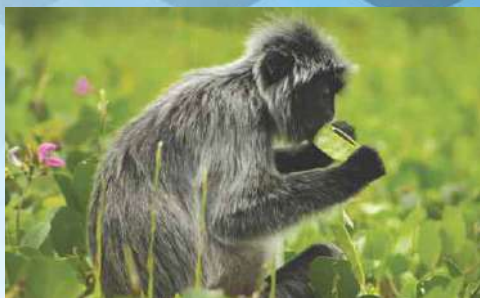
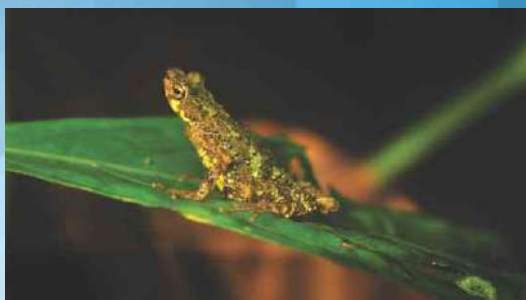


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## Short Notes

# Survey of Freshwater Fish in Kadamaian Area, Western Sabah

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## Abstract

The freshwater fish fauna was surveyed in forest streams located in the Kadamaian area, Sabah during the Borneo Geographic Expedition 2019. Fish samples were obtained from six stations by electrofishing from an upstream to downstream direction. A total of four families, 12 genera, and 14 species of fish were recorded from the present study. The most dominant family was Cyprinidae (10 species; 71.43%), followed by Gastromyzontidae (2 species; 14.29%), Nemacheilidae (1 species; 7.14%), and Mastacembelidae (1 species; 7.14%). The number of species was low in upstream stations at higher altitude but substantially higher in downstream stations with lower altitude. Economically valuable fish, *Tor tambra* was found at all stations. *Gastromyzon monticola* which is endemic to Sabah was also recorded at most stations.

**Keywords:** Freshwater fish, *Tor tambra*, *Gastromyzon monticola*, forest streams, Kadamaian, Sabah

## Introduction

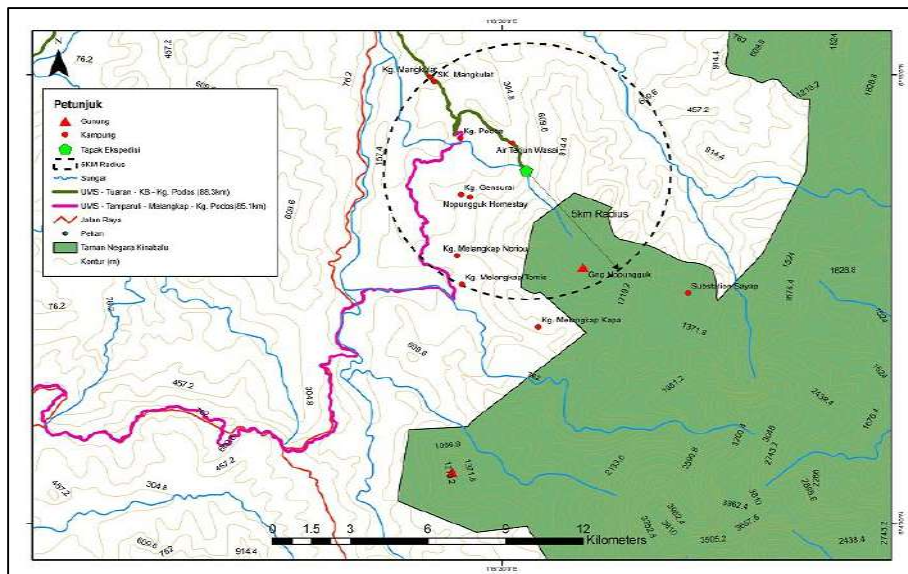
The survey and documentation of freshwater fishes have been continuously undertaken in Sabah (Ahmad et al., 2006; Jimmy et al., 2010; Kottelat & Tan, 2011; Wilkinson & Tan, 2018). Ng et al., (2017) reported that there are approximately 166 freshwater fish species in Sabah. However, documentation of fish fauna in Sabah's forest streams is still limited and patchy, mainly due to unequal sampling efforts and sampling difficulties in rural areas (Ng et al., 2017; Nyanti et al., 2019).

The Borneo Geographic Expedition 2019: Kadamaian, jointly organized by the Institute for Tropical Biology and Conservation (ITBC) and Sabah Parks was held from 14th to 25<sup>th</sup> October, 2019. One of the many objectives of the scientific expedition was to carry out an inventory of fish fauna in Kadamaian area of Kota

Belud, which is located immediately to the north west of Kinabalu Park. The baseline information obtained during the expedition can be utilised as a first step to develop an effective conservation strategy in the area adjacent to Kinabalu Park.

## **Methodology**

A three-day survey of freshwater fish fauna was conducted in Kadamaian area during the Borneo Geographic Expedition 2019 (Figure 1). Freshwater fish samples were obtained from a total of six sampling stations from an upstream to downstream direction covering a total streamline length of 10 km. Stream habitats changed from the upstream to downstream direction where stations in the upstream are shaded with a closed canopy and consist mostly of falls whereas the canopy cover of stations in the downstream are open (Figure 2). GPS coordinates and elevations were taken at each station by using a Portable Global Positioning System (Garmin GPSMAP® 645). Table 1 summarizes the details of sampling regime and station. The fish samples were collected using an electrofishing device powered by a 1000 watt portable generator (Elemex Honda SHX1000). The stunned fish were then collected using scoop nets (1 cm mesh size). The electroshocking process was carried out at a stream length of approximately 200 m. Fish samples were identified to the species level using available taxonomic keys (Inger & Chin, 2002; Kottelat & Tan, 2008; Mohsin & Ambak, 1983; Roberts, 1989; Tan, 2006). Samples that were unidentifiable in the field were first fixed in 10% formalin and later preserved in 70% ethanol for further identification in the laboratory. Finally, the taxonomic status was confirmed with online global databases of fish species (Froese & Pauly, 2019; van der Laan et al., 2020).



**Figure 1.** The map of Kadamaian and study area covered in the present expedition as indicated by dash circle.



**Figure 2.** Upstream habitat (a) with closed canopy cover that is mostly consisted of boulders and falls compared with downstream habitat (b) with open canopy cover.



**Table 1.** The details of the sampling regime and sampling locations in the present study.

Station no./stream	GPS Coordinates	Sampling Date	Elevation (m asl)
St 1: Sg. Malawan	N06°12'01.2" E116°30'50.3"	22 Oct 2019	615
St 2: Sg. Malawan	N06°12'45.3" E116°30'30.7"	22 Oct 2019	565
St 3: Sg. Malawan	N06°12'40.6" E116°30'30.6"	24 Oct 2019	379
St 4: Sg. Kopongian	N06°12'41.1" E116°30'31.2"	24 Oct 2019	381
St 5: Sg. Malawan	N06°13'22.8" E116°28'11.6"	23 Oct 2019	123
St 6: Sg. Kasiaan	N06°13'22.2" E116°28'12.0"	23 Oct 2019	111

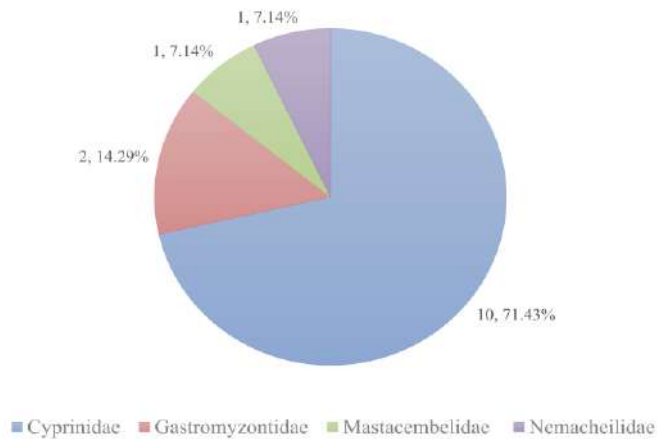
## Results and Discussion

A total of four families, 12 genera, and 14 species were recorded in forest streams located in the Kadamaian watershed. The low number of fish species recorded in the present study is not uncommon given this study has set out to document mainly the diversity of a small distance of forest streams within a short span of time during the expedition. Fish species numbers ranging from two to 19 species were also recorded in previous ichthyofauna surveys that were conducted in a short period of time (Ahmad et al., 2006; Jimmy et al., 2010; Rahim et al., 2002). Apart from the brief collection time, high elevation and low primary productivity of rivers may also be reasons for low number of fish species (Jimmy et al., 2010; Rahim et al., 2002).

Figure 3 shows that the most dominant family was Cyprinidae with 10 species (71.43%), followed by Gastromyzontidae (2 species; 14.29%), Nemacheilidae (1 species; 7.14%), and Mastacembelidae (1 species; 7.14%). Samat (1990) recorded a total of 22 fish species with Cyprinidae being the most dominant family in Kinabalu Park which is adjacent to the present study area. Dominance by the family Cyprinidae in the present forest stream is also similar to that in Maliau Basin Conservation Area, Sabah (Sade & Biun, 2012), Sarawak forest stream (Nyanti et al., 2019), Royal Belum Forest Reserve in Perak (Sharir et al., 2019), Pahang National Park (Farinordin et al., 2016) and Ulu Muda Forest Reserve in Kedah (Sah et al., 2012).

Table 2 shows that the species count was low in upstream stations at higher altitude, but there was substantially higher species count at downstream stations with lower altitude. This phenomenon was also observed in Maliau Basin, Sabah where fish was more diverse at downstream sites (Sade & Biun, 2012). The author attributed the increase of species diversity at the downstream area to the increase in habitat size, habitat diversity, and environmental stability. Nyanti et al., (2019) also demonstrated that elevation is one of the most significant factors related to fish assemblages in the upstream of Baleh River, Sarawak.





**Figure 3.** Number of fish species and percentage of the fish family recorded from six sampling stations in the study area.

*Tor tambra*, locally known as Pelian in Sabah, was recorded at all stations along forest streams. *Tor tambra*, which is known as a focal food species is expensive with high economic value (Ingram et al., 2007; Parenti & Lim, 2005; Pinder et al., 2019; Rachmatika et al., 2005). This fish is commonly found in mountainous forest streams with fast flowing water (Nyanti et al., 2019; Rachmatika et al., 2005; Rahim et al, 2002; Sharir et al., 2019). On the other hand, *Gastromyzon monticola* which is endemic to Sabah (Ng et al., 2017; Tan, 2006) was also found at almost all stations except station 2. Most of the fish species recorded in the present study is corroborated with previous records in Kinabalu Park (Samat, 1990).

## Conclusion

A total of four families, 12 genera and 14 species of fish were recorded in forest streams located in the Kadamaian area, Kota Belud, Sabah. The four families are Cyprinidae, Gastromyzontidae, Nemacheilidae and Mastacembelidae, with the most dominant family being Cyprinidae. Altitudinal changes of fish species number were observed. Further study should be conducted to investigate the altitudinal changes of fish composition and biodiversity associated with environmental variables changes.

Table 2. Occurrence of freshwater fish fauna in the forest streams located in Kadamaian area, Kota Belud, Sabah.

Family	Species	Station 1	Station 2	Station 3	Station 4	Station 5	Station 6
Cyprinidae	<i>Barbodes sealei</i>	-	-	-	-	+	+
Cyprinidae	<i>Garra borneensis</i>	-	-	-	-	+	+
Cyprinidae	<i>Lobacheilus ovalis</i>	-	-	-	-	+	+
Cyprinidae	<i>Nematabramis everetti</i>	-	-	-	-	+	+
Cyprinidae	<i>Osteochilus chini</i>	-	-	-	-	+	-
Cyprinidae	<i>Paracrossochilus acerus</i>	-	-	-	-	-	+
Cyprinidae	<i>Rasbora hubbsi</i>	-	-	-	-	+	+
Cyprinidae	<i>Rasbora argyrotaenia</i>	-	-	-	-	+	-
Cyprinidae	<i>Rasbora sp</i>	-	-	-	-	-	+
Cyprinidae	<i>Tor tambra</i>	+	+	+	+	+	+
Gastromyzontidae	<i>Gastromyzon monticola</i>	+	-	+	+	+	+
Gastromyzontidae	<i>Protomyzon griswoldi</i>	-	-	-	+	-	-
Mastacembelidae	<i>Macragnathus keithi</i>	-	-	-	-	-	+
Nemacheilidae	<i>Nemacheilus olivaceus</i>	-	-	-	-	+	+

Note: + indicated presence whereas - indicated absence.

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## Research Article

# What's flashing in Kadamaian? A Note on Fireflies (Coleoptera: Lampyridae) in Kadamaian, Sabah

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## Abstract

A firefly survey was conducted in Podos, Melangkap Noriou and Pinolobu in Kadamaian, Kota Belud during the Borneo Geographic Expedition Kadamaian 2019. A total of 48 fireflies were collected representing at least seven species consisting mainly of *Luciola* spp. and a single individual of *Pyrocoelia* sp. larva. Identification reveals that two samples are potentially new records to Borneo; cf. *Aquilonia* sp. and *Medeopteryx* sp., and also *Luciola niah* as a new record for Sabah. Furthermore, four samples have unique morphological characters including the potential *Aquilonia* and *Medeopteryx* samples and two *Luciola* samples. Fireflies were caught in various locations such as near rubber plantations, trails and rivers. Findings from this report expands the distributional knowledge about Lampyridae in Borneo.

**Keywords:** Borneo, *Luciola niah*, *Aquilonia* sp., *Medeopteryx* sp., *Pyrocoelia* sp.

## Introduction

One of the ecotourism magnets, the firefly, is a bioluminescent beetle (Coleoptera) with almost 2,000 species estimated worldwide (Branham & Wenzel, 2003; Lewis et al., 2020). Currently, there are limited reports on firefly species distribution in several parts of Asia (Jusoh et al., 2018; Ballantyne et al., 2019). Fireflies in the interior of Sabah are rarely explored. Most firefly research in the Southeast Asian region is mainly focused in mangrove areas, perhaps due to its economic importance in ecotourism (Mahadimenakbar et al., 2014, 2018; Mahadimenakbar & Fiffy Hanisdah, 2016; Siti Rozziana et al., 2020; Syazlina et al., 2016) or due to ease in access compared to fireflies in forested areas.

