, Baram, Sarawak, MALAYSIA (Charles Hose, collector; BM(NH) 1894.9.29.10).

Common English name: Dayak Roundleaf Bat

Barcode Index Number: BOLD:AAD0486 (1 DNA barcode is from Peninsular Malaysia; Figure 4.3)

Remarks: Murray *et al.* (2012) found little divergence in ND2 and RAG1 mtDNA (<1%) between specimens identified as *H. dyacorum* from Peninsular Malaysia and East Malaysia (Borneo). However, NJ analysis in this study showed 2.54% divergence between DNA barcodes from Peninsular Malaysia and Sabah (Borneo) (Figure 4.3). The name *H. dyacorum* is tentatively used in this checklist pending further research.

IUCN status: Least Concern

Recorded at: Perlis: Gua Tekong Siam (Hill & Zubaid, 1989), Wang Kelian State Park (Jayaraj *et al*., 2013a); **Pahang**: Krau Wildlife Reserve (Khan *et al*., 2008), Kenong (Lim *et al*., 2014); **Perak**: Temenggor Lake (Syaripuddin *et al*., 2014); **Terengganu**: Tasik Kenyir (Syaripuddin *et al*., 2014); **Kelantan**: Gunung Stong State Park (Lim *et al*., 2014), Gua Musang (ABBSI020-04).

H. dyacorum has been reported roosting in caves, under rocks and in tree hollows (Francis, 2008).

4.1.6.13 *Hipposideros galeritus* Cantor, 1846

Hipposideros galeritus Cantor, 1846: 183; Pulau Pinang, MALAYSIA (Collector unknown; Type unknown).

Common English name: Cantor's Roundleaf Bat

Barcode Index Number: BOLD:AAC3087 (2 DNA barcodes from Peninsular Malaysia; Figure 4.3)

Remarks: DNA barcodes recorded as *H. galeritus* are associated with two BINs: (i) BOLD:AAC3086 which contains DNA barcodes from Peninsular Malaysia and Thailand, and (ii) BOLD:AAC3087 which contains DNA barcodes from Vietnam and Laos (Figure 4.3). DNA barcodes from Peninsular Malaysia and Thailand are likely to represent *H. galeritus* sensu stricto as they cover the type locality.

IUCN status: Least Concern

Recorded at: Pulau Pinang: Unspecified (Cantor, 1846); **Selangor**: Batu Caves (Medway, 1969), Ulu Gombak (Medway, 1966; Heller & Volleth, 1984; Khan *et al.*, 2008), Bukit Kutu Wildlife Reserve (Lim *et al*., 1999a), Semangkok Forest Reserve (Joann *et al*., 2013); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990), Broga (Khan *et al*., 2008), Gunung Angsi Forest Reserve and Berembun Forest Reserve (Joann *et al*., 2013); **Pahang**: Krau Wildlife Reserve (Kingston *et al*., 2006; Khan *et al.*, 2008), Cameron Highland (Shahfiz *et al.*, 2008a), Kuala Atok, National Park (Tingga *et al*., 2012), Kenong and Gunung Aais (Lim *et al*., 2014); **Perak**: Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013), Maxwell Hill (Khan *et al*., 2008), Kledang Saiong Forest Reserve (Lim *et al*., 2014); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Kelantan**: Air Panas-Gua Musang (Hasan *et al*., 2012), Gunung Reng (Jayaraj *et al*., 2016b); **Perlis**: Wang Kelian State Park (Jayaraj *et al*., 2013a); **Johor**: Gunung Panti and Labis Forest Reserve (Lim *et al.*, 2014), Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al.*, 2015).

H. galeritus has been reported roosting in limestone caves and sighted near large rock boulders in mature lowland forest (Medway, 1969; Kingston *et al*., 2006; Francis, 2008).

4.1.6.14 *Hipposideros larvatus* [Horsfield, 1823]

Rhinolophus larvatus Horsfield, 1823: 6; Java, INDONESIA (Collector unknown; Type unknown).

Hipposideros larvatus Chasen, 1940.

Common English name: Intermediate Roundleaf Bat

Barcode Index Number: BOLD:AAA4092 (11 DNA barcodes from Peninsular Malaysia) and BOLD:AAA6227 (1 DNA barcode from Peninsular Malaysia; Figure 4.3)

Remarks: Thabah *et al.* (2006) reported variation in echolocation frequency (~82 kHz to ~100 kHz) among specimens identified as *H. larvatus* sensu lato from India, Myanmar, Malaysia, with those from Peninsular Malaysia emitted the highest frequency (100 – 102 kHz). They also reported size variation among the specimens with female specimens from Peninsular Malaysia having the lightest body mass and shortest forearm length. DNA barcodes recorded as *H. larvatus* formed five clusters, consistent with geographical origin of the sequences (see Figure 5 in Thabah *et al.*, 2006). The variations in echolocation, morphology and mtDNA suggest that *H. larvatus* is a species complex (Thabah *et al.*, 2006; Khan *et al.*, 2008; Murray *et al.*, 2012).

Here, DNA barcodes recorded as *H. larvatus* are associated with eleven BINs. DNA barcodes from Peninsular Malaysia fell into two BINs: (i) BOLD:AAA6227 comprises DNA barcodes from Perlis (northern Peninsular Malaysia), Thailand and Sumatera while (ii) BOLD:AAA4092 contains barcodes from across Peninsular Malaysia. Lim *et al.* (1999b) identified the specimens on Pulau Tioman (an island in Peninsular Malaysia) as *H. l. barbensis* (type locality: Sainte Barbe Island=Pulau Penjantan). In contrast, Thabah *et al.* (2006) stated that *H. larvatus* in Malaysia represents *H. larvatus* sensu stricto on the basis of specimens' shorter forearm length and type locality. NJ analysis suggested that at least two forms of *H. larvatus* occur in Peninsular Malaysia and clustered DNA barcodes of BIN, BOLD:AAA4092 with ABBSI021-04 which shares the same locality

with specimens examined by Thabah *et al.* (2006) (Figure 4.3). A single name, *H. larvatus* is tentatively used for this species complex in this checklist pending further research.

IUCN status: Least Concern

Recorded at: Selangor: Ulu Gombak (Medway, 1966), Bukit Kutu Wildlife Reserve (Lim *et al*., 1999a), Semangkok Forest Reserve,(Joann *et al*., 2013); **Kedah**: Pulau Langkawi (Medway, 1969), Ulu Muda Forest Reserve (Norsham *et al*., 1999), Gunung Angsi Forest Reserve (Joann *et al*., 2013; Lim *et al*., 2014), Bukit Hijau (Lim *et al*., 2014); **Pahang**: Pulau Tioman (Medway, 1969; Csorba *et al*., 1997), Krau Wildlife Reserve (Anan *et al*., 1998; Kingston *et al*., 2006), Kuala Atok, National Park (Tingga *et al*., 2012), Kemasul, Klau Besar, Kenong and Gunung Aais (Lim *et al*., 2014), Fraser Gill Forest Reserve (Mohd-Hanif *et al*., 2015); **Johor**: Pulau Aur (Medway, 1969), Gunung Panti and Labis Forest Reserve (Lim *et al*., 2014), Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al*., 2015); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990); **Terengganu**: Bukit Dendong (Yeap, 2003), Gunung Tebu Forest Reserve (Joann *et al*., 2013), Pasir Raja, Dunggun (Wilson *et al.*, 2014), Tasik Kenyir (Syaripuddin *et al*., 2014); **Melaka**: Unspecified (Shahfiz *et al*., 2009); **Perak**: Bukit Jerneh Cave and Tumang Lembing Cave (Douangboubpha *et al*., 2010b); Temenggor Lake (Syaripuddin *et al*., 2014), Kledang Saiong Forest Reserve (Lim *et al*., 2014); **Perlis**: Wang Kelian State Park (Jayaraj *et al*., 2013a); **Pulau Pinang**: Bukit Panchor (Lim *et al*., 2014); **Kelantan**: Air Panas-Gua Musang (Hasan *et al*., 2012), Gunung Stong State Park (Lim *et al*., 2014), Gunung Reng and Gua Musang (Jayaraj *et al.*, 2016b).

H. larvatus has been reported roosting in limestone caves, buildings, old mines rock and crevices in primary and secondary forests (Kingston *et al*., 2006; Francis, 2008).

4.1.6.15 *Hipposideros lekaguli* Thonglongya & Hill, 1974

Hipposideros lekaguli Thonglongya & Hill, 1974: 285; Phu Nam Tok Tap Kwang, Kaeng Khoi, Suraburi, THAILAND, c. 14°34'N, 101°9'E (Dr. Boonsong Lekagul, collector; TNRC 54-2200).

Common English name: Boonsoong's Roundleaf Bat

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

IUCN status: Near Threatened

Recorded at: Kedah: Gunung Keriang and Kodiang (Hill *et al.*, 1985). *H. lekaguli* roosts in caves and forages in both forested and disturbed areas (Francis, 2008).

4.1.6.16 *Hipposideros lylei* Thomas, 1913

Hipposideros lylei Thomas, 1913: 88; Chiendao Cave, 50 miles north of Chiang Mai, THAILAND, 350 meter (Th. H. Lyle, Esq., presenter; BM(NH) 1913.4.18.3).

Common English name: Shield-faced Roundleaf Bat

Barcode Index Number: BOLD:AAI8290 (1 DNA barcode from Peninsular Malaysia; Figure 4.3).

Remarks: *H. lylei* was once considered to be conspecific with *H. pratti* (Ellerman & Morrison-Scott, 1966). Although Tate (1947) commented that *H. pratti* is found in mountainous parts of lower Peninsular Malaysia, there are no other records of this species from Peninsular Malaysia. Therefore, *H. pratti* is excluded from this checklist.

IUCN status: Least Concern

Recorded at: Pahang: Bukit Chintamani (Medway, 1969), Krau Wildlife Reserve (Kingston *et al.*, 2006); **Kedah:** Unspecified caves (Medway, 1969); **Perlis**: Wang Tangga, Kaki Bukit (Hill, 1972); **Perak**: Gunung Tempurung (ABBSI053-04 – ABBSI055-04, Francis *et al.*, 2010).

H. lylei roosts primarily in limestone caves and has been recorded in lowland forests (Medway, 1969; Kingston *et al*., 2006; Francis, 2008).

4.1.6.17 *Hipposideros nequam* Andersen, 1918 (?)

Hipposideros nequam Andersen, 1918: 380, 381; Klang, Selangor, MALAYSIA (W. Davison, collector; BM(NH) 1885.8.1.369).

Common English name*:* Malay Roundleaf Bat

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

IUCN status: Data Deficient

Remarks: If valid, the species is extremely rare with only two records from Peninsular Malaysia: Klang (the holotype) and Krau Wildlife Reserve (Anan *et al.*,1998). However, the record from Krau Wildlife Reserve (Anan *et al.*,1998) is questionable due to the lack of details regarding the species identification and absence of any specimens in DWNP collection. In addition, Kingston *et al.* (2006) did not record this species in Krau Wildlife Reserve. The fact that the holotype is damaged remains another challenge to resolve the status of *H. nequam* (Medway, 1969; Francis, 2008). Tate (1941) commented that *H. nequam* resembles *H. bicolor* closely in forearm length but differs by having greatly reduced anterior lower premolar. Hill (1963) also noted the similarities in cranial structure between *H. nequam* and *H. (bicolor) atrox*; that *H. nequam* has a similar but slightly different cranial structure with "more inflated rostral eminences, shorter, broader premaxillae, blade-like vomer and greatly reduced anterior lower premolar" and larger than *H. bicolor*.

It is likely that *H. nequam* is a synonym of either *H. bicolor* or *H. atrox* (CM Francis, personal communication, March 8, 2017) but based on the slight differences between the type specimens of *H. nequam* and *H. bicolor* (Hill, 1963) and the locality of the holotype, the species is tentatively retained in this checklist. Morever, NJ analysis in this study suggests that *H. bicolor–H. atrox* species complex in Peninsular Malaysia is even more complicated with at least 4 forms (Figure 4.3; see remarks on *H. bicolor*

species complex). Whether *H. nequam* represents any of the forms in *H. bicolor–H. atrox* species complex remains to be determined.

Recorded at: Selangor: Klang (Andersen, 1918); **Pahang**: Krau Wildlife Reserve (Anan *et al.*,1998) (?).

4.1.6.18 *Hipposideros orbiculus* Francis, Kock & Habersetzer, 1999

Hipposideros orbiculus Francis, Kock & Habersetzer, 1999: 259; Abai Siat, southeast Kota Baru, 01°02’ S 101°43’ E, Sumatera Barat, Sumatra, INDONESIA (H. Stephan, collector; SMF 570902).

Common English name: Small Disc Roundleaf Bat

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

IUCN status: Vulnerable

Remarks: *H. orbiculus* is extremely rare and possibly has a limited distribution with only three known locations: Kota Baru in Sumatra Barat, Rawang-Kuala Selangor and Sungkai Wildlife Forest Reserve in Peninsular Malaysia (Francis *et al.*, 1999; Murray *et al.*, 2012).

Recorded at: Selangor: ~16 km from Rawang, on road between Rawang and Kuala Selangor, northwest Kuala Lumpur (Francis *et al.*, 1999); **Perak:** recorded at Sungkai Wildlife Reserve in the year 2007 (Francis *et al.*, 2016).

H. orbiculus has been reported roosting in drainage pipes and recorded in peat-swamp forest (Francis, 2008).

4.1.6.19 *Hipposideros pomona* Andersen, 1918

Hipposideros pomona Andersen, 1918: 380, 381; Haleri, North Coorg, INDIA (A few miles north of Mercara, Coorg District, Karnataka) (G. C. Shortridge; BM(NH) 1918.8.3.4).

Hipposideros pomona gentilis Hill *et al.*, 1986.

Common English name: Large-eared Roundleaf Bat

Barcode Index Number: DNA barcodes recorded as *H. pomona* are associated with eight BINs, BOLD:AAA4932, BOLD:AAA4933, BOLD: AAA4934, BOLD:AAA4935, BOLD:AAA4936, BOLD:AAA4937, BOLD:AAA4938 and BOLD:AAA4939, but there are no DNA barcodes from Peninsular Malaysia (Appendix E).

Remarks: Andersen (1918) first separated *H. pomona* and *H. gentilis* based on the noseleaf of *H. pomona* sensu stricto being broader than the noseleaf of *H. gentilis.* Corbet and Hill (1992) examined ethanol-preserved specimens identified as *H. pomona* sensu stricto and noted that these specimens lacked the lateral supplementary leaflets. Douangboubpha *et al.* (2010a) suggested that *H. pomona* sensu stricto (Corbet & Hill, 1992) may represent two species: *H. pomona* sensu stricto (restricted to Peninsular India) and *H. gentilis* (distributed from north-east India into Southeast Asia). Murray *et al.* (2012) reported that DNA sequences of *H. pomono* sensu lato from two mitochondrial genes: ND2 mtDNA and RAG1 fell into two distinct clades in a phylogenetic tree: (i) *H. pomona*, *H. rotalis* and *H. khaokhouayensis* from Laos, and (ii) *H. pomona* from Laos, China, Myanmar and Peninsular Malaysia. Specimens of *H. pomona* from both groups are morphologically similar. Three subspecies of *H. pomona* have been reported from China: *H. p. sinesis* (Min-Guang coastal region), *H. p. gentilis* (South Yunnan region) and an undescribed subspecies (Hainan Island), showing 6.0 – 8.5% divergence in cytochrome *b* mtDNA and 5.2 – 8.0% divergence in COI mtDNA (Zhao *et al.*, 2015). Due to the lack of DNA barcodes from Peninsular Malaysia and unresolved taxonomy across Southeast Asia, the name *H. pomona* is tentatively used in this checklist.

IUCN status: Least Concern

Recorded at: Perlis: Bukit Jerneh Cave and Tumang Lembing Cave (Douangboubpha *et al.*, 2010b), Bukit Lagi (Zubaid, 1988b).

H. pomona is a cave dweller and has been recorded from various forest types and disturbed areas (Francis, 2008).

4.1.6.20 *Hipposideros ridleyi* Robinson & Kloss, 1911

Hipposideros ridleyi Robinson & Kloss, 1911: 241; Botanic Gardens, SINGAPORE (H. N. Ridley, Esq., collector; MNM 2068/11).

Common English name: Ridley's Roundleaf Bat

Barcode Index Number: Two DNA barcodes recorded as *H. ridleyi* (BM470-04 and BM471-04) are not associated with any BIN due to their short sequence length (<500 bp) but are from Peninsular Malaysia (Figure 4.3).

IUCN status: Vulnerable

Recorded at: Pahang: Krau Wildlife Reserve (Anan *et al*., 1998; Kingston *et al*., 2006), Kuala Atok, National Park (Tingga *et al*., 2012), Bukit Ibam, Kemasul and Gunung Aais (Lim *et al*., 2014), Tasik Bera Forest Reserve (Mohd-Hanif *et al*., 2015); **Kedah:** Ulu Muda Forest Reserve (Norsham *et al*., 1999); **Johor**: Gunung Panti (Lim *et al*., 2014); **Kelantan**: Gunung Stong State Park (Lim *et al*., 2014).

H. ridleyi has been reported roosting in small groups in fallen tree hollows, culverts, and drainage pipes (Kingston *et al*., 2006; Francis, 2008).

4.1.7 Family: Rhinolophidae

4.1.7.1 *Rhinolophus acuminatus* Peters, 1871

Rhinolophus acuminatus Peters, 1871: 308; Gadok, Java, INDONESIA (Collector unknown; MNB 2548/1).

Common English name: Acuminate Horseshoe Bat

Barcode Index Number: DNA barcodes recorded as *R. acuminatus* are associated with two BINs, BOLD:AAB9238 and BOLD:ABY9249. The only DNA barcode from Peninsular Malaysia (RONP046-14) is excluded from the NJ analysis due to its short sequence length (<500bp) (Appendix F).

Remarks: Five subspecies are recognised by Simmons (2005): *R. a. acuminatus* in Java, *R. a. sumatranus* in Sumatra and Borneo, *R. a. circe* in Nias Island, *R. a. calypso* in

Enggano Island, and *R. a. audax* in Bali and Lombok. Corbet and Hill (1992) commented that specimens from mainland of Southeast Asia (i.e., Thailand, Laos, Cambodia and Peninsular Malaysia) resemble those from Java or Lombok.

IUCN status: Least Concern

Recorded at: Kedah: Ulu Muda Forest Reserve (Norsham *et al*., 1999); **Pahang**: Krau Wildlife Reserve (Kingston *et al*., 2006), Gunung Aais (Lim *et al*., 2014), Fraser Hill Forest Reserve (Mohd-Hanif *et al*., 2015); **Perak**: Royal Belum State Park (Shahfiz *et al*., 2008b), Temenggor Lake (Syaripuddin *et al*., 2014), Temengor Forest Reserve (Joann *et al*., 2011); **Terengganu**: Tasik Kenyir (Syaripuddin *et al*., 2014); **Kelantan:** Gunung Reng (Jayaraj *et al*., 2016b).

R. acuminatus has been reported roosting in caves, tree hollows, and sometimes roosts solitarily or in pairs under palm leaves in mature lowland forests and hills (Kingston *et al*., 2006; Francis, 2008).

4.1.7.2 *Rhinolophus affinis* Horsfield, 1823

Rhinolophus affinis Horsfield, 1823: 6; Java, INDONESIA (Collector unknown; BM(NH) 79.11.21.70, lectotype).

Rhinolophus affinis superans Andersen, 1905: 104; Pahang, MALAYSIA (MNM, presenter; BM(NH) 1900.7.3.2).

Common English name: Intermediate Horseshoe Bat

Barcode Index Number: BOLD:ACF0990 (8 DNA barcodes from Peninsular Malaysia; Figure 4.4)

Remarks: DNA barcodes recorded as *R. affinis* are associated with five BINs, BOLD:AAA3811, BOLD:ACF0988, BOLD:ACF0989, BOLD:ACF0990, and BOLD:ACQ4437. DNA barcodes from Peninsular Malaysia and southern Thailand (Songkhla and Hala Bala) fell into one BIN, BOLD:ACF0990 (Figure 4.4).

Nine subspecies are recognised by Simmons (2005): *R. a. affinis* (type locality: Java), *R. a. andamanensis* (type locality: South Andaman island), *R. a. himalayanus* (type locality: Mussoorie, Kumaon Division, northern India), *R. a. tener* (type locality: Pegu Division=Bago, Myanmar), *R. a. macrurus* (type locality: Taho, Karennee, Kyah State, Myanmar), *R. a. nesite* (type locality: Bunguran Island, north Natunas, Indonesia) , *R. a. princeps* (type locality: Lombok, Lesser Sunda Island), *R. a. hainanus* (type locality: Pouten, Hainan Island), and *R. a. superans* (type locality: Pahang, Peninsular Malaysia). Morphological (i.e., craniodental and baculum) and molecular (i.e., *COI* and *D-loop* regions mtDNA) characteristics provide support that the taxon occurring in Peninsular Malaysia is *R. a. superans* (Ith *et al.*, 2015).

IUCN status: Least Concern

Recorded at: Pahang: Pulau Tioman (Medway, 1969; Csorba *et al.*, 1997), Merapoh (Ratnam *et al*., 1989), Krau Wildlife Reserve (Zubaid, 1993; Anan *et al*., 1998; Kingston *et al.*, 2006), Tasik Chini (Lim & Ratnam, 1999), Cameron Highland (Shahfiz *et a*l., 2008a), National Park (Tingga *et al*., 2012), Kuala Atok, Bukit Ibam, Kemasul, Jengka, Klau Besar, Kenong and Gunung Aais (Lim *et al*., 2014), Tasik Bera Forest Reserve, Fraser Hill Forest Reserve and Lata Bujang Forest Reserve (Mohd-Hanif *et al*., 2015); **Terengganu**: Pulau Redang (Medway, 1969), Bukit Dendong (Yeap, 2003), Gunung Tebu Forest Reserve (Joann *et al*., 2013), Pasir Raja, Dungun (Wilson *et al.*, 2014), Tasik Kenyir (Syaripuddin *et al.*, 2014); **Perak**: Lenggong (Medway, 1969), Temengor Forest Reserve (Francis, 1995; Tamrin *et al.*, 2010; Shahfiz *et al*., 2013), Royal Belum State Park (Shahfiz *et al*., 2008b), Bukit Jerneh Cave and Tumang Lembing Cave (Douangboubpha *et al*., 2010b), Kledang Saiong Forest Reserve (Joann *et al*., 2013; Lim *et al.*, 2014), Temenggor Lake (Syaripuddin *et al*., 2014); **Selangor**: Batu Caves (Medway, 1969), Bukit Kutu Wildlife Reserve (Lim *et al*., 1999); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990), Gunung Angsi Forest Reserve (Joann *et al*., 2013;

Lim *et al*., 2014), Berembun Forest Reserve (Joann *et al*., 2013); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999; Lim *et al.*, 2014), Bukit Hijau (Lim *et al*., 2014); **Melaka**: Unspecified (Shahfiz *et al*., 2009); **Kelantan**: Air Panas-Gua Musang (Hasan *et al*., 2012), Gunung Stong State Park (Jayaraj *et al.*, 2012a), Gunung Stong State Park (Lim *et al*., 2014), Gunung Reng and Gua Musang (Jayaraj *et al*., 2016b); **Perlis**: Wang Kelian State Park, (Jayaraj *et al*., 2013a); **Johor**: Gunung Panti (Lim *et al.*, 2014), Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al*., 2015); **Pulau Pinang**: Bukit Panchor (Lim *et al.*, 2014).

R. affinis inhabits both primary and secondary forests, and roosts in limestone caves (Medway, 1969; Kingston *et al*., 2006; Francis, 2008).

4.1.7.3 *Rhinolophus borneensis* Peters, 1861

Rhinolophus borneensis Peters, 1861: 709; Labuan island, north Borneo, MALAYSIA (Collector unknown; Type unknown).

Rhinolophus chaseni Sanborn, 1939: 38; Pulo Condore=Con Son Island, south VIETNAM (C. B. Kloss, collector; BM(NH) 21.10.8.3).

Rhinolophus borneensis chaseni Corbet & Hill, 1992.

Common English name: Bornean Horseshoe Bat

Barcode Index Number: DNA barcodes recorded as *R. borneensis* are associated with a BIN, BOLD:AAC3741, but there are no barcodes from Peninsular Malaysia. DNA barcodes recorded as *R. chaseni* are not from Peninsular Malaysia and are associated with a BIN, BOLD:AAB4878, which also contains a single DNA barcode recorded as *R. shameli* (ABRVN329-06).

Remarks: *R. chaseni* was previously recognised as a subspecies of *R. borneensis* (Corbet & Hill, 1992); *R. b. chaseni* was geographically confined to Peninsular Malaysia whereas the nominal subspecies *R. b. borneensis* was confined to Borneo (Koopman, 1994). However, Francis *et al.* (2010) reported that DNA barcodes (COI mtDNA) of *R.*

borneensis and *R. chaseni* did not cluster together as conspecific (see Figure 3 in Francis *et al*., 2010). Likewise, Kruskop (2011) reported "about 13%" divergence in COI mtDNA between *R. chaseni* from Vietnam and *R. borneensis* from Borneo. Due to the lack of any DNA barcodes from Peninsular Malaysia, the taxonomic status of *R. borneensis* in the region could not be clarified. The name *R. borneensis* is tentatively retained in this checklist pending further research.

IUCN status: Least Concern

Recorded at: Pahang: Pulau Tioman (Csorba *et al*., 1997); **Perlis**: Wang Pinang (Zubaid, 1994).

R. borneensis is likely to be rare in Peninsular Malaysia (Khan *et al.*, 2008).

4.1.7.4 *Rhinolophus chiewkweeae* Yoshiyuki & Lim, 2005

Rhinolophus chiewkweeae Yoshiyuki & Lim, 2005: 29; Gunung Ledang, Tangkak, Muar, Johor, MALAYSIA, 1276 m (Boo-Liat Lim, collector; NSMT-M 33472).

Common English name: Chiewkwee's Horseshoe Bat

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

Remarks: *R. pearsonii* is reported to occur in Peninsular Malaysia (Corbet & Hill, 1992) but there are no any locality records. It is likely that the records of *R. pearsonii* from Peninsular Malaysia, if valid, may actually represent *R. chiewkweeae*, though NJ analysis revealed that DNA barcodes under these names (excluding specimens from Peninsular Malaysia) are 12% divergent in COI mtDNA (see Figure 3 in Morni *et al.*, 2016).

IUCN status: Not Evaluated

Recorded at: Melaka: Asahan Forest Reserve (Yoshiyuki & Lim, 2005); **Johor**: Gunung Ledang and Labis Forest Reserve (Yoshiyuki & Lim, 2005); **Kedah**: Lubok Semilan, Ulu Melaka in Pulau Langkawi and Weng Subcatchment Area in Ulu Muda Forest Reserve Forest Reserve (Yoshiyuki & Lim, 2005); **Perlis**: Wang Kelian State Park (Jayaraj *et al*.,

2013a); **Perak**: Temenggor Lake (Syaripuddin *et al*., 2014); **Terengganu**: Tasik Kenyir (Syaripuddin *et al*., 2014), Sungai Buweh (Morni *et al*., 2016).

R. chiewkweeae has been recorded in lowland, hill and submontane dipterocarp forests, and an island (Yoshiyuki & Lim, 2005; Morni *et al.*, 2016). In Peninsular Malaysia, all recorded individuals were from mature and secondary dipterocarp forests (Morni *et al*., 2016). The low capture rate of *R. chiewkweeae* in the region suggested that the population density of the species is very low (Jayaraj *et al*., 2013a; Morni *et al.*, 2016).

4.1.7.5 *Rhinolophus coelophyllus* Peters, 1867

Rhinolophus coelophyllus Peters, 1867: 426, pl. 35; Salween River=Thanlwin River, Burma=MYANMAR (Collector unknown; MNB 3143).

Common English name: Croslet Horseshoe bat

Barcode Index Number: DNA barcodes recorded as *R. coelophyllus* are associated with the BIN, BOLD:ACE9393, but there are no DNA barcodes from Peninsular Malaysia.

Remarks: *R. shameli* was previously considered as a subspecies of *R. coelophyllus* (Tate, 1943) but examination of specimens from Thailand and Cambodia suggested that they are distinct species on the basis of *R. shameli* having larger body size and differently shaped rostral part of the skull (Hill & Thonglongya, 1972).

The search of BOLD revealed that the two names are associated with different BINs. DNA barcodes recorded as *R. shameli* are associated with three BINs, BOLD:AAB4877, BOLD:AAB4878 and BOLD:ABY7284 (The BIN, BOLD:ABY7284 also contains DNA barcodes of *R. stheno* and therefore, may be erroneous) whereas DNA barcodes recorded as *R. coelophyllus* are associated with one BIN, BOLD:ACE9393. Specimens recorded as *R. shameli* from Kedah (BM(NH) 1898.10.1.1) and Pulau Langkawi (BM(NH) 1968.821 and BM(NH) 1968.822) are smaller and represent *R. coelophyllus* (Hill & Thonglongya, 1972).

IUCN status: Least Concern

Recorded at: Kedah: Pulau Langkawi and mainland Kedah (Medway, 1969); **Perlis**: mainland Perlis (Medway, 1969), Wang Kelian State Park, (Jayaraj *et al*., 2013a); **Selangor**: Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999).

R. coelophyllus has been recorded in forests near limestone hills and once in a house, and roosts in limestone caves in large colonies with hundreds of individuals (Medway, 1969; Francis, 2008).

4.1.7.6 *Rhinolophus convexus* Csorba, 1997

Rhinolophus convexus Csorba, 1997: 343; Gunung Jasar, Tanah Rata, Cameron Highlands, Pahang State, MALAYSIA, 4°28’ N, 101° 22’ E, 1600 m (G. Csorba and F. Zilahy, collector; HNHM 95.55.14).

Common English name: Convex Horseshoe Bat

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

IUCN status: Data Deficient due to the rarity of this species (Csorba *et al.*, 2016).

Recorded at: Pahang: Gunung Jasar at Tanah Rata in Cameron Highlands (Csorba, 1997); **Selangor:** Unspecified (SMF 84906). *R. convexus* is known by two specimens only from Peninsular Malaysia (Csorba *et al.*, 2016).

The holotype was caught in upper montane rainforest at elevation of 1600 m (Csorba, 1997).

4.1.7.7 *Rhinolophus lepidus* Blyth, 1844

Rhinolophus lepidus Blyth, 1844: 486; Calcutta, INDIA (Collector unknown; Type unknown).

Rhinolophus refulgens Andersen, 1905b: 124, pl. 4; Gunung Igari, Perak, MALAYSIA, 2000 ft. (A. L. Butlerm Esq., presenter; BM(NH) 1898.11.29.2).

Rhinolophus lepidus refulgens Corbet & Hill, 1992.

Common English name: Blyth’s Horseshoe Bat

Barcode Index Number: BOLD:AAB9127 (5 DNA barcodes from Peninsular Malaysia; Figure 4.4)

Remarks: DNA barcodes of *R. lepidus* are associated with three BINs (BOLD:AAB9127, BOLD:ABZ1016 and BOLD:ABZ2266; Figure 4.4). The BIN, BOLD:ABZ1016 contains DNA barcodes recorded as *R. lepidus* and *R. pusillus*.

Some authors considered *R. refulgens* as a subspecies of *R. lepidus* (Corbet & Hill, 1992; Csorba, 1997; Kingston *et al.*, 2006) while some considered them to be distinct (Jayaraj *et al.*, 2012a; Jayaraj *et al*., 2013a). Bumrungsri *et al.* (2008b) commented that *R. lepidus* from Peninsular Malaysia may represent a distinct taxon and the appropriate name would be *R. refulgens* based on the type locality. Here NJ analysis suggested that DNA barcodes recorded as *R. lepidus* from Peninsular Malaysia may be distinct from DNA barcodes recorded as *R. lepidus* from Indochina (Figure 4.4), congruent with Soisook *et al.* (2016) who reported 2.85% divergence in COI mtDNA between *R. refulgens* (from Peninsular Malaysia, south Thailand and Indonesia) and *R. lepidus* (from Vietnam and Cambodia). Soisook *et al.* (2016) also reported that female specimens of *R. refulgens* have shorter forearm and skull lengths compared to *R. lepidus*, though male specimens of both taxa are similar in size. Due to the lack of DNA barcodes from type locality of *R. lepidus* (India) for comparison, it remains to be determined whether the taxon occurring in Peninsular Malaysia represents the nominate *R. lepidus* or *R. refulgens*. The name *R. lepidus* is tentatively used in this checklist pending further research.

IUCN status: Least Concern

Recorded at: Pahang: Pulau Tioman (Csorba *et al.*, 1997), Krau Wildlife Reserve (Anan *et al*., 1998; Kingston *et al*., 2006), Cameron Highland (Shahfiz *et al*., 2008a), Kuala Atok, National Park (Tingga *et al.*, 2012), Bukit Ibam, Kemasul, Jengka, Klau Besar, Kenong and Gunung Aais (Lim *et al.*, 2014), Lata Bujang Forest Reserve (Mohd-Hanif *et al*., 2015); **Terengganu**: Gunung Tebu Forest Reserve (Joann *et al*., 2013), Pasir Raja,

Dungun (Wilson *et al*., 2014), Tasik Kenyir (Syaripuddin *et al*., 2014); **Perak**: Temengor Forest Reserve (Francis, 1995; Joann *et al*., 2011; Shahfiz *et al.*, 2013), Temenggor Lake (Syaripuddin *et al*., 2014); **Selangor**: Bukit Kutu Wildlife Reserve (Lim *et al*., 1999), Semangkok Forest Reserve (Joann *et al*., 2013); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al*., 1999; Lim *et al*., 2014), Bukit Hijau (Lim *et al*., 2014); **Melaka**: Unspecified (Shahfiz *et al*., 2009); **Kelantan**: Air Panas-Gua Musang (Hasan *et al*., 2012), Gunung Reng and Gua Musang (Jayaraj *et al*., 2016b); **Perlis**: Wang Kelian State Park, (Jayaraj *et al*., 2013a); **Johor**: Labis Forest Reserve (Lim *et al*., 2014), Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al.*, 2015); **Negeri Sembilan**: Gunung Angsi Forest Reserve (Joann *et al*., 2013; Lim *et al*., 2014), Berembun Forest Reserve (Joann *et al*., 2013); **Pulau Pinang**: Bukit Panchor (Lim *et al*., 2014).

As *R. refulgens*: **Perak**: Maxwell Hill (Medway, 1969); **Pahang**: Pulau Tioman (Medway, 1969), Krau Wildlife Reserve (Zubaid, 1993); **Johor**: Pulau Pemanggil and Pulau Aur (Medway, 1969); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990); **Kelantan**: Gunung Stong State Park (Jayaraj *et al*., 2012a; Lim *et al*., 2014).

R. lepidus inhabits mature lowland and hill forests. Individuals have been found roosting in caves and rock crevices, often with the congeneric *R. stheno* (Kingston *et al*., 2006; Francis, 2008).

4.1.7.8 *Rhinolophus morio* Gray, 1842

Rhinolophus morio Gray, 1842: 257; SINGAPORE (Collector unknown; BM(NH) 1840.5.14.36).

Rhinolophus luctus morio Lim *et al*., 1999.

Barcode Index Number: There are no DNA barcodes with this name on BOLD. However, DNA barcodes recorded as *R. luctus* are associated with the BIN, BOLD: AAD0380 but there are no DNA barcodes from Peninsular Malaysia.

Remarks: *R. morio* was recently recognised to be distinct from *R. luctus* based on the ratio of zygomatic width to mandible length in cranial measurements and the unique Y-autosomal translocation in karyotype (Volleth *et al.*, 2015).

IUCN status: Not Evaluated but Least Concern as *R. luctus*

Recorded at: Melaka: Unspecified (Gray, 1842); **Pahang**: Pulau Tioman (Lim *et al.*, 1999); **Kuala Lumpur**: Gombak Setia (Volleth *et al.*, 2015); **Selangor**: Templer Park-Rawang, (Volleth *et al.*, 2015).

Recorded as *R. luctus* at: **Pahang**: Bukit Renggit (Ratnam *et al.*, 1989), Krau Wildlife Reserve (Zubaid, 1993; Anan *et al.*, 1998; Kingston *et al.*, 2006), Tasik Chini (Lim & Ratnam, 1999), Cameron Highland (Shahfiz *et al.*, 2008a), Gunung Aais (Lim *et al.*, 2014); **Selangor**: Bangi Forest Reserve (Zubaid, 1993), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999), Ulu Gombak (Medway, 1966; Heller & Volleth, 1989; Lim *et al.*, 2014), Sungai Dusun Forest Reserve (Mohd-Hanif *et al.*, 2015); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Perak**: Royal Belum State Park (Shahfiz *et al.*, 2008b), Temengor Forest Reserve (Joann *et al.*, 2011), Temenggor Lake (Syaripuddin *et al.*, 2014); **Melaka**: Unspecified (Shahfiz *et al.*, 2009); **Kelantan**: Air Panas-Gua Musang (Hasan *et al.*, 2012); **Negeri Sembilan**: Gunung Angsi Forest Reserve (Joann *et al.*, 2013; Lim *et al.*, 2014), Berembun Forest Reserve (Joann *et al.*, 2013); **Terengganu**: Tasik Kenyir (Syaripuddin *et al.*, 2014).

Unlike other *Rhinolophus* species, *R. luctus* sensu lato roosts either solitarily or in pairs often in caves, rock crevices, tree hollows and among tree roots, and has been recorded in primary and secondary forests (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008). Specimens of *R. morio* were collected in lowland dipterocarp forest (Volleth *et al.*, 2015).

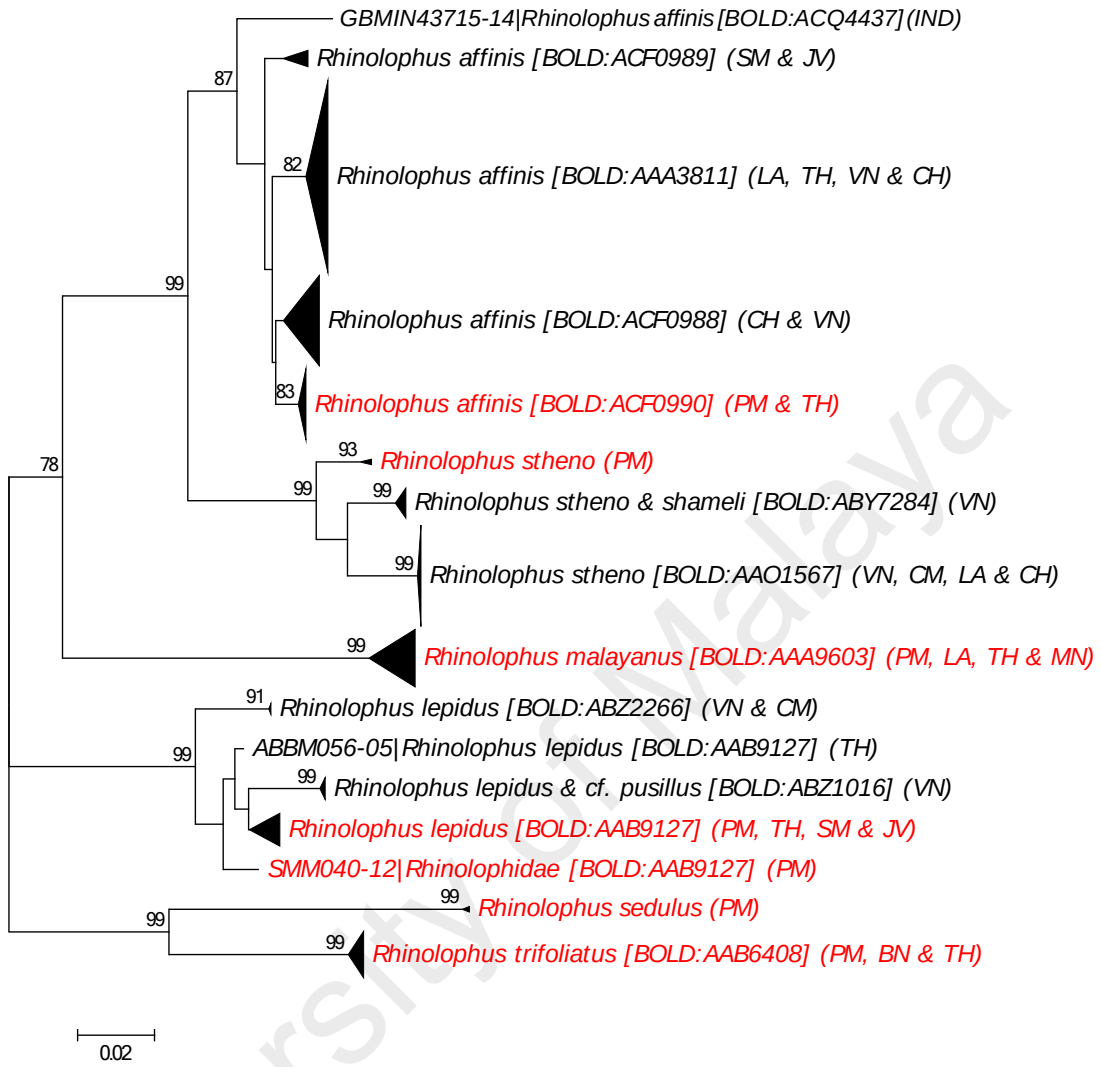

Figure 4.4: Neighbour-joining tree showing all available DNA barcodes for species in family Rhinolophidae reported from Peninsular Malaysia. The percentage of pseudoreplicate trees ($\geq$70%) in which the DNA barcodes clustered together in the bootstrap test (500 pseudoreplicates) are shown above the branches. Abbreviation as follows: PM=Peninsular Malaysia, VN=Vietnam, BN=Borneo (including Sabah & Sarawak of East Malaysia, Brunei and Kalimantan Indonesia), TH=Thailand, LA=Laos, SM=Sumatera Indonesia, JV=Java Indonesia, IND=India, CH=China, CM=Cambodia, MN=Myanmar.

4.1.7.9 *Rhinolophus luctoides* Volleth, Loidl, Mayer, Yong, Müller & Heller, 2015

Rhinolophus luctoides Volleth, Loidl, Mayer, Yong, Müller & Heller, 2015: 4; Ulu Gombak, Selangor, MALAYSIA, 600 m (K. -G. Heller and M. Volleth, collector; SMF 87483).

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

Remarks: *R. luctoides* and *R. morio* were previously synonymised under *R. luctus* but are now considered to be distinct from *R. luctus* on the basis of molecular and morphological characters. *R. luctoides* has a larger ratio of lower toothrow length to mandible length and larger baculum length compared to *R. morio* (Volleth *et al.*, 2015).

IUCN status: Not Evaluated but Least Concern as *R. luctus*.

Recorded at: Selangor: 5 km north-east of Ulu Gombak (Volleth *et al.*, 2015); **Pahang**: Cameron Highland and Genting Highland (Volleth *et al.*, 2015).

Specimens were caught in selectively logged dipterocarp forests at elevations above 600 m and in montane forests (Volleth *et al.*, 2015). See *R. morio* for records of *R. luctus*.

4.1.7.10 *Rhinolophus macrotis* Blyth, 1844

Rhinolophus macrotis Blyth, 1844: 485; NEPAL (Brian Houghton Hodgson, presenter; BM(NH) 45.1.8.416).

Common English name: Big-eared Horseshoe Bat

Barcode Index Number: DNA barcodes recorded as *R. macrotis* are associated with two BINs, BOLD:AAC2064 and BOLD:ACU9422, but there are no DNA barcodes from Peninsular Malaysia.

Remarks: The BIN, BOLD:AAC2064 includes DNA barcodes recorded as *R. macrotis* and *R. siamensis* which demonstrated very shallow genetic divergences (Appendix G; also see Figure 3 in Francis *et al.*, 2010). The BIN, BOLD:ACU9422 contains two DNA barcodes which are originally from GenBank and may be erroneous.

IUCN status: Least Concern

Recorded at: Pahang: Gunung Benom (Medway, 1969), Krau Wildlife Reserve (Zubaid, 1993; Anan *et al.*, 1998; Kingston *et al.*, 2006), Pulau Tioman (Csorba *et al.*, 1997), Klau Besar (Lim *et al.*, 2014); **Perlis**: Wang Kelian State Park (Jayaraj *et al.*, 2013a).

R. macrotis has been recorded in lowland and hill forests (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.7.11 *Rhinolophus malayanus* Bonhote, 1903

Rhinolophus malayanus Bonhote, 1903: 15; Biserat, Jalor, Patani, south THAILAND (Collector unknown: BM(NH) 1903.2.6.83).

Common English name: Malayan Horseshoe Bat

Barcode Index Number: BOLD:AAA9603 (1 DNA barcode from Peninsular Malaysia; Figure 4.4)

IUCN status: Least Concern

Recorded at: Kedah: Kisap Forest Reserve in Pulau Langkawi (Hill, 1972); **Perlis**: Wang Tangga at Kaki Bukit (Hill, 1972), Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Perak**: Bukit Jerneh Cave and Tumang Lembing Cave (Douangboubpha *et al.*, 2010b).

R. malayanus roosts in limestone caves in colonies of hundreds of individuals (Francis, 2008).

4.1.7.12 *Rhinolophus marshalli* Thonglongya, 1973

Rhinolophus marshalli Thonglongya, 1973: 590; foothills of Khao Soi Duo, Amphoe Pong Nam Ron, Chantthaburi, southeast THAILAND (Joe T. Marshall Jr. and Wandee Nong Ngok, collectors; TNRC 54-1669).

Common English name: Marshall's Horseshoe Bat

Barcode Index Number: DNA barcodes recorded as *R. marshalli* are associated with two BINs, BOLD:AAE7426; BOLD:ABZ6523, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Recorded at: Perlis: Guar Jentik (Zubaid & Davison, 1988).

R. marshalli has been recorded in lowland and hill forests at elevation of 800 m, roosting in limestone caves (Francis, 2008).

4.1.7.13 *Rhinolophus pusillus* Temminck, 1834

Rhinolophus pusillus Temminck, 1834: 29; Java, INDONESIA (Collector unknown; NMNL 35177, lectotype).

Common English name: Least Horseshoe Bat

Barcode Index Number: DNA barcodes recorded as *R. pusillus* are associated with three BINs, (BOLD:AAA9397, BOLD:ABZ1016, and BOLD:ABZ2360), but there are no DNA barcodes from Peninsular Malaysia (Appendix H).

Remarks: The BIN, BOLD:ABZ1016 contains DNA barcodes recorded as *R. pusillus* and *R. lepidus.* The DNA barcodes recorded as *R. pusillus* (ABBSI244-10, ABBSI253-10, ABBSI263-10 and ABRVN310-06) are likely to be cases of mis-identification (see remarks on *R. lepidus*). Based on analyses of *COI* mtDNA, Soisook *et al.* (2016) recently described a new species within the "*R. pusillus* group": *R. monticolus* (labelled as *R.* cf. *pusillus* under BIN, BOLD:ACE531) and reported five taxa within the specimens of "*R. pusillus*" from Thailand, Laos, Vietnam and China. Whether the taxon occurring in Peninsular Malaysia belongs to any of these BINs and forms remains to be determined.

IUCN status: Least Concern

Recorded at: Kedah: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Johor**: Gunung Ledang State Park (DWNP-M-08076, DWNP-M-08078, DWNP-M-08079, DWNP-M-08081, DWNP-M-08082); **Pahang**: Pulau Tioman (DWNP-M-08077, DWNP-M-08080, DWNP-M-08083); **Negeri Sembilan**: Berembun Forest Reserve (Joann *et al.*, 2013).

R. pusillus roosts in caves, bamboo clumps and buildings and has been reported foraging in primary and secondary forests (Francis, 2008).

4.1.7.14 *Rhinolophus robinsoni* Andersen, 1918

Rhinolophus robinsoni Andersen, 1918: 375; Khao Nawng, Bandon, THAILAND (Federated Malay States Museum, presenter; BM(NH) 1918.8.2.1).

Common English name: Peninsular Horseshoe Bat

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

Remarks: *R. robinsoni* was previously considered to be conspecific with *R. megaphyllus* (Corbet & Hill, 1992) but is now recognised as a distinct species (Simmons, 2005). Specimens recorded as *R. megaphyllus* from Peninsular Malaysia (Francis, 1995) should be updated to *R. robinsoni.*

IUCN status: Near Threatened

Recorded at: Pahang: Fraser Hill (Medway, 1969; Hill, 1972), Krau Wildlife Reserve (Kingston *et al.*, 2006) Pulau Tioman (Medway, 1969), Kenong and Gunung Aais (Lim *et al.*, 2014); **Johor**: Pulau Aur and Pulau Pemanggil (Medway, 1969), Gunung Panti and Labis Forest Reserve (Lim *et al.*, 2014); **Melaka**: Unspecified (Shahfiz *et al.*, 2009); **Perlis**: Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Kelantan:** Gua Musang (Jayaraj *et al.*, 2016b); **Negeri Sembilan**: Gunung Angsi Forest Reserve (Lim *et al.*, 2014); **Perak**: Kledang Saiong Forest Reserve (Lim *et al.*, 2014); **Pulau Pinang**: Bukit Panchor (Lim *et al.*, 2014).

As *R. megaphyllus*: **Perak**: Temengor Forest Reserve (Francis, 1995).

R. robinsoni is forest inhabitant and has been recorded in lowland and hill forests, roosting in rock crevices and palm leaves (Kingston *et al.*, 2006; Francis, 2008).

4.1.7.15 *Rhinolophus sedulus* Andersen, 1905b

Rhinolophus sedulus Andersen, 1905b: 247; Sarawak, MALAYSIA (A. R. Wallace, collector; Type specimen was previously recorded as no.19 in Robert Fisher Tome's private collection and is currently as BM(NH) 7.1.1.292).

Common English name: Lesser Woolly Horseshoe Bat

Barcode Index Number: DNA barcodes recorded as *R. sedulus* (BM141-03 and BM431-04) are not associated with any BINs due to their short sequence length (<500 bp). Both were collected in Peninsular Malaysia and share >99% similarity (Figure 4.4).

IUCN status: Near Threatened

Recorded at: Selangor: Ulu Gombak (Medway, 1966; Heller & Volleth, 1989), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999), Air Hitam Forest Reserve (Azlan *et al.*, 2000), Semangkok Forest Reserve (Joann *et al.*, 2013); **Pahang**: Kuala Tekah, (Medway, 1969), Krau Wildlife Reserve (Kingston *et al.*, 2006; Khan *et al.*, 2008), Kuala Atok, National Park (Tingga *et al.*, 2012), Bukit Ibam, Kemasul, Klau Besar and Gunung Aais (Lim *et al.*, 2014); **Negeri Sembilan**: Pasoh Forest Reserve, (Francis, 1990); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999), Bukit Hijau (Lim *et al.*, 2014); **Perak**: Kledang Saiong Forest Reserve (Lim *et al.*, 2014); **Johor**: Gunung Panti (Lim *et al.*, 2014), Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al.*, 2015).

R. sedulus has been reported roosting in caves, fallen tree hollows, and bushes either individually or in pairs (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.7.16 *Rhinolophus stheno* Andersen, 1905a

Rhinolophus stheno Andersen, 1905a: 91, pl. 3; Selangor, MALAYSIA (H. N. Ridley, Esq., presenter; BM(NH) 98.3.13.1).

Common English name: Lesser Brown Horseshoe Bat

Barcode Index Number: DNA barcodes recorded as *R. stheno* are associated with two BINs, BOLD:AAO1567 and BOLD:ABY7284, but there are no DNA barcodes from Peninsular Malaysia in these BINs. Two DNA barcodes recorded as *R. stheno* from Peninsular Malaysia (BM504-04 and BM505-04) are not placed in any BINs due to their short sequence length (<500bp). Neither of these barcodes from Peninsular Malaysia are associated with BOLD:AAO1567 or BOLD:ABY7284 based on NJ analysis (Figure 4.4).

Remarks: *R. microglobosus* was described as a subspecies of *R. stheno* based on its smaller skull and globular anterior median rostral swellings (Csorba & Jenkins, 1998). Soisook *et al*. (2008) reported that the taxa are morphometrically and acoustically distinct, and consequently raised *R. microglobosus* to full species with distribution covering Thailand, Myanmar, Cambodia, Vietnam and Laos, and restricted *R. stheno* to southern Thailand, Peninsular Malaysia and central Vietnam. DNA barcodes associated with the BIN, BOLD:AAO1567 may represent *R. microglobosus*, while DNA barcodes, BM504-04 and BM505-04 (Francis *et al.*, 2010) may represent *R. stheno* sensu stricto as they were collected from the type locality. The BIN, BOLD:ABY7284 which contains DNA barcodes recorded as *R. stheno* and *R. shameli* may be erroneous (Figure 4.4).

IUCN status: Least Concern

Recorded at: Pulau Pinang: Bukit Panchor (Medway, 1969; Lim *et al.*, 2014); **Selangor**: Ulu Gombak (Heller & Volleth, 1989; Lim *et al.*, 2014), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999), Semangkok Forest Reserve (Joann *et al.*, 2013); **Perak**: Temengor Forest Reserve (Francis, 1995; Joann *et al.*, 2011; Shahfiz *et al.*, 2013), Royal Belum State Park (Shahfiz *et al.*, 2008b), Kledang Saiong Forest Reserve (Joann *et al.*, 2013); **Pahang**: Pulau Tioman (Csorba *et al.*, 1997), Krau Wildlife Reserve (Anan *et al.*, 1998; Kingston *et al.*, 2006), Tasik Chini (Lim & Ratnam, 1999), Cameron Highland (Shahfiz *et al.*, 2008a), Bukit Ibam, Kemasul, Jengka, Klau Besar, Kenong and Gunung Aais (Lim *et al.*, 2014), Lata Bujang Forest Reserve (Mohd-Hanif *et al.*, 2015); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999; Lim *et al.*, 2014), Bukit Hijau (Lim *et al.*, 2014); **Kelantan**: Air Panas-Gua Musang (Hasan *et al.*, 2012), Gunung Stong State Park (Lim *et al.*, 2014), Gua Musang (Jayaraj *et al.*, 2016b); **Perlis**: Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Negeri Sembilan**: Gunung Angsi Forest Reserve (Joann *et al.*, 2013; Lim *et al.*, 2014), Berembun Forest Reserve (Joann *et al.*, 2013); **Terengganu**: Gunung Tebu Forest Reserve (Joann *et al.*, 2013); **Johor**: Gunung Panti and Labis Forest Reserve (Lim *et al.*, 2014).

R. stheno roosts in limestone caves, sometimes in smaller colonies in rock crevices and tree hollows (Kingston *et al.*, 2006; Francis, 2008), occassionally with *R. lepidus* (Kingston *et al.*, 2006).

4.1.7.17 *Rhinolophus trifoliatus* Temminck, 1834

Rhinolophus trifoliatus Temminck, 1834: 24, pl. 1 (and 1835: 27, pl. 31); Bantam, west Java, INDONESIA (Collector unknown; NMNL 35194).

Common English name: Trefoil Horseshoe Bat

Barcode Index Number: BOLD:AAB6408 (10 DNA barcodes from Peninsular Malaysia; Figure 4.4)

IUCN status: Least Concern

Recorded at: Pahang: Gunung Tahan (Bonhote, 1908), Krau Wildlife Reserve (Zubaid, 1993; Anan *et al.*, 1998; Kingston *et al.*, 2006), Tasik Chini (Lim & Ratnam, 1999), Kuala Atok, National Park (Tingga *et al.*, 2012), Bukit Ibam, Kemasul, Jengka, Klau Besar and Gunung Aais (Lim *et al.*, 2014), Tasek Bera Forest Reserve, Lata Bujang Forest Reserve and Fraser Hill Forest Reserve (Mohd-Hanif *et al.*, 2015); **Selangor**: Ulu Gombak (Ratnam *et al.*, 1989; Heller & Volleth, 1989; Lim *et al.*, 2014), Bangi Forest Reserve (Zubaid, 1993), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999), Air Hitam Forest Reserve (Azlan *et al.*, 2000), Semangkok Forest Reserve (Joann *et al.*, 2013), Sungai Dusun Forest Reserve (Mohd-Hanif *et al.*, 2015); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990), Gunung Angsi Forest Reserve (Joann *et al.*, 2013; Lim *et al.*, 2014), Berembun Forest Reserve (Joann *et al.*, 2013); **Perak**: Temengor Forest Reserve (Francis, 1995; Joann *et al.*, 2011; Shahfiz *et al.*, 2013), Royal Belum State Park (Shahfiz *et al.*, 2008b; Tamrin *et al.*, 2010), Kledang Saiong Forest Reserve (Joann *et al.*, 2013; Lim *et al.*, 2014), Temenggor Lake (Syaripuddin *et al.*, 2014); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999; Lim *et al.*, 2014); **Melaka**: Sungai Udang Forest Reserve (Shahfiz *et al.*, 2009); **Kelantan**: Air Panas-Gua Musang (Hasan *et al.*, 2012); **Perlis**: Wang Kelian State

Park (Jayaraj *et al.*, 2013a); **Johor**: Gunung Panti and Labis Forest Reserve (Lim *et al.*, 2014), Endau-Kluang Forest Reserve and Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al.*, 2015); **Terengganu**: Gunung Tebu Forest Reserve (Joann *et al.*, 2013), Tasik Kenyir (Syaripuddin *et al.*, 2014); **Pulau Pinang**: Bukit Panchor (Lim *et al.*, 2014).

R. trifoliatus roosts solitarily under leaves of palms, rattan and small trees, and has been recorded in mangroves, and primary and secondary forests at all elevations (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.8 Family: Vespertilionidae (Subfamily: Kerivoulinae)

4.1.8.1 *Kerivoula hardwickii* [Horsfield, 1824]

Vespertilio hardwickii Horsfield, 1824: part 8; Java, INDONESIA (Collector unknown; Type: BM(NH) 79.11.29.181).

Kerivoula hardwickii Chasen, 1940.

Common English name: Hardwicke's Woolly Bat

Barcode Index Number: BOLD:AAA6722 (5 DNA barcodes from Peninsular Malaysia; Figure 4.5)

Remarks: DNA barcodes recorded as *K. hardwickii* are associated with four BINs, BOLD:AAA6722, BOLD:AAA6725, BOLD:AAC5514 and BOLD:AAC5515. DNA barcodes from Peninsular Malaysia, Thailand, Vietnam and Laos formed a mainland group whereas DNA barcodes from Malaysian Borneo and Kalimantan, Indonesia formed a Bornean group in NJ tree (Figure 4.5). Francis *et al.* (2007) suggested *K. hardwickii* as a species complex based on *COI* mtDNA analysis, but Khan *et al.* (2010) recognised a single form only across Peninsular Malaysia and Borneo, and added that the Bornean form is a result of chromosomal polymorphism. Douangboubpha *et al.* (2016) reported that specimens from Thailand identified as *K. hardwickii* have either “flat” or “domed” skull. Specimens with “flat” skull did not show variation in size and morphology but were clustered into two clades: *K. hardwickii* A and *K. hardwickii* B which are 2.14% divergent

in *COI* mtDNA. Specimens with "domed" skull showed variation in size and morphology but were clustered together in *COI* mtDNA analysis as *K. hardwickii* C, and is 16.37% and 20.02% divergent from *K. hardwickii* A and *K. hardwickii* B in *COI* mtDNA.

Simmons (2005) did not recognise any subspecies under *K. hardwickii*, contradicting the older literature; Ellerman and Morrison-Scott (1966) recognised four subspecies: *K. h. hardwickii* (type locality: Java), *K. h. depressa* (type locality: southern Burma=Myanmar), *K. h. crypta* (type locality: southern India), and *K. h. malpasi* (type locality: Sri Lanka) whereas Hill (1965) recognised five including *K. h. engana* (type locality: southwest of Sumatra). Douangboubpha *et al.* (2016) proposed the names *hardwickii* and *depressa* for the specimens from Thailand with "domed" and "flat" skulls but recommended further research for assigning the Linnaean names conclusively. Whether the four BINs in the NJ tree (Figure 4.5) represent the four subspecies remains to be determined with the name *K. hardwickii* being tentatively used in this checklist.

IUCN status: Least Concern

Recorded at: Kelantan: Ulu Kelantan (Medway, 1969), Air Panas-Gua Musang, (Hasan *et al.*, 2012), Gunung Stong State Park (Lim *et al.*, 2014), Gua Musang (Jayaraj *et al.*, 2016b); **Perak**: Temengor Forest Reserve (Francis, 1995; Joann *et al.*, 2011; Shahfiz *et al.*, 2013), Royal Belum State Park (Shahfiz *et al.*, 2008b), Kledang Saiong Forest Reserve (Joann *et al.*, 2013; Lim *et al.*, 2014); **Melaka**: Sungai Udang Forest Reserve (Shahfiz *et al.*, 2009); **Perlis**: Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Selangor**: Semangkok Forest Reserve and Ulu Gombak (Joann *et al.*, 2013); **Terengganu**: Gunung Tebu Forest Reserve (Joann *et al.*, 2013); **Negeri Sembilan**: Gunung Angsi Forest Reserve (Joann *et al.*, 2013; Lim *et al.*, 2014), Berembun Forest Reserve (Joann *et al.*, 2013); **Johor**: Gunung Panti and Labis Forest Reserve (Lim *et al.*, 2014), Endau-Kluang Forest Reserve (Mohd-Hanif *et al.*, 2015); **Pahang**: Bukit Ibam, Klau Besar, and Gunung Aais (Lim *et al.*, 2014); **Pulau Pinang**: Bukit Panchor (Lim *et al.*, 2014).

K. hardwickii has been reported roosting in tree hollows, among clumps of dead leaves, and in dead and broken bamboo stems (Medway, 1969; Francis, 2008).

4.1.8.2 *Kerivoula krauensis* Francis, Kingston & Zubaid, 2007

Kerivoula krauensis Francis, Kingston & Zubaid, 2007: 3; Kuala Lompat, Krau Wildlife Reserve, Pahang, MALAYSIA, 3° 43' N 102° 10' E (Charles M. Francis, collector; BM(NH) 1999.294).

Common English name: Krau Woolly Bat

Barcode Index Number: BOLD:AAI8031 (1 DNA barcode from Peninsular Malaysia; Figure 4.5)

IUCN status: Data Deficient; Vulnerable to rapid deforestration (Francis, 2008).

Recorded at: Pahang: Krau Wildlife Reserve (Francis *et al.*, 2007); **Terengganu:** Sekayu Recreational Forest (Struebig *et al.*, 2017).

K. krauensis has been recorded in peat swamps, primary and logged lowland dipterocarp and montane forests. Its roosting ecology remains unknown (Francis *et al.*, 2007; Struebig *et al.*, 2017).

4.1.8.3 *Kerivoula intermedia* Hill & Fancis, 1984

Kerivoula intermedia Hill & Fancis, 1984: 323; Lumerau, Sabah, Borneo, MALAYSIA 5°12'N, 118°52'E (Charles M. Francis, collector; BM(NH) 1983.356).

Common English name: Small Woolly Bat

Barcode Index Number: BOLD:AAD4883 (5 DNA barcodes from Peninsular Malaysia; Figure 4.5)

Remarks: DNA barcodes recorded as *K. intermedia* are associated with two BINs, BOLD:AAD4883, and BOLD:AAM3704. The BIN, BOLD:AAD4883 comprises DNA barcodes from Peninsular Malaysia and Sarawak, Borneo while the BIN, BOLD:AAM3074 comprises a single DNA barcode (BM012-03) from Sabah, Borneo

(Figure 4.5). Whether the DNA barcode, BM012-03 represents a cryptic species, a case of mis-identification, or a case of high intraspecific variation remains to be determined.

IUCN status: Near Threatened

Recorded at: Pahang: Tekam Forest Reserve (Hill & Fancis, 1984), Krau Wildlife Reserve (Zubaid, 1993; Kingston *et al.*, 2006; Khan *et al.*, 2008), Bukit Ibam, Kenong and Gunung Aais (Lim *et al.*, 2014), Tasik Bera Forest Reserve (Mohd-Hanif *et al.*, 2015); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990), Gunung Angsi Forest Reserve and Berembun Forest Reserve (Joann *et al.*, 2013); **Selangor**: Air Hitam Forest Reserve (Azlan *et al.*, 2000), Semangkok Forest Reserve (Joann *et al.*, 2013); **Perak**: Royal Belum State Park (Shahfiz *et al.*, 2008b; Tamrin *et al.*, 2010), Temengor Forest Reserve (Joann *et al.*, 2011), Kledang Saiong Forest Reserve (Joann *et al.*, 2013; Lim *et al.*, 2014); **Melaka**: Sungai Udang Forest Reserve (Shahfiz *et al.*, 2009); **Perlis**: Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Johor**: Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al.*, 2015), Gunung Panti and Labis Forest Reserve (Lim *et al.*, 2014); **Kelantan**: Air Panas-Gua Musang (Hasan *et al.*, 2012), Gunung Stong State Park (Lim *et al.*, 2014); **Terengganu**: Gunung Tebu Forest Reserve (Joann *et al.*, 2013), Sungei Kelembang at Ulu Setiu in Besut (Hill & Fancis, 1984).

The roosting ecology of *K. intermedia* remains unknown but the species has been recorded in the understory of lowland forest (Kingston *et al.*, 2006; Francis, 2008).

4.1.8.4 *Kerivoula minuta* Miller, 1898

Kerivoula minuta Miller, 1898: 321; Lay Song Hong, Trang, south THAILAND (Dr. W. L. Abbott, collector; USNM 83547).

Common English name: Least Woolly Bat

Barcode Index Number: BOLD:AAC1298 (9 DNA barcodes from Peninsular Malaysia; Figure 4.5)

Remarks: DNA barcodes recorded as *K. minuta* are associated with six BINs, BOLD:AAC1296, BOLD:AAC1297, BOLD:AAC1298, BOLD:AAC1299, BOLD:ACF4510, and BOLD:ACF451 (Figure 4.5). Khan (2008) reported 4.44% of divergence in cytochrome *b* mtDNA between *K. minuta* from Peninsular Malaysia and Borneo (Sabah and Sarawak) with no shared haplotypes. NJ analysis also showed divergence between *K. minuta* from Peninsular Malaysia and Borneo (Figure 4.5). The taxon occurring in Peninsular Malaysia represents *K. minuta* based on the type locality.

IUCN status: Near Threatened

Recorded at: Perak: Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013), Maxwell Hill (Khan *et al.*, 2008), Royal Belum State Park (Shahfiz *et al.*, 2008b); **Kedah**: Bukit Hijau and Ulu Muda Forest Reserve (Norsham *et al.*, 1999; Khan *et al.*, 2008; Lim *et al.*, 2014); **Pahang**: Krau Wildlife Reserve and Lakum (Khan *et al.*, 2008), Kuala Atok, National Park (Khan *et al.*, 2008; Tingga *et al.*, 2012), Bukit Ibam, Kenong and Gunung Aais (Lim *et al.*, 2014); **Johor**: Endau Rompin National Park (BM422-04 and ABRSS347-06, Francis *et al.*, 2010), Gunung Panti and Labis Forest Reserve (Lim *et al.*, 2014); **Kelantan**: Gua Musang (Hasan *et al.*, 2012; Jayaraj *et al.*, 2016b); **Negeri Sembilan**: Gunung Angsi Forest Reserve (Lim *et al.*, 2014).

K. minuta has been recorded in understory of lowland forests and disturbed areas (Francis, 2008).

Kerivoula papillosa* and *K. lenis

K. lenis is closely associated with *K. papillosa* (Corbet & Hill, 1992) but the former has smaller skull and teeth, shorter muzzle and narrower palate (Vanitharani *et al.*, 2003; Khan *et al.*, 2010). The two species are 10.85% divergent in cytochrome *b* mtDNA and possess unique karyotypic characters: *K. papillosa* has a diploid number of chromosomes=38 and fundamental number=54 whereas *K. lenis* has a diploid number of chromosomes=38 and fundamental number=52 (Khan *et al.*, 2008). *COI* mtDNA analysis

by Francis *et al.* (2010) suggested that there are at least four distinct clusters among specimens recorded as *K. papillosa* and *K. lenis*. Douangboubpha *et al.* (2016) reported that specimens from Thailand identified as *K. papillosa* represent five morphological forms but only three distinct clusters based on COI mtDNA analyses.

4.1.8.5 *Kerivoula papillosa* Temminck, 1840

Kerivoula papillosa Temminck, 1840: 220, PL. 55; Bantam, west Java (restricted by Tate 1940), INDONESIA (Collector unknown; Type unknown).

Kerivoula malayana Chasen, 1940: 55; Ginting Bedai, Selangor-Pahang, MALAYSIA, 2300ft (Collector unknown; BM(NH) 1947.1483).

Kerivoula papillosa malayana Medway, 1969

Common English name: Papillose Woolly Bat

Barcode Index Number: BOLD:AAC9529 (8 DNA barcodes from Peninsular Malaysia; Figure 4.5)

Remarks: Chasen (1940) described *K. malayana* based on a specimen collected at the Selangor-Pahang border in Peninsular Malaysia. Kingston *et al.* (1999) reported two forms of *K. papillosa* which are different in forearm length and acoustic characters in Krau Wildlife Reserve, Peninsular Malaysia. Douangboubpha *et al.* (2016) reported five morphological forms within three distinct clusters (*K. papillosa* A, B and C) based on *COI* mtDNA analysis in Thailand. *K. papillosa* A corresponds with *K. p. malayana* based on the larger skull and higher braincase, and is 6.97% divergent from *K. papillosa* B which comprises two morphological forms. The larger *K. papillosa* B (forearm length: 42.1 – 42.3 mm and greatest skull length: 17.0 – 17.1 mm) and the smaller *K. papillosa* B (forearm: 39.4 – 40.2 mm and greatest skull length: 16.6 -17.0 mm) are only 1.99% divergent and may or may not represent a further undescribed species. *K. papillosa* C showed morphological variation and is only 0.55% divergent from *K. lenis* collected in

Peninsular Malaysia but is 13.06% and 14.86% divergent from *K. papillosa* A and B (Douangboubpha *et al*., 2016).

NJ analysis revealed three clusters of DNA barcodes recorded as *K. papillosa* corresponding to three BINs, BOLD:AAC9527, BOLD:AAC9528 and BOLD:AAC9529 (Figure 4.5). It is likely that the BIN, BOLD:AAC9529 (as *K. papillosa* Small in Khan *et al.*, 2010 and as *K. papillosa* A in Douangboubpha *et al.*, 2016) with DNA barcodes recorded as *K. papillosa* and *K.* cf. *papillosa* represent *K. p. malayana* based on type locality. The name *K. papillosa* is conservatively retained in this checklist pending further research to address the suggestion that "*malayana*" to be recognised as a distinct species.