However, these undiscovered locations may contain new lampyrid species or new geographical records for existing fireflies. For example, the new genus *Emasia* was erected in 2010 after an Otoretine firefly was found in Gunung Emas, Sabah (Bocakova & Janisova, 2010). While *Pygoluciola* was first thought

to be a rare firefly from the Bornean region (Ballantyne, 2008; Ballantyne & Lambkin, 2006), it was later found in several places transgressing biogeographic barriers such as *Pygoluciola cowleyi* from Australia (Ballantyne & Lambkin, 2013), *Pygoluciola qingyu* from China (Fu & Ballantyne, 2008) and *Pygoluciola hamulata* and *Pygoluciola satoi* from the Philippines (Ballantyne, 2008; Ballantyne & Lambkin, 2006). Recently, Nada and Ballantyne (2018) described a new species of *Pygoluciola* from the lowland dipterocarp forest as a result of an intricate sampling in Peninsular Malaysia. This shift into the inland fireflies brought a renewed look at the region's Lampyridae diversity with several new species being described recently (Ballantyne et al., 2019). Hence, we intend to build upon these efforts by exploring new areas to offer more insights on firefly distribution and diversity.

There is lack of information on the species of fireflies that reside in the interior of Sabah and Borneo at large. Via the Borneo Geographic Expedition in 2019, the authors took the opportunity to fill this knowledge gap. The specific purpose of this survey was to answer the question of what types of firefly dwell in Kadamaian, Kota Belud. The data on what firefly species and their number in certain areas served as our baseline for future scientific efforts, such as ecological study and taxonomy.

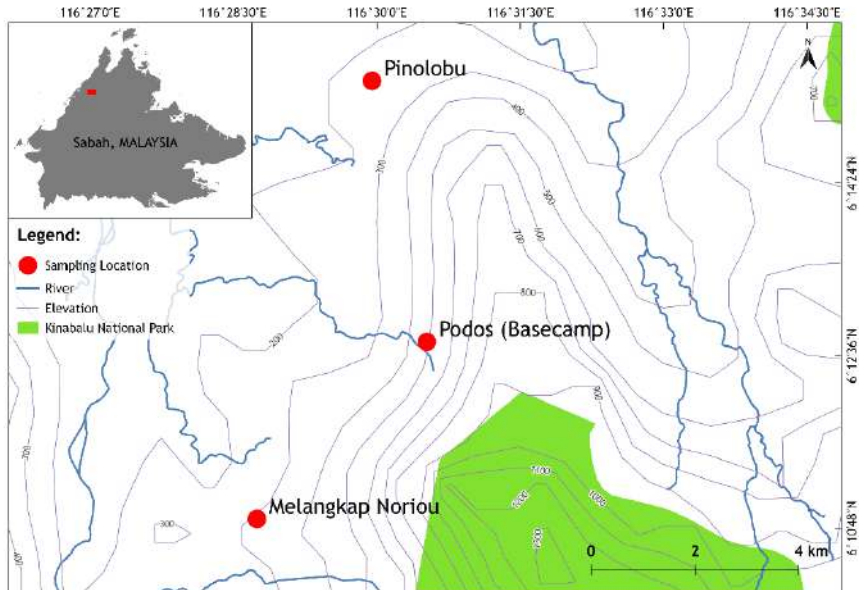
## Methodology

### *Sampling Locations and Method*

Fireflies were sampled using sweep net along the trail in Podos (basecamp), Melangkap Noriou (Noriou henceforth) and Pinolobu (Figure 1) during the Borneo Geographic Expedition from 20<sup>th</sup> to 24<sup>th</sup> October, 2019. Sampling lasted for three hours starting from 7 pm to 10 pm. Sampling locations, such as forest floor, tree, vines or river were noted. Samples were then stored in 95% ethanol and brought to the Institute for Tropical Biology and Conservation, Universiti Malaysia Sabah in Kota Kinabalu for identification.

### *Identification Process*

Male samples were identified by morphological characters using the key provided in Ballantyne et al., (2019). Male genitalia were extracted by soaking detached abdomen in 10% KOH for approximately 10 hours (Sasso Porto et al., 2016). Female specimens were identified to species level where description is available from the key or found mating with a known male. Specimens were photographed using Leica DFC495 attached to a Leica M165C stereomicroscope



**Figure 1.** Sampling location. Inset: Kadamaian area marked in red in the northwest part of Sabah, Malaysia.

and inspected using the software ImageJ for measurements (Bourne et al., 2019; Mobilim & Mahadimenakbar, 2020).

## Results and Discussion

After surveying three locations in Kadamaian, we found a total of 48 fireflies (Table 1) mainly dominated by *Luciola* spp. (66.66% out of total) including a single *Pyrocoelia* larva. Upon identification, we found new records of fireflies for Sabah and Borneo Island and four potentially new species. However, the suspected new species will be further investigated and will be described in detail elsewhere. Here, we only report the general descriptions of these potential new species. For the first time, the existence of cf. *Aquilonia* sp. and *Medeopteryx* sp. in Borneo Island as well as *Luciola niah* in Sabah are reported. In this survey, we observed the occurrence of cf. *Aquilonia* sp. in Pinolobu (Table 1) where the distribution was formerly recorded in Australia, New Guinea and Pacific islands (Australinea). Based on the dichotomous key by Ballantyne et al. (2019) for the identification of South East Asia and Australopacific fireflies, our sample falls into the *Atyphella* complex fireflies. This complex consists of five genera; *Atyphella*, *Aquilonia*, *Convexa*, *Lloydiella* and *Magnalata*, show emargination on the right side of their aedeagal sheath (Figure 2B and C). Interestingly, only *Aquilonia* fireflies have pale brownish or orange-yellow dorsal with dark elytral



Table 1. Fireflies caught in different areas of Kadamaian.

Species	Area			Total
	Noriou	Pinolobu	Podos	
<i>Luciola niah</i> (Jusoh 2019)	15 (6)	4 (3)	4 (2)	23 (11)
<i>Luciola pallidipes</i> (Pic 1928)	3 (1)	3	0	6 (1)
<i>Pygoluciola wittmeri</i> (Ballantyne 1968)	1	8 (3)	0	9 (3)
<i>Pyrocoelia</i> sp. larva (Gorham 1880)	0	0	1	1
cf. <i>Aquilonia</i> sp. (Ballantyne 2009)	0	1	0	1
<i>Medeopteryx</i> sp. (Ballantyne 2013)	2 (1)	1	0	3 (1)
<i>Luciola</i> sp. (Laporte 1833)	2	0	0	2
<i>Luciola</i> sp. (Laporte 1833)	1	0	0	1
Unidentified female	0	0	2	2
<b>Total</b>	<b>24 (8)</b>	<b>17 (6)</b>	<b>7 (2)</b>	<b>48 (16)</b>

Note: Firefly numbers indicate total individual and in brackets indicate numbers of female only.

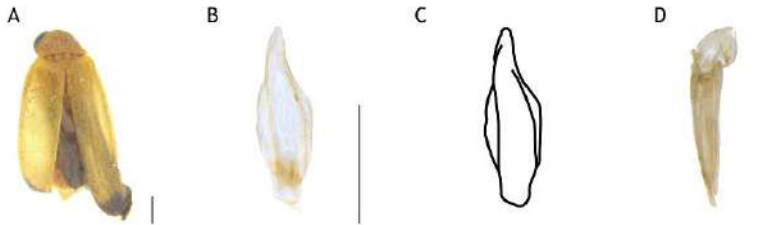
apices (Ballantyne & Lambkin, 2009) which fit the description of our sample (Figure 2A). The other four genera have orange pronotum with either dark or dark brown elytra.

Table 2. Firefly sampling locations.

Species	Area		
	Noriou	Pinolobu	Podos
<i>Luciola niah</i> (Jusoh 2019)	Rubber estate & trail	Trail	Near Basecamp
<i>Luciola pallidipes</i> (Pic 1928)	River	River	-
<i>Pygoluciola wittmeri</i> (Ballantyne 1968)	River	River	-
<i>Pyrocoelia</i> sp. larva (Gorham 1880)	-	-	Forest Floor
cf. <i>Aquilonia</i> sp. (Ballantyne 2009)	-	Trail	-
<i>Medeopteryx</i> sp. (Ballantyne 2013)	Rubber estate	Rubber estate	-
<i>Luciola</i> sp. (Laporte 1833)	Trail	-	-
<i>Luciola</i> sp. (Laporte 1833)	Trail	-	-
Unidentified female	-	-	Vines

Earlier, we suspected that this sample belongs to the *Luciola* genus based on its size; our sample measures 5.9 mm long as opposed to the smallest *Aquilonia* to be in the range of 7.2 - 9.7 mm (Ballantyne & Lambkin, 2009), and colour. However, microscopic investigation into its genitalia proved otherwise; the aedeagal median lobe (ML) of our sample is not curved and the apices of the lateral lobe (LL) is narrower (Figure 2D) while *Luciola* possesses strongly curved

ML and expanded LL apices (shown clearly in figures 277 - 279 in Ballantyne et al., 2019, p. 94). *Aquatica* was also considered in identification, however, our sample lacks toothed aedeagal sheath sternite (figure 22 in Fu et al., 2010, p. 10).



**Figure 2.** Dorsal part of the cf. *Aquilonia* sp. male (A); Aedeagal sheath ventral (B) with illustration (C); Aedeagus lateral (D). Scale lines are 1mm.

Although *Aquilonia* is restricted to the Australian region, they are now potentially distributed to the west of New Guinea. Additionally, a sample from Mantailang, Tenom was also collected by Mobilim in 2019 shared similar characteristics to cf. *Aquilonia similismessoria* (unpublished data). This genus has never been discovered on the west of the Wallace line, hence, *Aquilonia* existence in Borneo is unexpected. However, a growing number of literature using hyperdiverse animal shows these biogeographic lines are permeable. This explains the occurrences of the same taxonomic member in different biogeographic regions for example the *Trigonopterus* weevils (Letsch et al., 2020) and multiple genera of butterflies (Condamine et al., 2015; Toussaint et al., 2020) in Australinea, Wallacea and Sundaland. Our findings reflect this emerging data. In fact, in Lampyridae itself, current records show *Medeopteryx* fireflies have a wide distribution transgressing the biogeographic line. Ballantyne & Lambkin (2013) erected *Medeopteryx* from a bent-winged firefly that used to be in the *Pteroptyx* genus and proposed that species under *Medeopteryx* is distributed to the east of Wallace line (Australinea) (Jusoh et al., 2018). However, multiple discoveries have uncovered that member of this genus is distributed in the west of Australinea such as *Medeopteryx hongkongensis* in Hong Kong (Yiu, 2017), *Medeopteryx fraseri* in Peninsular Malaysia, *Medeopteryx timida* in Vietnam and a record from Thailand too (Ballantyne et al., 2019).

From this survey, we also reveal the wide distribution of *Luciola niah* (Figure 3A - D) in Borneo Island. In Kadamaian, *L. niah* is the most common species found in the trails of Noriou, Pinolobu and Podos. The species' first known date of collection is in 2010 from Niah National Park, Sarawak (Malaysian Borneo) and the species was only described by Ballantyne et al. (2019). The species is recognizable via its thin dark line on the base of abdominal ventrite 5 (Figure 3K) and entire light organ in ventrite 7. According to the same literature, there are only two species of *Luciola* that have dark elytral apices and yellowish dorsal; *Luciola jengai* and *Luciola niah*. Two of the other *Luciola* that we caught in Noriou have similar description but possess a unique pattern of dark colouration on their abdominal ventrites V3 to V5 which neither match *L. jengai* nor *L. niah*. Furthermore, the shape of their light organ in V7 is similar with *L. niah*; entire and longer instead of wide.

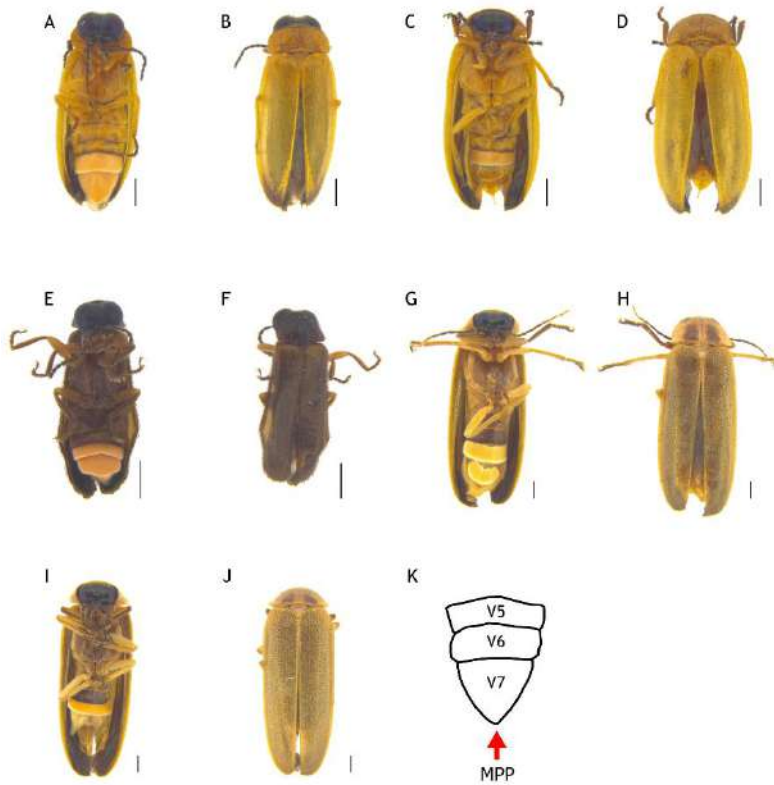
This potentially carries two conclusions; (1) the species are potentially new, or (2) phenotypic polymorphism. Deciding a taxonomical status based on minute morphological differences may be incorrect. This is because intraspecific phenotypic polymorphism may occur which is common in many species (Sánchez-Vialas et al., 2020; Yeong et al., 2018). In Lampyridae research, intraspecific variation was observed, such as the pronotum colour of *Pteroptyx bearni* from West versus East Malaysia and *Pt. tener* from different localities (Jusoh et al., 2018); multiple light spots on the body of *Phausis reticulata* females (De Cock et al., 2014) and small differences in elytral colouration of the new Brazilian *Uanauna angaporan* species (Campello-Gonçalves et al., 2019). This occurrence may be influenced by many factors during an insect development into adulthood such as sexual conflicts, food quality, temperature variations and other abiotic factors (Dillon & Lozier, 2019; Gering, 2017; Lin et al., 2018; Willink et al., 2020). Hence, to accurately arrive at any one of those two conclusions, the samples must undergo further morphological characterization coupled with DNA barcoding (Cognato et al., 2020; Lehmann et al., 2017; Sheth & Thaker, 2017).