IUCN status: Least Concern

Recorded at: Negeri Sembilan: Pasoh Forest Reserve (Francis, 1990), Gunung Angsi Forest Reserve (Joann *et al.*, 2013; Lim *et al.*, 2014), Berembun Forest Reserve (Joann *et al.*, 2013); **Pahang**: Krau Wildlife Reserve (Zubaid, 1993; Kingston *et al.*, 2006; Khan *et al.*, 2008), Kuala Atok, National Park (Tingga *et al.*, 2012), Bukit Ibam, Kemasul, Jengka, Klau Besar, Kenong and Gunung Aais (Lim *et al.*, 2014), Tasik Bera Forest Reserve and Fraser Hill Forest Reserve (Mohd-Hanif *et al.*, 2015); **Selangor**: Bangi Forest Reserve (Zubaid, 1993), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999), Air Hitam Forest Reserve (Azlan *et al.*, 2000), Sungai Dusun Forest Reserve (Mohd-Hanif *et al.*, 2015), Semangkok Forest Reserve (Joann *et al.*, 2013); **Perak**: Temengor Forest Reserve (Francis, 1995; Joann *et al.*, 2011; Shahfiz *et al.*, 2013), Royal Belum State Park (Shahfiz *et al.*, 2008b; Tamrin *et al.*, 2010), Kledang Saiong Forest Reserve (Joann *et al.*, 2013; Lim *et al.*, 2014); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999; Lim *et al.*, 2014); **Melaka**: Sungai Udang Forest Reserve (Shahfiz *et al.*, 2009);**Perlis**: Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Terengganu**: Gunung Tebu Forest Reserve (Joann *et al.*, 2013); **Pulau Pinang**: Bukit Panchor (Lim *et al.*, 2014); **Johor**: Gunung Panti and Labis Forest Reserve (Lim *et al.*, 2014), Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al.*,

2015); **Kelantan**: Gunung Stong State Park (Lim *et al.*, 2014), Gua Musang (Jayaraj *et al.*, 2016b).

K. papillosa roosts in pairs or small groups, with males tend to roost solitarily (Medway, 1969; Kingston *et al.*, 2006). The species has been recorded roosting in dead or broken bamboo stems and cavities in live standing trees (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008), and may have a small home range on the high recapture rate in Krau Wildlife Reserve (Kingston *et al.*, 2006).

4.1.8.6 *Kerivoula lenis* Thomas, 1916a

Kerivoula lenis Thomas, 1916a: 416; Calcutta, Bengal, INDIA (J. T. Pearson, presenter; BM(NH) 1879.11.21.126).

Common English name: Indian Woolly Bat

Barcode Index Number: BOLD:AAD4874 (3 DNA barcodes from Peninsular Malaysia; Figure 4.5)

Remarks: *K. lenis* was previously considered as a subspecies of *K. papillosa* Hill (1965) but was later raised as a distinct species (Vanitharani *et al.*, 2003; Simmons, 2005). Douangboubpha *et al.* (2016) reported three distinct clusters of *K. papillosa* (*K. papillosa* A, B and C) from Thailand based on NJ analyses of *COI* mtDNA which also clustered *K. lenis* from Peninsular Malaysia (BIN, BOLD:AAD4874) with *K. papillosa* C. *K. lenis* from Peninsular Malaysia has been reported to be 5.33% divergent from *K. lenis* from Borneo and >14% divergent from *K.* cf. *lenis* from Laos (Francis *et al.*, 2010; Khan *et al.*, 2010; Douangboubpha *et al.*, 2016). Here, NJ analysis revealed three clusters of DNA barcodes recorded as *K. lenis* and *K.* cf. *lenis* associated with three BINs, BOLD:AAC9530, BOLD:AAD4873 and BOLD:AAD4874 (Figure 4.5).Whether the taxon occurring in Peninsular Malaysia represents *K. lenis* sensu stricto remains to be determined due to the lack of comparative material from the type locality, India.

IUCN status: Least Concern

Recorded at: Pahang: Krau Wildlife Reserve (Khan *et al.*, 2008); **Negeri Sembilan**: Pasoh Forest Reserve (BM(NH) 1988.46).

K. lenis has been recorded in understory of forest (Francis, 2008).

4.1.8.7 *Kerivoula pellucida* [Waterhouse, 1845]

Vespertilio pellucidus Waterhouse, 1845: 6; PHILLIPINES (H. Cuming, Esq.; Type unknown).

Kerivoula pellucida Jentink, 1891.

Common English name: Clear-winged Woolly Bat

Barcode Index Number: BOLD:AAD1601 (8 DNA barcodes from Peninsular Malaysia; Figure 4.5)

IUCN status: Near Threatened

Recorded at: Pahang: Krau Wildlife Reserve (Kingston *et al.*, 2006), Kuala Atok, National Park (Tingga *et al.*, 2012), Bukit Ibam, Kemasul, Jengka, Kenong and Gunung Aais (Lim *et al.*, 2014), Tasik Bera Forest Reserve and Fraser Hill Forest Reserve (Mohd-Hanif *et al.*, 2015); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990), Gunung Angsi Forest Reserve (Joann *et al.*, 2013; Lim *et al.*, 2014), Berembun Forest Reserve (Joann *et al.*, 2013); **Perak**: Temengor Forest Reserve (Francis, 1995; Tamrin *et al.*, 2010; Shahfiz *et al.*, 2013), Royal Belum State Park (Shahfiz *et al.*, 2008b), Kledang Saiong Forest Reserve (Lim *et al.*, 2014); **Perlis**: Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Kelantan**: Air Panas-Gua Musang (Hasan *et al.*, 2012), Gunung Stong State Park (Lim *et al.*, 2014); **Selangor**: Semangkok Forest Reserve and Ulu gombak (Joann *et al.*, 2013); **Terengganu**: Gunung Tebu Forest Reserve (Joann *et al.*, 2013); **Johor**: Gunung Panti (Lim *et al.*, 2014), Endau-Kluang Forest Reserve (Mohd-Hanif *et al.*, 2015); **Kedah**: Bukit Hijau (Lim *et al.*, 2014).

K. pellucida has been reported foraging in understory of tall forests with dense vegetation and roosting in clumps of dried leaves (Medway, 1969; Kingston *et al.*, 2006;

Francis, 2008). Captured individuals were found roosting in tight clusters in harp traps, suggesting social bonds (Kingston *et al.*, 2006).

4.1.8.8 *Kerivoula picta* [Pallas, 1767] (?)

Vespertilio pictus Pallas, 1767: 7; probably Ternate Island, north Moluccas, INDONESIA (Collector unknown; Type unknown).

Kerivoula picta Cantor, 1846.

Common English name: Painted Woolly Bat

Barcode Index Number: DNA barcodes recorded as *K. picta* are associated with the BIN, BOLD:AAX0264, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Recorded at: Pulau Pinang: Unspecified (Cantor, 1846).

There are no recent locality reports for *K. picta* in Peninsular Malaysia, although the species is thought to occur in Peninsular Malaysia (Medway, 1969; Corbet & Hill, 1992; Nowak, 1994; Davison & Zubaid, 2007; Francis, 2008; DWNP, 2010). *K. picta* has been recorded from Thailand (Boonsong & McNeely, 1977) and therefore, may be restricted to northern Peninsular Malaysia. Individuals have been found roosting among dead leaves of trees and bananas (Francis, 2008).

4.1.8.9 *Kerivoula whiteheadi* Thomas, 1894 (?)

Kerivoula whiteheadi Thomas, 1894: 460; Molino, Isabella, northeast Luzaon Island, PHILLIPINES (J. Whitehead, collector; BM(NH) 1894.10.9.2).

Common English name: Whitehead's Woolly Bat

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

IUCN status: Least Concern

Remarks: Chasen (1940) listed *K. whiteheadi* as *Kerivoula bicolor* (=now *Kerivoula whiteheadi bicolor*) in his “Handlist of Malaysian Mammals”.

Recorded at: The holotype of *K. whiteheadi bicolor* (BM(NH) 3.2.6.91) was collected at Biserat, Jalor=Yala, Malay Peninsula (=now southern tip of Thailand) and is the only record from the mainland (Boonsong & McNeely, 1977; Francis, 2008). *K. whiteheadi* may be expected to occur in Peninsular Malaysia (Chasen, 1940; Medway, 1969; Corbet & Hill, 1992; Nowak, 1994) based on the type locality but is yet to be documented (Khan *et al.*, 2010).

K. whiteheadi has been recorded in secondary forests, shrubs and open grasslands, and found roosting in small groups of twenty to thirty individuals among dead leaves by a river (Francis, 2008).

4.1.8.10 *Phoniscus atrox* Miller, 1905

Phoniscus atrox Miller, 1905: 230; vicinity of the Kateman River, east Sumatra, INDONESIA (Dr. W. L. Abbott, collector; USNM 123141).

Common English name: Lesser Groove-toothed Bat

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

IUCN status: Near Threatened

Recorded at: Selangor: Ulu Gombak (Medway, 1966; 1969; Heller & Volleth, 1989); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990); **Pahang**: Krau Wildlife Reserve (Zubaid, 1993; Anan *et al.*, 1998; Kingston *et al.*, 2006), Bukit Ibam, Kemasul and Gunung Aais (Lim *et al.*, 2014): **Perak**: Temengor Forest Reserve (Francis, 1995; Joann *et al.*, 2011; Shahfiz *et al.*, 2013), Royal Belum State Park (Shahfiz *et al.*, 2008b); **Kelantan**: Air Panas-Gua Musang (Hasan *et al.*, 2012); **Terengganu**: Gunung Tebu Forest Reserve (Joann *et al.*, 2013), Pasir Raja-Dungun (Wilson *et al.*, 2014); **Johor**: Labis Forest Reserve (Lim *et al.*, 2014).

P. atrox has been recorded in primary lowland forest and disturbed areas near primary forest. Individuals have been reported roosting in abandoned hanging bird nests (Kingston *et al.*, 2006; Francis, 2008).

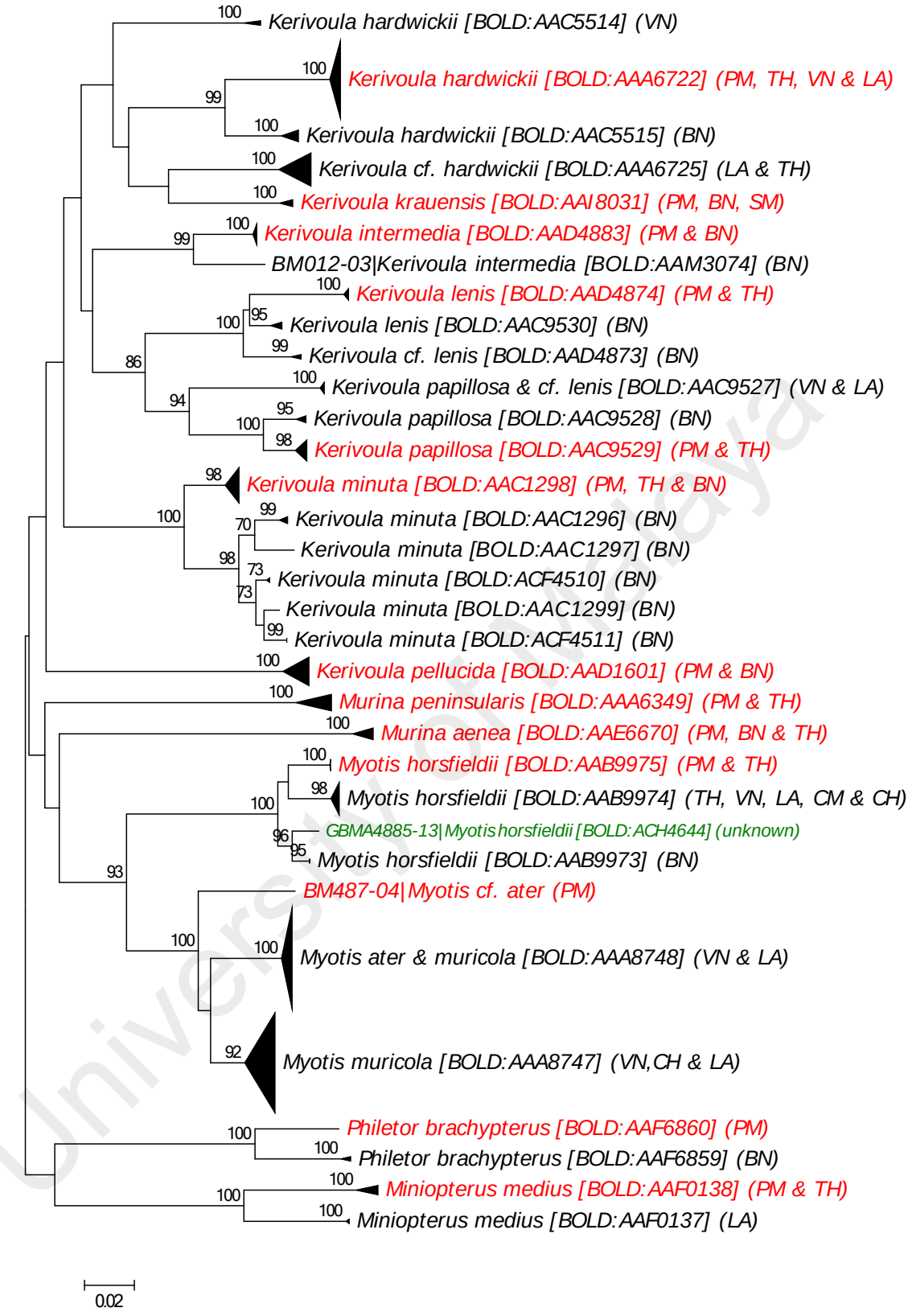

Figure 4.5: Neighbour-joining tree showing all available DNA barcodes for species in family Vespertilionidae reported from Peninsular Malaysia. The percentage of pseudoreplicate trees ($\geq$70%) in which the DNA barcodes clustered together in the bootstrap test (500 pseudoreplicates) are shown above the branches. Abbreviation as follows: PM=Peninsular Malaysia, VN=Vietnam, BN=Borneo (including Sabah & Sarawak of East Malaysia, Brunei and Kalimantan Indonesia), TH=Thailand, LA=Laos, SM=Sumatera Indonesia, JV=Java Indonesia, CH=China, CM=Cambodia.

4.1.8.11 *Phoniscus jagorii* [Peters, 1866a]

Vespertilio (Kerivoula) jagorii Peters, 1866a: 399; Samar Island, PHILLIPINES (Collector unknown; Type unknown).

Phoniscus jagorii Kingston *et al.*, 2006.

Common English name: Greater Groove-toothed Bat

Barcode Index Number: DNA barcodes recorded as *P. jagorii* are associated with the BIN, BOLD:AAC4331, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Recorded at: Pahang: Krau Wildlife Reserve (Kingston *et al.*, 2006).

P. jagorii is rare in understorey of forest and has been recoded in primary lowland forests (Kingston *et al.*, 2006; Francis, 2008).

4.1.9 Family: Vespertilionidae (Subfamily: Miniopterinae)

4.1.9.1 *Miniopterus magnater* Sanborn, 1931

Miniopterus schreibersii magnater Sanborn, 1931: 26; Marienburg, 40 miles up the Sepik River, PAPUA NEW GUINEA (Frank C. Wonder, collector; FMNH 31802).

Miniopterus magnater Corbet & Hill, 1992.

Common English name: Large Bent-winged Bat

Barcode Index Number: DNA barcodes recorded as *M. magnater* are associated with the BIN, BOLD:AAA9957, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Recorded at: Terengganu: Bukit Dendong (Yeap, 2003); **Pahang**: National Park (DWNP-M-07512).

M. magnater is a cave dweller and has been recorded near streams and small bodies of water (Francis, 2008).

4.1.9.2 *Miniopterus medius* Thomas &Wroughton, 1909

Miniopterus medius Thomas & Wroughton, 1909: 382; Kalipoetjang, Tji-Tandoei River, west Java, INDONESIA (G. C. Shortridge, collector; BM(NH) 1909.1.5.464).

Common English name: Medium Bent-winged Bat

Barcode Index Number: BOLD:AAF0138 (1 DNA barcode from Peninsular Malaysia; Figure 4.5)

Remarks: DNA barcodes recorded as *M. medius* are associated with two BINs, BOLD:AAF0137, and BOLD:AAF0138. The BIN, BOLD:AAF0138 comprises the only DNA barcode from Peninsular Malaysia (ABBSI031-04) and unidentified DNA barcodes from Thailand. None of the DNA barcodes were collected near the type locality. Based on NJ analysis, the two BINs are 8.1% divergent (Figure 4.5). No subspecies is described for the species at the moment.

IUCN status: Least Concern

Recorded at: Pahang: Panching and Fraser Hill (Medway, 1969), Bukit Cheras (Hill, 1972), Krau Wildlife Reserve (Kingston *et al.*, 2006); **Perak**: Maxwell Hill and Gunong Pondok (Medway, 1969); **Johor**: Kaban Island (Medway, 1969); **Selangor**: Ulu Gombak (Heller & Volleth, 1984); **Terengganu**: Bukit Dendong (Yeap, 2003).

M. medius roosts in caves and inhabits primary lowland, hill and montane forests (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.9.3 *Miniopterus schreibersii* [Kuhl, 1817]

Vespertilio schreibersii Kuhl, 1817: 185; 'Columbäzar Höhle', R Danube, ROMANIA (Collector unknown; Type unknown).

Miniopterus schreibersii blepotis Medway, 1969.

Miniopterus fuliginosus Francis & Eger, 2012.

Common English name: Common Bent-winged Bat

Barcode Index Number: DNA barcodes recorded as *M. schreibersii* are associated with four BINs, BOLD:AAC3658, BOLD:ACE8769, BOLD:AAX4032 and BOLD:AAA995, but there are no DNA barcodes from Peninsular Malaysia.

Remarks: Tian *et al.* (2004) reported a large divergence in cytochrome *b* mtDNA among specimens identified as *M. schreibersii* from Europe, Asia and Australia, congruent with Maeda (1982) and Appleton *et al.* (2004). The taxonomy of *M. schreibersii* was revised by Tian *et al.* (2004) based on molecular and geographical characteristics resulting in distribution of *M. schreibersii* sensu stricto in Europe, *M. oceanensis* in Australia and *M. fuliginosus* in Asia. However, Tian *et al.* (2004) included specimens from Japan and China only to represent "Asia". Therefore, the name *M. schreibersii* is retained in this checklist following Kingston *et al.* (2006) and Francis (2008) pending further research.

IUCN status: Near Threatened

Recorded at: Pahang: Fraser Hill (Medway, 1969), Krau Wildlife Reserve (Kingston *et al.*, 2006); **Perlis**: Kaki Bukit (Medway, 1969); **Perak**: Maxwell Hill (Medway, 1969), Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013); **Selangor**: Ulu Gombak (Heller & Volleth, 1989), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Melaka**: Sungai Udang Forest Reserve (Shahfiz *et al.*, 2009).

M. schreibersii has been recorded in primary hill and montane forests (Kingston *et al.*, 2006). The species roosts in caves in large colonies, sometimes with other *Miniopterus* bats (Medway, 1969; Francis, 2008).

4.1.10 Family: Vespertilionidae (Subfamily: Murininae)

4.1.10.1 *Harpiocephalus harpia* [Temminck, 1840]

Vespertilio harpia Temminck, 1840: 219, pls. 55; Southeast side of Mountain Gede, Java, INDONESIA (Collector unknown; Type unknown).

Harpiocephalus mordax Thomas, 1923c:88; Mogok, upper Burma=MYANMAR (Herbert Hampton, collector; BM(NH) 4.4.27.1)

Harpiocephalus harpia Dobson, 1876.

Harpiocephalus harpia mordax Ellerman & Morrison-Scott, 1966.

Common English name: Hairy-winged Bat

Barcode Index Number: DNA barcodes recorded as *H. harpia* are associated with the BIN, BOLD:AAB5424, but there are no DNA barcodes from Peninsular Malaysia.

Remarks: *H. mordax* was once considered a subspecies of *H. harpia* (Ellerman & Morrison-Scott, 1966) but was later recognised as a distinct species by having "a more robust skull and larger teeth" compared to *H. harpia* (Hill & Fancis, 1984). Corbet and Hill (1992) re-examined two male specimens and three female specimens from Java recorded as *H. harpia* and commented that the degree of dimorphism observed among the specimens is small when compared to the differences observed in rostral and tooth size between *H. harpia* and *H. mordax*. Matveev (2005) noted that all specimens of *H. mordax* used in earlier studies (including the type specimens) are female and added that a molecular analysis (Inter-SINE-PCR) of a male "*harpia*" and a female "*mordax*" from Cambodia indicated that the specimens were conspecific, consequently eliminating the occurrence of *H. mordax* in Cambodia. Two female specimens from Peninsular Malaysia (field ID.: CMF930806.7 and CMF930807.2) were initially identified as "*H. mordax*" based on their broader skull and large teeth by Francis (1995) who later stated that *H. harpia* is the only species that occurs in Southeast Asia with sexual dimorphism in size

(Francis, 2008). Following the current consensus, all records of *H. mordax* from Peninsular Malaysia should be updated to *H. harpia.*

IUCN status: Least Concern

Recorded at: Pahang: Fraser Hill Forest Reserve (Mohd-Hanif *et al.*, 2015).

Previously recorded as *H. mordax* at: **Perak**: Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013); **Pahang**: National Park (Yeap & Zubaid, 2002), Krau Wildlife Reserve (Kingston *et al.*, 2006).

The roosting ecology of *H. harpia* remains unknown due to its rarity but the species has been recorded in forests with hilly terrains (Kingston *et al.*, 2006). Two specimens were caught above a small manmade pond which is surrounded by secondary vegetation in the vicinity of pineapple plantation (Matveev, 2005).

4.1.10.2 *Murina aenea* Hill, 1964

Murina aenea Hill, 1964: 57, pls 54, 55; Ulu Chemperoh, near Janda Baik, Bentong District, Pahang, MALAYSIA, c. 3°18'N, 101°50'E, 2000 ft (Collector unknown; BM(NH) 1964.770).

Common English name: Bronzed Tube-nosed Bat

Barcode Index Number: BOLD:AAE6670 (2 DNA barcodes from Peninsular Malaysia; Figure 4.5)

IUCN status: Vulnerable

Recorded at: Pahang: Ulu Chemperoh (Hill, 1964), Bentong (Medway, 1969), Krau Wildlife Reserve (Kingston *et al.*, 2006), Bukit Ibam and Klau Besar (Lim *et al.*, 2014); **Selangor**: Ulu Gombak (Hill and Francis, 1984); **Perak**: Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013); **Terengganu**: Pasir Raja, Dungun (Wilson *et al.*, 2014); **Johor**: Gunung Panti (Lim *et al.*, 2014); **Kedah:** Bukit Hijau (Lim *et al.*, 2014).

M. aenea has been recorded in lowland and hill dipterocarp forests but its roosting ecology remains unknown (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.10.3 *Murina peninsularis* Hill, 1964

Murina cyclotis peninsularis Hill, 1964: 55; Ulu Chemperoh, near Janda Baik, Bentong District, Pahang, MALAYSIA (Collector unknown; BM(NH) 1964.771).

Murina peninsularis Soisook *et al.*, 2013.

Common English name: Peninsular Tube-nosed Bat

Barcode Index Number: BOLD:AAA6349 (2 DNA barcodes from Peninsular Malaysia; Figure 4.5)

Remarks: Three subspecies were previously described under *M. cyclotis* based on their geographical distributions: *M. c. cyclotis* from northeast India to Vietnam, the slightly darker and duller *M. c. eileenae* from Sri Lanka, and *M. c. peninsularis* from Peninsular Thailand to Malaysia and Indonesia (Corbet & Hill, 1992; Soisook *et al.*, 2013). However, the consistent medium-large body size and divergence in *COI* mtDNA supported the recognition of *M. c. peninsularis* as a distinct species (Francis *et al.*, 2010; Francis & Eger, 2012; Soisook *et al.*, 2013). Therefore, all records of *M. cyclotis* from Peninsular Malaysia should be updated to *M. peninsularis* following Soisook *et al.* (2013).

IUCN status: Not Evaluated but Least Concern as *M. cyclotis*.

Recorded at: Pahang: Ulu Chemperoh, near Janda Baik (Hill, 1964); **Perlis**: Wang Kelian State Park (Lim *et al.*, 1999); **Perlis**: Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Kelantan:** Lojing Highlands (Jayaraj *et al.*, 2016b).

As *M. cyclotis* at: **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990), Gunung Angsi Forest Reserve and Berembun Forest Reserve (Joann *et al.*, 2013); **Perak**: Temengor Forest Reserve (Francis, 1995; Joann *et al.*, 2011), Royal Belum State Park (Shahfiz *et al.*, 2008b), Kledang Saiong Forest Reserve (Joann *et al.*, 2013; Lim *et al.*, 2014); **Pahang**: Krau Wildlife Reserve (Anan *et al.*, 1998; Kingston *et al.*, 2006), Cameron Highland (Shahfiz *et al.*, 2008a), Kuala Atok-National Park (Tingga *et al.*, 2012), Bukit Ibam, Klau Besar and Kenong (Lim *et al.*, 2014); **Selangor**: Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999); **Johor**: Labis Forest Reserve (Lim *et al.*, 2014).

M. peninsularis has been recorded in wide variety of forest types (Francis, 2008) but its roosting ecology remains unknown (Kingston *et al.*, 2006).

4.1.10.4 *Murina huttoni* [Peters, 1872]

Harpyiocephalus huttonii Peters, 1872: 257; Dehra Dun, Kumaon, northwest INDIA (Collector unknown; BM(NH) 1879.11.21.685).

Murina huttoni Medway, 1969.

Common English name: Hutton's Tube-nosed Bat

Barcode Index Number: DNA barcodes of *M. huttoni* are associated with three BINs, BOLD:AAC6107, BOLD:AAC6108 and BOLD:AAC6109, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Remarks: Francis and Eger (2012) commented that *M. huttoni* may be the only *Murina* species that occurs in both Peninsular Malaysia and Indo-Burma after *M. peninsularis* was separated from *M. cyclotis*. The large divergence among the DNA barcodes in NJ tree suggested that *M. huttoni* is also a species complex (Appendix I). Simmons (2005) recognised two subspecies: *M. h. huttoni.* (type locality: India) and *M. h rubella* (type locality: Fokien, China). Whether the *M. huttoni* occurring in Peninsular Malaysia represents either of these subspecies remains to be determined (Francis & Eger, 2012).

Recorded at: Pahang: The only specimens of *M. huttoni* from Peninsular Malaysia were caught at Gunong Benom, Krau Wildlife Reserve at elevation of 1400 m (Medway, 1969).

4.1.10.5 *Murina rozendaali* Hill & Francis, 1984

Murina rozendaali Hill & Francis, 1984: 319; Gomantong, Sabah, Borneo, MALAYSIA 5°31’N, 118°4’E (Charles M. Francis, collector; BM(NH) 1983.360).

Common English name: Rozendaal’s Tube-nosed Bat

Barcode Index Number: DNA barcodes of *M. rozendaali* are associated with BIN, BOLD:AAK8797 but there are no DNA barcodes from Peninsular Malaysia.

Remarks: Specimens from Peninsular Malaysia are smaller than the specimens from Sabah (i.e., weight and forearm length), likely due to ecological factors (Francis, 1997).

IUCN status: Vulnerable

Recorded at: Pahang: Krau Wildlife Reserve (Francis, 1997; Kingston *et al.*, 2006; Khan *et al.*, 2008); **Negeri Sembilan**: Pasoh (Khan *et al.*, 2008); **Perak**: Temengor Forest Reserve (Joann *et al.*, 2011), Kledang Saiong Forest Reserve (Joann *et al.*, 2013); **Selangor**: Semangkok Forest Reserve and Ulu Gombak (Joann *et al.*, 2013); **Terengganu**: Gunung Tebu Forest Reserve (Joann *et al.*, 2013).

All specimens of *M. rozendaali* from Peninsular Malaysia were collected in primary forests (Kingston *et al.*, 2006) though the species has also been recorded in disturbed lowland forest in other regions (Francis, 2008).

4.1.10.6 *Murina suilla* [Temminck, 1840]

Vespertilio suillus Temminck, 1840: 224, pl. 56; Tapos, Java, INDONESIA (Collector unknown; Type unknown).

Murina suilla Gray, 1842.

Common English name: Lesser Tube-nosed Bat

Barcode Index Number: DNA barcodes recorded as *M. suilla* are associated with four BINs, BOLD:AAE0000, BOLD:AAE0001, BOLD:AAE0003 and BOLD:ABX8091 but there are no DNA barcodes from Peninsular Malaysia.

Remarks: Simmons (2005) recognises two subspecies: *M. s. suilla* (type locality: Java) and *M. s. canescens* (type locality: west Sumatra). Whether the two clusters suggested by NJ analysis (Appendix J) represent the two subspecies remains to be determined.

IUCN status: Least Concern

Recorded at: Pahang: Bentong (Medway, 1969), Krau Wildlife Reserve (Anan *et al.*, 1998; Kingston *et al.*, 2006), Cameron Highland (Shahfiz *et al.*, 2008a), Kuala Atok, National Park (Tingga *et al.*, 2012), Bukit Ibam, Kemasul, Jengka and Klau Besar (Lim *et al.*, 2014), Tasik Bera Forest Reserve (Mohd-Hanif *et al.*, 2015); **Selangor**: Ulu Gombak (Heller & Volleth, 1984; 1989), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999); **Perak**: Temengor Forest Reserve (Francis, 1995; Tamrin *et al.*, 2010; Shahfiz *et al.*, 2013), Royal Belum State Park (Shahfiz *et al.*, 2008b), Kledang Saiong Forest Reserve (Joann *et al.*, 2013); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999), Bukit Hijau (Lim *et al.*, 2014); **Melaka**: Unspecified (Shahfiz *et al.*, 2009); **Kelantan**: Air Panas-Gua Musang (Hasan *et al.*, 2012), Gunung Stong State Park (Lim *et al.*, 2014); **Perlis:** Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Johor**: Gunung Panti (Lim *et al.*, 2014); **Negeri Sembilan**: Gunung Angsi Forest Reserve (Lim *et al.*, 2014).

M. suilla has been recorded in lowland and hill forests (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.11 Family: Vespertilionidae (Subfamily: Vespertilioninae)

Arielulus circumdatus* and *A. societatis

Heller and Volleth (1984) reported that *Pipistrellus circumdatus* has a different structure of baculum and unique karyotypic characters (diploid number of chromosomes=50, fundamental number=48) compared to other *Pipistrellus* species which have a diploid number of chromosomes ranging from 26 to 44 and fundamental number=50; they considered *P. circumdatus* to be conspecific with *P. societatis* and transferred the taxon to the genus *Eptesicus.* However, Hill and Francis (1984) retained *circumdatus* and *societatis* under *Pipistrellus* as two distinct species on the basis of *societatis* having "shorter palate, bony post-palate and toothrows". Hill and Harrison (1987) later examined the os penis of all genera in Vepertilioninae and consequently established the subgenus *Arielulus* under *Pipistrellus.* Csorba and Lee (1999) concluded

that *Arielulus* is distinct from *Pipistrellus* based on the former's distinctive coloration, short and wide rostrum, high and globular braincase, tricuspid upper incisor (I^1), greatly reduced inner upper incisor (I^2), small (often missing) first upper premolar (PM^2), myotodont first and second lower molars (M_1 and M_2), very small Y-shaped baculum and the diploid number of chromosomes=50, and consequently raised *Arielulus* as a genus.

4.1.11.1 *Arielulus circumdatus* [Temminck, 1840]

Vespertilio circumdatus Temminck, 1840: 214; Tapos, Java, INDONESIA (Collector unknown; Type unknown).

Arielulus circumdatus Simmons, 2005.

Common English name: Black Gilded Pipistrelle

Barcode Index Number: DNA barcodes recorded as *A. circumdatus* are associated with a BIN, BOLD:AAD8838 but there are no DNA barcodes from Peninsular Malaysia.

Remarks: Sing *et al.* (2013) listed the species as *Eptesicus circumdatus* based on the nomenclature used by Heller and Volleth (1984) (see remarks on the genus *Arielulus*).

IUCN status: Least Concern

Recorded at: Selangor: Ulu Gombak (Heller & Volleth, 1984), **Pahang**: Unspecified (Csorba & Lee, 1999). *A. circumdatus* has been recorded in hill forest at an elevation of 1300 – 2000 m (Francis, 2008).

4.1.11.2 *Arielulus societatis* [Hill, 1972]

Pipistrellus societatis Hill, 1972: 34; Base Camp, Gunong Benom, Pahang, MALAYSIA, 3°51'N, 102°11'E, 800ft (Boo-Liat Lim and Hoi-Sen Yong, collector; BM(NH) 1967.1605).

Arielulus societatis Simmons, 2005.

Common English name: Benom Gilded Pipistrelle

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

Remarks: *A. societatis* resembles *A. circumdatus* closely but the former has shorter forearms, post-palatal extension, toothrows and rostrum (Hill, 1972). Heller and Volleth (1984) considered *A. societatis* and *A. circumdatus* to be conspecific with the former being the lowland subspecies of the latter but this was refuted by Hill and Francis (1984) on the basis of morphological characteristics (see remarks on the genus *Arielulus*). Simmons (2005) recognised *A. societatis* and *A. circumdatus* as two distinct species.

IUCN status: Vulnerable

Recorded at: Pahang: Gunong Benom (Hill, 1972); Fraser Hill Forest Reserve (Mohd-Hanif *et al.*, 2015); **Selangor**: Ulu Gombak (Heller & Volleth, 1984).

A. societatis has been recorded in primary lowland and hill forests, and secondary forests, and found roosting in a hole of a tree trunk beside a forest stream (Kingston *et al.*, 2006; Francis, 2008).

4.1.11.3 *Glischropus tylopus* [Dobson, 1875]

Vesperugo tylopus Dobson, 1875: 473; Sabah, north Borneo, MALAYSIA (Collector unknown; BM(NH) 70.2.10.2).

Glischropus tylopus Dobson, 1876.

Common English name: Thick-thumbed Pipistrelle

Barcode Index Number: Two DNA barcodes, RONP009-14 and RONP024-14 are from Perak, Peninsular Malaysia but are not associated with any BINs due to their short sequence length (<500 bp). Other DNA barcodes recorded as *G. tylopus,* which were collected in Vietnam and Laos, are associated with the BIN, BOLD:AAC0085.

Remarks: All DNA barcodes in the BIN, BOLD:AAC0085 which are from Vietnam and Laos (=Indochina) represent *G. bucephalus* which was recently described on the basis of longer forearm length and distinctive cranial features (Csorba, 2011). *G. aquilus* which was recently described from Sumatra, Indonesia is distinct from *G. tylopus* collected in Peninsular Malaysia on the basis of its darker colour and 12.4% divergence in cytochrome

b mtDNA (Csorba *et al.*, 2015). It is likely that the taxon occurs in Peninsular Malaysia represents *G. tylopus* sensu stricto based on the comparison of specimens from Peninsular Malaysia and Sabah=type locality (see Figure 6 in Csorba *et al.*, 2015). NJ analysis was not performed for *G. tylopus* as the DNA barcodes from Peninsular Malaysia (RONP009-14 and RONP024-14) are too short for comparison with other barcodes.