The fireflies were found in various locations. *Luciola niah* was found flying within the rubber estate, along the trail and cleared ground of the basecamp. Previously, *L. niah* was found dwelling in the forest track of Niah National Park and several remarks indicating general locations in Kapit, Long Aton in Ulu Baram and Lambir Hill (Ballantyne et al., 2019). This list of areas presumably in forested ground shows contrast with our findings that *L. niah* may be found in disturbed areas too. This is similar with *Medeopteryx* fireflies that were caught in the rubber estate of Noriou and Pinolobu. Though our data is insufficient to conclude ecological associations of land use type with the species distribution,

this observation is similar with several findings in which Lampyridae individuals exist in various habitats including secondary forests (Viviani, 2001; Viviani et al., 2012) and plantations. For example, *Inflata indica* was collected from rubber and palm plantations as well as banana orchard (Ballantyne et al., 2015); *Luciola parvula* from the Japanese cedar plantation (Takehashi et al., 2014); *Pyrocoelia tonkinensis* from rubber plantation (Senarat et al., 2019); and *Diaphanes* sp., *Pyrocoelia* sp. and *Trisinuata* sp. from coniferous plantation (Wattanachaiyingcharoen et al., 2016). Furthermore, our findings on the occurrence of the macropterous *Pygoluciola wittmeri* (Figure 3I - J) echoes the previous findings which was found near the river (Ballantyne & Lambkin, 2006; Kionsom River in Chey, 2008; Mobilim & Mahadimenakbar, 2020). Other than abiotic constraints (Nur Athirah et al., 2020), insect occurrence in certain locations can be influenced by several dispersal factors at different stages of their life cycle. For Lampyridae larva, such as *Pyrocoelia* sp. found in Podos leaf litter (Figure 4), food may be a factor at this phase in which they spend most of their time hunting snails on the forest floor (Jaikla et al., 2020; Kirton et al., 2006; Lewis et al., 2020) with limited dispersal range, as much as 100.7 to 245.4 cm for *Luciola parvula* flightless female in daily recapture rate of mark-release-recapture study (Takehashi et al., 2014). As they grow into adulthood, fireflies with flight ability will be able to disperse more to find mates such as *Luciola substriata* that have the ability to fly ~0.2 - 0.4 metres per second (Fu, 2005 found in Zhang et al., 2020). Hence, species existence in other ecotones can be expected as found in Kadamaian where the plantations are very near undisturbed forests and this potentially contributed to their dispersal capabilities.

## Conclusion

From our data, Kadamaian area holds an interesting array of Lampyridae species and they are found in various landscapes. It widens our distributional knowledge of cf. *Aquilonia* sp., *Medeopteryx* sp. and *Luciola niah*. Although our data is limited to providing a checklist, the accompanying location notes reveal that an important ecological question about Lampyridae distribution in various ecosystems should axis around these diverse land use types. This is to further understand their role in the natural and converted landscape or vice versa on how these changing landscapes impact firefly species. By diversifying Lampyridae research, conservation efforts could also be properly put in place for these areas.



**Figure 3.** *Luciola niah* male (A - B), female (C - D); *Luciola pallidipes* male (E - F); *Pygoluciola wittmeri* male (G - H); female (I - J); abdomen ventral (K). MPP = median posterior projection, V5 = ventrite number 5, V6 = ventrite number 6, V7 = ventrite number 7. Scale lines are 1mm.



**Figure 4.** *Pyrocoelia* sp. larva found on the forest floor along the trail in Podos.

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## **Short Communication**

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# **A Preliminary Survey on Edibles and Medicinal Plants Used By Dusun of Kampung Pinolobu, Kadamaian, Kota Belud, Sabah, Malaysia**

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## **Abstract**

This paper provides a brief enumeration of 22 species of plants and four species of fungi as wild edibles, and 13 species of plants that have been used for medicinal purposes by Dusun people of Kampung Pinolobu, Kadamaian, Kota Belud, Sabah. Seven informants were selected using snowball sampling technique, and data on edibles and medicinal plants were collected using semi structured interviews. This study reveals how Dusun people of Kampung Pinolobu used 13 species of medicinal plants to treat ailments and diseases like fever, high blood pressure, flatulence, rheumatism, cold, cough, and gastric pain. The flower of *Carica papaya* and fruit of *Passiflora foetida* are used for high blood pressure treatment, leaves of *Manihot esculenta* serve two purposes, as traditional vegetables and also consumed to avoid flatulence. Decoction made from the root of *Ficus septica* was given to women during postpartum recovery as they believed that it would help to keep the body warm, treat headache, and stomach pain. Paste made from leaves of *Melastoma malabathricum* is used to treat wounds. Meanwhile crushed leaves of *Hibiscus rosa-sinensis*, root decoction of *Imperata cylindrica*, sap from *Calamus* sp. are used to treat high fever, crushed leaves of *Hibiscus rosa-sinensis* is also used to subside carbuncle. Decoction of young leaves of *Psidium guajava* and *Leucosyke capitella* are used to treat stomach pain. Meanwhile decoction of crushed tuber of *Curcuma longa* is used to treat gastric pain.

**Keywords:** Dusun, Kadamaian, medicinal plants, wild edibles, Sabah.

## **Introduction**

Statistics on mortality and morbidity rates for Malaysia in 2016 indicate an increasing trend for both communicable and non-communicable diseases (Health, 2018). The urgency of producing valuable and innovative treatments have influenced skyrocketing drug discovery research activities. These are associated with a wide range of medicinal drugs testing activities that have been extensively conducted from various natural resources, including plants, animals,

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marine organisms, bacteria, and fungi (Baltz, 2019; Thomford et al., 2018). In fact, natural products remain relevant sources for producing new drugs, as most commercial products originate from natural resources and their derivatives (Heinrich, 2014; Chin et al., 2006).

Plant-derived medicine has proven its significance in drug development, as plant utilization has been rooted since ancient time. The presence of secondary metabolites in plants has not only been shown to be useful for defence systems, but also proven to harbour both preventative and curative effects (Li et al., 2020). As traditional knowledge of plant usage is usually passed from one generation to another, the pharmacological basis of these culturally important plants is what may relate the drug discovery study with ethnopharmacology (Patwardhan, 2005).

Ethnopharmacology research is popular as it always believes in promises for silver bullets. Ethnopharmacology based on Heinrich (2014) is a scientific study of any substances used by humans which can give pharmacological effects to consumers. Broader interpretation of this field of study has included traditional medicinal knowledge of a community as well as their documentation and systematic review. Bruhn & Rivier (2019) devoted reviews for Holmstedt's research in their paper by quoting ethnopharmacology as interdisciplinary exploration in evaluating the remedies in cultural heritage, which later aim for rescue and documentation of these material medica in their aboriginal form. Uniquely, discoveries from the ethnopharmacological side may not only open chances to advancement in medicinal aspects, but also may influence studies in other fields such as socio-culture, history and anthropology (Heinrich, 2014). These may be seen through the mushrooming of research papers related to biological potential of traditional knowledge based on demography or ethnicity. Hence, a scientific study was conducted among selected communities in Sabah, Malaysia to investigate their medicinal practices. The objective of this paper is to properly record edibles and medicinal plants that have been traditionally used by Dusun people in Kampung Pinolobu, Kadamaian, Kota Belud.

## **Methodology**

### *Study Area*

Kampung Pinolobu, Kedamaian (GPS: 6.282968376545483, 116.49234251189354) is located in Kota Belud District. Kota Belud is about an hour's drive from Kota Kinabalu and is a growing township located on the west coast of Sabah (Figure 1). According to Informant 1 (INF1) and also a local Dusun guide who assisted

this survey, Kampung Pinolobu is primarily inhabited by the Dusun tribe while Kota Belud is populated by several ethnic groups, including Bajau, Dusun, Irranun, and Rungus. Informant 1 (INF1) was also a village committee member, he verbally communicated that the villagers of Kampung Pinolobu are working as farmers, rubber tappers, or are self-employed. Kampung Pinolobu is located between the Kampung Podos and Wasai Waterfall Homestay area.

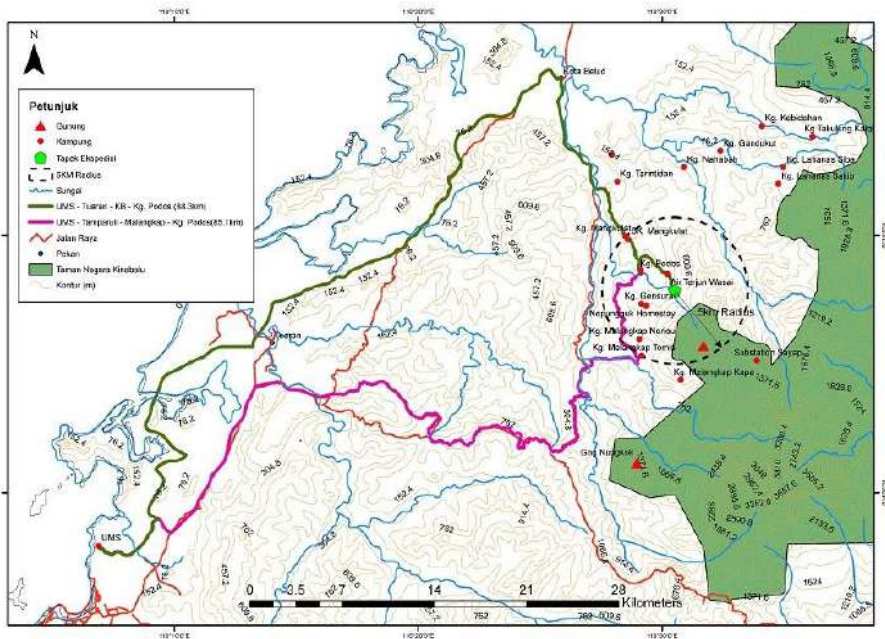


Figure 1. Map of Kota Belud and location of Borneo Geographic Expedition 2019 (in circle).

#### *Data collection and identification*

Data sampling through interviews was carried out during The Borneo Geographic Expedition 2019 on 21<sup>st</sup> October. In Figure 1, the site involved in the expedition is indicated in a dashed-circle-line. In this study, the interviews were conducted by using snowball sampling technique and through semi-structured interviews among seven Dusun informants from Kampung Pinolobu, Kedamaian, Kota Belud. In general, the informants have learned their traditional knowledge of edibles and medicinal plants from the older generation, i.e. their parents and grandparents, as well as through self-experience. None of the informants are qualified as herbalists nor are they formal practitioners of traditional herbal medicine. The interview session was conducted in their house compound using Bahasa Malaysia with a mix of Dusun language. Translation of the Dusun language was assisted by Informant 1 (INF1). The details of informants are shown in Table

1. After the interview, informants provided information about edibles and medicinal plants used by villagers by showing what was available in a nearby bush, just 3 to 5 metres away and around the village. Informants pointed out plants they use during an excursion around the village. Available plant species were photographed and the identification process was supported by Mr. Faiz and Mr. Razy, both of whom are staff from the Sabah Forestry Department, Sandakan. Meanwhile, the holding of the specimens was assisted by Mr. Bartwolomieus Jalius (PPST, UMS).

**Table 1.** Details of the informants from Kampung Pinolobu, Kedamaian, Kota Belud, Sabah

Informant	Age	Gender	Occupation	Education	Race/Religion
INF1	41	Male	Self-employed	University	Dusun/Christian (Seventh Day Adventist)
INF2	28	Female	Housewife	Secondary school (SPM)	Dusun/Christian (Seventh Day Adventist)
INF3	31	Female	Housewife	Primary school	Dusun/Christian (Seventh Day Adventist)
INF4	67	Female	Farmer	None	Dusun/Christian (True Jesus)
INF5	38	Male	Self-employed	Primary school	Dusun/Christian
INF6	50	Male	Farmer/Rubber tapper	Primary school	Dusun/Christian
INF7	54	Female	Farmer/Rubber tapper	None	Dusun/Christian

## Results and Discussion

The study found 22 species of plants and four species of fungi consumed by the Dusun people of Kg. Pinolobu, Kadamaian in their daily life as food plants. Meanwhile, 13 species have been used as medicinal plants. The medicinal plants were administered traditionally in the form of decoction, crushed leaves, paste, eaten, or ingested fresh from the sap. The details of these edible and medicinal plants are shown in Table 2.

The root sap of *Calamus* sp. or locally known as ‘lambah’ is taken orally to cool the body from fever. High fever is also treated by utilizing a decoction of *Imperata cylindrica* root, and by applying crushed young leaves of *Hibiscus rosa-sinensis* to the body. They also use crushed young leaves of *Hibiscus rosa-sinensis* to ease the swollen carbuncle. The practice of using *Imperata cylindrica* as traditional medicine was previously recorded by the Dusun of Tambunan and Murut from Kalabakan (Kulip, 2003; Kulip, 2014). The root of *Ficus septica* or known as ‘lintatobu’ is made into decoction and given to women during

postpartum confinement. The *Ficus septica* root decoction is used for headache and stomach pain treatment.

The boiled flower of *Carica papaya* has been used to reduce high blood pressure. Dusun in Penampang, Tambunan, Keningau have reported using decoction made from the root of *Carica papaya* for birth control after giving birth. This potion is also used to alleviate menstrual pain (Ahmad & Holdsworth, 2003). Meanwhile, decoction made from *Crotalaria pallida* plant stem and leaves is used for cold and cough treatment. Villagers from Kampung Pinolobu drink decoction made from tubers of *Curcuma longa* to treat gastric pain, while Sama Bajau from Kampung Menunggu, Kota Belud use tuber of *Curcuma longa* for post-partum treatment (Awang-Kanak et al., 2018b). Dusun of Kampung Pinolobu also eat fruit of *Passiflora foetida* to reduce high blood pressure. However, the Sama Bajau of Kampung Taun Gusi were previously reported to only consume the fruit (Awang-Kanak et al., 2018a).

## Conclusion

The study has listed 22 plant species and four fungi species consumed by the Dusun of Kampung Pinolobu, Kadamaian in their daily life as food plants. Meanwhile, 13 species have been used as medicinal plants. Vernacular names stated in this paper were based on oral communication with the Dusun of Pinolobu, it may have similarity with Dusun dialects from other districts in Sabah e.g. Tambunan and Keningau. The survey on traditional knowledge of Dusun people from Kampung Pinolobu, Kadamaian ensures that the traditional knowledge on edibles including some species of fungi, and medicinal plants used by the community can be preserved based on sustainable practice. The findings provide a screened set of useful plants for further potential ethnopharmacological research in plant-based medicine and may eventually unveils some valuable phytotherapeutic and traditional medicinal agents.



**Table 2.** List of wild edibles and medicinal plants used by Dusun of Kampung Pinolobu, Kedamaian, Kota Belud, Sabah.  
Fo: Food; Me: Medicinal plant; Asterisk\*: Fungi.