IUCN status: Least Concern

Recorded at: Selangor: Ulu Gombak (Heller & Volleth, 1984; 1989), Bukit Lanjan (Ratnam *et al.*, 1989), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999), Semangkok Forest Reserve (Joann *et al.*, 2013); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Pahang**: Tasik Chini (Lim & Ratnam, 1999), Krau Wildlife Reserve (Kingston *et al.*, 2006), Tasik Bera Forest Reserve (Mohd-Hanif *et al.*, 2015); **Perak**: Temengor Forest Reserve (Joann *et al.*, 2011); **Kelantan**: Air Panas-Gua Musang (Hasan *et al.*, 2012), Gua Musang (Jayaraj *et al.*, 2016b).

G. tylopus inhabits lowland forests but has been recorded in hill forests (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008). Individuals have been found roosting in small groups in internodes of dead and broken bamboo, sometimes in rock crevices and banana leaves, occasionally with *Tylonycteris* species (Kingston *et al.*, 2006; Francis, 2008).

4.1.11.4 *Nyctalus noctula* [Schreber, 1774] (?)

Vespertilios noctula Schreber, 1774: 166, pl. 52; FRANCE (Collector unknown; Type unknown).

Nyctalus noctula Chasen, 1940.

Common English name: Eurasian Noctule

Barcode Index Number: DNA barcodes recorded as *N. noctula* are associated with BIN, BOLD:AAC7411, but there are no DNA barcodes from Peninsular Malaysia.

Remarks: Chasen (1940) suggested that the taxon in Peninsular Malaysia may represents *N. n. labiate=labiatus* Hodgson, 1835 (type locality = Nepal) which also occurs in

Pakistan and India (Corbet & Hill, 1992). Csorba and Hutson (2016) considered *N. n. labiatus* to be morphologically distinct from *N. noctula* and suggested that the former to be treated as a distinct full species. They also added that the records of *N. noctula* from Peninsular Malaysia, if valid, may be referable to *"labiatus"* or *"plancyi"*.

IUCN status: Least Concern

Recorded at: The first record of *N. noctula* in this region is an old skin dated 1838 which was purchased in Singapore (Dobson, 1878) but its origin remains doubtful (Medway, 1969). Based on the purchased skin, Dobson (1878) included Peninsular Malaysia in the distribution range of *N. noctula*, which was followed by Corbet and Hill (1992) and Medway (1969). However, there are no recent records for the species although it is thought to occur in Peninsular Malaysia (Davison & Zubaid, 2007; DWNP, 2010). There are two old records which reported two specimens identified as *Nyctalus* sp. in Ulu Langat Forest Reserve, **Selangor** (Yatim, 1983) and National Park, **Pahang** (Yatim *et al.*, 1985). However, there are no specimens deposited in the DWNP collection.

N. noctula roosts in tree hollows and forages high above canopy (Medway, 1969; Francis, 2008).

4.1.11.5 *Philetor brachypterus* [Temmick, 1840]

Vespertilio brachypterus Temmick, 1840: 215, pl. 53; Padang district, Sumatra, INDONESIA (Collector unknown; Type unknown).

Philetor brachypterus Medway, 1969.

Common English name: Narrow-winged Brown Bat

Barcode Index Number: BOLD:AAF6860 (1 DNA barcode from Peninsular Malaysia; Figure 4.5)

Remarks: DNA barcodes recorded as *P. brachypterus* are associated with two BINs, BOLD: AAF6860 and BOLD:AAF6859. Hill and Francis (1984) noted that specimens from Borneo and Peninsular Malaysia are similar in size but Corbet and Hill (1992) later

commented that size variation occurs within the species. Based on NJ analysis, the DNA barcode from Peninsular Malaysia (BM434-04) may represents a cryptic species (Figure 4.5) but the name *P. brachypterus* is used in this checklist pending further research.

IUCN status: Least Concern

Recorded at: Perak Unspecified (Medway, 1969); **Selangor**: Unspecified (Medway, 1969), Ulu Gombak (Heller & Volleth, 1984; 1989); **Johor**: Endau-Rompin National Park (BM434-04 was collected in year 2001, Francis *et al.*, 2010).

P. brachypterus has been recorded in primary and secondary forests, and found roosting in tree hollows (Francis, 2008).

4.1.11.6 *Pipistrellus javanicus* [Gray, 1838]

Scotophilus javanicus Gray, 1838: 498; Java, INDONESIA (Collector unknown; Type unknown).

Pipistrellus javanicus Chasen, 1940.

Common English name: Javan Pipistrelle

Barcode Index Number: DNA barcodes recorded as *P. javanicus* are associated with two BINs, BOLD:AAC3383 and BOLD:AAL5777, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Recorded at: Perak and **Pulau Pinang:** Unspecified (Medway, 1969); **Pahang**: Gunung Benom, (Hill, 1972), Krau Wildlife Reserve (Zubaid, 1993); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Selangor**: Air Hitam Forest Reserve (Azlan *et al.*, 2000).

P. javanicus has been recorded in wide variety of habitats (i.e., mangroves, lowland and hill forests, towns and rubber plantations) and found roosting in treeferns, fallen logs and caves (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.11.7 *Pipistrellus stenopterus* [Dobson, 1875]

Vesperugo stenopterus Dobson, 1875: 470; Sarawak, Borneo, MALAYSIA (Collector unknown; Type unknown).

Pipistrellus stenopterus Medway, 1966.

Common English name: Narrow-winged Pipistrelle

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

IUCN status: Least Concern

Recorded at: Selangor: Ulu Gombak (Medway, 1966); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Pahang**: Krau Wildlife Reserve (Kingston *et al.*, 2006; Khan *et al.*, 2008).

P. stenopterus has been recorded foraging in open areas and over rivers in forest and rubber plantations, and has been reported roosting in tree hollows and under house roofs with *Scotophilus kuhlii* (Kingston *et al.*, 2006; Francis, 2008).

4.1.11.8 *Pipistrellus tenuis* [Temminck, 1840]

Vespertilio tenuis Temminck, 1840: 229; Sumatra, INDONESIA (Collector unknown; Type unknown).

Pipistrellus tenuis Chasen, 1940.

Common English name: Least Pipistrelle

Barcode Index Number: DNA barcodes recorded as *P. tenuis* are associated with the BIN, BOLD:AAB2554, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Recorded at: Pahang and **Pulau Pinang:** Unspecified (Medway, 1969); **Selangor**: Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Melaka**: Sungai Udang Forest Reserve (Shahfiz *et al.*, 2009).

P. tenuis has been reported roosting in buildings in highly disturbed areas and in hollowed branches and among dead leaves in forests (Francis, 2008).

4.1.11.9 *Hesperoptenus blanfordi* [Dobson, 1877]

Vesperugo blanfordi Dobson, 1877: 312; Tenasserim, east of Moulmein, south Burma=MYANMAR (Limborg, collector; Type unknown).

Hesperoptenus blanfordi Chasen, 1940.

Common English name: Least False-serotine

Barcode Index Number: DNA barcodes recorded as *H. blanfordi* are associated with the BIN, BOLD:AAD5793, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Recorded at: Pahang: Jengka in Temerloh (Hill, 1972), Krau Wildlife Reserve (Zubaid, 1993; Kingston *et al.*, 2006); **Selangor**: Ulu Gombak (Heller & Volleth, 1984; 1989).

H. blanfordi has been reported foraging in open areas, in gaps created by fallen trees, and above rivers, and found roosting at the entrances of limestone caves in small colonies (Kingston *et al.*, 2006; Francis, 2008).

4.1.11.10 *Hesperoptenus doriae* [Peters, 1868]

Vesperus (H.) doriae Peters, 1868: 626; Sarawak, Borneo, MALAYSIA (Collector unknown; Type unknown).

Hesperoptenus doriae Peters, 1868.

Common English name: Doria's False-serotine

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

IUCN status: Data Deficient

Recorded at: Selangor: Air Hitam Forest Reserve (Ratnam *et al.*, 1989), Ulu Gombak (Heller & Volleth, 1989).

H. doriae has been reported roosting in a small colony of eight to ten individuals at overhanging rocks near a stream (Ratnam *et al.*, 1989) and in leaves of palm trees (Francis, 2008).

4.1.11.11 *Hesperoptenus tomesi* Thomas, 1905

Hesperoptenus tomesi Thomas, 1905: 575; Malacca=Melaka, MALAYSIA (Collector unknown; Originally No. 190A in the collection of Mr. R. F. Tomes but currently as BM(NH) 1907.1.1.428).

Common English name: Tome's False-serotine

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

IUCN status: Vulnerable

Recorded at: Melaka: Unspecified (Thomas, 1905); **Selangor**: Ulu Gombak (Heller & Volleth, 1984; 1989).

H. tomesi has been recorded in mature lowland forests (Francis, 2008).

4.1.11.12 *Hypsugo macrotis* [Temminck, 1840]

Vespertilio macrotis Temminck, 1840: 218, pl. 54; Padang, Sumatra, INDONESIA (Collector unknown; Type unknown).

Pipistrellus imbricatus Medway, 1969.

Pipistrellus macrotis Francis & Hill, 1986.

Hypsugo macrotis Simmons, 2005.

Common English name: Big-eared Pipistrelle

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

Remarks: *H. macrotis* was previously known as *Pipistrellus macrotis* (Simmons, 2005) and was first reported from Peninsular Malaysia as *Pipistrellus imbricatus macrotis* (Chasen, 1940). Francis and Hill (1986) later concluded that specimens recorded as *P. imbricatus macrotis* from Peninsular Malaysia represent *P. macrotis=H. macrotis.*

IUCN status: Data Deficient

Recorded at: Selangor: Kuala Selangor (Hill & Francis, 1984); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Negeri Sembilan:** Seremban (Lim *et al.*, 2016).

As *P. imbricatus macrotis*: **Selangor**: Unspecified lowland forest (Medway, 1969).

H. macrotis has been recorded in lowland forests and coastal lagoons near mangroves (Medway, 1969; Francis, 2008) and recently at a school located in an urbanised habitat with small secondary forest fragments, suggesting that the species has adapted to human modified habitats (Lim *et al.*, 2016). The lack of recent records may be due to sampling bias (i.e., surveys primarily targeting forested habitats).

4.1.11.13 *Scotophilus kuhlii* Leach, 1821

Scotophilus kuhlii Leach, 1821: 72; INDIA (Collector unknown; Type unknown).

Scotophilus temminckii Medway, 1969.

Scotophilus kuhlii temminckii Corbet & Hill, 1992.

Common English name: Lesser Asian House Bat

Barcode Index Number: DNA barcodes recorded as *S. kulii* are associated with the BIN, BOLD:AAC0094, but there are no DNA barcodes from Peninsular Malaysia.

Remarks: Medway (1969) reported *S. temminckii* from Peninsular Malaysia but Corbet and Hill (1992) considered *S. temminckii* a synonym of *S. kuhlii*.

IUCN status: Least Concern

Recorded at: Selangor: Bukit Kemandul (Ratnam *et al.*, 1989), Ulu Gombak (Heller & Volleth, 1989), Bangi Forest Reserve (Zubaid, 1993), Air Hitam Forest Reserve (Azlan *et al.*, 2000); **Pahang**: Krau Wildlife Reserve (Kingston *et al.*, 2006); **Melaka**: Sungai Udang Forest Reserve (Shahfiz *et al.*, 2009); **Perak**: Selama (Shafie *et al.*, 2011); **Kelantan:** Gunung Reng (Jayaraj *et al.*, 2016b).

S. kuhlii is associated with humans, often sighted hunting insects at lamp posts in urban areas (Kingston *et al.*, 2006) and roosts in large colonies under roofs of buildings, under the fronds of palms, in hollowed dead trees in forests, and in hollowed old rubber trees in rubber plantations (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.11.14 *Tylonycteris pachypus* [Temminck, 1840]

Vespertilio pachypus Temminck, 1840: 217; Bantam, west Java, INDONESIA (Collector unknown; Type unknown).

Tylonycteris pachypus Chasen, 1940.

Common English name: Lesser Bamboo Bat

Barcode Index Number: DNA barcodes of *T. pachypus* are associated with two BINs, BOLD:AAC1209 and BOLD:AAC1210, but none of them are from Peninsular Malaysia.

Remarks: The BIN, BOLD:AAC1210 contains DNA barcodes recorded as *T. pachypus* and a DNA barcode recorded as *T. robustula* (ABBSI217-10) (Appendix K). It is likely that the DNA barcode, ABBSI217-10 is a case of mis-identification as *T. pachypus* and *T. robustula* are distinct in body size and coloration. Simmons (2005) recognised five subspecies: *T. p. pachypus* (type locality: Java, Indonesia), *T. p. aurex* (type locality: India), *T. p. fulvidus* (type locality: Burma=Myanmar), *T. p. meyeri* (type locality: Philippines), and *T. p. bhakti* (type locality: Lombok Island, Indonesia).

IUCN status: Least Concern

Recorded at: Selangor: Ulu Gombak (Medway, 1966; Heller & Volleth, 1984; 1989), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999), Air Hitam Forest Reserve (Azlan *et al.*, 2000); **Pahang**: Krau Wildlife Reserve (Zubaid, 1993; Kingston *et al.*, 2006), Tasik Bera Forest Reserve (Mohd-Hanif *et al.*, 2015); **Perak**: Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013), Royal Belum State Park (Shahfiz *et al.*, 2008b); **Kelantan**: Air Panas-Gua Musang (Hasan *et al.*, 2012), Gunung Stong State Park (Jayaraj *et al.*, 2012a), Gunung Reng and Gua Musang (Jayaraj *et al.*, 2016b); **Johor**: Labis Forest Reserve (Lim *et al.*, 2014).

T. pachypus roosts in small colonies in internodes of live standing bamboo stems by entering through slits created by stem-boring beetle larvae (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.11.15 *Tylonycteris robustula* Thomas, 1915

Tylonycteris robustula Thomas, 1915: 227; Upper Sarawak, Borneo, MALAYSIA (Cecil J. Brooks, collector; BM(NH) 1911.1.18.8).

Tylonycteris malayana Chasen, 1940: 52; Jor, Batang Padang Dist., Perak, MALAYSIA (Frederick N. Chasen, collector; BM(NH) 47.1433).

Tylonycteris robustula malayana Simmons, 2005.

Common English name: Greater Bamboo Bat

Barcode Index Number: DNA barcodes recorded as *T. robustula* are associated with three BINs, BOLD:AAB3205, BOLD:AAB3206 and BOLD:AAC1210, but there are no DNA barcodes from Peninsular Malaysia (Appendix K).

IUCN status: Least Concern

Remarks: The BIN, BOLD:AAC1210 comprises seven DNA barcodes of *T. pachypus* and a DNA barcode of *T. robustula* (ABBSI217-10) which is likely to be mis-identified (see remarks on *T. pachypus*). Simmons (2005) recognised two subspecies: *T. r. robustula* (type locality: Borneo) and *T. r. malayana* (type locality: Peninsular Malaysia).

Recorded at: Selangor: Ulu Gombak (Medway, 1966; 1969; Heller & Volleth, 1984; 1989), Bukit Lanjan (Ratnam *et al.*, 1989), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999), Air Hitam Forest Reserve (Azlan *et al.*, 2000), Semangkok Forest Reserve (Joann *et al.*, 2013); **Pahang**: Krau Wildlife Reserve (Zubaid, 1993; Kingston *et al.*, 2006), Tasik Chini (Lim & Ratnam, 1999), Tasik Bera Forest Reserve and Fraser Hill Forest Reserve (Mohd-Hanif *et al.*, 2015); **Perak**: Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013), Royal Belum State Park (Shahfiz *et al.*, 2008b); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Kelantan**: Gunung Stong State Park, (Jayaraj *et al.*, 2012a); Gunung Reng (Jayaraj *et al.*, 2016b).

T. robustula roosts in internodes of large dead bamboo stems by entering through slits made by chrysomelid beetles and has been reported roosting in small harem groups,

with one adult male and up to six females in one group (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008). Solitary males have also been reported (Medway, 1969).

4.1.12 Family: Vespertilionidae (Subfamily: Myotinae)

4.1.12.1 *Myotis adversus* [Horsfield, 1824] (?)

Vespertilio adversus Horsfield, 1824: part 8; Java, INDONESIA (Collector unknown; Type unknown).

Myotis adversus Chasen, 1940.

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

IUCN status: Least Concern

Recorded at: Perak: Unspecified (Medway, 1969).

4.1.12.2 *Myotis ater* [Peters, 1866b]

Vespertilio ater Peters, 1866b: 18; Ternate Island, Moluccas, INDONESIA (Collector tunknown; Type unknown).

Myotis ater Corbet & Hill, 1992.

Common English name: Peters's Myotis

Barcode Index Number: A DNA barcode recorded as *M.* cf. *ater* (BM487-04) is from Peninsular Malaysia but is not associated with any BINs due to its short sequence length (<500 bp). Other DNA barcodes recorded as *M. ater* are from Vietnam and are associated with a BIN, BOLD:AAA8748 which also contains DNA barcodes recorded as *M. muricola* (Figure 4.5)

Remarks: Hill (1962) considered *M. ater* and *M. muricola* to be conspecific but Francis and Hill (1998) recognised them as two distinct species which occur in sympatry in Malaysia and added that *M. ater* from Peninsular Malaysia are larger than *M. ater* from elsewhere. As *M. muricola* is putatively a species complex (see remarks on *M. muricola*), the relationship between *M. ater* and *M. muricola* remains to be determined.

IUCN status: Least Concern

Recorded at: Kedah: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Pahang**: Krau Wildlife Reserve (Kingston *et al.*, 2006), Cameron Highland (Shahfiz *et al.*, 2008a); **Perak**: Bukit Jerneh Cave and Tumang Lembing Cave (Douangboubpha *et al.*, 2010b).

M. ater has been reported roosting in caves, either solitarily or in small colonies and recorded foraging in open areas such as gaps created by fallen trees, midstorey openings and forest edge (Kingston *et al.*, 2006; Francis, 2008).

4.1.12.3 *Myotis federatus* Thomas, 1916a

Myotis peytoni federatus Thomas, 1916a: 3; Semangko Paas, MALAYSIA, 2700 ft (Collector unknown; BM(NH) 1916.4.20.5).

Myotis montivagus federatus Corbet & Hill, 1992.

Myotis federatus Görföl *et al.*, 2013.

Barcode Index Number: There are no DNA barcodes with this name on BOLD. DNA barcodes recorded as *M. montivagus* are associated with two BINs, BOLD:AAC5917 and BOLD:AAU0309, but there are no DNA barcodes from Peninsular Malaysia.

Remarks: *M. federatus* was previousy considered as a subspecies of *M. montivagus* (Corbet & Hill, 1992) on the basis of dental characteristics (Hill, 1962). However, Görföl *et al.* (2013) noted that *M. federatus* has smaller forearms, a larger skull and smaller middle upper premolars (P3), and added that the taxa have distinct geographical ranges: *M. federatus* is confined to Peninsular Malaysia whereas *M. montivagus* is distributed from south China to northern Myanmar. Previous records of *M. montivagus* from Peninsular Malaysia (Davison & Zubaid, 2007; Joann *et al.*, 2011; Sing *et al.*, 2013; Jayaraj *et al.*, 2013a) should be updated to *M. federatus* following Görföl *et al.* (2013).

IUCN status: Not Evaluated but Least Concern as *M. montivagus.*

Recorded at: As *M. montivagus*: **Selangor**: Genting Semangkok (Medway, 1969), Ulu Gombak (Heller & Volleth, 1989), Batu Caves (HNHM 98.14.31); **Pahang**: Genting

Highland (Heller & Volleth, 1989); **Perak**: Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013); **Perlis**: Wang Kelian State Park (Jayaraj *et al.*, 2013a).

M. federatus has been recorded in primary and secondary forests with elevations up to 1,000 m (Francis, 2008).

4.1.12.4 *Myotis hasseltii* [Temminck, 1840]

Vespertilio hasseltii Temminck, 1840: 225; Bantam, Java, INDONESIA (Collector unknown; Type unknown).

Myotis hasseltii Chasen, 1940.

Common English name: Hasselt's Myotis

Barcode Index Number: DNA barcodes recorded as *M. hasseltii* are associated with the BIN, BOLD:AAC1504, but there are no DNA barcodes from Peninsular Malaysia.

IUCN status: Least Concern

Recorded at: Selangor: Unspecified mangrove forest (Medway, 1969); **Kedah**: Kuah in Pulau Langkawi (Medway, 1969; Hill, 1972); **Perlis**: Kangar (Hill, 1972); **Perak**: Kuala Gula (Hill, 1972); **Pahang**: Krau Wildlife Reserve (Kingston *et al.*, 2006).

M. hasseltii has been found roosting in limestone caves and rock crevices, and reported foraging near coastal areas, mangroves and water bodies (e.g., rivers, lakes and seashores), presumed to skim small fishes and insects from water surface (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.12.5 *Myotis horsfieldii* [Temminck, 1840]

Vespertilio horsfieldii Temminck, 1840: 226; Mount Gede, Java, INDONESIA (Collector unknown; Type unknown).

Myotis horsfieldii Chasen, 1940.

Common English name: Horsfield’s Myotis

Barcode Index Number: BOLD:AAB9975 (1 DNA barcode from Peninsular Malaysia; Figure 4.5)

Remarks: DNA barcodes recorded as *M. horsfieldii* are associated with four BINs, BOLD:AAB9973, BOLD:AAB9974, BOLD:AAB9975, and BOLD:ACH4644 (Figure 4.5). Simmons (2005) recognised five subspecies: *M. h. horsfieldii* (type locality: Java), *M. h. dryas* (type locality: Andaman Islands), *M. h. peshwa* (type locality: India), *M. h. jeannei* (type locality: Philippines) and *M. h. deignani* (type locality: Thailand). The form occurring in Peninsular Malaysia represents the *M. h. horsfieldii* (Corbet & Hill, 1992).

IUCN status: Least Concern

Recorded at: Pulau Pinang: Pulau Pinang (Medway, 1969); **Kuala Lumpur**: Ampang (Medway, 1969; Hill, 1972); **Pahang**: Merapoh (Ratnam *et al.*, 1989), Krau Wildlife Reserve (Kingston *et al.*, 2006), Cameron Highland (Shahfiz *et al.*, 2008a); **Perak**: Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013); **Terengganu**: Bukit Dendong (Yeap, 2003); **Kelantan:** Gunung Reng (Jayaraj *et al.*, 2016b).

M. horsfieldii has been recorded roosting in limestone caves, in crevices of rocks and boulders, and foraging near forest streams, presumably to skim insects from water surface (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.12.6 *Myotis hermani* Thomas, 1923a

Myotis hermani Thomas, 1923a: 252; Sabang, northwest Sumatra, INDONESIA (G. Herman, collector; BM(NH) 1923.1.2.13)

Common English name: Herman's Myotis

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

IUCN status: Data Deficient

Recorded at: Perak: Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013).

M. hermani has been recorded in lowland forests (Francis, 2008).

4.1.12.7 *Myotis muricola* [Gray, 1846]

Vespertilio muricola Gray, 1846: 4; NEPAL (Brian Houghton Hodgson, collector; Type unknown).

Myotis muricola Chasen, 1940.

Common English name: Asian Whiskered Myotis

Barcode Index Number: DNA barcodes recorded as *M. muricola* are associated with two BINs, BOLD:AAA8747 and BOLD:AAA8748 but none of them are from Peninsular Malaysia. A DNA barcode recorded as *M.* cf. *muricola* (RONP037-14) is from Peninsular Malaysia but is not associated with any BIN due to its short sequence length (<500 bp).

Remarks: Francis and Hill (1998) commented that specimens of *M. muricola* from across Southeast Asia showed moderate morphological variation. Wiantoro *et al.* (2012) revealed that *M. muricola* is a species complex with two groups which are 31.5% divergent in cytochrome *b* mtDNA: (i) *M. muricola* Western (with DNA sequences from Krakatau, Bali, Lombok, Sumba, Sumbawa, Flores Lembata and Pantar) and (ii) *M. muricola* Eastern (with DNA sequences from Sumatra, Peninsular Malaysia, Phillipines and Asian mainland). Wiantoro *et al.* (2012) reported 9.5% divergence in cytochrome *b* mtDNA within *M. muricola* Eastern and 8% divergence within *M. muricola* Western, and further segregated *M. muricola* Western into two subgroups which are 7.2% divergent in cytochrome *b* mtDNA: (i) Sumatra-Asian subgroup (Sumatra, Peninsular Malaysia and Asian mainland) and (ii) Bornean subgroup (Sarawak, Sabah and Kalimantan).

M. muricola was previously considered a subspecies of *M. mystacinus* (Hill, 1962; Medway, 1969). Hill (1983) reviewed the taxonomy of *M. mystacinus* and concluded that the taxon occurring in Peninsular Malaysia represents *M. muricola* sensu stricto, which was followed by Corbet and Hill (1992) who excluded Malaysia from the distribution range of *M. mystacinus*. Wiantoro *et al.* (2012) reported that *M. mystacinus* is 17.1% and 26% divergent from *M. muricola* Western and *M. muricola* Eastern in cytochrome *b*

mtDNA. After the separation of *M. muricola* from *M. mystacinus,* the latter was thought to occur only in Europe until Bates *et al.* (2005) recorded the taxon in Myanmar. Further surveys are required to determine whether the records of *M. mystacinus* from Peninsular Malaysia represents *M. muricola* or whether both occur in sympatry in the region.

The BIN, BOLD:AAA8748 contains DNA barcodes recorded as *M. muricola* and *M. ater* (see remarks on *M. ater* and Figure 4.5). The two species were previously considered to be conspecific (Hill, 1962) but were later considered to be distinct based on variation in body size (Hill, 1983) which occur in sympatry in Malaysia (Francis & Hill, 1998). The DNA barcode recorded as *M.* cf. *muricola* (RONP037-14) from Peninsular Malaysia was excluded from the NJ analysis due to its short sequence length (<500 bp), but note that the DNA barcode did not cluster with DNA barcodes of *M. muricola* on a taxon ID tree generated in BOLD (hence "cf. *muricola*").

IUCN status: Least Concern

Recorded at: Negeri Sembilan: Pasoh Forest Reserve (Francis, 1990); **Selangor**: Ulu Gombak (Heller & Volleth, 1984; 1989), Bukit Kutu Wildlife Reserve (Lim *et al.*, 1999), Air Hitam Forest Reserve (Azlan *et al.*, 2000); **Perak**: Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013); **Pahang**: Krau Wildlife Reserve (Kingston *et al.*, 2006); **Kelantan**: Air Panas-Gua Musang (Hasan *et al.*, 2012), Gunung Stong State Park (Jayaraj *et al.*, 2012a); **Johor:** Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al.*, 2015).

As *M. mystacinus* at: **Selangor**: Batu Caves (Thomas, 1916b), Ulu Gombak (Medway, 1966); Air Hitam Forest Reserve (Ratnam *et al.*, 1989); **Pulau Pinang**: Unspecified (Medway, 1969); **Pahang**: Krau Wildlife Reserve (Zubaid, 1993), Tasik Chini (Lim & Ratnam, 1999).

M. muricola has been reported roosting in small colonies of up to ten individuals at vegetated cave entrances and in tightly rolled central leaves of banana plants, in both forested and agricultural areas (Medway, 1969; Kingston *et al.*, 2006; Francis, 2008).

4.1.12.8 *Myotis ridleyi* [Thomas, 1898b]

Pipistrellus ridleyi Thomas, 1898b: 361; Selangor, MALAYSIA (H. N. Ridley, collector; BM(NH) 1898.3.13.5).

Myotis ridleyi Hill, 1972.

Common English name: Ridley's Myotis

Barcode Index Number: There are no DNA barcodes with this name on BOLD.

IUCN status: Near Threatened

Recorded at: Pahang: Gunong Benom (Hill, 1972) in Krau Wildlife Reserve (Kingston *et al.*, 2006; Khan *et al.*, 2008); **Selangor**: Ulu Gombak (Heller & Volleth, 1989); **Negeri Sembilan**: Pasoh Forest Reserve (Francis, 1990), Gunung Angsi Forest Reserve (Lim *et al.*, 2014); **Perak**: Temengor Forest Reserve (Francis, 1995; Shahfiz *et al.*, 2013), Bukit Jerneh Cave and Tumang Lembing Cave (Douangboubpha *et al.*, 2010b), Kledang Saiong Forest Reserve (Joann *et al.*, 2013); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999); **Johor**: Gunung Panti (Lim *et al.*, 2014), Endau-Kluang Forest Reserve and Endau-Kota Tinggi Forest Reserve (Mohd-Hanif *et al.*, 2015).

M. ridleyi has been recorded only at understory of lowland forests, suggesting that the species is confined to forest interior. Individuals have been reported roosting in caves and under fallen logs and rocks (Kingston *et al.*, 2006; Francis, 2008).

4.1.12.9 *Myotis siligorensis* [Horsfield, 1855]

Vespertilio siligorensis Horsfield, 1855: 102; Siligori, NEPAL (Brian Houghton Hodgson, collector; Type unknown).

Myotis siligorensis Medway, 1969.

Common English name: Small-toothed Myotis

Barcode Index Number: DNA barcodes recorded as *M. siligorensis* are associated with five BINs, BOLD:AAA9718, BOLD:AAA9719, BOLD:AAA9720, BOLD:AAA9721, and BOLD:ACF1046, but there are no DNA barcodes from Peninsular Malaysia.

Remarks: NJ analysis suggested that *M. siligorensis* may be a species complex (Appendix L). Simmons (2005) recognised four subspecies: *M. s. siligorensis* (type locality: Nepal), *M. s. sowerbyi* (type locality: China), *M. s. alticraniatus* (type locality: Vietnam) and *M. s. thaianus* (type locality: Thailand). Whether the five BINs correspond to the described subspecies remains to be determined.

IUCN status: Least Concern

Recorded at: Pahang: Kuantan (Hill, 1972); Cheras Cave (Medway, 1969), Krau Wildlife Reserve (Zubaid, 1993; Kingston *et al.*, 2006), Tasik Chini (Lim & Ratnam, 1999); **Perlis**: Wang Kelian State Park (Jayaraj *et al.*, 2013a); **Kedah**: Ulu Muda Forest Reserve (Norsham *et al.*, 1999).

M. siligorensis has been recorded roosting in rock crevices and fissures in caves, often in small colonies at forest edges, in primary and secondary forests (Medway, 1969; Francis, 2008) and foraging near street lights at research station (Kingston *et al.*, 2006).

4.2 Diet of *C. brachyotis* as revealed by DNA barcoding

4.2.1 Recovery of plant DNA barcodes from faecal samples

Of the 95 faecal samples analysed, 65 samples (68.4%; seeds=43; pulps=22) produced both *rbcL* and *ITS2* DNA barcodes, 7 samples (7.4%; seeds=5; pulps=2) produced only *ITS2* barcodes, 8 samples (8.4%; seeds=1; pulps=7) produced only *rbcL* barcodes and the remaining 15 samples (15.8%; seeds=2; pulps=13) failed to produce any DNA barcodes (See supplementary file). Two *ITS2* barcodes were discarded: one from the urban site due to the short length of usable sequence (57 bp) and one from secondary forest which was suspected to be a contaminant due to its similarity (96%) to algal sequences (*Chlorella angustoellipsoidea* and *Chloroidium ellipsoideum*).

4.2.2 Taxonomic assignation

Seventeen plant species were detected in the faecal samples using DNA barcoding (Figure 4.6; Table 4.1). Seven plant species were detected from 26 faecal samples collected at the urban site, seven from 25 samples collected at the agricultural site and seven from 28 samples collected at the secondary forest site (Figure 4.6). Of the 17 plant species, nine was identified as native plants and four as introduced plants (Table 4.1).

4.2.3 Species richness and sampling completeness ratio

Based on the 78 faecal samples which produced usable DNA barcodes, the estimated plant species richness in the faecal samples ranged from 8.933 to 16.643 (Table 4.2). The estimated sampling completeness ratios were 0.936 for both secondary forest and urban sites and 0.825 for agricultural site (Table 4.2). The lower sampling completeness ratio and higher plant species richness for agricultural site suggested that larger faecal sample size and longer sampling period will reveal more plant species in faeces of *C. brachyotis* at agricultural site. Note that the faecal sampling at all sites were not conducted simultaneously and therefore, the results should be interpreted cautiously.

4.2.4 Dietary resource overlap

Ficus fistulosa was detected at all sampling sites with the highest detection frequency at agricultural and urban sites (Figure 4.6). Overall, a moderate dietary resource overlap was observed between the bats at the three sampling sites (O_{ij}=0.59). Moderate dietary resource overlap was observed between *C. brachyotis* at urban and secondary forest sites (O_{ij}=0.52) although bats at both sites shared *F. fistulosa* and *F. lepicarpa*. Similarly, moderate dietary resource overlap was observed between *C. brachyotis* at secondary forest and agricultural sites (O_{ij} = 0.45) although both shared *D. zibethinus* and *F. fistulosa*. In contrast, very high dietary resource overlap was observed between *C. brachyotis* at urban and agricultural sites (O_{ij} = 0.84) which shared only *F. fistulosa*.

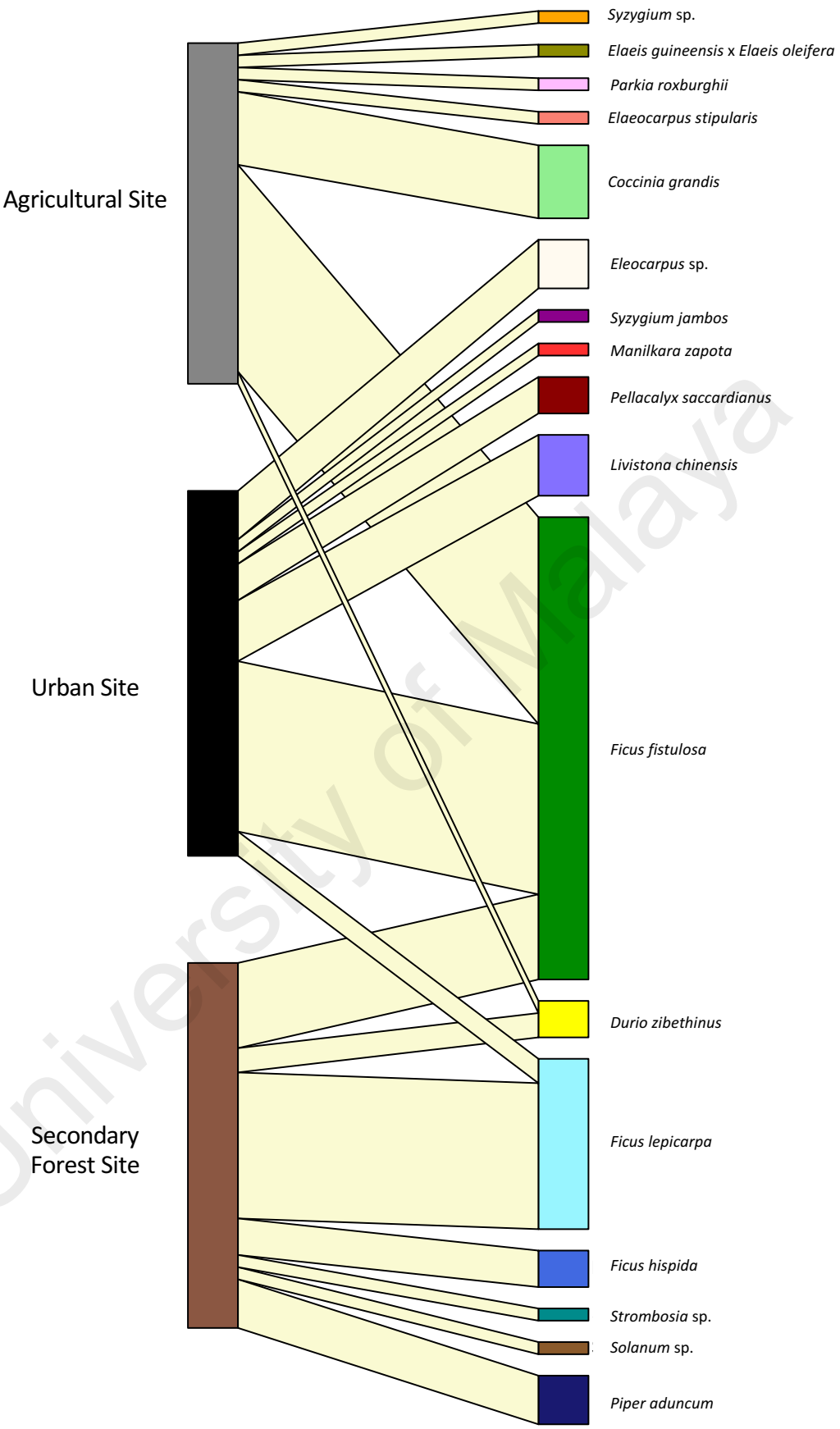

Figure 4.6: The interaction between C*ynopterus brachyotis* and plant species detected from faecal samples collected at three sites in Peninsular Malaysia. The width of the interaction bar corresponds to the number of fruit bats and occurrence of plants in the faeces of fruit bats.