Family	Scientific Name	Local Name (Dusun)	Uses	Informant(s)	Remark	Previous record in Sabah
Amaranthaceae	<i>Amaranthus oleraceus</i>	Bayam kampung	Fo	INF4		Jualang et al. (2016).
Amaryllidaceae	<i>Allium tuberosum</i>	Losun/Lokio	Fo	INF1		Kulip, (2014).
Araceae	<i>Schismatoglottis achmadii</i>	Dukaruk	Fo	INF6, INF7		Kulip, (2014); Kulip, (2003)
Araceae	<i>Colocasia esculenta</i>	Ubi ketadi	Fo	INF7		Nassir & Low, (2015)
Araceae	<i>Calamus</i> sp.	Lambah/Lamba	Me	INF5	Cooling the body from fever	Kulip, (2014).
Areaceae	<i>Arenga undulatifolia</i>	Polud (young shoot)	Fo	INF6, INF7		Kulip, (2014)
Dryopteridaceae	<i>Diplazium esculantum</i>	Pakis	Fo	INF5, INF7		Awang-Kanak et al. (2020); Awang-Kanak et al. (2018a); Jualang et al. (2016).
Asteraceae	<i>Cosmos caudatus</i>	Ransa ransa	Fo	INF1		Kulip, (2014)
Blechnaceae	<i>Stenochalena palustris</i>	Lemiding	Fo	INF1		Awang-Kanak et al. (2018a); Awang-Kanak et al. (2018b)
Caricaceae	<i>Carica papaya</i>	Tepayas	Fo, Me	INF1	Flower is used to lower blood pressure	Nassir & Low, (2015); Kulip, (2014); Ahmad & Holdsworth, (2003)
Convolvulaceae	<i>Ipomoea batatas</i>	Ubi manis	Fo	INF7		Awang-Kanak et al. (2018a); Kulip, (2014)

(Continued on next page)

Table 2. (Continued)

Family	Scientific Name	Local Name (Dusun)	Uses	Informant(s)	Remark	Previous record in Sabah
Cucurbitaceae	<i>Cucumis sativus</i>	Timun	Fo	INF1		Awang-Kanak et al. (2018a)
Dryteridaceae	<i>Diplazium esculantum</i>	Pakis	Fo	INF5, INF7		Kulip, (2014)
Euphorbiaceae	<i>Manihot esculenta</i> (leaf)	Daun ubi kayu	Fo, Me	INF1, INF2, INF3, INF4	To avoid flatulence	Awang-Kanak et al. (2018a); Kulip, (2014)
Euphorbiaceae	<i>Homalanthus populneus</i>	Mato/Dolimato	Me	INF1	For swollen feet/rheumatism treatment	Kulip, (2014); Kulip, (2003)
Fabaceae	<i>Crotalaria pallida</i>	Kirik kirik/ Ngirik ngrik	Me	INF1, INF4	Decoction made from plant for cold and cough treatment	Latiff et al. (2001)
Malvaceae	<i>Hibiscus rosa-sinensis</i> (young leaf)	Pucuk bunga raya	Me	INF1	Crushed leaves can be used to treat carbuncle and fever	Kulip, (2003).
Melastomataceae	<i>Melastoma malabathricum</i>	Gosing	Me	INF4, INF5	Crushed leave to treat wound	Ahmad & Holdsworth, (2003)
Moraceae	<i>Ficus septica</i>	Lintotobou/ Hintotobou	Me	INF2, INF3, INF4	Drink root decoction during post-partum recovery to warm body, to treat stomach pain, and headache	Kulip, (2014)
Musaceae	<i>Musa</i> sp. (inner pith)	Batang pisang (umbut)	Fo	INF1		Awang-Kanak et al. (2018b)
Myrtaceae	<i>Psidium guajava</i> (young leaf)	Pucuk jambu	Fo, Me	INF4	Decoction of use to treat diarrhea	Nassir & Low, (2015); Kulip, (2003)

(Continued on next page)

Table 2. (Continued)

Family	Scientific Name	Local (Dusun)	Name	Uses	Informant(s)	Remark	Previous record in Sabah
Passifloraceae	<i>Passiflora foetida</i> (fruit)	Lapak lapak		Me	INF2	Treat high blood pressure	Awang-Kanak et al. (2018a)
Phyllanthaceae	<i>Baccaurea lanceolata</i>	Liposu/Limposu		Fo	INF1, INF5		Kulip, (2003).
Poaceae	<i>Imperata cylindrica</i>	Paka (lalang)		Me	INF4	Root decoction use to treat fever/anti-pyretic medicine	Kulip, (2014); Kulip, (2003)
Unknown	Unknown	Melopau/Molopau/Malapau		Fo	INF1, INF5		
Urticaceae	<i>Leucosyke capitella</i>	Mandahasih		Me	INF4, INF7	Abdominal pain	Ahmad Holdsworth, (2003) &
Zingiberaceae	<i>Curcuma longa</i>	Kunyit		Fo, Me	INF1, INF7	Tuber decoction use for gastric treatment	Awang-Kanak et al. (2018a); Ahmad Holdsworth, (2003) &
Zingiberaceae	<i>Etilingera coccinea</i>	Tuhau		Fo	INF1		Kulip, (2014)
*Lyophyllaceae	<i>Termitomyces eurhizus</i>	Kulat tamburong		Fo	INF7		Foo et al. 2018
*Schizophyllaceae	<i>Schizophyllum commune</i>	Kulat kodop		Fo	INF7		Foo et al. 2018
*Unknown	Unknown	Kulat purak mata		Fo	INF7		
*Unknown	Unknown	Kulat sorukan		Fo	INF7		

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## Research Article

# Diversity of Herpetofauna in Kadamaian, Kota Belud, Sabah, Malaysia

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## Abstract

A survey on amphibians and reptiles or herpetofauna in Kadamaian, Kota Belud was carried out on 21<sup>st</sup> to 25<sup>th</sup> October, 2019 during the Borneo Geographic Expedition jointly organized by the Institute for Tropical Biology and Conservation (ITBC) of Universiti Malaysia Sabah (UMS) and Sabah Parks. The main purpose of the survey is to update the list of herpetofauna in the study area. Visual Encounter Survey (VES) method was applied and transects were established at the waterfall near the expedition base camp, Pinolobu River, Kipungit River in Melangkap Noriou and Meliawa River, resulting in 15 species of amphibians from five families and six species of reptiles from three families recorded. Significantly, the survey recorded 10 species of herpetofauna that are endemic to Borneo, including one Endangered (EN) frog species namely *Leptobrachella sabahmontana* of the family Megophryidae and one new record for Sabah which is *Ansonia minuta* of the family Bufonidae. Two reptiles out of the recorded list were Bornean endemics namely *Cyrtodactylus baluensis* and *Tropidophorus micropus*. *Fejervarya limnocharis* and *Polypedates leucomystax* seemed to be the common species of amphibians recorded whereas *Eutrophis rudis* was the common reptile species found in the area. The result, in term of species number was low especially for reptiles due to the short survey period. However, this report managed to update the list for herpetofauna species in Kadamaian, Kota Belud. Nevertheless, this study contributes to Borneo's herpetofauna database specifically in Sabah and serves as reference for more studies and research in future.

**Keywords:** Herpetofauna, Amphibians, Reptiles, Kadamaian, Kota Belud, Sabah, Visual Encounter Survey

## Introduction

Herpetofauna is defined as amphibians and reptiles in a particular region, habitat or geological period and Borneo's tropical rainforest is home to a variety of this group. Inger et al., (2017) documented over 180 species of frogs in

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Borneo, of these 139 are endemic to the island and about 100 species had been recorded in Sabah whereas there were about 289 species of reptiles that have been recorded in Borneo (Das, 2006). Amphibians and reptiles inhabit diverse types of environments including disturbed areas, mangroves, rivers, streams, swamps, waterfalls, lakes, primary forests, plantations, caves and mountains (Shahriza Shahrudin et al., 2011). In tropical forests, amphibians may utilize a wide range of microhabitat types such as rocky streams, hilly terrain and non-riparian forested area (Inger et al., 2002; Pui & Das, 2016).

The hilly sub-montane and montane forests of Mount Kinabalu are home to 77 species of amphibians and 112 species of reptiles. They are found in various environments or microhabitats such as trees at various storeys, leaf-littered forest floor and habitats dominated by mountain streams (Malkmus et al., 2002). Mount Kinabalu which is situated within the Kinabalu Park, is one of the most crucial water catchments in Sabah and many streams originate from the massif including Kadamaian River that flows to the west. As a protected area, several studies or surveys related to herpetofauna have been performed in Mount Kinabalu and its herpetofauna diversity is well-documented compared to areas outside the park especially within its buffer zone. Thus, it would be interesting to explore the environments beyond the border of the protected area.

To date, so far data or records on herpetofauna in Kadamaian have been documented in a miscellaneous collection of publications, i.e in Malkmus et al., (2002). Therefore, this study aims to record amphibians and reptiles that occur in Kadamaian and to update the list of herpetofauna in the area.

## **Methods**

### *Study site*

Kadamaian area is located in Kota Belud District within the west coast of Sabah and the study site (N 06° 12' 25.6" E 116° 30' 34.1") borders Kinabalu Park at the eastern part (Figure 1). This survey was conducted for a period of five days, from 21<sup>st</sup> to 25<sup>th</sup> October, 2019 during the Borneo Geographic Scientific Expedition in Kadamaian - Kinabalu Park which was jointly organized by the Institute for Tropical Biology and Conservation (ITBC) of Universiti Malaysia Sabah (UMS) and Sabah Parks. The expedition base camp was located at an elevation of approximately 600 meters above sea level (m a.s.l). The Kadamaian expedition area comprised of hill submontane forest type and montane forest with the elevation ranging from 500 to 1430m a.s.l, with the highest elevation being Mount Nopungguk (Figure 2).



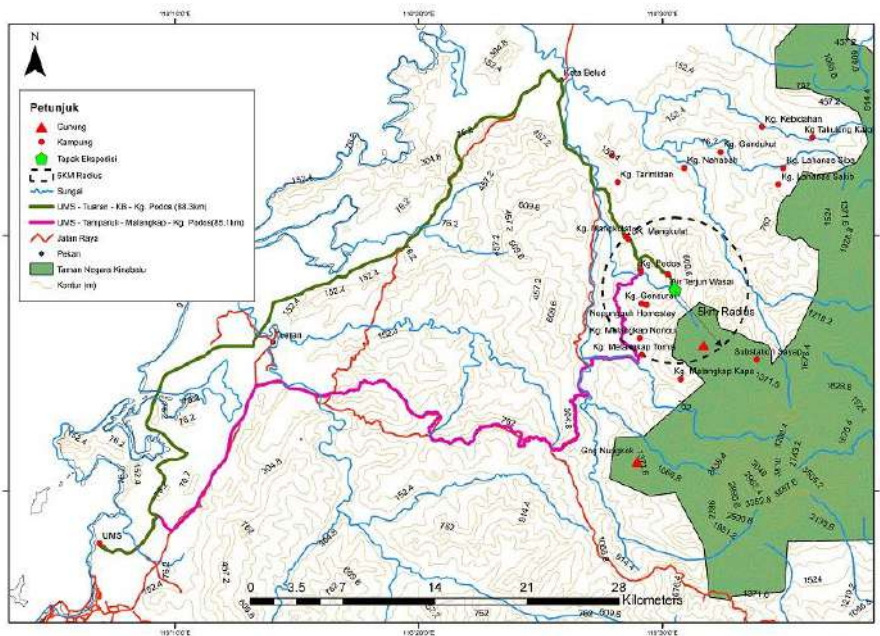


Figure 1. The Borneo Scientific Expedition site map in Kadamaian, Kota Belud which borders Kinabalu Park and a small part of the park included in the scientific expedition study area (Source: ITBC, UMS).

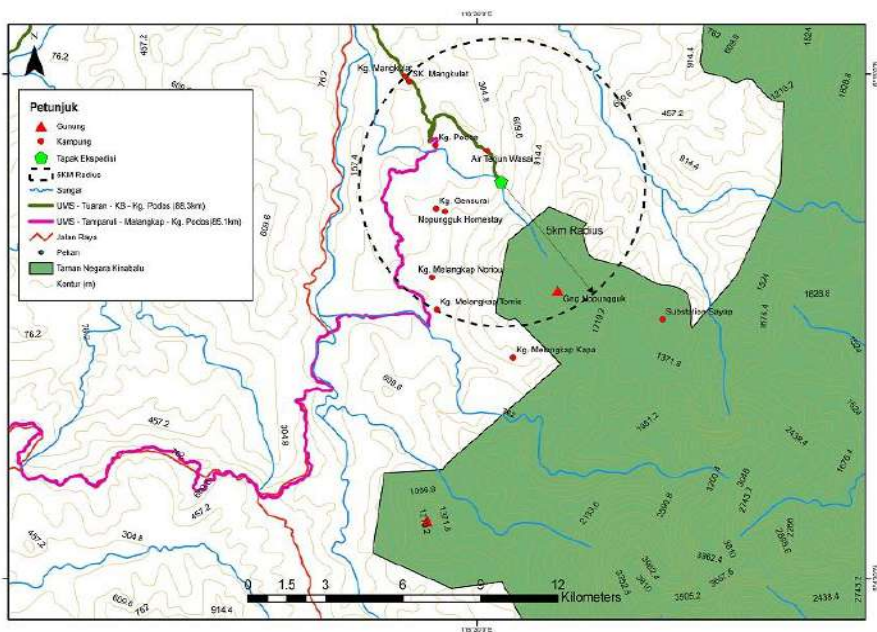


Figure 2. Topography Map of the study area (Source: ITBC, UMS)

Four localities were selected namely the waterfall near the base camp, Pinolobu River, Kipungit River in Melangkap Noriou and Meliawa River at the base camp. All localities comprise of riparian forests within 300-700m a.s.l, and night transects were established along the riparian area. Only localities near the base camp namely waterfall and Meliawa River can be considered as good forests. Meanwhile, localities adjacent to human settlements are old secondary forests or have been converted into rubber tree plantations. Samplings were conducted at 1900 hour until 2200 hour and Visual Encounter Survey (VES) method was applied. Photos of the captured herpetofauna were taken before release for documentation and identification. Global Positioning System (GPS), Garmin device was used to mark the coordinates and the 'A field guide to the Frogs of Borneo' book by Inger et al., (2017) was referred for identification purpose.