Table 4.1: List of plants consumed by *Cynopterus brachyotis* in Southeast Asia. References: 1. Phua & Corlett (1989) recorded 21 plant species by observing *C. brachyotis* feeding at botanical garden; 2. Tan *et al*. (1998) recorded 53 plant species by morphologically identifying the plant remains in ejecta from *C. brachyotis* at secondary forests; 3. Hodgkison et al. (2004) recorded 15 plant species by morphologically identifying the plant remains in faeces, on bodies and under roosts of *C. brachyotis* at primary forest; 4. This study recorded 17 plant species using DNA barcoding.

Family	Species	Status[a]	Reference(s)
Moraceae	*Artocarpus fulvicortex*	Native	2
	Artocarpus maingayi	Native	2
	Ficus fistulosa	Native	1, 2, 4
	Ficus benjamina	Native	2
	Ficus globosa	Native	3
	Ficus hispida	Native	4
	Ficus lepicarpa	Native	4
	Ficus magnoliifolia	Native	3
	Ficus religiosa	Introduced	2
	Ficus scortechinii	Native	3
	Ficus (Unidentified)		2
Leguminosae	*Bauhinia purpurea*	Introduced	2
	Cassia fistula	Introduced	2
	Parkia roxburghii	Native	4
	Peltophorum pterocarpum	Native	2
	Senna spectabilis	Introduced	2
	Erythrina subumbrans	Native	2
	Erythrina variegata	Native	2
	Erythrina fusca	Introduced	2
	Erythrina (Unidentified)		2
Sapotaceae	*Manilkara zapota*	Introduced	2, 4
	Mimusops elengi	Native	2
	Palaquium clarkeanum	Native	2
	Palaquium gutta	Native	1, 2
	Palaquium obovatum	Native	1, 2, 3
	Payena selangorica	Native	2
	Payena lucida	Introduced	2, 3
	Payena maingayi	Native	2
	Pouteria malaccensis	Native	2
Myrtaceae	*Psidium guajava*	Introduced	1, 2
	Syzygium jambos	Native	1, 2, 4
	Syzygium chloranthum	Native	3
	Syzygium grande	Native	1, 2
	Syzygium aqueum	Native	2
	Syzygium malaccense	Native	1, 2
	Syzygium lineatum	Native	1

Table 4.1, continued.

Family	Species	Status	References
	Syzygium (Unidentified)		4
	Eugenia (Unidentified)		2, 3
Arecaceae	*Dypsis lutescens*	Introduced	2
	Elaies guineensis x *Elaies oleifera*	Introduced	4
	Ptychosperma macarthurii	Introduced	2
	Roystonea regia	Introduced	2
	Saribus rotundifolius	Introduced	2
	Licuala grandis	Introduced	2
	Livistona chinensis	Introduced	2, 4
Annonaceae	*Annona squamosa*	Introduced	2
	Cyathocalyx scortechinii	Native	3
	Polyalthia longifolia	Introduced	2
Anacardiaceae	*Campnosperma auriculatum*	Native	1
	Mangifera indica	Introduced	2
Pentaphylacaceae	*Adinandra dumosa*	Native	1
	Adinandra sarosanthera	Native	3
Elaeocarpaceae	*Elaeocarpus stipularis*	Native	2, 3, 4
	Elaeocarpus (Unidentified)		2, 4
Malvaceae	*Grewia tomentosa*	Native	2
	Durio zibethinus	Native	4
Clusiaceae	*Calophyllum inophyllum*	Native	1, 2
Combretaceae	*Terminalia catappa*	Native	1, 2
Cucurbitaceae	*Coccinia grandis*	Native	4
Euphorbiaceae	*Hevea brasiliensis*	Introduced	2
Gentianaceae	*Fagraea fragrans*	Native	1, 2
Lamiaceae	*Vitex pinnata*	Native	1
Melastomataceae	*Pternandra echinata*	Native	2, 3
Muntingiaceae	*Muntingia calabura*	Introduced	1, 2
Olacaceae	*Strombosia javanica*	Native	3
	Strombosia (Unidentified)		4
Piperaceae	*Piper aduncum*	Introduced	2, 4
Podocarpaceae	*Podocarpus rumphii*	Native	1
Rhizophoraceae	*Pellacalyx saccardianus*	Native	1, 2, 3, 4
Rosaceae	*Prunus polystachya*	Native	3
Rubiaceae	*Nauclea officinalis*	Native	3
Salicaceae	*Flacourtia inermis*	Introduced	2
Sapindaceae	*Nephelium malaiense*	Native	1, 2
Urticaceae	*Cecropia peltata*	Introduced	1
Ebenaceae	*Diospyros* (Unidentified)		1, 2
Musaceae	*Musa* (Unidentified)		1, 2
Solanaceae	*Solanum* (Unidentified)		4

Table 4.2: Estimated plant richness in faecal samples of *Cynopterus brachyotis*. The number of observed species for each sampling site is seven. The number of faecal samples for urban, agricultural and secondary forest sites are 26, 25 and 28 respectively.

Sampling Site	Sampling Completeness Ratio	Species Richness Model	Estimate	Standard Error	Lower Limit of 95% Confidence Interval	Upper Limit of 95% Confidence Interval
Agricultural site	0.825	Homogenous Model[a]	12.000	6.258	7.746	40.526
		Chao1[b]	16.643	9.750	8.862	56.949
		Chao1-bc[c]	16.643	9.750	8.862	56.949
		iChao1[d]	16.643	9.750	8.862	56.949
Urban site	0.936	Homogenous Model[a]	7.857	1.245	7.106	13.912
		Chao1[b]	8.933	3.626	7.174	28.456
		Chao1-bc[c]	7.483	1.262	7.029	15.034
		iChao1[d]	9.308	2.467	7.417	19.774
Secondary forest site	0.936	Homogenous Model[a]	7.750	1.139	7.088	13.393
		Chao1[b]	8.933	3.626	7.174	28.456
		Chao1-bc[c]	7.483	1.262	7.029	15.034
		iChao1[d]	9.308	2.467	7.417	19.774

[a]This model assumes that all species have same detection probabilities

[b]This approach is derived from the lower bound of undetected species richness in terms of the numbers of singletons and doubletons

[c]A bias-corrected form for the Chao1 estimator

[d]Improved Chao1 estimator

4.3 Diet of *E. spelaea* as revealed by DNA metabarcoding

4.3.1 Recovery of plant OTU from bat faeces and taxonomic assignation

A total of 47 OTU (~320 bp) were detected using *ITS2* primers and 13 OTU (~200 bp) were detected using *rbcL* primers. *RbcL* OTU which were assigned with a genus and/or family name that was also assigned to an *ITS2* OTU were discarded as likely duplicates. This resulted in 55 OTU (*ITS2*=47, *rbcL*=8), of which 37 OTU were assigned with a species name (*ITS2*=36, *rbcL*=1), fourteen were assigned a genus name (*ITS2*=11, *rbcL*=3) and the remaining four were assigned a family name (*rbcL*=4) (Figure 4.7; Appendix B). An average of 18 OTU were recovered each week (sd=5.103, min=12, max=30).

4.3.2 Species richness and sampling completeness ratio

The plant species richness in faecal samples of *E. spelaea* estimated by different models were within the range of 65.260 to 68.961 (Table 4.3). The sampling completeness ratio was estimated to be 0.912 (Figure 4.8). Both asymptotic (species richness estimation) and non-asymptotic analyses (rarefaction/extrapolation) suggested that a longer sampling period and larger faecal sample size would detect more plant species in the diet of *E. spelaea* (Figure 4.8). Of the 55 plant species, 24 were native (*ITS2* OTU=23; *rbcL* OTU=1) while 16 were introduced to Peninsular Malaysia (*ITS2* OTU=16) (Table 4.4). The status of the remaining 15 plant species is unknown (*ITS2* OTU=8; *rbcL* OTU=7) as I could not assign them to a species name. I detected 49 plant species which have not been reported by previous dietary studies of *E. spelaea* conducted during the same sampling months (i.e., December to March) (Table 4.4).

4.3.3 Relative detection rate of plants consumed by *E. spelaea*

Two native plant species, *Duabanga grandiflora* and *Musa balbisiana*, and an introduced species, *Artocarpus heterophyllus*, were detected from all ten faecal samples

(i.e. every week) and were flowering during the sampling period (Figure 4.7; Table 4.4). The native *Musa acuminata* and the introduced *Ceiba pentandra* were detected in nine faecal samples, and were flowering during the sampling period. The native *Urophyllum leucophlaeum* and a fern, *Dicranopteris* sp., were detected in eight faecal samples. Ten of the 55 plant species moderately detected in the ten faecal samples while the remaining 38 plant species were infrequently detected (Figure 4.7).

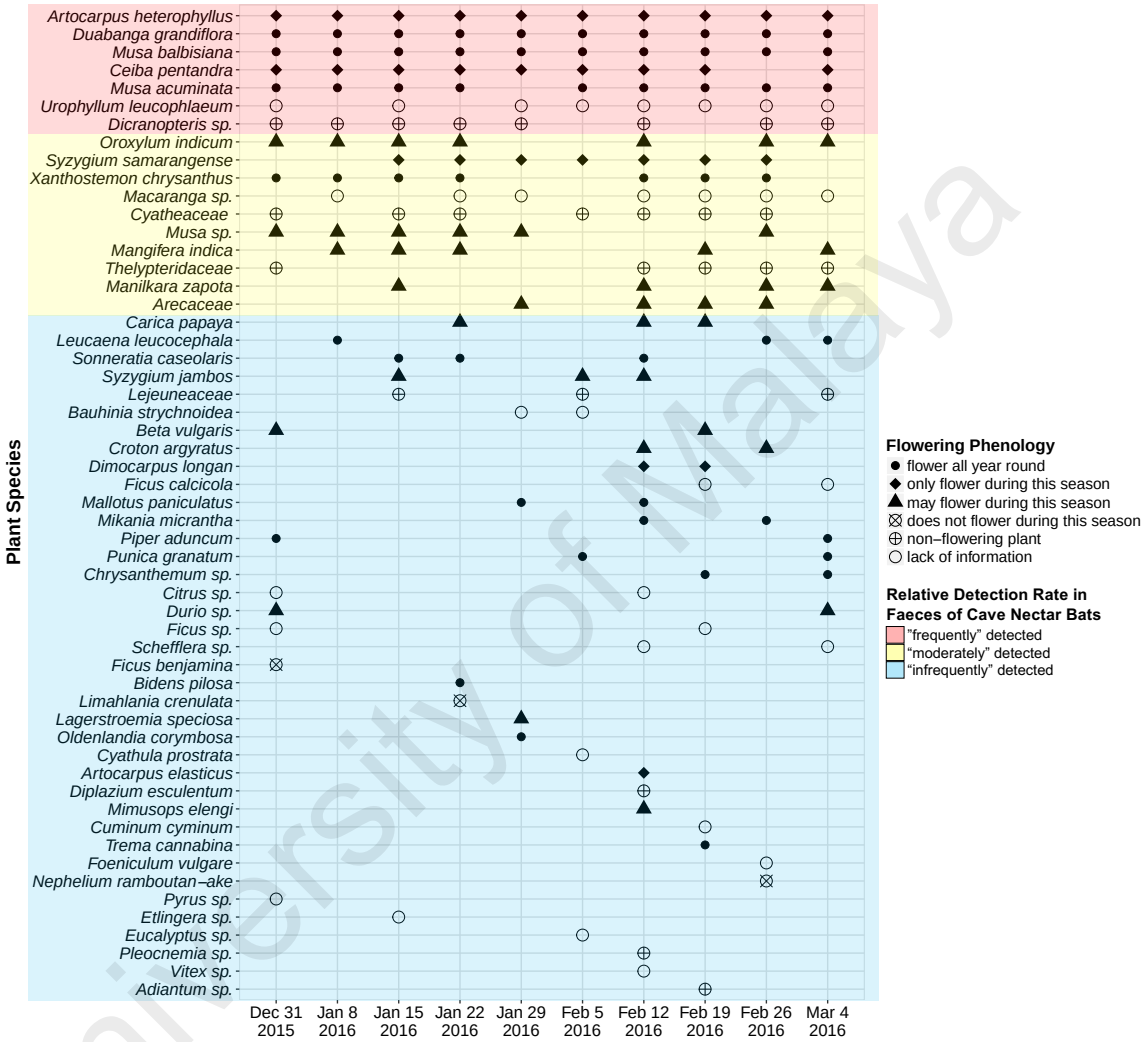

Figure 4.7: Plant species detected from faecal samples of *Eonycteris spelaea* using DNA metabarcoding for ten weeks (31st of December 2015 to 4th of March 2016). Order of y-axis is based on (i) number of detection, (ii) taxonomic rank (i.e., species, genus and family), (iii) alphabetical order and (iv) date of detection.

Table 4.3: Estimated plant richness in the faecal samples of *Eonycteris spelaea*. The number of observed species is 55, the number of faecal sample is 10 and the total number of incidences is 185.

Species Richness Model	Estimate	Standard Error	Lower Limit of 95% Confidence Interval	Upper Limit of 95% Confidence Interval
Homogenous Model[a]	60.324	2.996	56.904	69.882
Chao2[b]	66.604	7.040	58.873	89.766
Chao2-bc[c]	65.260	6.342	58.365	86.286
iChao2[d]	68.961	4.356	62.682	80.372

[a]This model assumes that all species have same incidence of detection probabilities

[b]This approach uses the frequencies of uniques and duplicates to estimate the number of undetected species

[c]A bias-corrected form for the Chao2 estimator

[d]Improved Chao2 estimator

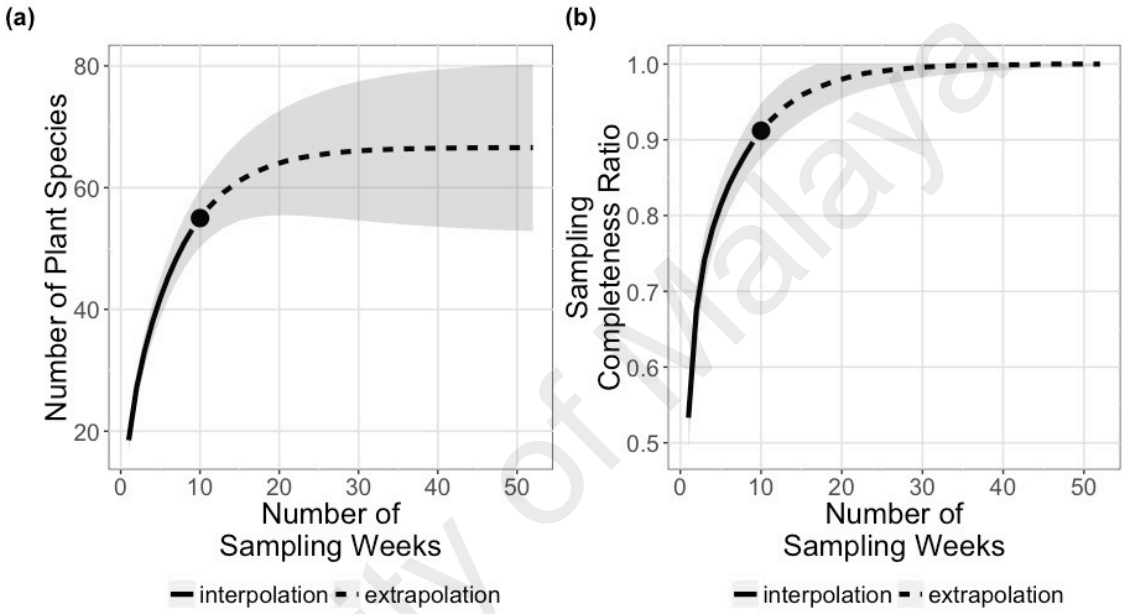

Figure 4.8: Rarefaction and extrapolation sampling curves for this study (from 31st of December 2015 to 4th of March 2016) showing estimated species richness using Chao2. Sampling curves are extrapolated to one year (52 weeks) with 95% confidence interval, number of replications=100, and sampling completeness ratio=0.912. (a) Sample-sized-based rarefaction and extrapolation curve (b) Sample completeness-based rarefaction and extrapolation curve.

Table 4.4: Checklist of plants consumed by *Eonycteris spelaea* between December and March. References: 1= Start (1974) reported fourteen plant species; 2= Bumrungsri *et al.* (2013) reported nine plant species; 3=Thavry *et al.* (2017) reported seven plant species; 4=This study detected 55 plant species.

Family	Species	Status	Type of detection [a]	Month(s) of detection [b]	References
Amaranthaceae	*Beta vulgaris*	Introduced	DNA	Dec, Feb	4
	Cyathula prostrata	Native	DNA	Feb	4
Anacardiaceae	*Mangifera indica*	Introduced	DNA	Jan - Mar	4
Apiaceae	*Cuminum cyminum*	Introduced	DNA	Feb	4
	Foeniculum vulgare	Introduced	DNA	Feb	4
Araliaceae	*Schefflera* (Unidentified)		DNA	Feb - Mar	4
Arecaceaea	*Cocos nucifera*	Native	P	Dec - Mar	1, 2
	Arenga (Unidentified)		P	Jan - Mar	1, 2
	(Unidentified)		DNA	Jan - Feb	4
Asteraceae	*Bidens pilosa*	Native	DNA	Jan	4
Anacardiaceae	*Chrysanthemum* (Unidentified)	Introduced	DNA	Feb - Mar	4
	Mikania micrantha	Introduced	DNA	Feb	4
Bignoniaceae	*Oroxylum indicum*	Native	P, DNA	Dec - Mar	1, 2, 3, 4
Cannabaceae	*Trema cannabina*	Native	DNA	Feb	4
Caricaceae	*Carica papaya*	Introduced	DNA	Jan - Feb	4
Compositae	(Unidentified)		P	Dec	1
Euphorbiaceae	*Croton argyratus*	Native	DNA	Feb	4
	Macaranga (Unidentified)		DNA	Jan - Mar	4
	Mallotus paniculatus	Native	DNA	Jan - Feb	4
Fabaceae	*Bauhinia strychnoidea*	Native	DNA	Jan - Feb	4
	Leucaena leucocephala	Introduced	DNA	Jan - Mar	4
Gentianaceae	*Limahlania crenulata*	Native	DNA	Jan	4
Lamiaceae	*Vitex* (Unidentified)		DNA	Feb	4
Leguminosae	*Parkia* spp.		P	Dec - Mar	1, 2, 3
Lythraceae	*Duabanga grandiflora*	Native	Fl, P, DNA	Dec - Mar	1, 4
	Lagerstroemia speciosa	Native	DNA	Jan	4
	Punica granatum	Introduced	DNA	Feb - Mar	4
	Sonneratia alba	Native	Fl, P	Dec - Feb	1
	Sonneratia caseolaris	Native	Fl, P, DNA	Dec – Feb	1, 4
	Sonneratia (Unidentified)		P	Dec - Mar	2, 3
Malvaceae	*Bombax anceps*	Native	Fl, P	Dec – Feb	1, 3
	Bombax (Unidentified)		P	Feb	2
	Ceiba pentandra	Introduced	Fl, P, DNA	Dec - Mar	1, 2, 3, 4
	Durio spp.	Native	P, DNA	Dec - Mar	1, 2, 3, 4

Table 4.4, continued.

Family	Species	Status	Type of detection [a]	Month(s) of detection [b]	References
Moraceae	*Artocarpus elasticus*	Native	DNA	Feb	4
	Artocarpus heterophyllus	Introduced	DNA	Dec - Mar	4
	Artocarpus (Unidentified)		P	Jan - Mar	1
	Ficus benjamina	Native	DNA	Dec	4
	Ficus calcicola	Native	DNA	Feb - Mar	4
	Ficus (Unidentified)		DNA	Dec, Feb	4
Musaceae	*Musa acuminata* (previously reported as *malaccensis* and *truncata*)	Native	Fl, DNA	Dec - Mar	1, 4
	Musa balbisiana	Native	DNA	Dec - Mar	4
	Musa (Unidentified)		Fl, P, DNA	Dec - Mar	1, 2, 3, 4
Myrtaceae	*Syzygium jambos*	Native	DNA	Jan - Feb	4
	Syzygium malaccensis (previously reported as *Eugenia malaccensis*)	Native	Fl	Dec - Feb	1
	Syzygium samarangense	Native	DNA	Jan - Feb	4
	Syzygium (Unidentified)		P	Dec - Mar	1, 2
	Xanthostemon chrysanthus	Introduced	DNA	Jan - Feb	4
	Eucalyptus (Unidentified)		P, DNA	Feb	3, 4
Piperaceae	*Piper aduncum*	Introduced	DNA	Dec, Mar	4
Rosaceae	*Pyrus* (Unidentified)		DNA	Dec	4
Rubiaceae	*Oldenlandia corymbosa*	Introduced	DNA	Jan	4
	Urophyllum leucophlaeum	Native	DNA	Dec - Mar	4
Rutaceae	*Citrus* (Unidentified)		DNA	Dec, Feb	4
Sapindaceae	*Dimocarpus longan*	Native	DNA	Feb	4
	Nephelium ramboutan-ake	Native	DNA	Feb	4
Sapotaceae	*Manilkara zapota*	Introduced	DNA	Jan - Mar	4
	Mimusops elengi	Native	DNA	Feb	4
	(Unidentified)		P	Feb - Mar	1
Zingiberaceae	*Etlingera* (Unidentified)		DNA	Jan	4

Table 4.4, continued.

Family	Species	Status	Type of detection [a]	Month(s) of detection [b]	References
Athyriaceae	*Diplazium esculentum*	Native	DNA	Feb	4
Pteridaceae	*Adiantum* (Unidentified)		DNA	Feb	4
Dryopteridaceae	*Pleocnemia* (Unidentified)		DNA	Feb	4
Gleicheniaceae	*Dicranopteris* (Unidentified)		DNA	Jan - Mar	4
Thelypteridaceae	(Unidentified)		DNA	Dec, Feb - Mar	4
Cyatheaceae	(Unidentified)		DNA	Jan - Feb	4
Lejeuneaceae	(Unidentified)		DNA	Jan - Mar	4

a = Type of detection (Fl = sighted on and/or caught near flowers, P = pollen found in faeces and/or on body, Fr = caught near fruiting trees, DNA=DNA metabarcoding)

b = Month of the year (Jan = January, Feb = February, Mar = March, Dec = December)

CHAPTER 5: DISCUSSION

5.1 Diversity in bats of Peninsular Malaysia

Verifying the taxonomic status of bats is necessary for creating checklist of bats and framing suitable conservation approaches for particular species and their habitats (Tsang *et al.*, 2016; Wilson, 2017). Yet studying the taxonomy of bats remains a challenge often due to the poor preservation of type specimens and indistinctive morphological characteristics of certain bat species (Tsang *et al.*, 2016). As discussed below, DNA barcoding could resolve the problems in taxonomy of bats with implications for conservation of particular species and the localities where they were last recorded.

This literature review has produced a checklist of 110 bat species for Peninsular Malaysia. In comparison, Kingston *et al.* (2006) reported 69 species for Krau Wildlife Reserve and estimated ~125 species for Peninsular Malaysia, while Davison and Zubaid (2007) reported 106 species for Peninsular Malaysia. Of the 110 bat species in this checklist, 105 species have precise locality records from Peninsular Malaysia whereas the remaining five lack recent and/or precise locality records, raising the need for intensive surveys to confirm their presence and distribution in the region. This checklist includes records of bats previously reported under informal names, also known as "dark taxa" (Page, 2016) such as *Cynopterus* cf. *brachyotis* SUNDA, *C.* cf. *brachyotis* FOREST, *Hipposideros* bicolor131, *H.* bicolor142, and *Myotis muricola* "Eastern". Whether these species (or the form occurring in Peninsular Malaysia) deserve to be recognised as distinct species remains to be determined. In addition, the findings of this research also support the view of previous studies which proposed several subspecies to be recognised as distinct species: *Balionycteris seimundi* (Khan *et al.*, 2008), *Murina peninsularis* (Francis & Eger, 2012; Soisook *et al.*, 2013), *Myotis federatus* (Görföl *et al.*, 2013) and *Rhinolophus morio* (Volleth *et al.*, 2015). Therefore, the name of these should be updated.

The search of BOLD (Ratnasingham & Hebert, 2007) revealed that 86 species (78%) have public records on BOLD, of which 48 of them (44%) have DNA barcodes from Peninsular Malaysia. This means 62 species (56%) did not have DNA barcodes from Peninsular Malaysia, leaving their taxonomic status and presence in the region somewhat unresolved. *Nyctalus noctula* did not have DNA barcodes from Southeast Asia. Eighty (73%) of the 86 species with public DNA barcodes were associated with BINs. Moreover, the search of BOLD uncovered several cases where DNA barcodes recorded under the same species name were assigned to different BINs. NJ analyses of available DNA barcodes from bats sampled in Peninsular Malaysia uncovered several cases of taxonomic uncertainty (i.e., *Hipposideros larvatus, H. cervinus, H. galeritus, H. armiger, Macroglossus minimus, M. sobrinus, Rhinolophus lepidus, R. stheno, Kerivoula hardwickii, K. minuta, Philetor brachypterus* and *Miniopterus medius*) which were highlighted here for further investigation.

COI mtDNA was proposed as the standard DNA barcode marker for animals on the basis of high variation within the region which allows the discrimination of closely related species (Hebert *et al.*, 2003). Here, *COI* mtDNA was observed to be not sufficiently variable for *Macroglossus minimus* and *M. sobrinus* which are likely to have diverged recently (van Velzen *et al.*, 2012). Hence, the use of *COI* mtDNA alone may limit the identification of species boundaries and investigation of deep phylogenetic relationships (Hajibabaei *et al.*, 2007; Wilson *et al.*, 2016). Therefore, the use of BINs as arbitrary division of "species" in BOLB may or may not correspond to the species differences. Further molecular analyses for resolving the taxonomy of highlighted species should include other regions of mtDNA (e.g., cytochrome *b* and *D-loop*) and nuclear DNA to complement the use of *COI* mtDNA and minimise the error in interpretating population history caused by specific gene genealogies (see Hajibabaei *et al.*, 2007). However, note that there is no perfect gene as DNA marker for resolving deep phylogenetic relationships

including cytochrome *b* which is insufficiently variable in first and second codon positions (Meyer, 1994). In addition, several studies have uncovered cases where a taxon showed variations in mtDNA but did not show any variations in nuclear DNA (Kuo *et al.,* 2015; Dool *et al.,* 2016; Mao *et al.,* 2017). Such incongruences may be explained by several factors such as homoplasy selection, between species' variation in mutation rates, introgression, incomplete lineage sorting and even female philopatry associated with male biased dispersal (Kuo *et al.,* 2015; Dool *et al.,* 2016; Mao *et al.,* 201). Altogether these findings raise the importance of interpretating results from mtDNA-based analysis cautiously.

A total of 18 species (16%) were retrieved from old records dated before year 2000 and seven of them have been recorded only once in Peninsular Malaysia (Figure 5.1). The lack of recent records for these species may be due to sampling biases (Douangboubpha *et al.,* 2010b; Lim *et al.,* 2016) as some of them appear to be exclusively dependent on certain habitat structures (e.g., *Hipposideros halophyllus* in limestone areas, *Aethalops alecto* in hill and montane forests, and *Pteropus hypomelanus* on islands) and restricted to certain localities (e.g., *Phoniscus jagorii* in Krau Wildlife Reserve, and *Myotis hermani* in Temenggor Forest Reserve). Several species are seemingly restricted to northern Peninsular Malaysia (i.e., *Hipposideros lekaguli, H. halophyllus, H. pomona, Rhinolophus acuminatus, R. marshalli, R. malayanus*). Three species (2.8%) have been reported from Peninsular Malaysia but without any precise localities (Figure 5.1).

The main threat to bats in Peninsular Malaysia is habitat loss due to expansion of agricultural land and urbanisation (Francis, 2008). Yet only *Pteropus vampyrus* and *P. hypomelanus* are currently receiving conservation protection from the federal government of Malaysia under Wildlife Conservation Act 2010. According to IUCN (2016), ten species: *Megaerops wetmorei, Chaerephon johorensis, Coelops robinsoni, Hipposideros halophyllus, H. orbiculus, H. ridleyi, Murina aenea, M. rozendaali, Arielulus societatis*

and *Hesperoptenus tomesi* are listed as “Vulnerable”, 15 species are listed as “Near Threatened”, and 71 species are listed as “Least Concern”. Six species: *Hipposideros nequam*, *Rhinolophus convexus*, *Kerivoula krauensis*, *Hesperoptenus doriae*, *Myotis hermani* and *Hypsugo macrotis* are listed as “Data Deficient” while eight species have yet to be assessed. Many species were listed as “Least Concern” by IUCN (2016) (e.g., *Aselliscus stoliczkanus*, *Chaerephon plicatus*, *R. marshalli*, *Kerivoula picta*, *Arielulus circumdatus*), but the lack of recent records for these species suggests the need for reconsideration of their conservation status in Peninsular Malaysia.

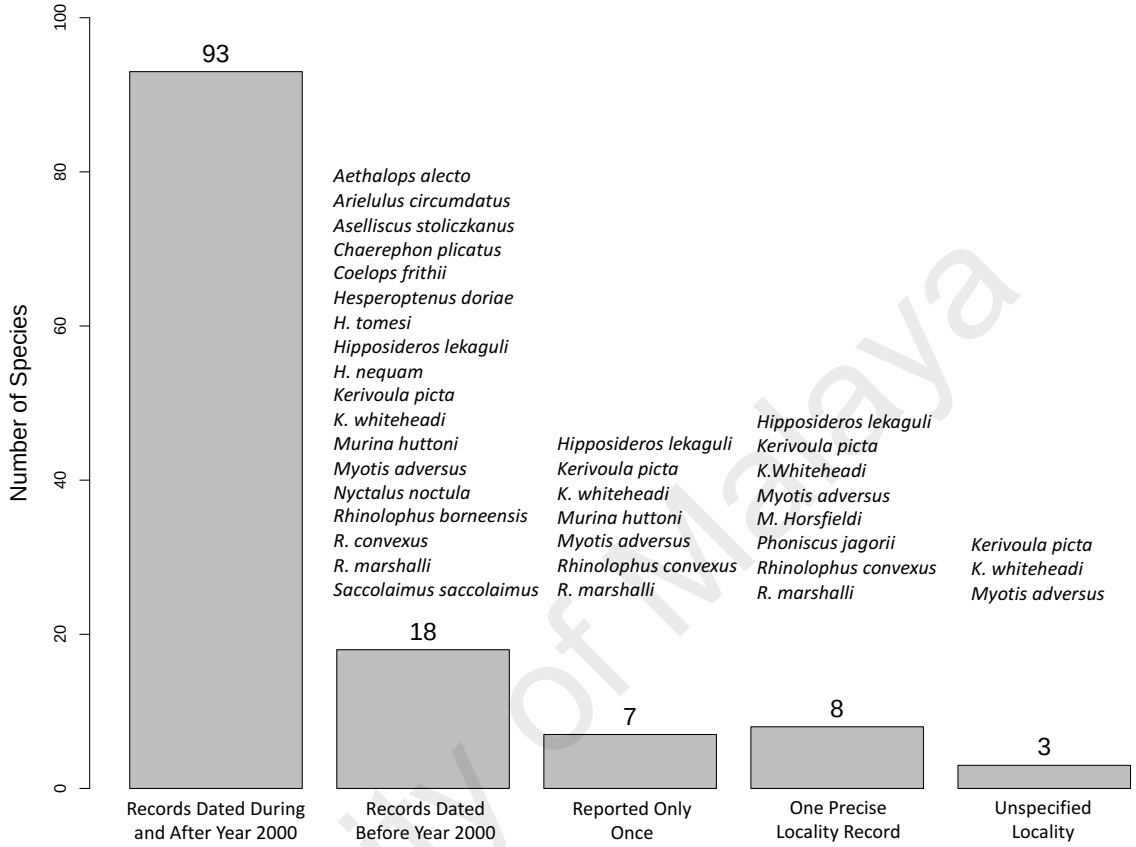

Figure 5.1: Bat species with recent (dated during or after the year 2000) and old (dated before year 2000) records from Peninsular Malaysia.

5.2 Impact of urbanisation and agriculture on diet of *C. brachyotis*

Unambiguous identification of plant material present in faeces of frugivorous bats is necessary for understanding their diet but remains a challenge due to the difficulties in identifying the fragmented plant material and morphologically similar seeds of different species. Here, most of the digested pulp and seeds in the faeces of frugivorous bat, *C. brachyotis* could be identified using DNA barcoding, consequently revealed previously unknown food resources for the fruit bats and their potential ecological roles.

It is likely that *C. brachyotis* feeds predominantly on pioneer and forest plants. The pioneer plant genus *Ficus* which often dominate regenerating forest (Muscarella & Fleming, 2007) emerged as the dominant component of the diet of *C. brachyotis* at all sampling sites and is responsible for the high dietary resource overlap observed between the bats in urban and agricultural sites, although bats at both sites shared only *F. fistulosa* with very high detection frequency (note that the faecal sampling at all sites were not conducted simultaneously). Many *Ficus* species including *F. fistulosa*, *F. lepicarpa* and *F. hispida* have multiple fruiting periods throughout the year (Phillipps & Phillipps, 2016), making *Ficus* spp. a stable resource compared to more transient species (e.g., *Syzygium jambos* and *Manilkara zapota*) (Tan *et al.*, 1998; Fukuda *et al.*, 2009), consequently promoting stable population dynamics in consumers (Tan *et al.*, 2000).