## Results and Discussion

Overall, a total of 15 species of amphibians from five families and six species of reptilians from three families were found inhabiting the study area (Table 1). The recorded frogs were *Ansonia minuta*, *Ansonia spinulifer* and *Ansonia longidigata* from the family Bufonidae (20%); *Staurois guttatus*, *Meristogenys poecilus*, *Huia cavitympanum* and *Hylarana picturata* from the family Ranidae (26.7%); *Limnonectes kuhlii* and *Fejevaryia limnocharis* from the family Dicroglossidae (13.3%); *Leptotalax gracilis*, *Leptobrachella sabahmontana* and *Leptobrachium abbottii* from the family Megophryidae (20%); and *Philautus hosii*, *Polypedates leucomystax* and *Polypedates macrotis* from the family Rhacophoridae (20%). Meanwhile, the six reptiles were *Gonocephalus liogaster* and *Branchocela cristatella* from the family Agamidae (33.3%); *Cyrtodactylus baluensis* from the family Gekkonidae (16.7%); and finally *Tropidophorus micropus*, *Eutropis rudis* and *Sphenomorphus* sp. from the family Scincidae (50%). Most of these were considered as Least Concern (LC) in the IUCN Red List of Threatened Species data and *Leptobrachella sabahmontana* was the only Endangered (EN) frog species that was documented over the four-night survey. There was one skink species classified as Data Deficient (DD) namely *T. micropus*. Common species of frogs in the list were *F. limnocharis* and *P. leucomystax* and *E. rudis* was the most common reptile encountered during the survey.

Ten (10) species out of the recorded herpetofauna list were endemic to Borneo namely *Ansonia minuta*, *A. spinulifer*, *A. longidigata*, *Meristogenys poecilus*, *Huia cavitympanum*, *Leptotalax gracilis*, *Leptobrachella sabahmontana*, *Philautus hosii*, *Cyrtodactylus baluensis* and *Tropidophorus micropus*.

Significantly, two specimens of *A. minuta* were spotted on leaf near the stream (riparian area) during the night survey at Kipungit River in Melangkap Noriou. The vegetation of the locality comprised of lowland hill forest with the elevation of approximately 600m a.s.l. The snout-vent length of the specimens are 17mm and 23mm, respectively. Both with a weight of 1 gram, the specimens are kept in Sabah Parks Zoological Collection. According to Inger et al., (2017), *A. minuta* has long hind limbs, its snout projecting well beyond the mouth, and tips of the outer fingers are widened. The first finger is distinctly shorter than the second and the toes are fully webbed in males, about three-quarters webbed in females. The upper surfaces of its body and limbs are covered with small, round warts. Adult males have two or three rows of yellowish spines under the chin. This toad has brown with irregular small orange or yellow spots or streaks on the back, its belly is pale with black spots and yellowish dots. The side of the head have dark and light bars, with a whitish area below the eye. This toad is a common lowland stream bufonid that was known only from several localities in Sarawak (Malaysia) and Kalimantan (Indonesia) such as in Kubah National Park, Gading National Park and Mount Penrissen of Sarawak (IUCN SSC Amphibian Specialist Group, 2018). In fact, Inger et al. (2017) stated that *A. minuta* may occur more widely than the current published records which is in line with our finding. This finding in Kadamaian is a new record for Sabah. Meanwhile, the other two *Ansonia* species are dispersing widely within the lowland, hilly and submontane rainforests in Sabah.

Interestingly, *Leptobrachella sabahmontana* which is classified as EN in the IUCN Red List appears to be restricted to Sabah (Northern Borneo) and can be found in the sub-montane to montane forests with the altitude ranges between 750 to 1500m a.s.l (Matsui et al., 2014; IUCN SSC Amphibian Specialist Group, 2019). *L. sabahmontana* was found at two localities within the study area namely at the waterfall near the expedition base camp with an altitude of approximately 600m a.s.l and Kipungit River in Melangkap Noriou. The frogs were spotted on leaf near water sources or streams within the riparian area. The river near the base camp is wide and rocky; and the water current was moderate whereas Kipungit River is small/narrow with slower water current. This species is documented to occur at several localities within Kinabalu Park and Crocker Range Park including Sayap Substation in Kota Belud (Matsui et al., 2014).

Table 1. The amphibians and reptiles recorded from Kadamaian, Kota Belud.

FAMILY	SPECIES	COMMON NAME	LOCALITIES				CONSERVATION STATUS (IUCN)
			Waterfall near base camp	Pinolobu River	Kipungit River, Melangkap Noriou	Meliawa River	
Bufonidae	<i>Ansonia minuta</i> *,**	Dwarf Slender Toad			x		Least concern
	<i>Ansonia spinulifer</i> *	Spiny Slender Toad	x	x	x	x	Least concern
	<i>Ansonia longidigita</i> *	Long-fingered Slender Toad	x		x	x	Least concern
Ranidae	<i>Staurois guttatus</i>	Black-spotted Foot-Flagging Frog	x	x	x	x	Least concern
	<i>Meristogenys poecilus</i> *	Speckle-legged Torrent Frog	x		x	x	Least concern
	<i>Huia cavitimpanum</i> *	Hole-in-the-head Frog	x			x	Least concern
Dicroglossidae	<i>Hylarana picturata</i>		x	x	x		Least concern
	<i>Limnonectes kuhlii</i>	Kuhl's Creek Frog	x	x	x	x	Least concern
	<i>Fejervarya limnocharis</i>			x			Least concern
Megophryidae	<i>Leptotalax gracilis</i> *	Sarawak Slender Litter Frog		x	x	x	Least concern
	<i>Leptobrachella sabahmontana</i> *		x		x		Endangered
	<i>Leptobrachium abbotti</i>	Lowland Large-eyed Litter Frog			x	x	Least concern
Rhacophoridae	<i>Phyllautus hosii</i> *		x		x		Least concern
	<i>Polypedates leucomystax</i>	Hose's Bush Frog		x		x	Least concern
	<i>Polypedates macrotis</i>	Four-lined Tree Frog		x			Least concern
Agamidae	<i>Gonocephalus liogaster</i>	Dark-eared Tree Frog				x	Least concern
	<i>Branchocela cristatella</i>					x	
	<i>Cyrtodactylus baluensis</i> *					x	
Gekkonidae	<i>Tropidophorus micropus</i> *		x				Least concern
	<i>Eutropis rudis</i>		x				Data deficient
	<i>Sphenomorphus sp.</i>					x	

\*Endemic to Borneo, \*\*New record to Sabah, x-Present

*Cyrtodactylus baluensis* and *Trodiphorus micropus* are both endemic reptiles to Borneo. *C. baluensis* has been reported to occur in several sites near Mount Kinabalu such as at Kadamaian River and near Kiau in Kota Belud (Iskandar & McGuire, 2018). It prefers hilly, submontane and montane rainforest and is suspected to be restricted to high elevations (Sah et al., 2016; Iskandar & McGuire, 2018). Therefore, the occurrence of this lizard within the study site can be expected. *T. micropus* usually inhabits rocky streams and it was found near the waterfall not far from expedition's base camp. However, this species has limited information compared to *C. baluensis* and classified as DD in the IUCN Red List. This skink was documented in Long Bloe Upper Mahakkam River and Murung Raya of Indonesian Borneo and is suspected to be present in protected areas in Borneo (Iskandar et al., 2019).

The remaining endemic herpetofauna that were recorded in the survey, namely *M. poecilus*, *H. cavitympanum*, *L. gracilis* and *P. hosii* are common inhabitants of primary lowland, hilly forests and old growth secondary forests in Borneo. Although these frogs are considered as LC in the IUCN Red List, their populations are declining as a result from habitat loss and forest degradation, yet survive in protected areas in Borneo (IUCN SSC Amphibian Specialist Group, 2018; IUCN SSC Amphibian Specialist Group, 2019; IUCN SSC Amphibian Specialist Group, 2020)

## Conclusion

The number of species was low due to the short survey period. However, the team managed to update the list of herpetofauna in Kadamaian area with one Endangered (EN) frog species namely *Leptobrachella sabahmontana* of the family Megophryidae and one new record for Sabah which is *Ansonia minuta* of the family Bufonidae. Nevertheless, this study contributes to Sabah's herpetofauna database and serves as reference for more studies and research in future.

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## Appendices

### Photos of habitat in selected localities during expedition



Photo 1. Sungai Kipunyit at Melangkap Noriou



Photo 2. Sungai Marangoi Pinolobu



Photo 3. Waterfall near base camp

### Photos of Herpetofauna in Kadamaian, Kota Belud



Photo 1. *Ansonia minuta*, which was spotted on a leaf at Kipungit river in Melangkap Noriou



Photo 2. *Ansonia spinulifer* was found in all localities during the survey



Photo 3. *Ansonia longidigata* was found in 3 survey localities except Pinolobu river



Photo 4. *Meristogenys poecilus* was recorded in 3 survey localities except Pinolobu river





**Photo 5.** *Huia cavitympanum* was found in 2 localities namely waterfall near base camp and Meliawa river



**Photo 6.** *Leptolalax gracilis* was found in 3 localities except waterfall near base camp



**Photo 7.** *Leptobrachella sabahmontana* was found in 2 localities namely waterfall near base camp and Kipungit river, Melangkap Noriou



**Photo 8.** *Leptobrachium abbotti* was found in 2 localities namely Kipungit river and Meliawa river. Usually spotted on the ground leaf litter.



**Photo 9.** *Philautus hosii* was recorded in 3 localities except Pinolobu river



**Photo 10.** *Gonocephalus liogaster* was found in Kipungit river, Melangkap Noriou



**Photo 11.** *Branchocela cristatella* was found in 2 localities namely Kipungit river and Meliawa River



**Photo 12.** *Tropidophorus micropus* was recorded in the waterfall area near base camp



**Photo 13.** *Cyrtodactylus baluensis* was found in Kipungit river, Melangkap Noriou

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## Research Article

# A Preliminary Survey and Chemical Profiling of Wild Ginger Species in Kadamaian, Kota Belud, Sabah

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## Abstract

A preliminary survey of the diversity of gingers (Zingiberaceae) was conducted in Kadamaian, Kota Belud from 14<sup>th</sup> to 19<sup>th</sup> October, 2019. Wild ginger species is utilized widely as one of the most important material in traditional medicine among indigenous people of Sabah. However, few of these plant species have been studied for their chemical constituents and beneficial properties. In order to investigate the compound composition, the essential oil from *Etlingera brevilabrum*, *Alpinia nieuwenhuizii* and *Hornstedtia havilandii* were screened. The essential oil was obtained from leaves, stems and rhizomes of the plant through hydro-distillation and analysed for their chemical composition through Gas Chromatography-Mass Spectrometry (GC-MS). The result of this study indicated that the chemical constituents of all three parts for all species are similar; all have terpenoids (monoterpene and sesquiterpene), aldehyde, hydrocarbon, ketone and alcohol in the essential oil extracts. GC-MS analyses of the oils led to the identification of 35 compound constituents from the leaves, stems and rhizomes of *E. brevilabrum*, which is the highest. Meanwhile, *A. nieuwenhuizii* displayed 34 chemical compositions from all parts (leaf, stem and rhizome) of the plant. *H. havilandii* showed the lowest number of volatiles from all plant parts (24 compounds). Monoterpene is dominant in all wild ginger studied, except for rhizome of *E. brevilabrum*. On the contrary, *E. brevilabrum* showed sesquiterpene as the most abundant compound in its composition. This shows that the volatile oil composition of wild ginger species is extremely variable. This study provides preliminary key chemical information for evaluating the quality of local wild gingers in Kadamaian, Kota Belud, Sabah.

**Keywords:** Chemical composition, essential oil, GC-MS, Ginger, Monoterpene, Sesquiterpene.

## Introduction

The ginger family, Zingiberaceae, is native to tropical and subtropical Southeast Asia and mainly distributed in Asia. The family consists of pseudo-stem and tuberous rhizomes, in which the latter are also known as ginger root or commonly known as ginger. Due to its strong odour and pungent taste, the most common custom usage of ginger is as a flavouring agent, ingredient for culinary and as traditional medicine (Mahomoodally et al., 2019). Zingiberaceae consists of 50 genera with approximately over 1,600 species (Christenhusz & Byng, 2016). Within the species itself, there are at least 172 species recorded from the Indo-Malayan region, with 80% found in Borneo (Poulsen, 2006).

As a natural product, ginger is a complex spice composed of standard food nutrients and volatile essential oils. Several terpene components that made up the compositions are bisabolene, farnesene, curcumene and quiphellandrene (Mahomoodally et al., 2019). The presence of high quality essential oils in many wild ginger species attributed its potential to pharmaceutical and nutraceutical values (Liu et al., 2012).

Due to the potential of wild gingers as an alternative source for health-related treatment, further scientific study has sought to reveal the chemical composition from different parts of ginger such as leaf, stems and rhizomes. Vairappan et al. (2012) has studied the chemical composition from the rhizomes of *Etlingera* species namely *E. pyramidosphaera*, *E. megalochelos* and *E. brevibrum* from Kimanis, Sabah, *E. coccinea* from Ranau, Sabah and *E. elatior* from Tambunan, Sabah. Four of the species, except for *E. pyramidosphaera* contained high concentrations of hydrocarbon sesquiterpenes that ranged from 21.4% to 50.0%. Oxygenated sesquiterpene contents ranged from 3.0% to 28.6% in *E. megalochelos*, *E. pyramidosphaera* and *E. coccinea*, while diterpene hydrocarbon were only detected in *E. elatior* (12.5%). Mahdavi et al. (2017) extracted essential oil through hydrodistillation from *Etlingera sayapensis* (leaf, stem and rhizome) collected from Kipandi, Sabah. The leaves exhibited high content of carveol (21.38%), the rhizome showed high linalool formate content (25.47%), while the stem was dominated by  $\alpha$ -terpineol (39.86%). The essential oils of rhizomes from *etlingera* sp. were further studied by Nagapoan et al. (2017), and these include *E. pyramidosphaera*, *E. megalochelos*, *E. coccinea* and *E. elatior* from Ranau, Sabah. A total of 39 volatile chemicals were detected from the said species consisting mixtures of oxygenated monoterpenes,

sesquiterpenes, oxygenated diterpenes and diterpenes. Although all rhizomes revealed terpenoids as major compounds, only *E. coccinea* and *E. elatior* showed terpenoid at the highest abundance, with borneol from *E. coccinea* at 28.2% abundance and aromadendrene oxide from *E. elatior* at 46.2% abundance. Owing to the wide range of beneficial effects of this herb, studies are necessary to investigate the variation in the production of phytochemicals throughout the plant organs. It is important to gather relevant evidence regarding herbs with high level of potentially beneficial components (Ghasemzadeh et al., 2016). At the time when the present survey was conducted, there was no information available on ginger species from Kadamaian. Therefore, this study is important in providing valuable baseline data for conservation purposes through inventory, and information on the chemical properties of essential oils from different plant tissues of selected wild gingers through Gas Chromatography-Mass Spectrometry (GC-MS) technique.

## Materials and Methods

### *Sampling and Study Area*

Twelve species of wild gingers from six different trails/site (Malangkap Noriou (a), Gensurai (b), Wasai waterfall (c), Basecamp (d), Ulu Malawa (e) and Pinolobu (not shown)) were collected in Kadamaian, Kota Belud, Sabah from 14th to 19th October, 2019 (Figure 1). The plants (with or without floral structures) were then brought back to Universiti Malaysia Sabah (UMS) for plant material preparation. The plants were preserved using a standard herbarium method.

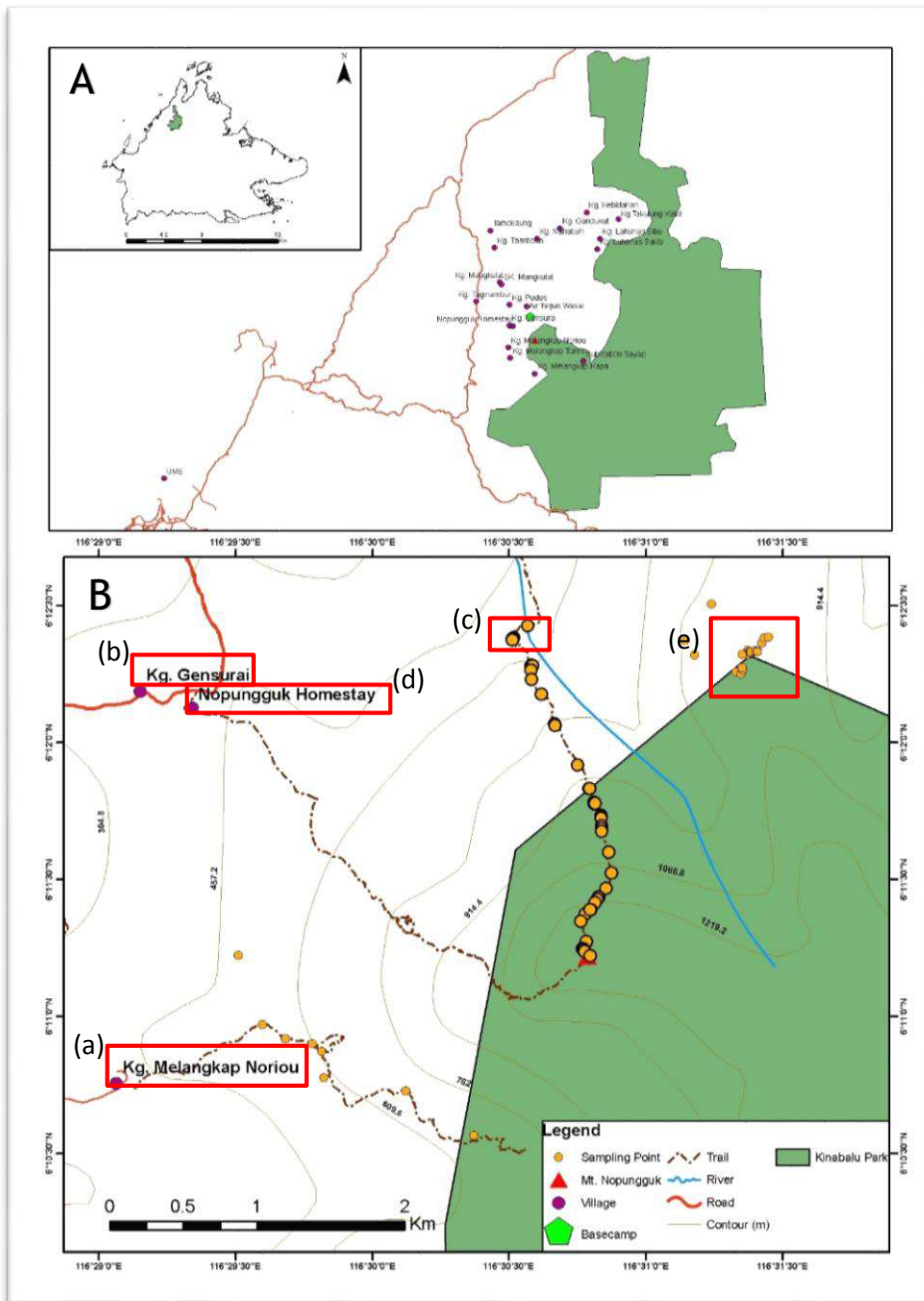
### *Plant materials*

Three selected fresh samples of *Etlingera brevilabrum*, *Alpinia nieuwenhuizii* and *Hornstedtia havilandii* were collected from three different trails in Kadamaian, Kota Belud, Sabah from 14<sup>th</sup> to 19<sup>th</sup> October, 2019. Samples were brought back to ITBC, cut separately (leaves, stems and rhizomes) and stored in -80°C prior to further use. The samples were authenticated by Mr. Johny Gisil during sampling collection. However, no voucher specimens were deposited due to limited availability of samples during collection.

### *Extraction of essential oil from plant*

Briefly, about 250-300g of fresh leaves and stems were chopped into small pieces and crushed using a blender to increase the surface area. Then, 400mL of distilled water was added and placed into a 500-1000mL round flask. The 10mL of 99% (v/v) n-pentane (BDH, Germany) were added to trap the condensed oil,

through the top of the condenser into the glass round flask containing samples of leaves and stems. Hydro-distillation of each sample was carried out in a



**Figure 1.** The location of the study area in Kadamaian, Kota Belud District in Sabah (A) and locations of the study plots (B). shown in the map are sampling sites/trails of Malangkap Noriou (a), Gensurai (b), Wasai waterfall (c), Basecamp (d) and Ulu Malawa (e).

modified Clevenger apparatus separately with a water-cooled oil receiver to reduce the potential of hydro-distillation over-heating artifacts. The extraction was carried out at 100°C for 6-7 hours. The light-yellow colour with pleasant aroma was obtained, then separated and dried over with 10g of anhydrous sodium sulphate ( $\text{Na}_2\text{SO}_4$ ) overnight, and then filtered. Finally, the essential oil was concentrated by blowing with pure nitrogen gas. The oils were then stored in sealed vials at 4°C prior to further use (GC-MS analysis). The yields were calculated based on dry weight of plant materials (Tajidin et al., 2012).

#### *Identification of volatile composition of essential oils*

The GC-MS analysis was carried out using Shimadzu QP-2010 gas chromatograph equipped with SH-Stabil wax-DA capillary column (30m x 0.25mm x 0.25 $\mu\text{m}$ ) coupled with mass chromatography (MS) detector. The initial oven temperature was programmed from 60°C to 250°C at a rate of 5°C/min and was held at 250°C for 5 min. The injections of an ion source were adjusted to 270°C and 280°C, respectively. Helium was used as a carrier gas at 1mL/min. The detector interface temperature was set to 280°C, with the actual temperature in the MS source reaching approximately 230°C, and the ionization energy was 70eV. A 1  $\mu\text{L}$  volume of the essential oil extract mixed with hexane at 1:1 ratio was injected in spitless mode. Acquisition mass range was set to 39-600 amu. Total volatile production was estimated by summing all the GC peak areas in the chromatogram and individual compounds were quantified as relative percent area. Chemical composition was identified by comparing the mass spectra of the samples with the data system (NIST 08 and Flavor and Fragrance 2.0).

## **Results and Discussions**

### *The diversity of wild gingers*

A total of 12 Zingerberaceae species were collected and identified up to species level (Table 1). The survey revealed that there are at least 12 species from six genus representing three tribes. The species commonly believed to be indicators of disturbed forests such as *Etlingera coccinea* were the most abundant, implying that the trails surveyed had been disturbed (Larsen et al., 1999). According to Larsen et al. (1999) some species of *Etlingera* rapidly inhabit disturbed secondary forest or newly opened areas and subsequently spread like weed. From the 12 species documented in this report, most of them have high potential to be developed into ornamental plants such as in the genera of *Boesenbergia* and *Ammomum* (Lamb et al., 2013).

**Table 1.** A checklist of wild gingers collected from Kadamaian, Kota Belud.

Site	Tribe	Genera	Species name	GPS	Elevation (m)
Malangkap Noriou	Alpiniae	<i>Hornstedtia</i>	<i>havilandii</i>	N: 06 10.696; E: 116 29.899	578
Gansurai	Zingiberaceae	<i>Zingiber</i>	N/A	N: 06 11.231; E: 116 30.332	901
	Alpinioideae	<i>Alpinia</i>	<i>nieuwenhuizii</i>	N: 06 11.479; E: 116 29.975	767
	Alpiniae	<i>Etlingera</i>	<i>probescence</i>	N: 06 11.348; E: 116 30.137	682
	Zingiberaceae	<i>Zingiber</i>	N/A	N: 06 11.348; E: 116 30.137	682
Wasai Waterfall	Alpiniae	<i>Etlingera</i>	<i>brevilabrum</i>	N: 06 11.319; E: 116 30.147	290
Basecamp	Alpiniae	<i>Etlingera</i>	N/A	N: 06 15.217; E: 116 30.382	245
Pinolobu	Alpiniae	<i>Etlingera</i>	N/A	N: 06 15.238; E: 116 30 314	207
	Alpiniae	<i>Etlingera</i>	N/A	N: 06 15. 341; E: 116 30 169	163
Ulu Maluwa	Globbeae	<i>Globba</i>	N/A	N: 06 12. 164; E: 116 31.212	933
	Alpiniae	<i>Ammomum</i>	<i>kinabaluensis</i>	N: 06 12.159; E: 116 81.213	934
	Zingiberaceae	<i>Boesenbergia</i>	N/A	N: 06 12. 192; E: 116 31.211	982

### *Chemical composition of selected wild gingers*

Of the 12 species of gingers reported in this study, not much work has been done to explore their bioactive and antioxidant potential. Due to limited number of samples, only three selected gingers were submitted to chemical profiling study. The identification of *Hornstedtia havilandii* (Figure 2a), *Alpinia nieuwenhuizii* (Figure 2b) and *Etlingera brevilabrum* (Figure 2c) were based on their morphology during sample collection. In the present study, the GC-MS analysis of the essential oil extracts from leaves, stems and rhizomes of selected wild gingers resulted in the identification of 52 chemical compounds of which a majority belonged to the terpenoid group (Table 2). These included monoterpenes such as  $\alpha$ -pinene,  $\alpha$ -myrcene, borneol, camphene and camphor, and sesquiterpenes such as  $\alpha$ -caryophyllene, alloaromadendrene, copaene, cubenene and nerolidol. The essential oil also showed the presence of other volatile groups such as phenylpropanoids methyleugenol and asarone, and fatty acid derivative, benzeneacetyldehyde. In terms of yield, the leaf of *Alpinia nieuwenhuizii* contained the highest oil yield (0.21%), followed by the rhizome of *A. nieuwenhuizii* (0.19%) and rhizome of *Hornstedtia havilandii* (0.18%). Synthesis and accumulation of essential oils can accumulate in all plant organs, but in varying amounts. This is contributed to the distribution secretory structures such as glandular trichomes outside the plant and secretory cells and intercellular spaces inside the plant (Sarac & Butnariu, 2018). The findings from Dodoš et al. (2021) revealed variation of essential oil accumulation across plant organs (leaf, calyx, corolla and herba) of *Satureja montana*, *S. subspicata* and *S. Kitaibelii* contributed by peltate and capitate glandular trichomes.





**Figure. 2** Selected wild ginger species that were used in this study. (a) *Etlingera brevilabrum*; (b) *Alpinia nieuwenhuizii*; (c) *Hornstedtia havilandii*

GC-MS analyses of the oils extracted from the stem, leaves and rhizomes combined of *Etlingera brevilabrum* resulted in the identification of 35 different chemical constituents, the highest compared to *A. nieuwenhuizii* and *H. havilandii*. The stem of *E. brevilabrum* recorded the highest number of chemical constituents (25 compounds), whereas, the leaves and rhizomes comprised the same number of chemical compositions (18 compounds). On the other hand, *A. nieuwenhuizii* exhibited a total of 34 compound constituents, in which the rhizome displayed the highest number of chemical compositions (21 compounds), followed by its leaf (17 compounds) and stem (15 compounds). *H. havilandii* and *A. nieuwenhuizii* leaves showed similar number of volatile constituents, while displaying as the highest among all parts of *H. havilandii*. The rhizome of *H. havilandii* contained 13 compounds, whereas the stem of *H. havilandii* displayed the lowest number of volatiles (10 compounds).

Monoterpenes dominated all plant parts of *H. havilandii*, *A. nieuwenhuizii*, and *E. brevilabrum*, except for the rhizome of *E. brevilabrum* which exhibited sesquiterpenes as the major compounds (Figure 3). This was followed by alcohol, aldehyde, hydrocarbons and ketones. The stem and leaf of *E. brevilabrum* revealed eucalyptol as the most abundant chemical compound (18.59% and 16.63%, respectively). Meanwhile, the rhizome of *E. brevilabrum*, leaves of *A. nieuwenhuizii* and *H. havilandii* recorded sesquiterpene caryophyllene as the compound with highest abundance at 16.23%, 34.53% and 22.54% respectively. On the other hand, the rhizome of *A. nieuwenhuizii* and stem of *H. havilandii* showed (-)- $\beta$ -pinene as their highest abundant volatiles, at 28.07% and 46.68%.

Cis-sabinene is recorded at highest abundance (28.81%) in the stem of *A. nieuwenhuizii*. Finally, the rhizome of *H. havilandii* recorded sabinene as the most abundant constituent at 46.30%. Apart from monoterpenes and sesquiterpenes, other notable volatiles were also detected, such as phenylpropanoid methyleugenol in the stems and leaf of *E. brevilabrum* at 4.83% and 1.07%, and benzeneacetyldehyde in the leaf of *A. nieuwenhuizii* (0.04%).

Similar variations of chemical constituents across plant organs were also observed. Feng et al. (2021) observed the accumulation of terpenoids as the major compound in stems, leaves, flowers and fruits of *Alpinia zerumbet*. While monoterpene eucalyptol dominated the constituents in leaves, stems and fruits, camphor exist as the most abundant compound in the flowers. On the other hand, Jusoh et al. (2020) reported the constituents of essential oils extracted from the leaf and pseudo-stems of *Alpinia malaccensis*. As the leaf revealed  $\beta$ -pinene, 1,8-cineol, trans-caryophyllene and  $\alpha$ -pinene as the major compounds, while 1,8-cineol,  $\beta$ -pinene,  $\alpha$ -terpineol, trans-caryophyllene and  $\alpha$ -terpinolene as the major components in the pseudo stems. Ramos et al. (2020) suggested that the variations in volatile compositions and abundance were affected by the structure development of secretory structures within the plant organs and the gene expression profile across plant organs and species for the particular compound synthesis.