Native forest plants and cultivated plants were detected in faecal samples collected from urban and agriculture sites although these plants were not observed at these locations. Seeds belong to *Ficus* spp. were found in faecal samples collected from all sites. During the sampling at urban site, an individual was captured with a fruit of *Ficus* sp. in its mouth. This suggests that the bats are moving and depositing seeds away from parent plants, implying the role of *C. brachyotis* in seed dispersal. In Thailand, *C.*

brachyotis have been reported to travel up to 14.5 km per day (Bumrungsri, 2002). By transporting seeds across habitats, *C. brachyotis* could promote plant diversity, particularly in disturbed habitats (i.e., urban and agricultural areas) which often lack seed resources and succession (Hodgkison *et al.*, 2003; McConkey *et al.*, 2012).

Introduced plants were detected from pulps in faecal samples at all sampling sites. The fairly high detection rate of introduced plants particularly *P. aduncum* and *L. chinensis* shows that *C. brachyotis* can exploit novel food resources and potentially aid invasion of introduced plants through dispersal activities (Muscarella & Fleming, 2007). Although the seeds of introduced plants were not visually observed in the faecal samples nor the feeding behaviour of *C. brachyotis* (i.e., carrying fruits away from parent trees to feeding perches) was visually assessed, the next step would be to determine the relative role of *C. brachyotis* in facilitating succession of native plants and/or promoting invasions of introduced plants.

The low detection rate of oil palm (*Elaies guineensis x Elaies oleifera*) in faecal samples collected at agricultural site suggests that the bats are not predominantly feeding on oil palm fruits and their presence in oil palm plantations could be explained by other factors. The diverse diet of *C. brachyotis* at the oil palm plantation (a monoculture) suggests that the bats may have used the plantation as connecting flyway to travel to forest fragments and agricultural plantations nearby which provide more diverse food resources. This is similar to the findings of Heer *et al.* (2015) which detected large number of frugivorous bats in rubber-cacao plantations that offered little food resources to the bats, but obviously served as corridors. However, it is also possible that oil palm may not be detected if it is ingested by the bats just before they depart from this area though the low detection everywhere suggests this possibility is remote. The detection of other cultivated plants indicates that *C. brachyotis* feed on other readily available food crops which consequently may lead to conflict between the bats and fruit growers. Although the extent

of the damage to food crops caused by *C. brachyotis* is significantly smaller than that of other larger mammals (i.e., *Macaca nemestrina, Arctictis binturong, Cervus timorensis,* and *Sus barbatus*), the bats are often killed in large numbers as they are generally of lower concern to the wildlife authorities (Fujita & Tuttle, 1991; Aziz *et al.*, 2016).

Plants (i.e., *Ficus fistulosa, Szygium jambos,* and *Pellacalyx saccardianus)* which have been reported in previous dietary studies of *C. brachyotis* conducted at secondary forest and urban areas (Phua & Corlett, 1989; Tan *et al.*, 1998) were also detected in this study. However, the failure in detecting plants which were reported to be seasonally dominant in the diet of *C. brachyotis* in this study is likely due to the short sampling period, as supported by the estimated sampling completeness ratio and plant species richness which indicated that larger faecal sample size and longer sampling period will detect more plant species in faeces of *C. brachyotis* at all sampling sites particularly agricultural site. Nevertheless, this study has detected cultivated (i.e., *Parkia roxburghii, Elaies guineensis* x *Elaies oleifera,* and *Coccinia grandis*) and pioneer plants (i.e., *Ficus hispida* and *F. lepicarpa*) which have not been reported in other dietary studies of *C. brachyotis*.

The advantage of using DNA barcoding to identify the diet of *C. brachyotis* is that most of the seeds and digested plant pulp in the faeces could be assigned with species names. Most of the seeds were assigned with the species name *Ficus fistulosa* which also has been reported as the most common *Ficus* species eaten by *C. brachyotis* at secondary forest and urban areas (Phua & Corlett, 1989; Tan *et al.*, 1998). Seeds belong to genus *Ficus* can be easily assigned to this plant genus based on the morphology of the seeds. However, assigning *Ficus* seeds accurately to a species based on the morphological characteristics of seeds is often time-consuming and requires high level of plant taxonomic expertise. Phua and Corlett (1989) failed to assign species name to six types of *Ficus* remains due to the difficulty in identifying the remnants of seeds and fruits while

Hodgkison *et al.* (2004) germinated the seeds collected from faeces of bats for species identification based on the morphological characteristics of the seedlings.

The use of DNA barcoding has detected plants with seeds that are too large to be ingested by *C. brachyotis* (i.e., *Elaies guineensis* x *Elaies oleifera* and *Coccinia grandis*) which consequently are not visually observed in the faeces. Although the bats may not be able to disperse large seeds through defecation, *C. brachyotis* may still serve as important seed disperser by carrying the heavy fruits with large seeds to feeding perches away from parent trees (Funakoshi & Zubaid, 1997). Together these findings highlighed the utility of DNA barcoding in dietary studies of frugivorous bats as the reliance on morphological identification of seeds in faeces may overlook plants with large seeds where only pulp is present and consequently overlook the potential seed dispersal role of the bats.

In this study, seeds were preferentially selected rather than fruit pulp for sequencing. If a bat had consumed a large fruit (and dropped the large seed) along with a small fruit (and swallowed the small seeds), it may potentially cause a bias in the detection of plants with small seeds. However, the gut passage time of most frugivorous bats is fast enough that multiple fruit types were not frequently observed in a faecal sample (E Clare, personal observation) and thus the effect of the bias is likely minimal.

One limitation of using DNA barcoding to identify the plant pulp is that it cannot determine which part of the plant that the bats are feeding on. For example, the most important pollinator of economically important *Durio zibethinus* is *Eonycteris spelaea* which feeds on nectar (Bumrungsri *et al.,* 2009), whereas *C. brachyotis* is reported to feed on the flowers (Funakoshi & Zubaid, 1997). Although *D. zibethinus* is also detected from plant pulp isolated from bat faeces in this study, it is unknown whether *C. brachyotis* feed on the nectar and consequently pollinate the economically important crops, or consume the fruits and/or flowers which would inhibit the development of the crops. However, DNA barcoding may still be able to provide information regarding the feeding preference

of bats if seeds were preferentially isolated from the bat faeces for Sanger sequencing. Direct observation of feeding behaviour of bats such as using camera traps (Aziz *et al.*, 2017b) remains necessary for identifying the feeding preference of bats and to determine whether the interactions between bats and plants are mutualistic or antagonistic.

The reliance on existing databases and local botanical records for taxonomic assignation of the plant DNA barcodes leaves these names as provisional. Most of the *ITS2* barcodes were assigned with species names as the region can distinguish closely related species within same genus when comprehensive reference libraries are available (Braukmann *et al.*, 2017). However, *ITS2* region also produced some ambiguous results in rapidly radiating groups (e.g., *Ficus*) and local botanical records were used to refine these cases. The *ITS2* region also detected fewer plant families compared to *rbcL*. In contrast, most of the *rbcL* barcodes matched to sequences in GenBank recorded under multiple species names with 100% similarity. These observations supported the importance of using paired markers for plant identification as single marker could not completely discriminate among species in certain plant groups (Kress & Erickson, 2007; CBOL, 2009; Kress, 2017). High-throughput NGS (DNA metabarcoding) could be utilised for future dietary study of frugivorous bats, which may help to distinguish mixed signals in individuals consuming multiple species.

5.3 Diverse diet of *E. spelaea* in an urban environment

By using DNA metabarcoding to identify the plant species present in faeces of *E. spelaea* collected over ten weeks, this study has detected 55 plant species, many of which had not been reported in previous studies of the diet of *E. spelaea* (including studies conducted during the same time of year; Table 4.4). In this study, most of the detected plants could be assigned to a species name. For example, the two OTU belonging to the economically important genus *Artocarpus* could be identified as *Artocarpus elasticus* and

A. heterophyllus, whereas Start and Marshall (1976) could only identify pollen grains to the genus *Artocarpus* but could not assign to a species name. In addition, the failure of previous studies (which examined the morphology of pollen grains) to detect pollen grains of species recorded in this study may be due to degradation of the pollen grains in the bats' gastrointestinal tract (Herrera & Martinez del Rio, 1998). Therefore, it is difficult to conclude if the detection of these species in this study is due to the changing landscape or a result of the better detection capability of DNA metabarcoding.

In contrast, this study failed to detect several plant species that were previously recorded in the diet of *E. spelaea*. This may be due to the plant DNA barcoding primers used in this study which could be biased towards the detection of particular plant families (García-Robledo *et al.*, 2013; Prosser & Hebert, 2017). Furthermore, using BLAST (against NCBI GenBank) for OTU identification is limited to plant species which have already been sequenced and submitted to the database (Bell *et al.*, 2016; Bell *et al.*, 2017). Consequently, this study may have failed to detect some of the previously reported diet species that are not currently in NCBI GenBank (e.g., *Bombax anceps* and *Syzygium malaccensis*).

The short sampling period of this study (31 December 2015 – 4 March 2016) may also account for the failure to detect certain plant species. Although the relatively high sampling completeness ratio and estimated plant species richness support the adequacy of the sampling effort for this study, both estimates only apply for the particular sampling period (when only certain plant species were flowering). As floral community changes over time (Boulter *et al.*, 2006; Delaney *et al.*, 2015), especially in Peninsular Malaysia where many species flower at irregular intervals (Appanah, 1993; Chen *et al.*, 2018), a longer sampling period and larger faecal sample size will likely reveal more plant species in the diet of *E. spelaea*.

The native plant species, *Duabanga grandiflora* and *Musa* spp. were frequently detected in this study and, considering these species flower year-round, likely represent a stable food resource for cave nectar bats throughout the year. Two other native plant species: *Urophyllum leucophlaeum*, which has been recorded in hill and montane forests in Peninsular Malaysia (Wong, 1989), and *Bauhinia strychnoidea*, a calciphile plant which has been recorded from Batu Caves (Ridley, 1922), were frequently and infrequently detected in the bat faeces. Little is known about the flowering phenology and pollination ecology of these plants. The infrequent detection of a mangrove plant, *Sonneratia caseolaris*, which flowers year-round, suggests that the species is not an important food resource for these particular cave nectar bats, yet indicates that some bats likely travelled ~40 km from Batu Caves to the nearest mangrove forest at Kuala Selangor. This is congruent with the finding of Start (1974) who observed that individuals of *E. spelaea* roosting at Batu Caves travelled 38 km to Rantau Panjang to feed on *Sonneratia alba*. Interestingly, Acharya *et al.* (2015b) estimated the foraging range for *E. spelaea* in southern Thailand to be 17.9 km only, though this could be due to the fact that the cave roosts in that particular study were located in agricultural areas where the cultivated fruit orchards nearby provided an easy source of food. In contrast, *E. spelaea* in Batu Caves appears to travel long distances from the roost to different habitats (i.e., mangrove, limestone and montane forests) where it feeds and consequently may promote genetic diversity among plant populations by dispersing pollen (Fleming *et al.*, 2009). Radio-tracking the cave nectar bats at Batu Caves remains a highly desirable approach to determine their foraging distances, and assess whether the long-distance travelling behaviour (i) is sex-specific where female tend to forage further while male tend to forage closer to roost as observed in *E. spelaea* in Thailand (Bumrungsri *et al.*, 2013) and *Pteropus rufus* in Madagascar (Oleksy *et al.*, 2015), and (ii) whether it is a strategy to reduce extreme competition for food which may be a consequence of the gregarious

roosting behaviour of *E. spelaea* as recognised for *Leptonycteris curasoae* in Mexico (Horner *et al.*, 1989). Together these findings support the view that *E. spelaea* remain a crucial pollinator of native plants in highly disturbed habitats.

Many of the plant species detected from the bat faeces were introduced to Peninsular Malaysia and have since naturalised in the region. This include the *Artocarpus heterophyllus* and *Ceiba pentandra* which are commonly planted in human settlements for fruits (Corner, 1997) and were flowering during the sampling period. The high detection rate of these introduced plants in the bat faeces suggests that these plants may be important food resource for the cave nectar bats in human-dominated habitats. On the other hand, the moderate and infrequent detection rate of other introduced plant species which are often planted for urban beautification and shade (e.g., *Chrysanthemum* sp., *Leucaena leucocephala* and *Xanthostemon chrysanthus*) suggests that these plants may be supplement food resources for the bats (Corlett, 2005; Nakamoto *et al.*, 2007). However, consumption and potential pollination of these introduced plants by cave nectar bats may have an adverse impact on the reproductive success of native plants (Morales & Traveset 2009) and on other dependant urban wildlife (Corlett, 2005; Grimm *et al.*, 2008). Therefore, the status of a plant species should be considered carefully prior to gardening and landscaping activities. Planting native plants instead of introduced plants could help to promote the consumption and hence pollination of native plants by the cave nectar bats, which consequently could maintain healthy ecosystems in urban areas.

Many of the plant species detected in this study are grown as commercial food crops including jackfruit (*Artocarpus heterophyllus*), banana (*Musa* spp.), water apple (*Syzygium samarangense*), mango (*Mangifera indica*) and papaya (*Carica papaya*); most of these plants were likely to be flowering during the sampling period. One of the commercial food crops which was frequently detected and flowers seasonally is jackfruit; a fruit with an estimated production value of RM 55 million for year 2011 (Abd-Aziz *et*

al., 2016). The previous study in Peninsular Malaysia also reported pollen grains of genus *Artocarpus* in faeces of *E. spelaea* (Start & Marshall, 1976). Altogether it is likely that *E. spelaea* play an important role in pollination of this economically important plant species.

Plant species which are pollinated and/or dispersed by wind and/or insects were detected in the bat faeces including ferns (e.g., *Adiantum* sp. and *Pleocnemia* sp.), weeds (e.g., *Bidens pilosa, Cuminum cyminum, Cyathula prostrata, Oldenlandia corymbosa*), figs (*Ficus* spp.) and *Artocarpus elasticus* (Corner, 1997; Boo *et al.*, 2014). The infrequent detection of these plant species suggests they form a relatively minor part of the cave nectar bat's diet or were unintentionally consumed. It could also be likely that the spores and pollen grains of these plant species may have adhered to the fur of the cave nectar bats when they were foraging (Corbet *et al.*, 1982) and consequently were ingested when they groomed themselves later (Fleming *et al.*, 2009). Another potential explanation (though unlikely given the protocol in this study) is that the spores and pollen grains of these plant species may have been unintentionally collected when sampling the bat faeces directly from the cave floor.

One limitation of DNA metabarcoding is the inability to identify which part of the plant is being consumed by the bats. Previous studies have observed remains of fruits and leaves in faeces and under the day roosts of *E. spelaea*, and consequently suggested that fruits and leaves may form a part of the cave nectar bat's diet (Start & Marshall, 1976; Bumrungsri *et al.*, 2013). Similarly, this study has detected ferns and figs (which were either not flowering during the sampling period or have unknown flowering phenology) in the faeces of *E. spealea* but could not determine whether the bats were feeding on the fronds and fruits or ingesting the spores and pollen grains inadvertently. It is possible that *E. spelaea* chew the fronds and fruits, ingest the juice (and possibly fragments of the fronds and fruits), and spit out the fibres later; a feeding behaviour which is common in pteropodid bats including *Cynopterus brachyotis* (Phua & Corlett, 1989; Tan *et al.*, 1998)

and *Pteropus* spp. (Nakamoto *et al.,* 2007; Scanlon *et al.,* 2014; Win & Mya, 2015; Aziz *et al.,* 2017a). The ability of pteropodid bats to eat fronds and disperse the spores of the bird-nest fern (*Asplenium setoi*) has also been demonstrated in a feeding experiment with *Pteropus pselaphon,* an endemic to islands in Japan (Sugita *et al.*, 2013). Whether *E. spelaea* is specialised nectarivore or feeds opportunistically on other parts of plants remains to be determined. Observations of *E. spelaea*'s feeding behaviour, possibly using camera traps as demonstrated in a study of the locally endangered *P. hypomelanus* (Aziz *et al.*, 2017b), is a promising further avenue of research to determine (i) which part of the plants are being consumed by the bats and (ii) the interactions between the bats and plants (e.g., bats dispersing spores and seeds).

CHAPTER 6: CONCLUSION

DNA barcoding (in general) has proved to be a useful tool for studying the diversity of bats and the diet of plant-visiting bats. Several limitations of this molecular approach exist in this research which may have impacted the interpretation of the results. Nevertheless, the use of DNA barcoding has highlighted the taxonomic uncertainty of many bat species and revealed previously unknown food resources for plant-visiting bats; both findings have significant impacts on ecosystem and associated ecological services, particularly the agricultural industry. Altogether these findings have important implications for conservation of bats, plants and their habitats in the region.

6.1 Assessing the diversity of bats using DNA barcoding

Knowing which species are present in Peninsular Malaysia and their distributions across the region are crucial for developing suitable conservation plans for the bats. Through this study, 110 bat species have been documented in Peninsular Malaysia. However, many of the bat species lack recent and/or precise locality records and were listed as “Least Concerned” by IUCN, consequently suggests the need for further bat surveys and reconsideration of conservation status of these species in the region. More than half of the 110 species did not have DNA barcodes from bats sampled in Peninsular Malaysia, leaving their taxonomic status and presence in the region somewhat unresolved. Based on NJ analyses and the allocation of DNA barcodes to BINs by BOLD, several DNA barcodes recorded under the same species name are likely to represent distinct taxa. Therefore, this study has demonstrated the utility of a comprehensive DNA barcode reference library for assessing the diversity of bats in a particular region. Future work includes (i) more ground surveys to determine the presence of particular bat species in Peninsular Malaysia and collect comparison material (e.g., specimens and DNA

barcodes), and (ii) resolving the taxonomy of highlighted bat species with implications for conservation approaches needed for bats and their habitats in this region.

6.2 Understanding the diet of frugivorous bats using DNA barcoding

The diet of *C. brachyotis* at secondary forest, urban and agricultural sites was compared using DNA barcoding. The high detection rate of *Ficus* seeds in the faeces of *C. brachyotis* indicates that the bats rely heavily on this native food source in all habitats. The fairly high detection rate of introduced and cultivated plants in the faeces suggests that *C. brachyotis* is flexible and can exploit these plants as novel food resource. The diverse diet of *C. brachyotis* at oil palm plantation suggests that the bats are not predominantly feeding on oil palm fruits but cultivated plants nearby. Together these observations suggest an interesting dual role of *C. brachyotis* in dispersing (i) native pioneer plants which aid in forest regeneration, and (ii) non-native plants which potentially facilitate their invasion, consequently suggesting a research avenue that deserves further investigation. The use of DNA barcoding in this study facilitated the detection of plant species that have not been reported in previous dietary studies of *C. brachyotis* but could not identify which part of the plant was consumed by the bats. Nevertheless, the utility of DNA barcoding in dietary studies of frugivorous bats was demonstrated here, consequently revealed the extent to which *C. brachyotis* is capable of adapting to changing landscapes and plant resources. Future research avenues for diet of *C. brachyotis* and other frugivorous bats in the region include (i) the assembly of comprehensive DNA barcode library for plants to facilitate the identification of plants consumed by the bats, and (ii) the use of high-throughput NGS (DNA metabarcoding) which may help to distinguish mixed signals in individuals consuming multiple species.

6.3 Understanding the diet of nectarivorous bats using DNA metabarcoding

This study has demonstrated the utility of DNA metabarcoding in documenting the diet of cave nectar bats roosting in an urban environment. As a result, many native plants which are found in specialised habitats (e.g., mangrove and montane forests) and commercial food crops were detected in the bat faeces. This consequently supports the significant role of *E. spelaea* in pollination of these plants and raises the need for protecting the cave nectar bats in Peninsular Malaysia which are mainly threatened by habitat loss due to rapid urbanisation and limestone mining. The detection of commonly planted introduced (i.e., non-native) plants in the bat faeces indicates that *E. spelaea* in the urban area are exploiting these plants as novel food resources which may adversely impact the reproductive success of native plants. Therefore, it is necessary to consider the status of plant species (i.e., native or non-native) prior planting for urban beautification and agriculture. To date, this study is the first to use DNA metabarcoding to identify the plant material present in faeces of tropical nectarivorous bat and consequently provided important baseline data for future research avenues in diet of tropical nectarivorous bats.

REFERENCES

Abd-Aziz, N. B., Abd-Rahman, M., & Razali, M. Z. (2016). Fruit morphology description of seven jackfruit clones from farmers collection. In *Regional conference on science, technology and social sciences (RCSTSS 2014): Science and Technology* (pp. 549-556). Springer International Publishing.

Aida, N., Sasidhran, S., Kamarudin, N., Aziz, N., Puan, C. L., & Azhar, B. (2016). Woody trees, green space and park size improve avian biodiversity in urban landscapes of Peninsular Malaysia. *Ecological Indicators, 69*, 176-183.

Acharya, P.R., Racey, P.A., Sotthibandhu, S. & Bumrungsri, S. (2015a). Feeding behaviour of the dawn bat (*Eonycteris spelaea*) promotes cross-pollination of economically important plants in Southeast Asia. *Journal of Pollination Ecology 15*(7), 44-50.

Acharya, P. R., Racey, P. A., Sotthibandhu, S., & Bumrungsri, S. (2015b). Home-range and foraging areas of the dawn bat *Eonycteris spelaea* in agricultural areas of Thailand. *Acta Chiropterologica, 17*(2), 307-319.

AMNH. (2012). *Wing punch and hair sampling protocols. American Museum of Natural History, New York.* Retrieved 11 February 2018 from: http://research.amnh.org/vz/mammalogy/donating-bat-tissue-and-hair-samples-genomic-and-stable-isotope-studies/wing-punch-and-hair-sampling

Andersen, K. (1905a). On some bats of the genus *Rhinolophus*, with remarks on their mutual affinities, and descriptions of twenty-six new forms. *Journal of Zoology, 75*(3), 75-144.

Andersen, K. (1905b). On the bats of the *Rhinolophus philippinensis* group, with descriptions of five new species. *The Annals and Magazine of Natural History, 16*, 546 – 550.

Andersen, K. (1911). LXXV.—Six new fruit-bats of the genera *Macroglossus* and *Syconycteris*. *Journal of Natural History, 7*(42), 641-643.

Andersen, K. (1912a). *Catalogue of the Chiroptera in the collection of the British Museum.* London: Trustees.

Andersen, K. (1912b). LXIV.—Brief diagnoses of eight new Petalia, with a list of the known forms of the genus. *Annals and Magazine of Natural History, 10*(59), 546-550.

Andersen, K. (1918). Diagnoses of new bats of the families Rhinolophidae and Megadermatidae. *The Annals and Magazine of Natural History, 2*, 374 – 384.

Anan, S., Rashdi, A. M., Abdul, J., & Lim, B. L. (1998). A survey of small mammals at Sungai Chenderoh, Perlok, Krau Wildlife Reserve, Pahang. *Journal of Wildlife and Parks, 16,* 104–109.

Appanah, S. (1993). Mass flowering of dipterocarp forests in the aseasonal tropics. *Journal of Biosciences, 18*(4), 457-474.

Appleton, B. R., McKenzie, J. A., & Christidis, L. (2004). Molecular systematics and biogeography of the bent-wing bat complex *Miniopterus schreibersii* (Kuhl, 1817) (Chiroptera: Vespertilionidae). *Molecular Phylogenetics and Evolution, 31*(2), 431-439.

Aziz, S. A., Olival, K. J., Bumrungsri, S., Richards, G. C., & Racey, P. A. (2016). The conflict between pteropodid bats and fruit growers: species, legislation and mitigation. In C. Voigt & T. Kingston (Eds.), *Bats in the nthropocene: Conservation of bats in a changing world* (pp. 377-426). Switzerland: Springer International Publishing.

Aziz, S. A., Clements, G. R., Peng, L. Y., Campos-Arceiz, A., McConkey, K. R., Forget, P. M., & Gan, H. M. (2017a). Elucidating the diet of the island flying fox (*Pteropus hypomelanus*) in Peninsular Malaysia through Illumina Next-Generation Sequencing. *PeerJ*, 5, e3176.

Aziz, S. A., Clements, G. R., McConkey, K. R., Sritongchuay, T., Pathil, S., Yazid, A., ... Bumrungsri, S. (2017b). Pollination by the locally endangered island flying fox (*Pteropus hypomelanus*) enhances fruit production of the economically important durian (*Durio zibethinus*). *Ecology and Evolution, 7*(21), 8670–8684.

Azlan, J. M., Sharma, R. S. K., & Zakaria, M. (2000). Species diversity and relative abundance of understorey bats at Air Hitam forest reserve, Selangor, Malaysia. *Malayan Nature Journal, 54*(1), 69-75.

Bates, P. J., Nwe, T., Bu, S. S. H., Mie, K. M., Swe, K. M., Nyo, N., ... Thi, M. M. (2005). A review of the genera *Myotis, Ia, Pipistrellus, Hypsugo,* and *Arielulus* (Chiroptera: Vespertilionidae) from Myanmar (Burma), including three species new to the country. *Acta Chiropterologica, 7*(2), 205-236.

Bates, P. J., Rossiter, S. J., Suyanto, A., & Kingston, T. (2007). A new species of *Hipposideros* (Chiroptera: Hipposideridae) from Sulawesi. *Acta Chiropterologica, 9*(1), 13-26.

Beck, A. J., & Lim, B. L. (1972). Reproductive biology of *Eonycteris spelaea,* Dobson (Megachiroptera) in West Malaysia. *Acta Tropica, 30*(4), 251-260.

Bell, K. L., de Vere, N., Keller, A., Richardson, R. T., Gous, A., Burgess, K. S., & Brosi, B. J. (2016). Pollen DNA barcoding: Current applications and future prospects. *Genome, 59*(9), 629-640.

Bell, K. L., Loeffler, V. M., & Brosi, B. J. (2017). An rbcL reference library to aid in the identification of plant species mixtures by DNA metabarcoding. *Applications in Plant Sciences, 5*(3), 1600110.

Bickford, D., Lohman, D. J., Sodhi, N. S., Ng, P. K., Meier, R., Winker, K., ... Das, I. (2007). Cryptic species as a window on diversity and conservation. *Trends in Ecology & Evolution, 22*(3), 148-155.

Blainville, H. M. D. (1840). *Ostéographie, ou description iconographique des mammiferes récents et fossiles (Carnivores).* Paris.

Blankenberg, D., Kuster, G. V., Coraor, N., Ananda, G., Lazarus, R., Mangan, M., ... Taylor, J. (2010). Galaxy: A web-based genome analysis tool for experimentalists. *Current Protocols in Molecular Biology,* 19.10.

Blyth, E. (1844). Notices of various Mammalia with descriptions of many new species. *Journal of the Asiatic Society of Bengal, 13*, 463 – 494.

Blyth, E. (1848). Report of curator, Zoological Department. *Journal of the Asiatic Society of Bengal, 17*, 247 – 255.

Blyth, E. (1853). Report of curator, Zoological Department. *Journal of the Asiatic Society of Bengal, 22*, 408–417.

Bohmann, K., Evans, A., Gilbert, M. T. P., Carvalho, G. R., Creer, S., Knapp, M., ... De Bruyn, M. (2014). Environmental DNA for wildlife biology and biodiversity monitoring. *Trends in Ecology & Evolution*, *29*(6), 358-367.

Bonhote, J. L. (1903). Report on the mammals. *Fasciculi malayenses, 1*, 1 – 45.

Bonhote, J. L. (1908). Report on the mammals. Report on the Gunong Tahan Expedition, May-September 1905. *Journal of the Federated Malay States Museums*, *3*, 1-11.

Boo, C. M., Chew, S. Y. J., & Yong, J. W. H. (2014). *Plants in tropical cities* (1st Ed.). Singapore.

Boonsong, L., & McNeely, J. A. (1977). *Mammals of Thailand.* Thailand: Association for the Conservation of Wildlife.

Boratyn, G. M., Camacho, C., Cooper, P. S., Coulouris, G., Fong, A., Ma, N., … Zaretskaya, I. (2013). BLAST: A more efficient report with usability improvements. *Nucleic Acids Research, 41,* W29–W33.

Boulter, S. L., Kitching, R. L., & Howlett, B. G. (2006). Family, visitors and the weather: Patterns of flowering in tropical rain forests of northern Australia. *Journal of Ecology*, *94*(2), 369-382.

Braukmann, T. W., Kuzmina, M. L., Sills, J., Zakharov, E. V., & Hebert, P. D. (2017). Testing the efficacy of DNA barcodes for identifying the vascular plants of Canada. *PLoS ONE, 12*(1), e0169515.

Buchannan, F. (1800). XXVII. Description of the *Vespertilio plicatus*. *Transactions of the Linnean Society of London, 5*(1), 261-263.

Bumrungsri, S. (2002). *The foraging ecology of the short-nosed fruit bat, Cynopterus brachyotis (Muller, 1838), in lowland dry evergreen rain forest, southeast Thailand.* (Unpublished doctoral dissertation). University of Aberdeen, Scotland.

Bumrungsri, S., Harbit, A., Benzie, C., Carmouche, K., Sridith, K., & Racey, P. (2008a). The pollination ecology of two species of *Parkia* (Mimosaceae) in southern Thailand. *Journal of Tropical Ecology*, *24*(5), 467-475.

Bumrungsri, S., Francis, C., Csorba, G. (2008b). *Rhinolophus lepidus. The IUCN red list of threatened species 2008:* e.T19547A8971569. Retrieved 22 May 2016 from: http://dx.doi.org/10.2305/IUCN.UK.2008.RLTS.T19547A8971569.en

Bumrungsri, S., Sripaoraya, E., Chongsiri, T., Sridith, K., & Racey, P. A. (2009). The pollination ecology of durian (*Durio zibethinus*, Bombacaceae) in southern Thailand. *Journal of Tropical Ecology*, *25*(1), 85-92.

Bumrungsri, S., Lang, D., Harrower, C., Sripaoraya, E., Kitpipit, K., & Racey, P. A. (2013). The dawn bat, *Eonycteris spelaea* Dobson (Chiroptera: Pteropodidae) feeds mainly on pollen of economically important food plants in Thailand. *Acta Chiropterologica*, *15*(1), 95-104.

Burland, T. M., & Worthington Wilmer, J. (2001). Seeing in the dark: Molecular approaches to the study of bat populations. *Biological Reviews*, *76*(3), 389-409.

Butler, R. (2013a, November 15). *Malaysia has the world's highest deforestation rate, reveals Google forest map.* Retrieved from https://news.mongabay.com/2013/11/malaysia-has-the-worlds-highest-deforestation-rate-reveals-google-forest-map/

Butler, R. (2013b, November 12). *3.5 million ha of Indonesian and Malaysian Forest Converted for Palm Oil in 20 Years.* Retrieved 11 February 2018 from https://news.mongabay.com/2013/11/3-5-million-ha-of-indonesian-and-malaysian-forest-converted-for-palm-oil-in-20-years/

Buth, D. G. (1984). The application of electrophoretic data in systematic studies. *Annual Review of Ecology and Systematics*, *15*(1), 501-522.

Campbell, P., Schneider, C. J., Adnan, A. M., Zubaid, A., & Kunz, T. H. (2004). Phylogeny and phylogeography of Old World fruit bats in the *Cynopterus brachyotis* complex. *Molecular Phylogenetics and Evolution*, *33*(3), 764-781.

Campbell, P., Schneider, C. J., Zubaid, A., Adnan, A. M., & Kunz, T. H. (2007). Morphological and ecological correlates of coexistence in Malaysian fruit bats (Chiroptera: Pteropodidae). *Journal of Mammalogy*, *88*(1), 105-118.

Cantor, T. (1846). Catalogue of mammalia inhabiting the Malayan Peninsula and islands. *Journal of the Asiatic Society of Bengal*, *15*, 171–203.

CBOL Plant Working Group. (2009). A DNA barcode for land plants. *Proceedings of the National Academy of Sciences, 106*(31), 12794-12797.

Chao, A. & Shen, T.J. (2010). *User's guide for program SPADE (Species Prediction and Diversity Estimation).* Retrieved 13 August 2017 from http://chao.stat.nthu.edu.tw/

Chao, A. & Chiu, C. H. (2016). Species richness: Estimation and comparison. *Wiley StatsRef: Statistics Reference Online*, 1-26.

Chasen, F. N. (1940). A handlist of Malaysian mammals: A systematic list of the mammals of the Malay Peninsula, Sumatra, Borneo and Java, including the adjacent small islands. *Bulletin of the Raffles Museum, Singapore, Straits Settlements no. 15, April, 1940.*

Chen, S., Yao, H., Han, J., Liu, C., Song, J., Shi, L., ... Luo, K. (2010). Validation of the ITS2 region as a novel DNA barcode for identifying medicinal plant species. *PLoS ONE*, *5*(1), e8613.

Chen, Y. Y., Satake, A., Sun, I. F., Kosugi, Y., Tani, M., Numata, S., ... Wright, S. J. (2018). Species-specific flowering cues among general flowering Shorea species at the Pasoh Research Forest, Malaysia. *Journal of Ecology*, *106*(2), 586-598.

Clare, E. L., Fraser, E. E., Braid, H. E., Fenton, M. B., & Hebert, P. D. (2009). Species on the menu of a generalist predator, the eastern red bat (*Lasiurus borealis*): using a molecular approach to detect arthropod prey. *Molecular Ecology*, *18*(11), 2532-2542.

Clare, E. L., Symondson, W. O., Broders, H., Fabianek, F., Fraser, E. E., MacKenzie, A., ... Menzies, A. K. (2014). The diet of *Myotis lucifugus* across Canada: Assessing foraging quality and diet variability. *Molecular Ecology*, *23*(15), 3618-3632.

Cooper S. J. B., Reardon T. B., & Skilins J. (1998) Molecular systematics of Australian rhinolophid bats (Chiroptera: Rhinolophidae). *Australian Journal of Zoology, 46,* 203-220.

Corbet, S. A., Beament, J., & Eisikowitch, D. (1982). Are electrostatic forces involved in pollen transfer?. *Plant, Cell & Environment*, *5*(2), 125-129.

Corbet, G. B., & Hill, J. E. (1992). *The mammals of the Indomalayan region: A systematic review (Vol. 488)*. Oxford: Oxford University Press.

Corlett, R. T. (2005). Interactions between birds, fruit bats and exotic plants in urban Hong Kong, South China. *Urban Ecosystems*, *8*(3), 275-283.

Corner, E. J. H. (1997). *Wayside trees of Malaya*. Malaysia: United Selangor Press.

Csorba, G. (1997). Description of a new species of *Rhinolophus* (Chiroptera: Rhinolophidae) from Malaysia. *Journal of Mammalogy*, *78*(2), 342-347.

Csorba, G., Fuisz, T., & Kelen, B. (1997). New birds and bats from Pulau Tioman, Malaysia. *Malayan Nature Journal*, *50*, 197-200.

Csorba, G., & Jenkins, P. D. (1998). First records and a new subspecies of *Rhinolophus stheno* (Chiroptera, Rhinolophidae) from Vietnam. *Bulletin of the Natural History Museum, London (Zoology), 64,* 207 – 211.

Csorba, G., & Lee, L. L. (1999). A new species of vespertilionid bat from Taiwan and a revision of the taxonomic status of *Arielulus* and *Thainycteris* (Chiroptera: Vespertilionidae). *Journal of Zoology*, *248*(3), 361-367.