Many of the chemical compounds detected in wild gingers of this study were observed to exhibit various pharmaceutical potential. The high abundance of sabinene produced from *H. havilandii* was reported to exhibit anti-fungal and anti-inflammatory properties (Cao et al., 2017). The monoterpene eucalyptol was also known to display anti-inflammatory, anti-nociceptive and reduce the interferon-gamma levels in mice (Júnior et al., 2017). Besides,  $\alpha$ -pinene that can be found in all three wild ginger species in this research were also known to exhibit neuroprotective effect by attenuating neuroinflammation and inhibit apoptosis (Khoshnazar et al., 2020).

**Table 2.** Volatile profiling and percentage yield of stems, leaves and rhizomes of three selected wild gingers species by using Gas Chromatography-Mass Spectrometry (GC-MS).

No	Compound Name <sup>a</sup>	Compound group	<i>Etlingera brevilabrum</i> (%) <sup>b</sup>			<i>Alpinia nieuwenhuizii</i> (%) <sup>b</sup>			<i>Hornstedtia havlani</i> (%) <sup>b</sup>		
			leaf	stem	rhizome	leaf	stem	rhizome	leaf	stem	rhizome
1	δ-Cadinene	monoterpene	7.26	3.53	-	-	-	-	9.00	0.41	-
2	(-)-borneol	monoterpene	1.32	-	-	-	-	-	-	-	-
3	(-)-linalool	monoterpene	5.41	-	-	-	-	-	-	-	-
4	(-)-β-pinene	monoterpene	-	-	-	18.10	-	28.07	12.35	46.48	-
5	α-Caryophyllene	sesquiterpene	6.15	2.47	4.34	9.23	-	0.28	3.32	-	-
6	α-Phellandrene	monoterpene	0.91	-	-	-	-	-	0.59	-	-
7	α-Pinene	monoterpene	9.25	12.89	13.19	18.38	5.10	7.75	7.14	29.81	28.02
8	α-Myrcene	monoterpene	4.50	-	1.01	-	-	-	-	-	-
9	1-Decanol	alcohol	-	3.27	0.75	-	-	9.11	-	1.09	0.05
10	1-Hexanol	alcohol	-	-	-	0.10	-	-	-	-	-
11	1-Nonanol	alcohol	-	-	-	-	0.23	0.10	-	-	-
12	1-Octanol	alcohol	-	-	-	0.02	-	-	-	-	-
13	1-Undecanol	alcohol	-	-	-	0.13	-	-	-	-	-
14	2-Butanone	ketone	-	-	-	-	-	0.05	-	-	-
15	2-Nonanone	ketone	-	-	0.20	-	-	-	-	-	0.26
16	Alloaromadendrene	sesquiterpene	-	-	-	0.30	-	-	-	-	-
17	Asarone	Phenylpropanoid	-	7.10	15.03	-	22.02	-	7.53	-	-
18	Benzeneacetaldehyde	Fatty acid derivatives	-	-	-	0.04	-	-	-	-	-

(Continued on next page)

Table 2. (Continued)

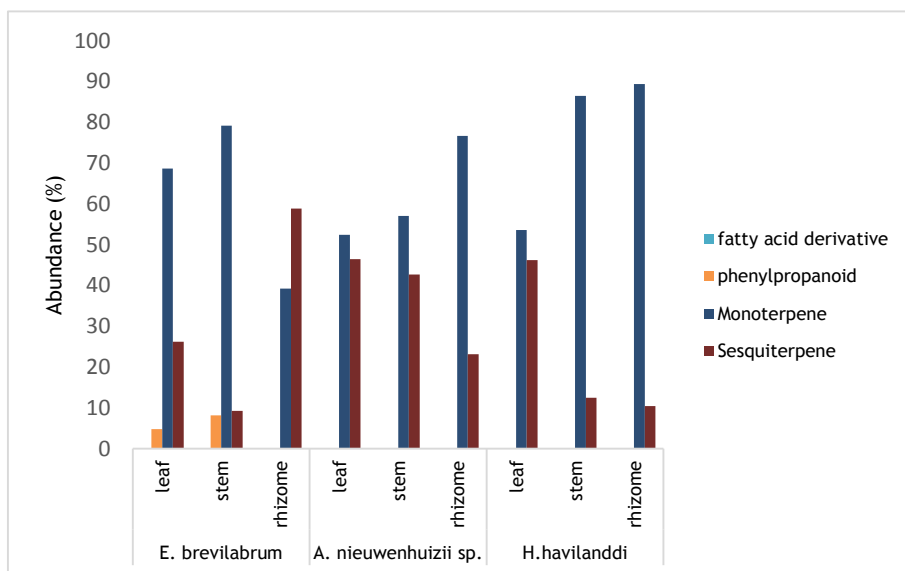
19	Borneol	monoterpene	-	1.92	-	-	1.15	2.94	0.99	0.41	0.80
20	Camphene	monoterpene	10.76	7.61	14.06	0.38	3.71	3.65	8.06	-	4.45
21	Camphor	monoterpene	8.18	5.52	8.63	-	3.16	1.05	4.06	-	0.35
22	Caryophyllene	sesquiterpene	1.13	2.15	16.23	34.53	11.71	2.49	22.54	-	-
23	Caryophyllene oxide	Sesquiterpene	6.07	-	9.12	2.44	7.39	14.38	1.88	-	-
24	cis-sabinene	monoterpene	-	-	-	-	28.81	0.28	-	-	-
25	Copaene	sesquiterpene	5.60	1.14	2.12	-	-	1.25	1.95	-	3.45
26	Cubenene	sesquiterpene	-	-	0.85	-	-	-	-	-	-
27	Decanal	aldehyde	-	-	1.00	-	-	18.24	-	-	-
28	Eucalyptol	monoterpene	16.63	18.59	-	-	6.69	-	-	-	-
29	Fenchol	monoterpene	0.26	0.30	0.24	1.20	0.54	0.39	-	-	-
30	Fenchone	monoterpene	10.99	3.54	-	-	-	-	9.87	-	-
31	Gurjunene	sesquiterpene	-	-	-	-	1.56	-	-	-	-
32	Hexanal	aldehyde	0.26	0.10	-	0.79	-	-	0.21	-	0.12
33	L-4-terpineol	monoterpene	-	-	-	-	-	-	-	-	-
34	Limonene	monoterpene	-	3.96	-	6.64	2.01	2.20	-	4.88	5.15
35	Linalool	monoterpene	-	3.39	1.13	-	-	0.73	-	-	-
36	Linalyl alcohol	monoterpene	-	-	-	-	1.26	-	-	-	-
37	Methyleugenol	phenylpropanoid	4.83	1.07	-	-	-	-	-	-	-
38	Nerolidol	sesquiterpene	-	-	-	-	-	-	-	12.05	6.10
39	Nonane	hydrocarbon	-	-	-	-	-	0.12	-	-	-
40	Nopinone	monoterpene	-	0.19	-	0.86	-	-	-	-	-
41	Sabinene	monoterpene	-	-	-	-	-	-	10.17	-	46.30
42	Terpineol	monoterpene	-	-	-	-	4.66	-	-	-	-
43	Terpinolene	monoterpene	0.50	0.12	-	-	-	-	0.23	-	-

(Continued on next page)

Table 2. Continued

44	Trans-bornyl acetate	monoterpene	-	-	-	-	-	0.28	-	-	-
45	Tricyclene	monoterpene	-	0.13	0.42	-	-	-	0.11	0.34	-
46	Verbenone	monoterpene	-	0.18	-	-	-	-	-	-	-
47	$\alpha$ -gurjunene	sesquiterpene	-	-	11.16	-	-	4.78	-	-	0.85
48	$\alpha$ -sabinene	monoterpene	-	-	0.53	-	-	-	-	-	-
49	$\alpha$ -terpinene	monoterpene	-	0.10	-	0.75	-	-	-	0.41	-
50	$\alpha$ -terpineol	monoterpene	-	1.26	-	6.13	-	1.86	-	4.12	4.09
51	$\beta$ -myrcene	monoterpene	-	1.07	-	-	-	-	-	-	-
52	$\beta$ -pinene	monoterpene	-	18.41	-	-	-	-	-	-	-
Terpene compound abundance (%)		Total monoterpene	68.70	79.17	39.21	52.43	57.09	76.65	53.57	86.45	89.43
		Total sesquiterpene	26.21	9.29	58.85	46.49	42.68	23.18	46.22	12.46	10.40
		Total terpene	99.74	88.46	98.05	98.93	99.77	99.83	99.79	98.91	99.83
		Total abundance	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00
		Yield (%)	0.17	0.06	0.14	0.21	0.10	0.19	0.13	0.06	0.18

<sup>a</sup>Volatile compounds were listed in elution time order from capillary VF-WAXms column<sup>b</sup>Note: (%) - Relative abundance



**Figure. 3.** Relative abundance of three major groups of volatiles (Terpenoids monoterpene and sesquiterpene, phenylpropanoid and fatty acid derivatives).

## Conclusion

The chemical profile from the leaf, stem and rhizomes of selected wild gingers exhibited dissimilar volatile profile between and even within the species. There are still many wild ginger species in the forests of Sabah that require attention, waiting to be discovered and documented. It is of utmost importance to conserve Sabah's forests not only to protect the habitat, but also to maintain the existence of this plant family that serves as a potential reservoir for development into a variety of pharmaceutical and nutraceutical products.

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## Research Article

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# Rapid Assessment of Nocturnal Sciurid and Avifauna Diversity in Kadamaian - Kinabalu Park for Ecotourism Potential

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## Abstract

A rapid assessment of sciurids and avifauna was done from 14<sup>th</sup> October 2020 - 20<sup>th</sup> October 2020 in Kadamaian - Kinabalu Park, Kota Belud, Sabah. Two sets of binoculars (Swarovski 8x30 and Bushnell 10x42) and two sets of Digital SLR cameras affixed with telephoto lens (Canon 7D; Tamron 150-600mm G2 F/4-6.3 and Nikon D500; Nikkor 200-500mm F/5.6) were used to aid in data collections. This assessment managed to record 5 species of Sciurids and 58 species of Avifauna in and around the Kadamaian-Kinabalu Park. Continuous biodiversity surveys are crucial in Kadamaian - Kinabalu, Sabah to maximize the potential ecotourism opportunity.

**Keywords:** canopy mammals, birds, gliding squirrels, ecotourism potential

## Introduction

Kadamaian-Kinabalu Park is located in Kota Belud district, west coast of Sabah. It comprises lowland forest and montane forest with pristine rivers, hills and waterfalls. In 2015, Kadamaian Tourism Association (KATA) was established in order to empower Kadamaian as a tourism product. And in early 2019, the Asean Community Based Tourism Standard 2019-2021 award was given to KATA at the Asean Tourism Ministers' Conference in Hanoi, Vietnam (New Straits Times, 2019).

Sciurids are classified under the order Rodentia and Family Sciuridae. Southeast Asia held the greatest diversity of squirrels followed by Africa (Koprowski & Nandini, 2008). Squirrels can be divided into three categories; diurnal tree squirrels, nocturnal gliding squirrels and ground squirrels. Tropical squirrels are seed predators and seed dispersers (Hallwachs 1986; Smythe 1989; Paschoal & Galetti, 1995) as well as agents of pollination. Apart from that, tree squirrels are included in the diet of large avian predators and small carnivores (Datta & Nandini, 2015).

Gliding squirrel is elusive, cryptic and unique animals that are often overlooked by people as they are nocturnal and arboreal. Gliding squirrel's watching is an activity that can be done in wildlife tourism package. There are at least 8 species of gliding squirrels that can be found in Kadamaian - Kinabalu Park area (Phillipps & Phillipps, 2018). Thus, this rapid assessment was conducted to assess ecotourism potential through gliding squirrels' watching activity in the Kadamaian - Kinabalu Park, while avifaunal survey is a secondary objective of this sampling event. The assessment will be followed with a few other visits in order to i) create a checklist on nocturnal non-volant small mammals diversity; and ii) to add them to the growing checklist of avifauna in and around Kadamaian - Kinabalu Park.

## Methodology

This rapid assessment was conducted for a total of 7 days; from the 14<sup>th</sup> to 20<sup>th</sup> October, 2019. Sampling was carried out via line transect with a total 1.5 km using direct observation method either through sight and/or vocalisation to record species presence. The trail is accessible and can be access by local communities and tourists. The assessment was focused on nocturnal sciurids, hence surveyors started the assessment before dark at 1600 hours to late night at 0200 hours each day. Two sets of binoculars (Swarovski 8x30 and Bushnell 10x42) and two sets of Digital SLR cameras affixed with telephoto lens (Canon 7D; Tamron 150-600mm G2 F/4-6.3; Nikon D500; Nikkor 200-500mm F/5.6) were used to aid in data collection. Sciurids were identified according to Payne et al. 1985; Carleton and Musser, 2005; Thorington Jr et al., 2012; Phillipps and Phillipps, 2016 while Phillips and Phillips, 2016 was used for bird identifications and its corresponding naming system. As for bird vocalisation, unfamiliar bird calls were recorded and compared to online repository (xeno-canto.org) for further identification off-site. As all birds have unique and distinct calls, the surveyor could easily distinguish and differentiate the birds up to species level as well.

## Results and Discussion

### *Sciuridae*

The rapid assessment managed to record a total of 5 species of sciurid from 4 genera which includes two species of gliding squirrels and three species of non-gliding squirrels. Four of these sciurid were listed as Least Concern (LC) while one species was listed as Data Deficient (DD).

Table 1. Taxonomic checklist for sciurids

Taxonomy	Common name	IUCN
Order: Rodentia		
Family: Sciuridae		
<i>Petaurista petaurista</i>	Red giant gliding squirrel	LC
<i>Aeromys tephromelas</i>	Black gliding squirrel	DD
<i>Callosciurus prevostii</i>	Prevost's squirrel	LC
<i>Callosciurus notatus</i>	Plaintain squirrel	LC
<i>Sundasciurus lowii</i>	Low's squirrel	LC

\*Note:

Observation Type;

S: Direct sighting on the individual using naked eyes/binoculars/cameras

V: Observation through vocalisation of the individuals

IUCN Red List Categories:

EX - Extinct, EW - Extinct in the Wild, CR - Critically Endangered, EN - Endangered, VU -

Vulnerable, LR/cd - Lower Risk/conservation dependent, NT - Near Threatened (includes LR/nt

- Lower Risk/near threatened), DD - Data Deficient, LC - Least Concern (includes LR/lc - Lower Risk, least concern)

As there are about 13 species of gliding squirrels in Borneo and 3 species are endemic to Borneo (Phillips & Phillips, 2016), this rapid assessment shed light on the distribution of these mammals in Borneo. Weather was a strong factor in influencing the sampling results. Weather conditions such as rain affect the rate of capture of non-volant small mammals (Gentry et al., 1966). The survey was done during the rainy season in Kadamaian - Kinabalu Park. Sciurids and avifauna were inactive and have a higher tendency to be in their nests during this weather condition.