Csorba, G., Ujhelyi, P., & Thomas, N. (2003). *Horseshoe bats of the world (Chiroptera: Rhinolophidae)*. Shropshire: Alana books.

Csorba, G. (2011). A new species of *Glischropus* from the Indochinese Subregion (Mammalia: Chiroptera: Vespertilionidae). *Zootaxa*, *2925*(1), 41-48.

Csorba, G., Bumrungsri, S., Francis, C., Bates, P., Ong, P., Gumal, M., … Srinivasulu, C. (2014). *Chaerephon plicatus. The IUCN red list of threatened species 2014:* e.T4316A67361960. Retrieved 17 March 2017 from http://dx.doi.org/10.2305/IUCN.UK.2014-3.RLTS.T4316A67361960.en

Csorba, G., Görföl, T., Wiantoro, S., Kingston, T., Bates, P. J., & Huang, J. C. C. (2015). Thumb-pads up—a new species of thick-thumbed bat from Sumatra (Chiroptera: Vespertilionidae: *Glischropus*). *Zootaxa*, *3980*(2), 267-278.

Csorba, G., & Hutson, A. M. (2016) *Nyctalus noctula. The IUCN red list of threatened species 2016:* e.T14920A22015682. Retrieved 16 January 2017 from http://dx.doi.org/10.2305/IUCN.UK.2016-2.RLTS.T14920A22015682.en

Csorba, G., Bumrungsri, S., Francis, C., Görföl, T., Bates, P. J. J. (2016). *Rhinolophus convexus. The IUCN red list of threatened species 2016*: e.T40037A22060825. Retrieved 16 January 2017 from: http://dx.doi.org/10.2305/IUCN.UK.2016-2.RLTS.T40037A22060825.en

Cuvier, F. (1824). *Des dents des mammiferes.* Strasbourg, France: FG Levrault.

Davison, G. W. H., & Zubaid, A. (2007). The status of mammalian biodiversity in Malaysia. In *Proceedings of the seminar and workshop status of biological diversity in Malaysia and threat assessment of plant species in Malaysia, 28-30 June 2005* (pp. 3-27). Malaysia: Forest Research Institute Malaysia.

de Vere, N., Rich, T. C., Ford, C. R., Trinder, S. A., Long, C., Moore, C. W., ... Tatarinova, T. (2012). DNA barcoding the native flowering plants and conifers of Wales. *PLoS ONE*, *7*(6), e37945.

de Vere, N., Jones, L. E., Gilmore, T., Moscrop, J., Lowe, A., Smith, D., ... Ford, C. R. (2017). Using DNA metabarcoding to investigate honey bee foraging reveals limited flower use despite high floral availability. *Scientific Reports*, *7*, 42838.

Delaney, J. T., Jokela, K. J., & Debinski, D. M. (2015). Seasonal succession of pollinator floral resources in four types of grasslands. *Ecosphere*, *6*(11), 1-14.

DeNiro, M. J., & Epstein, S. (1978). Influence of diet on the distribution of carbon isotopes in animals. *Geochimica Et Cosmochimica Acta*, *42*(5), 495-506.

DeNiro, M. J., & Epstein, S. (1981). Influence of diet on the distribution of nitrogen isotopes in animals. *Geochimica Et Cosmochimica Acta*, *45*(3), 341-351.

Desmarest, A. G. (1820). Mammalogie ou description des espèces de mammifères. In *Encyclopédie méthodique part 1*. Paris: Agasse.

Dobson, G. E. (1871). On some new species of Malayan bats from the collection of Dr. Stoliczka. In *Proceedings of the Asiatic Society of Bengal* (pp. 105-108). Calcutta: Baptist Mission Press.

Dobson, G. E. (1873a). Notes on the Pteropidae of India. In *Proceedings of the Asiatic Society of Bengal* (pp. 147–148). Calcutta: Baptist Mission Press.

Dobson, G. E. (1873b). Description of a remarkable new species of *Molossu*s (Nyctinomus) from Johore in the Malay Peninsula. In *Proceedings of the Asiatic Society of Bengal* (pp. 22 – 23). Calcutta: Baptist Mission Press.

Dobson, G. E. (1875). Descriptions of new or little known species of bats. *Proceedings of the Zoological Society of London*, 470 – 474.

Dobson, G. E. (1876). Monograph of the Asiatic Chiroptera and catalogue of the species of bats in the collection of the Indian Museum, Calcutta. *The Annals and Magazine of Natural History, 18*(4), 266 – 268.

Dobson, G. E. (1877). Notes on a collection of Chiroptera from India and Burma. *Journal of the Asiatic Society of Bengal, 46*(2), 310 – 313.

Dobson, G. E. (1878). *Catalogue of Chiroptera in the collection of the British Museum*. London: Bristish Museum.

Dobson, G. E. (1880). XXI.—Description of a new species of *Cynopterus (Ptenochirus)* from Saráwak. *Journal of Natural History, 6*(32), 163-164.

Dool, S. E., Puechmaille, S. J., Foley, N. M., Allegrini, B., Bastian, A., Mutumi, G. L., ... Jacobs, D. S. (2016). Nuclear introns outperform mitochondrial DNA in inter-specific phylogenetic reconstruction: Lessons from horseshoe bats (Rhinolophidae: Chiroptera). *Molecular Phylogenetics and Evolution, 97*, 196-212.

Dormann, C. F., Gruber, B., & Fruend, J. (2008). Introducing the bipartite Package: Analysing Ecological Networks. *R News, 8*(2), 8-11.

DOSM. (2017). *Anggaran penduduk semasa (current population estimates)*. Malaysia: Department of Statistics Malaysia.

Douangboubpha, B., Bumrungsri, S., Soisook, P., Satasook, C., Thomas, N. M., & Bates, P. J. (2010a). A taxonomic review of the *Hipposideros bicolor* species complex and *H. pomona* (Chiroptera: Hipposideridae) in Thailand. *Acta Chiropterologica, 12*(2), 415-438.

Douangboubpha, B., Bumrungsri, S., Soisook, P., Murray, S. W., Puechmaille, S. J., Satasook, C., ... Bates, P. J. (2010b). A taxonomic review of *Hipposideros halophyllus*, with additional information on *H. ater* and *H. cineraceus* (Chiroptera: Hipposideridae) from Thailand and Myanmar. *Acta Chiropterologica, 12*(1), 29-50.

Douangboubpha, B., Bumrungsri, S., Satasook, C., Wanna, W., Soisook, P., & Bates, P. J. (2016). Morphology, genetics and echolocation calls of the genus *Kerivoula* (Chiroptera: Vespertilionidae: Kerivoulinae) in Thailand. *Mammalia, 80*(1), 21-47.

DWNP. (2010). *Red list of mammals for Peninsular Malaysia.* Malaysia: Department of Wildlife and National Parks Peninsular Malaysia.

Ellerman, J. R., & Morrison-Scott, T. C. S. (1966). *Checklist of Palaearctic and Indian Mammals: 1758 to 1946* (*2nd ed*.). London:Trustees of British Museum (Natural History).

Epstein, J. H., Olival, K. J., Pulliam, J. R., Smith, C., Westrum, J., Hughes, T., ... Field, H. E. (2009). *Pteropus vampyrus*, a hunted migratory species with a multinational home-range and a need for regional management. *Journal of Applied Ecology*, *46*(5), 991-1002.

Faeth, S. H., Warren, P. S., Shochat, E., & Marussich, W. A. (2005). Trophic dynamics in urban communities. *AIBS Bulletin, 55*(5), 399-407.

Faure, P. A., Re, D. E., & Clare, E. L. (2009). Wound healing in the flight membranes of big brown bats. *Journal of Mammalogy, 90*(5), 1148-1156.

Fitzherbert, E. B., Struebig, M. J., Morel, A., Danielsen, F., Brühl, C. A., Donald, P. F., & Phalan, B. (2008). How will oil palm expansion affect biodiversity? *Trends in Ecology & Evolution*, *23*(10), 538-545.

Fletcher, C., Akbar, Z., & Kunz, T. H. (2012). Fruit diet of frugivorous bats (*Cynopterus brachyotis* and *Cynopterus horsfieldii*) in tropical hill forests of Peninsular Malaysia. *Mammalia, 76*(4), 389–397.

Fleming, T. H., Geiselman, C., & Kress, W. J. (2009). The evolution of bat pollination: A phylogenetic perspective. *Annals of Botany, 104(*6), 1017-1043.

Francis, C. M. (1989). Notes on fruit bats (Chiroptera, Pteropodidae) from Malaysia and Brunei, with the description of a new subspecies of *Megaerops wetmorei* Taylor, 1934. *Canadian Journal of Zoology, 67*(12), 2878-2882.

Francis, C. M. (1990). Trophic structure of bat communities in the understorey of lowland dipterocarp rain forest in Malaysia. *Journal of Tropical Ecology*, *6*(4), 421-431.

Francis, C. M. (1995). The diversity of bats in Temengor Forest Reserve, Hulu Perak, Malaysia. *Malayan Nature Journal*, *48*, 403-408.

Francis, C. M. (1997). First record for peninsular Malysia of the Gilded Tube-nosed Bat, *Murina rozendaali*. *Malayan Nature Journal*, *50*, 359-362.

Francis, C. M. (2008). *A field guide to the mammals of South-East Asia.* Australia: New Holland Publishers.

Francis, C. M., & Hill, J. E. (1986). A review of the Bornean *Pipistrellus* (Mammalia: Chiroptera). *Mammalia*, *50*(1), 43-56.

Francis, C. M., & Hill, J. E. (1998). New records and a new species of *Myotis* (Chiroptera, Vespertilionidae) from Malaysia. *Mammalia*, *62*(2), 241-252.

Francis, C. M., & Eger, J. L. (2012). A review of tube-nosed bats (*Murina*) from Laos with a description of two new species. *Acta Chiropterologica*, *14*(1), 15-38.

Francis, C. M., Kock, D., & Habersetzer, J. (1999). Sibling species of *Hipposideros ridleyi* (Mammalia, Chiroptera, Hipposideridae). *Senckenbergiana Biologica*, *79*(2), 255-270.

Francis, C. M., Kingston, T., & Zubaid, A. (2007). A new species of *Kerivoula* (Chiroptera: Vespertilionidae) from Peninsular Malaysia. *Acta Chiropterologica*, *9*(1), 1-12.

Francis, C. M, Bates, P. J. J., Kingston, T., & Senawi, J. (2016). *Hipposideros orbiculus. The IUCN red list of threatened species 2016:* e.T136192A22008477. Retrieved 16 January 2017 from: http://dx.doi.org/10.2305/IUCN.UK.2016-2.RLTS.T136192A22008477.en

Francis, C. M., Borisenko, A. V., Ivanova, N. V., Eger, J. L., Lim, B. K., Guillén-Servent, A., ... Hebert, P. D. (2010). The role of DNA barcodes in understanding and conservation of mammal diversity in Southeast Asia. *PLoS ONE*, *5*(9), e12575.

Fu, L., Niu, B., Zhu, Z., Wu, S., & Li, W. (2012). CD-HIT: Accelerated for clustering the next-generation sequencing data. *Bioinformatics*, *28*(23), 3150-3152.

Fujita, M. S., & Tuttle, M. D. (1991). Flying foxes (Chiroptera: Pteropodidae): Threatened animals of key ecological and economic importance. *Conservation Biology*, *5*(4), 455-463.

Fukuda, D., Tisen, O. B., Momose, K., & Sakai, S. (2009). Bat diversity in the vegetation mosaic around a lowland dipterocarp forest of Borneo. *Raffles Bulletin of Zoology*, *57*(1), 213-221.

Funakoshi, K., & Zubaid, A. (1997). Behavioural and reproductive ecology of the dog-faced fruit bats, *Cynopterus brachyotis* and *C. horsfieldi*, in a Malaysian rainforest. *Mammal Study*, *22*, 95-108.

García-Robledo, C., Erickson, D. L., Staines, C. L., Erwin, T. L., & Kress, W. J. (2013). Tropical plant–herbivore networks: Reconstructing species interactions using DNA barcodes. *PLoS ONE*, *8*(1), e52967.

Geoffroy (-Saint Hilaire), E. (1810a). Description des rousettes et des cephalotes. *Annales du Muséum National d'Histoire Naturelle*, *15*, 86–108.

Geoffroy (-Saint Hilaire) E. (1810b). Sur les phyllostomes et les mégadermes. *Annales du Muséum National d'Histoire Naturelle, Paris*, *15*, 157–198.

Geoffroy (-Saint Hilaire), E. (1813). De l'organisation et de la determination des Nycteres une des familles de chauves-souris. *Annales du Muséum National d'Histoire Naturelle, Paris*, *20*, 11 – 20.

Giardine, B., Riemer, C., Hardison, R. C., Burhans, R., Elnitski, L., Shah, P., ... Miller, W. (2005). Galaxy: A platform for interactive large-scale genome analysis. *Genome Research, 15*(10), 1451-1455.

Görföl, T., Estók, P., & Csorba, G. (2013). The subspecies of *Myotis montivagus*-taxonomic revision and species limits (Mammalia: Chiroptera: Vespertilionidae). *Acta Zoologica Academiae Scientiarum Hungaricae, 59*(1), 41-59.

Gotelli, N. J., & Ellison, A. M. (2013). *EcoSimR 1.00.* Retrieved 4 August 2016 from http://www.uvm.edu/~ngotelli/EcoSim/EcoSim.html

Gould, J. (1854). *Mammals of Australia* (vol 3). London.

Gould, E. (1978). Foraging behavior of Malaysian nectar-feeding bats. *Biotropica*, 184-193.

Gould, E. (1988). Wing-clapping sounds of *Eonycteris spelaea* (Pteropodidae) in Malaysia. *Journal of Mammalogy, 69*(2), 378-379.

Gray, J. E. (1838). A revision of the genera of bats. *Magazine of Zoology and Botany, Edinburgh and Dublin, 2*, 483 – 505.

Gray, J. E. (1842). Descriptions of some new genera and fifty unrecorded species of Mammalia. *The Annals and Magazine of Natural History (Series 1), 10*, 255–267.

Gray, J. E. (1843). *List of the specimens of the Mammalia in the collection of the British Museum*. London: British Museum.

Gray, J. E. (1846). *Catalogue of the specimens and drawings of Mammalia and birds of Nepal and Thibet.* London: British Museum.

Grimm, N. B., Faeth, S. H., Golubiewski, N. E., Redman, C. L., Wu, J., Bai, X., & Briggs, J. M. (2008). Global change and the ecology of cities. *Science, 319*(5864), 756-760.

Grismer, L. L., Onn, C. K., Anuar, S., & Muin, M. A. (2014). Cyrts in the city: A new Bent-toed Gecko (Genus *Cyrtodactylus*) is the only endemic species of vertebrate from Batu Caves, Selangor, Peninsular Malaysia. *Zootaxa, 3774*(4), 381-394.

Hajibabaei, M., Singer, G. A., Hebert, P. D., & Hickey, D. A. (2007). DNA barcoding: How it complements taxonomy, molecular phylogenetics and population genetics. *Trends in Genetics, 23*(4), 167-172.

Hardwicke, T. (1825). Description of a new species of tailed bat (*Taphozous*) found in Calcutta. *Transactions of the Linnean Society of London, 14*, 525 – 526.

Hasan, N. H., Khan, F. A. A., Senawi, J., Ketol, B., Sait, I., & Abdullah, M. T. (2012). A report on bats survey at Air Panas-Gua Musang, Kelantan, Malaysia. *Journal of Tropical Biology & Conservation (JTBC), 9*(2), 156-162.

Hawkins, J., de Vere, N., Griffith, A., Ford, C. R., Allainguillaume, J., Hegarty, M. J., ... Adams-Groom, B. (2015). Using DNA metabarcoding to identify the floral composition of honey: A new tool for investigating honey bee foraging preferences. *PLoS ONE*, *10*(8), e0134735.

Hayes, J. P., Ober, H. K., & Sherwin, R. E. (2009). Survey and monitoring of bats. In T. H. Kunz & S. Parsons (Eds.), *Ecological and behavioral methods for the study of bats* (pp. 112-129). Baltimore: The Johns Hopkins University Press.

Hayward, C. E. (2013). *DNA barcoding expands dietary identification and reveals dietary similarity in Jamaican frugivorous bats* (Master's dissertation, University of Western Ontario, Canada). Retrieved from http://ir.lib.uwo.ca/etd/1697

Hebert, P. D., Cywinska, A., & Ball, S. L. (2003). Biological identifications through DNA barcodes. *Proceedings of the Royal Society of London B: Biological Sciences*, *270*(1512), 313-321.

Heer, K., Helbig-Bonitz, M., Fernandes, R. G., Mello, M. A., & Kalko, E. K. (2015). Effects of land use on bat diversity in a complex plantation–forest landscape in northeastern Brazil. *Journal of Mammalogy*, *96*(4), 720-731.

Heller, K. G., & Volleth, M. (1984). Taxonomic position of "*Pipistrellus societatis*" Hill, 1972 and the karyological characteristics of the genus *Eptesicus* (Chiroptera: Vespertilionidae). *Journal of Zoological Systematics and Evolutionary Research*, *22*(1), 65-77.

Heller, K. G., & Volleth, M. (1989). Fledermäuse aus Malaysia. 1. Beobachtungen zur biologie, morphologie und taxonomie. *Senckenbergiana Biologica*, *69*(4-6), 243-276.

Herrera, M., Gerardo, L., & Martínez Del Río, C. (1998). Pollen digestion by New World bats: Effects of processing time and feeding habits. *Ecology*, *79*(8), 2828-2838.

Herrera M, L. G., Hobson, K. A., Manzo A, A., Estrada B, D., Sánchez-Cordero, V., & Méndez C, G. (2001). The role of fruits and insects in the nutrition of frugivorous bats: Evaluating the use of stable isotope models. *Biotropica*, *33*(3), 520-528.

Hill, J. E. (1962). Notes on some insectivores and bats from Upper Burma. *Proceedings of the Zoological Society of London 139*(1), 119-137.

Hill, J. E. (1963). A review of the genus *Hipposideros*. *Bulletin of British Museum (Natural History): Zoology, 1963-1965 (II*), 1–129.

Hill, J. E. (1964). Notes on some tube-nosed bats, genus *Murina,* from southeastern Asia, with descriptions of a new species and a new subspecies. *Federation Museums Journal*, *8*, 48-59.

Hill, J. E. (1965). Asiatic bats of the genera *Kerivoula* and *Phoniscus* (Vespertilionidae), with a note on *Kerivoula aerosa* Tomes. *Mammalia*, *29*(4), 524 – 556.

Hill, J. E. (1972). The Gunong Benom Expedition 1967: New records of Malayan bats, with taxonomic notes and the description of a new *Pipistrellus*. *Bulletin of the British Museum (Natural History), Zoology, 23*, 21 – 42.

Hill, J. E. (1983). Bats (Mammalia: Chiroptera) from Indo-Australia. *Bulletin of the British Museum (Natural History), Zoology, 45*, 103 – 208.

Hill, J. E., & K. Thonglongya. (1972). Bats from Thailand and Cambodia. *Bulletin of British Museum Natural History, 22*, 173 – 196.

Hill, J. E., & S. Yenbutra. (1984). A new species of the *Hipposideros bicolor* group (Chiroptera: Hipposideridae) from Thailand. *Bulletin of British Museum Natural History, 47*, 77 – 82.

Hill, J. E., & Francis, C. M. (1984). New bats (Mammalia: Chiroptera) and new records of bats from Borneo and Malaya. *Bulletin of the British Museum (Natural History), Zoology Series, 47*(5), 305 – 329.

Hill, J. E., A. Zubaid, & Davison, G. W. H. (1985). *Hipposideros lekaguli*, a new leaf-nosed bat recorded from Peninsular Malaysia. *Malayan Nature Journal, 39*, 147–148.

Hill, J. E., Zubaid, A., & Davison, G. W. H. (1986). The taxonomy of leaf-nosed bats of the *Hipposideros bicolor* group (Chiroptera: Hipposideridae) from southeastern Asia. *Mammalia, 50*(4), 535-540.

Hill, J. E., & Harrison, D. L. (1987). The baculum in the Vespertilioninae (Chiroptera: Vespertilionidae) with a systematic review, a synopsis of *Pipistrellus* and *Eptesicus*, and the descriptions of a new genus and subgenus. *Bulletin of the British Museum (Natural History), Zoology, 52*, 225 – 305.

Hill, J. E., & Zubaid, A. (1989). The Dyak leaf-nosed bat, *Hipposideros dyacorum* Thomas, 1902 (Chiroptera: Hipposideridae) in Peninsular Malaysia. *Mammalia, 53*(2), 307-308.

Hillis, D.M., Moritz, C., & Mable, B. K. (1996). *Molecular systematics*. Canada: Sinauer Associates.

Hodgkison, R., Balding, S. T., Zubaid, A., & Kunz, T. H. (2003). Fruit bats (Chiroptera: Pteropodidae) as seed dispersers and pollinators in a lowland Malaysian rain forest. *Biotropica, 35*(4), 491-502.

Hodgkison, R., Balding, S. T., Zubaid, A., & Kunz, T. H. (2004). Temporal variation in the relative abundance of fruit bats (Megachiroptera: Pteropodidae) in relation to the availability of food in a lowland Malaysian rain forest. *Biotropica, 36*(4), 522-533.

Hodgson, B. H. (1835). Synopsis of the Vespertilionidae of Nepal. *Journal Asiatic Society of Bengal, 4*, 699–701.

Horner, M. A., Fleming, T. H., & Sahey, C. T. (1998). Foraging behaviour and energetics of a nectar-feeding bat, *Leptonycteris curasoae* (Chiroptera: Phyllostomidae). *Journal of Zoology, 244*(4), 575-586.

Horsfield, T. (1823–1824). *Zoological researches in Java.* London: Kingbury, Parbury and Allen.

Horsfield, T. (1855). Brief notices of several new or little-known species of Mammalia, lately discovered and collected in Nepal. *The Annals and Magazine of Natural History (Series 2), 16*, 101 – 114.

Hsieh, T.C., Ma, K.H. & Chao, A. (2016). iNEXT: An R package for rarefaction and extrapolation of species diversity (Hill numbers). *Methods in Ecology and Evolution, 7*(12), 1451-1456.

Huang, J. C. C., Jazdzyk, E. L., Nusalawo, M., Maryanto, I., Wiantoro, S., & Kingston, T. (2014). A recent bat survey reveals Bukit Barisan Selatan Landscape as a chiropteran diversity hotspot in Sumatra. *Acta Chiropterologica, 16*(2), 413-449.

Hughes, A. C., Satasook, C., Bates, P. J., Bumrungsri, S., & Jones, G. (2012). The projected effects of climatic and vegetation changes on the distribution and diversity of Southeast Asian bats. *Global Change Biology, 18*(6), 1854-1865.

Ith, S., Bumrungsri, S., Furey, N. M., Bates, P. J., Wonglapsuwan, M., Khan, F. A. A., ... Thomas, N. M. (2015). Taxonomic implications of geographical variation in *Rhinolophus affinis* (Chiroptera: Rhinolophidae) in mainland Southeast Asia. *Zoological Studies, 54*(1), 31.

IUCN. (2016). *The IUCN red list of threatened species. International Union for Conservation of Nature, Gland.* Retrieved 16 January 2017 from www.iucnredlist.org

Ivanova, N., Kuzmina, M., & Fazekas, A. (2011). *CCDB Protocols: Manual protocol employing centrifugation: Glass fiber plate DNA extraction protocol for plants, fungi, echinoderms and mollusks. Canadian Centre for DNA Barcoding.* Retrieved 10 August 2016 from http://ccdb.ca/site/wp-content/uploads/2016/09/CCDB_DNA_Extraction-Plants.pdf

Ivanova, N. V., & Grainger, C. M. (2008). *CCDB Protocols: Sequencing. Canadian Centre for DNA Barcoding.* Retrieved 10 August 2016 from http://ccdb.ca/site/wp-content/uploads/2016/09/CCDB_Sequencing.pdf

Jayaraj, V. K., Tahir, N. F. D. A., Udin, N. A., Baharin, N. F. K., Ismail, S. K., & Zakaria, S. N. A. (2012a). Species diversity of small mammals at Gunung Stong State Park, Kelantan, Malaysia. *Journal of Threatened Taxa, 4*(6), 2617-2628.

Jayaraj, V. K., Laman, C. J., & Abdullah, M. T. (2012b). A predictive model to differentiate the fruit bats *Cynopterus brachyotis* and *C.* cf. *brachyotis* Forest (Chiroptera: Pteropodidae) from Malaysia using multivariate analysis. *Zoological Studies, 51*(2), 259-271.

Jayaraj, V. K., Daud, S. H. M., Azhar, M. I., Sah, S. A. M., Mokhtar, S. I., & Abdullah, M. T. (2013a). Diversity and conservation status of mammals in Wang Kelian State Park, Perlis, Malaysia. *Check List*, *9*(6), 1439-1448.

Jayaraj, V. K., Azhar, I. S. H. A. M., & Daud, S. H. M. (2013b). A new record of *Chaerephone johorensis* at Mount Jerai, Kedah, Malaysia. *Malayan Nature Journal*, *64*(4), 233-235.

Jayaraj, J. V. K., Tingga, R. C. T., Struebig, M. (2016a). *Aethalops alecto*. *The IUCN red list of threatened species 2016.* Retrieved 16 January 2017 from http://dx.doi.org/10.2305/IUCN.UK.2016-2.RLTS.T565A22028716.en

Jayaraj, J. V. K., Khan, F. A. A., Azhar, I., Wee Chen, E., Ali, M. R. M., Ahmad, A., & Yusoff, A. M. (2016b). Diversity and conservation status of small mammals in Kelantan, Malaysia. *Songklanakarin Journal of Science & Technology*, *38*(2), 213-220.

Jenkins, P. D, & Hill, J E. (1981). The status of *Hipposideros galeritus* Cantor, 1846 and *Hipposideros cervinus* (Gould, 1854) (Chiroptera: Hipposideridae). *Bulletin of the British Museum (Natural History): Zoology*, *41*(5), 279–294.

Jentink, F. A. (1891). Some observations relating *Cynopterus brachyotis* Müller and *Kerivoula pellucida* Waterhouse. *Notes from the Leyden Museum*, *13*(3), 202-206.

Jisming-See, S. W., Sing, K. W., & Wilson, J. J. (2016). DNA barcodes and citizen science provoke a diversity reappraisal for the “ring” butterflies of Peninsular Malaysia (*Ypthima*: Satyrinae: Nymphalidae: Lepidoptera). *Genome*, *59*(10), 879-888.

Joann, C. L., Fletcher, C., Salim, H. M., Rahman, A., Harrison, R. D., & Potts, M. D. (2011). Insectivorous bat assemblage in the hill dipterocarp forest of Temengor Forest Reserve, Peninsular Malaysia. *Malayan Nature Journal*, *63*(3), 569-576.

Joann, C. L., Fletcher, C., & Rahman, K. A. (2013). Spatial effects of virgin jungle reserves (VJR) on the community of insectivorous bats in Peninsular Malaysia. *Journal of Tropical Forest Science*, *25*(1), 118-130.

Kasim, A. (2011). Balancing tourism and religious experience: Understanding devotees' perspectives on Thaipusam in Batu Caves, Selangor, Malaysia. *Journal of Hospitality Marketing & Management*, *20*(3-4), 441-456.

Khan, F. A. A. (2008). *Diversification of Old World Bats in Malaysia: An evolutionary and phylogeography hypothesis tested through the Genetic Species Concept* (Master’s dissertation, Texas Tech University). Retrieved from https://ttu-ir.tdl.org/ttu-ir/handle/2346/19879?show=full

Khan, F. A. A., Vicki, J., Solari, S., Peter, A., Ketol, B., Marni, W., ... Robert, J. (2008). Using genetics and morphology to examine species diversity of Old World bats: Report of a recent collection from Malaysia. *Natural Science Research Laboratory*, *281*, 1-28.

Khan, F. A. A., Solari, S., Swier, V. J., Larsen, P. A., Abdullah, M. T., & Baker, R. J. (2010). Systematics of Malaysian woolly bats (Vespertilionidae: *Kerivoula*) inferred from mitochondrial, nuclear, karyotypic, and morphological data. *Journal of Mammalogy*, *91*(5), 1058-1072.

Kimura, M. (1980). A simple method for estimating evolutionary rates of base substitutions through comparative studies of nucleotide sequences. *Journal of Molecular Evolution*, *16*(2), 111-120.

Kingston, T., Jones, G., Akbar, Z., & Kunz, T. H. (1999). Echolocation signal design in Kerivoulinae and Murininae (Chiroptera: Vespertilionidae) from Malaysia. *Journal of Zoology*, *249*(3), 359-374.

Kingston, T., Lara, M. C., Jones, G., Akbar, Z., Kunz, T. H., & Schneider, C. J. (2001). Acoustic divergence in two cryptic *Hipposideros* species: a role for social selection? *Proceedings of the Royal Society of London B: Biological Sciences*, *268*(1474), 1381-1386.

Kingston, T., Lim, B. L., & Akbar, Z. (2006). *Bats of Krau wildlife reserve*. Malaysia: Penerbit Universiti Kebangsaan Malaysia.

Kingston, T. (2010). Research priorities for bat conservation in Southeast Asia: A consensus approach. *Biodiversity and conservation*, *19*(2), 471-484.

Kingston, T. (2013). Response of bat diversity to forest disturbance in Southeast Asia: Insights from long-term research in Malaysia. In S. C. Pedersen & R. A. Adams (Eds.), *Bat evolution, ecology, and conservation* (pp. 169-185). New York: Springer.

Kitchener, D. J., How, R. A., Cooper, N. K., & Suyanto, A. (1992). *Hipposideros diadema* (*Chiroptera hipposideridae*) in the Lesser Sunda Islands, Indonesia: Taxonomy and geographic morphological variation. *Records of the Western Australian Museum*, *16*(1), 1-60.

Kloss, C. B. (1921). Seven new Malayan mammals. *Journal of the Federated Malay States Museums*, *10*, 229-234.

Kock, D., Altmann, J., & Price, L. (2000). A fruit bat new to West Malaysia: *Rousettus leschenaultii* (Desmarest 1820) in Batu Caves. *Malayan Nature Journal*, *54*(1): 63–67.

Koopman, K. F. (1994). Chiroptera: Systematics. *Handbook of Zoology*. *Mammalia, part 60*. Berlin: Walter de Gruyte.

Kuo, H. C., Chen, S. F., Fang, Y. P., Cotton, J. A., Parker, J. D., Csorba, G., ... Rossiter, S. J. (2015). Speciation processes in putative island endemic sister bat species: False impressions from mitochondrial DNA and microsatellite data. *Molecular Ecology*, *24*(23), 5910-5926.

Kowarik, I. (2011). Novel urban ecosystems, biodiversity, and conservation. *Environmental Pollution*, *159*(8), 1974-1983.

Kress, W. J., & Erickson, D. L. (2007). A two-locus global DNA barcode for land plants: The coding rbcL gene complements the non-coding trnH-psbA spacer region. *PLoS ONE, 2*(6), e508.

Kress, W. J., García-Robledo, C., Uriarte, M., & Erickson, D. L. (2015). DNA barcodes for ecology, evolution, and conservation. *Trends in Ecology & Evolution, 30*(1), 25-35.

Kress, W. J. (2017). Plant DNA barcodes: Applications today and in the future. *Journal of Systematics and Evolution, 55,* 291–307.

Kruskop, S. V. (2011). New data on the bat fauna of Con Dao Islands (Ba Ria—Vung Tau Province, Vietnam). *Russian Journal of Theriology, 10,* 37-46.

Kuhl, H. 1817. Die Deutschen Fledermäuse. *Annalen der Wetter auischen Gesellschaft für die Gesammte Naturkunde. Frankfurt am Main, 4,* 11–49 and 185–215.

Kumar, S., Stecher, G., & Tamura, K. (2016). MEGA7: Molecular Evolutionary Genetics Analysis version 7.0 for bigger datasets. *Molecular Biology and Evolution, 33*(7), 1870-1874.

Kuzmina, M., & Ivanova, N. (2011a). *CCDB Protocols: Primer sets for plant and fungi. Canadian Centre for DNA Barcoding.* Retrieved 10 August 2016 from http://ccdb.ca/site/wp-content/uploads/2016/09/CCDB_PrimerSets-Plants.pdf

Kuzmina, M., & Ivanova, N. (2011b). *CCDB Protocols: PCR amplification for plants and fungi. Canadian Centre for DNA Barcoding.* Retrieved 10 August 2016 from http://ccdb.ca/site/wp-content/uploads/2016/09/CCDB_Amplification-Plants.pdf

Kuzmina, M. L., Johnson, K. L., Barron, H. R., & Hebert, P. D. (2012). Identification of the vascular plants of Churchill, Manitoba, using a DNA barcode library. *BMC Ecology, 12*(1), 25.

Leach, W. E. (1821). The characters of three new genera of bats without foliaceous appendages to the nose; The characters of seven genera of bats with foliaceous appendages to the nose. *Transactions of the Linnean Society of London, 13,* 69-82.

Lee, P. S., Gan, H. M., Clements, G. R., & Wilson, J. J. (2016). Field calibration of blowfly-derived DNA against traditional methods for assessing mammal diversity in tropical forests. *Genome, 59*(11), 1008-1022.

Lim, B. L., & Ratnam, L. (1999). A study of the vertebrate fauna in the Tasek Chini Nature Reserve, Pahang in relation use. *Malayan Nature Journal, 53,* 217-238.

Lim, B. L., Majid, R. A., & Norsham, Y. (1999a). Studies on the mammal fauna of Bukit Kutu Wildlife reserve, Hulu Selangor. *Journal of Wildlife and* Parks, *17,* 1-6.

Lim, B. L., Lim, K. K. P., & Yong, H. S. (1999b). The terrestrial mammals of Pulau Tioman, peninsular Malaysia, with a catalogue of specimens at the Raffles Museum, National University of Singapore. *Raffles Bulletin of Zoology, 47,* 101-124.

Lim, L. S. (2012). *Assemblage and genetic structure of insectivorous bats in Peninsular Malaysia* (Doctoral dissertation, Queen Mary University of London, UK). Retrieved from http://qmro.qmul.ac.uk/jspui/handle/123456789/8544

Lim, L. S., Mohd-Adnan, A., Zubaid, A., Struebig, M. J., & Rossiter, S. J. (2014). Diversity of Malaysian insectivorous bat assemblages revisited. *Journal of Tropical Ecology, 30*(2), 111-121

Lim, L. S., Csorba, G., Wong, C. M., Zubaid, A., Rahman, S. P. H., Kumaran, J. V., ... Görföl, T. (2016). The systematic position of *Hypsugo macrotis* (Chiroptera: Vespertilionidae) and a new record from Peninsular Malaysia. *Zootaxa*, *4170*(1), 169-177.

Lim, V. C., Ramli, R., Bhassu, S., & Wilson, J. J. (2017). A checklist of the bats of Peninsular Malaysia and progress towards a DNA barcode reference library. *PLoS ONE, 12*(7), e0179555.

Linnaeus, C. (1758). *Systema naturae* (10th ed., vol. 1). Holmiae: Impensis Direct. Laurentii Salvii.

Maeda, K. (1982). Studies on the classification of Miniopterus in Eurasia, Australia and Melanesia. *Honyurui Kagaku (Mammalian Sci.), 1*, 176.

Mao, X., Tsagkogeorga, G., Bailey, S. E., & Rossiter, S. J. (2017). Genomics of introgression in the Chinese horseshoe bat (*Rhinolophus sinicus*) revealed by transcriptome sequencing. *Biological Journal of the Linnean Society, 121*(3), 698-710.

Matveev, V. A. (2005). Checklist of Cambodian bats (Chiroptera), with new records and remarks on taxonomy. *Russian Journal of Theriology*, *4*(1), 43-62.

McConkey, K. R., Prasad, S., Corlett, R. T., Campos-Arceiz, A., Brodie, J. F., Rogers, H., & Santamaria, L. (2012). Seed dispersal in changing landscapes. *Biological Conservation*, *146*(1), 1-13.

Medway, L. (1966). The Ulu Gombak field studies centre. *Malayan Scientist*, *2*, 1-16.

Medway, L. (1969). *The wild mammals of Malaya*. London: Oxford University Press.

Meyer, A. (1994). Shortcomings of the cytochrome b gene as a molecular marker. *Trends in Ecology & Evolution, 9*(8), 278-280.

Meyer, C. P., & Paulay, G. (2005). DNA barcoding: Error rates based on comprehensive sampling. *PLoS Biology, 3*(12), e422.

Miller, G. S. (1898). List of bats collected by Dr. W. L. Abbot in Siam. In *Proceedings of the Academy of Natural Sciences of Philadelphia* (pp. 316 – 325). Logan Square: Academy of Natural Sciences.

Miller, G. S. (1905). A new genus of bats from Sumatra. *Proceedings of Biological Society Washington, 18,* 229 – 230.

Mohd, R. A., & Abdullah, M. T. (2012). Population genetics of the cave-dwelling dusky fruit bat, *Penthetor lucasi*, based on four populations in Malaysia. *Pertanika Journal of Tropical Agricultural Science*, *35*(3), 459-483.

Mohd-Hanif, R. M. D., Nur-Aida, M. T., Zahirunisa, A. R., Mohd-Ridwan, A. R., & Abdullah, M. T. (2015). Contribution of regenerated forest in conservation of bats in Peninsular Malaysia. *Journal of Tropical Forest Science*, *27*(4), 506-516.

Morales, C. L., & Traveset, A. (2009). A meta-analysis of impacts of alien vs. native plants on pollinator visitation and reproductive success of co-flowering native plants. *Ecology Letters*, *12*(7), 716-728.

Morni, M. A., Tahir, A., Diyana, N. F., Rosli, Q. S., Dee, J. W., Azhar, I., ... Anwarali Khan, F. A. (2016). New record of *Rhinolophus chiewkweeae* (Chiroptera: Rhinolophidae) from the east coast of Peninsular Malaysia with new information on their echolocation calls, genetics and their taxonomy. *Raffles Bulletin of Zoology*, *64*, 242-249.

Moseley, M. (2009). Estimating diversity and ecological status of cave invertebrates: Some lessons and recommendations from Dark Cave (Batu Caves, Malaysia). *Cave and Karst Science*, *35*(1), 47-52.

Moseley, M., Lim, T. W., & Lim, T. T. (2012). Fauna reported from Batu Caves, Selangor, Malaysia: Annotated checklist and bibliography. *Cave and Karst Science*, *39*(2), 77-92.

Müller, S. (1838). Over eenige nieuwe zoogdieren van Borneo. *Tijdschrift voor Naturlike Geschiedenis en Physiologie*, *1838*, 134-150.

Murray, S. W., Campbell, P., Kingston, T., Zubaid, A., Francis, C. M., & Kunz, T. H. (2012). Molecular phylogeny of hipposiderid bats from Southeast Asia and evidence of cryptic diversity. *Molecular Phylogenetics and Evolution*, *62*(2), 597-611.

Muscarella, R., & Fleming, T. H. (2007). The role of frugivorous bats in tropical forest succession. *Biological Reviews*, *82*(4), 573-590.

Nakamoto, A., Kinjo, K., & Izawa, M. (2007). Food habits of Orii's flying-fox, *Pteropus dasymallus inopinatus*, in relation to food availability in an urban area of Okinawa-jima Island, the Ryukyu Archipelago, Japan. *Acta Chiropterologica*, *9*(1), 237-249.

NCBI, R. C. (2016). Database resources of the National Center for Biotechnology Information. *Nucleic Acids Research*, *45*(D1), D12.

Nor Zalipah, M., Anuar, S., Sah, M., & Jones, G. (2016). The potential significance of nectar-feeding bats as pollinators in mangrove habitats of Peninsular Malaysia. *Biotropica*, *48*(4), 425-428.

Norsham, Y., Shariff, F., Norhayati, A., Nordin, M., & Lim, B. L. (1999). Pre-logging survey of mammal fauna at Sungai Weng sub-catchment, Ulu Muda Forest Reserve Forest Reserve, Kedah. *Journal of Wildlife and Parks, 17*, 28–43.

Nowak, R. M. (1994). *Walker's bats of the world.* USA: Johns Hopkins University Press.

Oleksy, R., Racey, P. A., & Jones, G. (2015). High-resolution GPS tracking reveals habitat selection and the potential for long-distance seed dispersal by Madagascan flying foxes *Pteropus rufus*. *Global Ecology and Conservation, 3*, 678-692.

Page, R. D. (2016). DNA barcoding and taxonomy: Dark taxa and dark texts. *Philosophical Transaction of the Royal Society B, 371*(1702), 20150334.

Pallas, P. S. (1767). *Spicilegia zoologica quibus novae imprimis et obscurae animalium species iconibus, descriptionibus atque commentariis illustrantur*. Berlin.

Palsbøll, P. J. (1999). Genetic tagging: Contemporary molecular ecology. *Biological Journal of the Linnean Society, 68*(1-2), 3-22.

Peters, W. (1861). Uber die von Hrn. F. Jagor bisher auf Malacca, Borneo, Java und den Philippinen gesammelten Säugethiere aus den Ordnungen der Halbaffen, Pelzflatterer und Flederthiere. *Monatsberichte Koniglichen Akademie der Wissenschaften, 6*, 706 – 712.

Peters, W. (1866a). Neue oder ungenügend bekannte Flederthiere (*Vampyrops, Uroderma, Chiroderma, Ametrida, Tylostoma, Vespertilio, Vesperugo*) und Nager (*Tylomys, Lasiomys*). *Monatsberichteder Königlich Preussischen Akademie der Wissenschaften zu Berlin* (pp. 392 – 411).

Peters, W. (1866b). Über einige neue oder weniger bekannte Flederthiere. *Monatsberichte der Königlichen Preussische Akademie der Wissenschaften zu Berlin, 1866*, 16-24.

Peters, W. (1867). On some mammals collected by Capt. AC Beavan, CMZS, at Moulmein, Burma. *Proceedings of the Zoological Society of London, 1866*, 426 – 430.

Peters, W. (1868). Mittheilung u¨ber die von dem hon. marquis giacomo doria in Sarawak aud Borneo gesammelten Flederthiere. *Auszug aus dem Monatsbericht Ko¨niglichen Akademie der Wissenschaften zu Berlin, 1868*, 626 – 627.

Peters, W. (1871). Über die gattungen und arten der hufeisennasen, Rhinolophi. *Monatsberichte der Königlich Preussischen Akademie der Wissenschaften zu Berlin, 1871*, 301-332.

Peters, W. (1872). Mitteilung über neue Flederthiere. *Monatsberichteder Königlich Preussischen Akademie der Wissenschaften zu Berlin, 1872,* 256–264.

Phillipps, Q., & Phillipps, K. (2016). *Phillipps' field guide to the mammals of Borneo and their ecology: Sabah, Sarawak, Brunei, and Kalimantan*. New Jersey: Princeton University Press.

Phua, P. B., Corlett. R. T. (1989). Seed dispersal by the lesser short-nosed fruit bat (*Cynopterus brachyotis*, Pteropodidae, Megachiroptera). *Malayan Nature Journal*, *42*, 251-256.

Pianka, E. R. (1974). Niche overlap and diffuse competition. *Proceedings of the National Academy of Sciences*, *71*(5), 2141-2145.

Piggott, M. P., & Taylor, A. C. (2003). Remote collection of animal DNA and its applications in conservation management and understanding the population biology of rare and cryptic species. *Wildlife Research*, *30*(1), 1-13.

Pompanon, F., Deagle, B. E., Symondson, W. O., Brown, D. S., Jarman, S. N., & Taberlet, P. (2012). Who is eating what: Diet assessment using next generation sequencing. *Molecular Ecology*, *21*(8), 1931-1950.

Price, L. (2002). Dark Cave surveys. *Malaysian Naturalist*, *55*(3), 38-39.

Prosser, S. W., & Hebert, P. D. (2017). Rapid identification of the botanical and entomological sources of honey using DNA metabarcoding. *Food Chemistry*, *214*, 183-191.

R Core Team. (2017). *R: A language and environment for statistical computing.* Vienna, Austria: R Foundation for Statistical Computing.

Ratnam, L., Anan, S., Muda, H. (1989). Studies on a collection of bats. *Journal of Wildlife and Parks*, *8*, 9–16.

Ratnasingham, S., & Hebert, P. D. (2007). BOLD: The Barcode of Life Data System (http://www. barcodinglife. org). *Molecular Ecology Resources*, *7*(3), 355-364.

Ratnasingham, S., & Hebert, P. D. (2013). A DNA-based registry for all animal species: The Barcode Index Number (BIN) system. *PLoS ONE*, *8*(7), e66213.

Richardson, B. J., Baverstock, P. R., & Adams, M. (1986). *Allozyme electrophoresis: A handbook for animal systematics and population studies*. London: Academic Press Limited.

Ridley, H.N. (1922). *The flora of Malay Peninsula* (vol 1). Suffolk: Richard Clay & Sons.

Robinson, H. C., & C. B. Kloss. (1911). On new animals from the Malay Peninsula and adjacent islands. *Journal of the Federated Malay States Museums, 4*, 241 – 246.

Rödder, D., & Engler, J. O. (2011). Quantitative metrics of overlaps in Grinnellian niches: Advances and possible drawbacks. *Global Ecology and Biogeography*, *20*(6), 915-927.

Rognes, T., Mahé, F., & Xflouris (2015, February 19). *vsearch: VSEARCH version 1.0.16.* Retrieved from https://zenodo.org/record/15524#.W01y1NgzZQI

Rossiter, S. J., Burland, T. M., Jones, G., & Barratt, E. M. (1999). Characterization of microsatellite loci in the greater horseshoe bat *Rhinolophus ferrumequinum*. *Molecular Ecology*, *8*(11), 1959-1960.

Rossiter, S. J. (2009). Parentage and kinship analysis in bats. In T. H. Kunz & S. Parsons (Eds.), *Ecological and behavioral methods for the study of bats* (pp. 695-714). Baltimore: The Johns Hopkins University Press.

Roslan, A., David, G., Muhamad-Aidil, Z., Fathihi-Hakimi, R., Nur Izzah, I. A., & Abdullah, M. T. (2016). A new distributional record of *Chaerephon johorensis* (Chiroptera: Mollosidae) at Belukar Bukit, Hulu Terengganu, Terengganu, Malaysia. *Journal of Wildlife and Parks, 31*, 61-65.

Sanborn, C. C. (1931). Bats from Polynesia, Melanesia, and Malaysia. *Publications of the Field Museum of Natural History Zoological Series, 18*, 7–29.

Sanborn, C. C. (1939). Eight new bats of the genus *Rhinolophus. Publications of the Field Museum of Natural History Zoological Series, 24*, 37 – 43.

Scanlon, A.T., Petit, S., Tuiwawa, M. & Naikatini, A. (2014). High similarity between a bat-serviced plant assemblage and that used by humans. *Biological Conservation, 174*, 111-119.

Schneider, A., Mertes, C. M., Tatem, A. J., Tan, B., Sulla-Menashe, D., Graves, S. J., … Dastur, A. (2015). A new urban landscape in East–Southeast Asia, 2000–2010. *Environmental Research Letters, 10*(3), 034002.

Schreber, J. C. (1775). *die säugethiere. abbildungen nach der natur mit beschreibungen* (vol 1). Erlangen: Wolfgang Walther.

Sessions, S. K. (1996). Chromosomes: Molecular cytogenetics. In D.M. Hillis, C. Moritz, & B. K Mable (Eds.), *Molecular systematics* (pp.121-168). Sunderland, MA: Sinauer Associates.

Shafie, N. J., Sah, S. A. M., Latip, N. S. A., Azman, N. M., & Khairuddin, N. L. (2011). Diversity pattern of bats at two contrasting habitat types along Kerian River, Perak, Malaysia. *Tropical Life Sciences Research*, *22*(2), 13.

Shahfiz, M. A., Sahrul-Anuar, M. S., Shukor, M. N., Zalipah, M. N., Pan, K. A., Muin, M. A., ... Nik-Fathul, R. (2008a). Survey of small mammals at two forest reserves in Cameron Highlands, Pahang, Peninsular Malaysia. In *Proceedings for Forestry Department of Peninsular Malaysia, 3-5 August 2008* (pp. 293–298). Malaysia: Department of Wildlife and National Parks.

Shahfiz, M. A., Sahrul-Anuar, M. S., Nor-Zalipah, M., Nurul-Ain, E., Pan, K. A., Muin, M. A., … Fadhil, A. (2008b). Surveys of small mammals at selected sites in Royal Belum State Park. In *Proceedings for Forestry Department of Peninsular Malaysia, 4-5 November 2008* (pp. 299 – 308). Malaysia: Department of Wildlife and National Parks.

Shahfiz, M. A., Sahrul-Anuar, M. S., Nor-Zalipah, M., Kaviarasu, M., Pan, K. A., Yusof, M. O., … Nordin, A. (2009). Surveys of bats at selected sites in Melaka Forest Reserves, Peninsular Malaysia. *In Proceedings for Forestry Department of Peninsular Malaysia, 1-2 April 2009* (pp. 265 – 270). Malaysia: Department of Wildlife and National Parks.

Shahfiz, M. A., Shahrul-Anuar, M. A., Kaviarasu, M., Fauzi, M. Z., Rizal, M. R. (2013). Preliminary checklist of mammals at Sungai Enam in Temengor Forest Reserve, Perak, Peninsular Malaysia. In *Proceedings of the 2nd Temengor Scientific Expedition 2012, 22-25 August 2013* (pp. 197-214). Malaysia: Pulau Banding Foundation.

Sheherazade, Yasman, Pradana, D. H. & Tsang, S. M. (2017). The role of fruit bats in plant community changes in an urban forest in Indonesia. *Raffles Bulletin of Zoology*, *65*, 497–505.

Shokralla, S., Spall, J. L., Gibson, J. F., & Hajibabaei, M. (2012). Next-generation sequencing technologies for environmental DNA research. *Molecular Ecology, 21*(8), 1794-1805.

Simmons, N. B. (2005). Order chiroptera. In D. E. Wilson & D. M. Reeder (Eds.), *Mammal species of the world: A taxonomic and geographic reference* (pp. 312-529). Baltimore: Johns Hopkins University Press.

Sing, K. W., Syaripuddin, K., & Wilson, J. J. (2013). Changing perspectives on the diversity of bats (Mammalia: Chiroptera) at Ulu Gombak since the establishment of the Field Study Centre in 1965. *The Raffles Bulletin of Zoology. 29,* 211-217.

Soisook, P., Bumrungsri, S., Satasook, C., Thong, V. D., Bu, S. S. H., Harrison, D. L., & Bates, P. J. (2008). A taxonomic review of *Rhinolophus stheno* and *R. malayanus* (Chiroptera: Rhinolophidae) from continental Southeast Asia: An evaluation of echolocation call frequency in discriminating between cryptic species. *Acta Chiropterologica*, *10*(2), 221-242.

Soisook, P., Karapan, S., Satasook, C., Thong, V. D., Khan, F. A. A., Maryanto, I., ... Bates, P. J. (2013). A review of the *Murina cyclotis* complex (Chiroptera: Vespertilionidae) with descriptions of a new species and subspecies. *Acta Chiropterologica*, *15*(2), 271-292.

Soisook, P., Karapan, S., Srikrachang, M., Dejtaradol, A., Nualcharoen, K., Bumrungsri, S., ... Buś, M. M. (2016). Hill forest dweller: A new cryptic species of *Rhinolophus* in the '*pusillus* group'(Chiroptera: Rhinolophidae) from Thailand and Lao PDR. *Acta Chiropterologica*, *18*(1), 117-139.

Soltis, P. S., & Soltis, D. E. (2003). Applying the bootstrap in phylogeny reconstruction. *Statistical Science, 18*(2), 256-267.

Srithongchuay, T., Bumrungsri, S., & Sripao-raya, E. (2008). The pollination ecology of the late-successional tree, *Oroxylum indicum* (Bignoniaceae) in Thailand. *Journal of Tropical Ecology, 24*(05), 477-484.

Start, A. N. (1974). *The feeding biology in relation to food sources of nectarivorous bats (Chiroptera: Macroglossinae) in Malaysia*. (Unpublished doctoral dissertation). University of Aberdeen, Scotland.

Start, A. N., & Marshall, A. G. (1976). Nectarivorous bats as pollinators of trees in West Malaysia. In J. Burley & B. T. Styles (Eds.), *Tropical trees: Variation, breeding and conservation* (pp. 141–149). London: Academic Press.

Stewart, A. B., & Dudash, M. R. (2017). Flower-visiting bat species contribute unequally toward agricultural pollination ecosystem services in southern Thailand. *Biotropica, 49*(2), 239-248.

Struebig, M. J., Huang, J. C. C., Mohamed, N. Z., Noerfahmy, S., Schöner, C. R., Schöner, M. G., & Francis, C. M. (2017). Forest surveys extend the range of the Krau woolly bat (*Kerivoula krauensis*) in the Malay-Thai Peninsula, Borneo and Sumatra. *Mammalia, 81*(2), 211-215.

Sugita, N., Ootsuki, R., Fujita, T., Murakami, N., & Ueda, K. (2013). Possible spore dispersal of a bird-nest fern *Asplenium setoi* by Bonin flying foxes *Pteropus pselaphon*. *Mammal study, 38*(3), 225-229.

Syaripuddin, K., Kumar, A., Sing, K. W., Halim, M. R. A., Nursyereen, M. N., & Wilson, J. J. (2014). Mercury accumulation in bats near hydroelectric reservoirs in Peninsular Malaysia. *Ecotoxicology, 23*(7), 1164-1171.

Tamrin, N. A. M., Rahman, M. R. A., Daud, M. H. R. M., Hassan, N., Abdullah, M. T. (2010). A survey on small mammals in the Royal Belum Park, Perak, Malaysia. In *Proceeding of National Biodiversity Seminar 2008, 7–14 December 2008* (pp. 27–35). Malaysia: Department of Wildlife and National Parks.

Tan, K. H., Zubaid, A., Kunz, T. H. (1998). Food habits of *Cynopterus brachyotis* (Müller)(Chiroptera: Pteropodidau) in Peninsular Malaysia. *Journal of Tropical Ecology, 14*, 299-307

Tan, K. H., Zubaid, A., & Kunz, T. H. (2000). Fruit dispersal by the lesser dog-faced fruit bat, *Cynopterus brachyotis* (Müller)(Chiroptera: Pteropodidae). *Malayan Nature Journal*, *54*(1), 57-62.

Tate, G. H. H. (1941). A review of the genus *Hipposideros* with special reference to Indo-Australian species. *Bulletin of the American Museum of Natural History, 78*, 353 – 393.

Tate, G. H. H. (1943). Further notes on the *Rhinolophus philippinensis* group (Chiroptera). *American Museum Novitates, 1219*, 1 – 5.

Tate, G. H. H. (1947). *Mammals of Eastern Asia*. New York: The Macmillan Company.

Taylor, E. H. (1934). *Philippine land mammals. Monograph No. 30*. Manila: Bureau of Science, Department of Agriculture and Commerce, Government of the Philippine Islands.

Temminck, C. J. (1824-1827). Vues générales sur l'ordre des cheiroptères. In *Monographies de mammalogie, ou description de quelques genres de mammifères, dont les espèces dont été observées dans les différens musées de l'Europe* (pp. 157-204). Paris: G. Dufour and E. D'Ocagne.

Temminck, C. J. (1834). Over een geslack der vleugelhandige zoogdieren. *Tijdschrift voor Natuurlijke Geschiedenis en Physiologie, 1*, 1–30.

Temminck, C. J. (1837-1841). *Monographies de mammalogie.* Leiden, Paris: Tome 2.

Temminck, C. J. (1853). *Esquisses zoologiques sur la côte de Guiné... le partie, les mammifères vol 1. Mammiferes.* Leiden: Brill.

Thabah, A., Rossiter, S. J., Kingston, T., Zhang, S., Parsons, S., Mya, K. M., ... Jones, G. (2006). Genetic divergence and echolocation call frequency in cryptic species of *Hipposideros larvatus* sl.(Chiroptera: Hipposideridae) from the Indo-Malayan region. *Biological Journal of the Linnean Society*, *88*(1), 119-130.

Thavry, H., Cappelle, J., Bumrungsri, S., Thona, L., & Furey, N. M. (2017). The diet of the Cave Nectar Bat (*Eonycteris spelaea* Dobson) suggests it pollinates economically and ecologically significant plants in Southern Cambodia. *Zoological Studies*, *56*(17), 1-7.

Thomas, O. (1890). XXXVI.—Diagnosis of a new *Cynopterus* from Borneo. *Journal of Natural History*, *5*(27), 235-236.

Thomas, O. (1894). Descriptions of two new bats of the genus *Kerivoula*. *The Annals and Magazine of Natural History, 14*, 460 – 462.

Thomas, O. (1898a). Description of a new bat from north Borneo. *The Annals and Magazine of Natural History, 1*, 243.

Thomas, O. (1898b). Description of a new bat from Selangor. *The Annals and Magazine of Natural History, 7*(1), 360 – 362.

Thomas, O. (1902). A new *Hipposideros* from Borneo. *The Annals and Magazine of Natural History, 9*, 271 – 272.

Thomas, O. (1904). In Schneider, G. Ergebnisse zoologischer Forschungsreisen in Sumatra. *Zoologischer Anzeiger, 27*, 722 – 724.

Thomas, O. (1905). A new genus and two new species of bats. *The Annals and Magazine of Natural History, 16*, 572 – 576.

Thomas, O., & Wroughton, R. C. (1909). On a collection of mammals from western Java presented to the National Museum by Mr. WE Balston. *Proceedings of the Zoological Society of London, 79*(2), 371 – 393.

Thomas, O. (1913). Some new Ferae from Asia and Africa. *The Annals and Magazine of Natural History, 12*, 88 – 92.

Thomas, O. 1915. On bats of the genera *Nyctalus, Tylonycteris* and *Pipistrellus. Annals and Magazine of Naural History, 15*, 225–232.

Thomas, O. (1916a). Scientific results from the mammal survey, XIII. *Journal of the Bombay Natural History Society*, *24*, 404-430.

Thomas, O. (1916b). List of Microchiroptera, other than leaf-nose bats, in the collection of the Federated Malay States Museums. *Journal of the Federated Malay States Museums, 7*(1), 1–6.

Thomas, O. (1923a). XXVII.—On some small mammals, chiefly bats, from the East Indian Archipelago. *Journal of Natural History*, *11*(62), 250-255.

Thomas O. (1923b). Exhibition of the skill of a pygmy fruit-bat from Sumatra. In *Proceedings Zoological Society of London.* London: Academic Press.

Thomas O. (1923c). Scientific results from the mammal survey. XLI. On the forms contained in the genus *Harpiocephalus. Journal of the Bombay Natural History Society, 29*, 88 – 89.

Thonglongya, K. (1973). First record of *Rhinolophus paradoxolophus* from Thailand. *Mammalia, 37*, 587 – 597.

Thonglongya, K., & Hill, J. E. (1974). A new species of *Hipposideros* (Chiroptera) from Thailand. *Mammalia*, *38*(2), 285-294.

Tian, L., Liang, B., Maeda, K., Metzner, W., & Zhang, S. (2004). Molecular studies on the classification of *Miniopterus schreibersii* (Chiroptera: Vespertilionidae) inferred from mitochondrial cytochrome *b* sequences. *Folia Zoologica*, *53*(3), 303-311.

Tingga, R. C. T., Anwarali, F. A., Ridwan, A. M., Senawi, J., & Abdullah, M. T. (2012). Small mammals from Kuala Atok, Taman Negara Pahang, Malaysia. *Sains Malaysiana*, *41*(6), 659-669.

Tomes, R. F. (1858). Notice of five species of bats in the collection of L. L. Dillwyn, ESQ., M.P.; collected in Labuan by Mr. James Motley. In *Proceeding of the Zoological Society of London* (pp. 536–540). London: Taylor and Francis.

Tsang, S. M., Cirranello, A. L., Bates, P. J., & Simmons, N. B. (2016). The roles of taxonomy and systematics in bat conservation. In C. Voigt & T. Kingston (Eds.), *Bats in the anthropocene: Conservation of bats in a changing world* (pp. 503-538). Switzerland: Springer International Publishing.

Tuler, A. C., Carrijo, T. T., Nóia, L. R., Ferreira, A., Peixoto, A. L., & da Silva Ferreira, M. F. (2015). SSR markers: A tool for species identification in *Psidium* (Myrtaceae). *Molecular Biology Reports*, *42*(11), 1501-1513.

Uni, S., Udin, A. S. M., Agatsuma, T., Saijuntha, W., Junker, K., Ramli, R., ... Martin, C. (2017). Morphological and molecular characteristics of *Malayfilaria sofiani* Uni, Mat Udin & Takaoka ng, n. sp.(Nematoda: Filarioidea) from the common treeshrew *Tupaia glis* Diard & Duvaucel (Mammalia: Scandentia) in Peninsular Malaysia. *Parasites & Vectors*, *10*(1), 194.

Vahl, M. (1797). Beskrivelse paa tre nye Arter Flagermuse. Skrivt. Naturhist. *Selskabet Kjobenhavn, 4*, 121-38.

Van Cakenberghe, V., & De Vree, F. (1993). The systematic status of Southeast Asian *Nycteris* (Chiroptera: Nycteridae). *Mammalia, 57*(2), 227-244.

van Velzen, R., Weitschek, E., Felici, G., & Bakker, F. T. (2012). DNA barcoding of recently diverged species: Relative performance of matching methods. *PLoS ONE*, *7*(1), e30490.

Vanitharani, J., Rajendran, A., Bates, P. J., Harrison, D. L., & Pearch, M. J. (2003). A taxonomic reassessment of *Kerivoula lenis* Thomas, 1916 (Chiroptera: Vespertilionidae) including a first record from peninsular India. *Acta Chiropterologica, 5*(1), 49-60.

Voigt, C. C., Matt, F., Michener, R., & Kunz, T. H. (2003). Low turnover rates of carbon isotopes in tissues of two nectar-feeding bat species. *Journal of Experimental Biology*, *206*(8), 1419-1427.

Voigt, C. C., Kelm, D. H., Bradley, B., & Ortmann, S. (2009). Dietary analysis of plant-visiting bats. In T. H. Kunz & S. Parsons (Eds.), *Ecological and behavioral methods for the study of bats* (pp. 593-609). Baltimore: The Johns Hopkins University Press.

Voigt, C. C., & Kingston, T. (2016). Bats in the anthropocene. In C. Voigt & T. Kingston (Eds.) *Bats in the anthropocene: Conservation of bats in a changing world* (pp. 1-9). New York: Springer International Publishing.

Volleth, M., Loidl, J., Mayer, F., Yong, H. S., Müller, S., & Heller, K. G. (2015). Surprising genetic diversity in *Rhinolophus luctus* (Chiroptera: Rhinolophidae) from Peninsular Malaysia: description of a new species based on genetic and morphological characters. *Acta Chiropterologica, 17*(1), 1-20.

Waterhouse, G. R. (1845). Descriptions of bats collected in the Phillipine Islands. In *Proceedings of the Zoological Society of London, 13,* 3–10.

White, T. J., Bruns, T., Lee, S. J. W. T., & Taylor, J. W. (1990). Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. *PCR Protocols: A Guide to Methods and Applications, 18*(1), 315-322.

Wiantoro, S., Maryanto, I., & Abdullah, M. T. (2012). Phylogeny and phylogeography of *Myotis muricola* (Gray, 1846)(Chiroptera: Vespertilionidae) from the West and East of Wallace's Line inferred from partial mtDNA cytochrome *b* gene. *Pertanika Journal of Tropical Agricultural Science*, *35*(2), 271-292.

Wilson, J. J., Sing, K. W., & Sofian-Azirun, M. (2013). Building a DNA barcode reference library for the true butterflies (Lepidoptera) of Peninsula Malaysia: What about the subspecies?. *PLoS ONE*, *8*(11), e79969.

Wilson, J. J., Sing, K. W., Halim, M. R. A., Ramli, R., Hashim, R., & Sofian-Azirun, M. (2014). Utility of DNA barcoding for rapid and accurate assessment of bat diversity in Malaysia in the absence of formally described species. *Genetic Molecular Research*, *13*(1), 920-925.

Wilson, J. J., Sing, K. W., Lee, P. S., & Wee, A. K. (2016). Application of DNA barcodes in wildlife conservation in tropical East Asia. *Conservation Biology*, *30*(5), 982-989.

Wilson, E. O. (2017). Biodiversity research requires more boots on the ground. *Nature Ecology & Evolution*, *1*(11), 1590.

Win, S.S. & Mya, K.M. (2015). The diet of the Indian Flying Fox *Pteropus giganteus* (Brünnich. 1782) (Chiroptera: Pteropodidae) in Myanmar – conflicts with local people? *Journal of Threatened Taxa*, *7*(9): 7568–7572.

Wong, K. M. (1989). Rubiaceae. In T. C. Whitemore (Ed.), *Tree flora of Malaya: A manual for foresters* (vol 4, pp. 472-478). Malaysia: Longman.

Yatim, S. H. (1983). A preliminary survey on inventory habitat and wildlife in the Ulu Langat Forest Reserve and Sungai Dusun Game Reserve. *Journal of Wildlife and Parks, 2,* 119–140.

Yatim, S. H., Baatu, Z., Saaidun, K. A., & Marzuki, M. I. (1985). Survey of mammals and bird species in six different habitats: Mangrove, lowland dipterocarp, hill dipterocarp, upper dipterocarp, montane oak and montane ericaceous forest. *Journal of Wildlife and Parks, 4,* 65–91.

Yeap, C. A., & Zubaid, A. (2002). *Harpiocephalus mordax* (Thomas 1923), a new record for Taman Negara, Peninsular Malaysia. *Malayan Nature Journal, 56*(2), 195–197.

Yeap, C. A. (2003). A brief survey of Microchiropterans at Bukit Dendong and Bukit Keluang, Tembila, Terengganu. *Malayan Nature Journal*, *56*(4), 397–401.

Yoshiyuki, M., & Lim, B. L. (2005). A new horseshoe bat, *Rhinolophus chiewkweeae* (Chiroptera, Rhinolophidae), from Malaysia. *Bulletin of the National Science Museum, Tokyo*, *Series A, 31*, 29-36.

Zhao, L. Z., Bu, Y. Z., Zhou, H. X., Zhou, H. W., Zhang, Z. X., & Niu, H. X. (2015). Differences in *Hipposideros pomona* from three geographical regions in China based on morphology and molecular sequences data. *Journal of Mammalogy*, *96*(6), 1305-1316.

Zubaid, A. (1988a). Food habits of *Hipposideros armiger* (Chiroptera: Rhinolophidae) from Peninsular Malaysia. *Mammalia, 52*(4), 585-588.

Zubaid, A. (1988b). Food habits of *Hipposideros pomona* (Chiroptera: Rhinolophidae) from peninsular Malaysia. *Mammalia*, *52*(1), 134-137.

Zubaid, A. (1988c). The second record of a trident horseshoe bat, *Aselliscus stoliczkanus* (Hipposideridae) from Peninsular Malaysia. *Malayan Nature Journal, 42,* 29 – 30.

Zubaid, A., & Davison, G. W. H. (1988). *Rhinolophus marshalli*, a new horshoe bat recorded from Peninsular Malaysia. *Sains Malayana, 17*(3), 367 – 368.

Zubaid, A. (1993). A comparison of the bat fauna between a primary and fragmented secondary forest in Peninsular Malaysia. *Mammalia*, *57*(2), 201-206.

Zubaid, A. (1994). Bornean Horseshoe Bat *Rhinolophus borneensis* newly recorded in Peninsular Malaysia. *Malayan Nature Journal, 48,* 37 -38.

LIST OF PUBLICATIONS AND PAPERS PRESENTED

1. **Lim, V. C.**, Ramli, R., Bhassu, S., & Wilson, J. J. (2018). Pollination implications of the diverse diet of tropical nectar-feeding bats roosting in an urban cave. *PeerJ. 6*, e4572.

2. **Lim, V. C.**, Clare, E. L., Littlefair, J. E., Ramli, R., Bhassu, S., & Wilson, J. J. (2018). Impact of urbanisation and agriculture on the diet of fruit bats. *Urban Ecosystems*, *21*(1), 61-70.

3. **Lim, V. C.**, Ramli, R., Bhassu, S., & Wilson, J. J. (2017). A checklist of the bats of Peninsular Malaysia and progress towards a DNA barcode reference library. *PLoS ONE*, *12*(7), e0179555.

4. **Lim, V.C.** (2018, July*). Utilisation of DNA Barcoding in Assessing the Diversity of Bats and Their Phytophagous Diet in Peninsular Malaysia*. Paper presented at the 55th Annual Meeting of the Association for Tropical Biology and Conservation, Malaysia. (Awarded Travel Grant)

5. **Lim, V.C.**, R. Ramli., S. Bhassu & J.J. Wilson. (2017, December). *Diverse diet of cave nectar bats in an urban environment as revealed by DNA metabarcoding.* Paper presented at the 22nd Biological Sciences Graduate Congress 2017, Singapore. (Awarded Best Oral Presentation for theme Biodiversity, Ecology and Environmental Biology).

6. **Lim, V.C.**, R. Ramli., S. Bhassu & J.J. Wilson. (2017, November). *A checklist of the bats of Peninsular Malaysia.* Paper presented at the 8th Biodiversity Seminar 2017, Malaysia.

7. **Lim, V.C.**, R. Ramli., S. Bhassu & J.J. Wilson. (2017, July). *Using DNA metabarcoding to understand the diet and pollination role of Eonycteris spelaea.* Paper presented at the 19th International Botanical Congress, China.

8. **Lim, V.C.**, R. Ramli., S. Bhassu & J.J. Wilson. (2016, November*). A checklist of the bats of Peninsular Malaysia and progress towards a DNA barcode reference library*. Paper presented at the Student Conference on Conservation Science, China. (Awarded Travel and Accommodation Scholarship)

9. **Lim, V.C.**, E. Clare, J. Littlefair, R. Ramli & J.J. Wilson (2016, June). *Fruit for thought: how human activities affect the diet of the frugivorous bat, Cynopterus brachyotis in Peninsular Malaysia.* Paper presented at the Conservation Asia 2016, Singapore.

10. **Lim, V.C.** & J.J. Wilson. (2015, August*). Progress in DNA barcoding the bats of Peninsular Malaysia*. Paper presented at the 6th International Barcode of Life Conference, Canada.