### *Species Account of the Sciurids*

#### 1. Red giant gliding squirrel - *Petaurista petaurista* (Pallas, 1766)

Phillips and Phillips (2016) mentioned this species is the most common gliding squirrel that can be found in all types of lowlands to 900m altitude forests in Borneo. Throughout Malaysia, it is also the species that can be found easily in forest. They are easily spotted because of their size and colour. Recent findings on the ecology of this species include limestone habitat usage (Miard et al., 2020). Although this species is still listed as Least Concern (LC) by the IUCN Red List of Threatened Species (Duckworth, 2016), threats such as habitat loss is still

looming throughout their distribution. This species was found in Kipungit trail (No.2).

2. Black gliding squirrel - *Aeromys tephromelas* (Günther, 1873)

Although this species is widespread in Borneo, its occurrence is scarce and little is known about it. This species is smaller in size compared to the red giant gliding squirrel and the colour of its body is full black. It is listed as Data Deficient under the IUCN Red List of Threatened Species (Lee, 2016). This species was found in Nopungguk trail (No.1).

3. Prevost's squirrel - *Callosciurus prevostii* (Desmarest, 1822)

One of the beautiful squirrels in Borneo, this species is diverse in its subspecies throughout Borneo and all have orange or rufous belly. Occasionally found in tall secondary forests and orchards near the forest, this study found five individuals on the same tree foraging with Plainain squirrel. The squirrel was seen on 0536 in the evening. It can be found in tall and secondary forests, orchards that are close to forest. It is diurnal and arboreal, rarely seen on the ground. Its favoured diet is ripe fruit. The IUCN Red List of Threatened Species list them as Least Concern (Cassola, 2016).

4. Plainain squirrel - *Callosciurus notatus* (Boddaert, 1785)

One of the most common squirrels in Peninsular Malaysia and Borneo and is well adapted to habitat changes. This species can be found in cultivated areas, disturbed forests, secondary forests, coastal forests, gardens and plantations. This is also the only squirrel that can easily adapt to oil palm and rubber plantations (Phillipps & Phillipps, 2016). In Borneo, it can only be distinguished from *C. adamsi* by the absence of white spot behind the ear and pale rufous underparts. It was found on 0534 on the neighbouring tree of which Prevost's squirrel was found. It is listed as Least Concern by the IUCN Red List of Threatened Species (Duckworth, 2016).

5. Low's squirrel - *Sundasciurus lowii* (Thomas, 1892)

The most common resident in primary and secondary lowland forests of Borneo, this species is always found foraging from the ground to subcanopy. Its distribution overlaps with Brooke's squirrel in the montane forest. It can be differentiated from Brooke's squirrel by its white belly and shorter, bushy, plain thicker tail. It was found on 0534 on the same tree with *C. Notatus*. The IUCN Red List of Threatened Species list it as Least Concern (Meijaard, 2016).

***Avifauna***

A total of 58 species of birds from 28 families were recorded during this assessment in and around Kadamaian-Kinabalu Park. Fifty four species are resident, one migrant and three are endemic to Borneo. Forty seven species are recognised as Least Concern (LC) while nine are listed as Near-Threatened (NT) and two Vulnerable (VU) species by the International Union for Conservation of Nature (IUCN) RedList.

As most birds are more active and forage during the early morning and late evenings, surveyors were only able to sample birds during the evening session, right before nightfall.

***Selected Species Accounts for Avifauna***

Most of the birds recorded during this assessment are common resident in Borneo and have Least Concern (LC) status globally. However, 19% of the total birds recorded are of global conservation concern based on the IUCN Red List of Threatened Species. This includes two species of Hornbills (Bucerotidae) i.e the Wreathed Hornbill and Rhinoceros Hornbill which are listed as Vulnerable (VU); and the rest are classified as Near-Threatened (NT) including the Bornean Falconet, Chestnut-collared Kingfisher, Yellow-crowned Barbet, Red-crowned Barbet, Black-and-Yellow Broadbill, Green Broadbill, Streaked Bulbul, Short-tailed Babbler and the Rail-Babbler.

Apart from globally threatened and Near-threatened, those species that were listed above are among the birds that are desirable by bird-watchers and bird photographers (pers.obs.). Bird enthusiasts will travel at great-length to complete their life list which will contribute to the ecotourism potential of the study area.

Table 2. Taxonomic checklist for Avifauna

No	Family	Species	Common Name	Obs	Status	IUCN
1	Accipitridae	<i>Spizaetus cirrhatus</i>	Changeable Hawk-Eagle	S	R	LC
2		<i>Spilornis cheela</i>	Crested Serpent Eagle	S	R	LC
3		<i>Accipiter trivirgatus</i>	Crested Goshawk	S	R	LC
4	Falconidae	<i>Microhierax latifrons</i>	Bornean Falconet	S	E	NT
5	Columbidae	<i>Streptopelia chinensis</i>	Spotted Dove	S,V	R	LC
6		<i>Chalcophaps indica</i>	Emerald Dove	S	R	LC
7		<i>Treron vernans</i>	Pink-necked Green Pigeon	S,V	R	LC
8	Psittaculidae	<i>Loriculus galgulus</i>	Blue-crowned Hanging Parrot	S,V	R	LC
9	Cuculidae	<i>Cacomantis sonneratii</i>	Banded Bay Cuckoo	V	R	LC
10		<i>Cacomantis merulinus</i>	Plaintive Cuckoo	V	R	LC
11		<i>Surniculus lugubris</i>	Drongo Cuckoo	V	R	LC
12		<i>Centropus bengalensis</i>	Lesser Coucal	S	R	LC
13		<i>Phaenicophaeus chlorophaeus</i>	Raffles's Malkoha	S	R	LC
14	Strigidae	<i>Ketupa ketupu</i>	Buffy Fish Owl	V	R	LC
15		<i>Ninox scutulata</i>	Brown Hawk-Owl	V	R	LC
16		<i>Caprimulgus macrurus</i>	Large-tailed Nightjar	V	R	LC
17	Caprimulgidae	<i>Hemiprocne longipennis</i>	Grey-rumped Treeswift	S	R	LC
18	Apodidae	<i>Actenoides concretus</i>	Chestnut-collared Kingfisher	V	R	NT
19	Alcedinidae	<i>Rhyticeros undulatus</i>	Wreathed Hornbill	S	R	VU
20	Bucerotidae	<i>Buceros rhinoceros</i>	Rhinoceros Hornbill	S,V	R	VU
21	Ramphastidae	<i>Megalaima henricii</i>	Yellow-crowned Barbet	V	R	NT
22		<i>Megalaima chrysopsis</i>	Golden-faced Barbet	V	E	LC
23		<i>Megalaima rafflesii</i>	Red-crowned Barbet	V	R	NT
24	Picidae	<i>Picus miniaceus</i>	Banded Woodpecker	S	R	LC

(Continued on next page)

Table 2. (continued)

25	Eurylaimidae	<i>Eurylaimus ochromalus</i>	Black-and-Yellow Broadbill	V	R	NT
26		<i>Eurylaimus javanicus</i>	Banded Broadbill	V	R	LC
27		<i>Cymbirhynchus macrorhynchos</i>	Black-and-Red Broadbill	V	R	LC
28		<i>Calptomena viridis</i>	Green Broadbill	V	R	NT
29	Campephagidae	<i>Hemipus hirundinaceus</i>	Black-winged Flycatcher-Shrike	S	R	LC
30	Sittidae	<i>Sitta frontalis</i>	Velvet-fronted Nuthatch	S	R	LC
31	Laniidae	<i>Lanius cristatus</i>	Brown Shrike	S	M	LC
32	Irenidae	<i>Irena puella</i>	Asian Fairy Bluebird	S	R	LC
33	Dicruridae	<i>Dicrurus aeneus</i>	Bronzed Drongo	S	R	LC
34	Cisticolidae	<i>Orthotomus sericeus</i>	Rufous-tailed Tailorbird	S,V	R	LC
35	Pycnonotidae	<i>Pycnonotus atriceps</i>	Black-headed Bulbul	S	R	LC
36		<i>Pycnonotus goiavier</i>	Yellow-vented Bulbul	S	R	LC
37		<i>Pycnonotus brunneus</i>	Red-eyed Bulbul	S	R	LC
38		<i>Ixos malaccensis</i>	Streaked Bulbul	S	R	NT
39	Timaliidae	<i>Trichastoma malaccense</i>	Short-tailed Babbler	V	R	NT
40		<i>Trichastoma sepiarium</i>	Horsfield's Babbler	V	R	LC
41		<i>Malacopteron magnirostre</i>	Moustached Babbler	S	R	LC
42		<i>Stachyris erythroptera</i>	Chestnut-winged Babbler	V	R	LC
43		<i>Macronus bornensis</i>	Bold-striped Tit-Babbler	V	R	LC
44	Eupetidae	<i>Eupetes macrocerus</i>	Rail-Babbler	V	R	NT
45	Vireonidae	<i>Erpornis zantholeuca</i>	White-bellied Erpornis	S	R	LC
46	Zosteropidae	<i>Zosterops everetti</i>	Everett's White-Eye	S	R	LC
47	Muscicapidae	<i>Copsychus saularis adamsi</i>	Oriental Magpie Robin	S	R	LC
48		<i>Copsychus stricklandi</i>	White-crowned Shama	S	R	LC

(Continued on next page)

Table 2. (continued)

49	Monarchidae	<i>Terpsiphone paradisi</i>	Asian Paradise-Flycatcher	S,V	R	LC
50		<i>Rhipidura perlata</i>	Spotted Fantail	S	R	LC
51		<i>Hypothymis azurea</i>	Black-naped Monarch	S,V	R	LC
52	Dicaeidae	<i>Dicaeum cruentatum</i>	Scarlet-backed Flowerpecker	S	R	LC
53		<i>Dicaeum trigonostigma</i>	Orange-bellied Flowerpecker	S	R	LC
54		<i>Prionochilus maculatus</i>	Yellow-breasted Flowerpecker	S	R	LC
55	Nectariniidae	<i>Nectarinia jugularis</i>	Olive-backed Sunbird	S	R	LC
56		<i>Arachnothera hypogrammicum</i>	Purple-naped Spiderhunter	S	R	LC
57		<i>Arachnothera longirostra</i>	Little Spiderhunter	S	R	LC
58		<i>Arachnothera everetti</i>	Bornean Spiderhunter	S	E	LC

\*Note:

Observation Type (Obs):

S: Direct sighting on the individual using naked eyes/binoculars/cameras

V: Observation through vocalisation of the individuals which are either identified on site, or recorded and compared to online repository (*Xeno-canto.org*).

Migratory Status (Status):

R- Resident, breed and spend life history within Borneo;

M-Migrant, visits Borneo during cold months in area of origin, or passage migrant in which they spend time in Borneo before departing further south or north;

E-Endemic, can only be found on the island of Borneo or part of it.

IUCN Red List Categories (IUCN):

EX - Extinct, EW - Extinct in the Wild, CR - Critically Endangered, EN - Endangered, VU -

Vulnerable, LR/lcd - Lower Risk/conservation dependent, NT - Near Threatened (includes LR/nt

- Lower Risk/near threatened), DD - Data Deficient, LC - Least Concern (includes LR/lc - Lower

Risk, least concern)



1. Wreathed Hornbill *Rhyticeros undulatus* Shaw, 1811

A local resident species, widespread but scarce and may be locally common in suitable habitats in Borneo. This is one of the larger species of Hornbills found in the region apart from Wrinkled Hornbill (*Aceros corrugatus*), Rhinoceros Hornbill (*Buceros rhinoceros*) and Helmeted Hornbill (*Rhinoplax virgil*) (Phillips & Phillips, 2016). This species of Hornbill is semi-nomadic and has a wide range, foraging in big flocks and roosts communally (Myers, 2009). This is also the only hornbill species found at higher elevations up to 3,300m a.s.l in Mount Kinabalu, with preference at hill forests (Phillips & Phillips, 2016). An arboreal frugivores which feeds primarily on figs and other fruits, as well as invertebrates including snails and other small vertebrates (Smythies et al., 2000). This species is protected in Sabah under the Wildlife Conservation Enactment.

2. Rhinoceros Hornbill- *Buceros rhinoceros* Linnaeus, 1758

Another larger species of resident hornbill, the Rhinoceros Hornbill is locally common throughout forests in Borneo, but are heavily hunted traditionally by the locals (Phillips & Phillips, 2016). This species prefer the lowland areas of the primary forest, but can be found up to 1,750m a.s.l (Myers, 2009). This species are usually found in pairs, or with a juvenile. However they do flock outside of breeding season. They usually forage at the top canopy layer and the emergent, and call with a distinct barking-like sound usually before taking off-flights. Feeds primarily on figs and other fruits but also consume large insects, spiders, lizards and eggs (Myers, 2009). Like the Wreath Hornbill, this species is also protected in Sabah under the Wildlife Conservation Enactment.

3. Borneon Falconet - *Microhierax latifrons* Sharpe, 1879

One of the smallest birds-of-prey in the world, the Borneon Falconet is also endemic to Sabah. This species prefers forest edges and open-areas, and can also be found in primary and secondary lowland forests (Myers, 2009). Phillips and Phillips (2016) claim this species is widespread in Sabah, and is more commonly found in hilly areas. As the name suggests, this bird is a predator and prefers to sally-hunt insects and small vertebrates (Smythies et al., 2000).

4. Chestnut-collared Kingfisher - *Actenoides concretus* (Temminck, 1825)

A species of forest kingfisher that inhabits dense primary lowland forest up to 1650 m a.s.l (Smythies, 1999; Myers, 2009). This species is usually found in the middle to lower storey (Smythies, 1999), by still-perching and usually unobtrusive while hunting for large invertebrates and vertebrates usually from the ground (Myers, 2009). It is the commonest Kingfisher in the hills and submontane areas (Phillips & Phillips, 2016) and is one of the kingfisher species

that can be found far from water-bodies such as streams and rivers (Smythies et al., 2000; Myers, 2009).

5. Yellow-crowned Barbet - *Megalaima henricii* (Temminck, 1831)

This species of Barbet is more commonly found in hills and submontane areas throughout Borneo up to 1,200m a.s.l before being replaced by the Golden-naped Barbet (*Megalaima pulcherrima*) at the ecotone and montane forest (Phillips & Phillips, 2016). They are more often heard rather than seen due to their behaviour of spending time on the crown of trees at the upper storey foraging for food while giving out loud distinct calls.

6. Red-crowned Barbet - *Megalaima rafflesii* (Lesson, 1839)

A locally common resident throughout Borneo especially at disturbed areas and degraded forests (Smythies et al., 2000; Phillips & Phillips, 2016). Similar to most other species of Barbets, they are typically known as arboreal frugivores where diets may includes figs, berries and occasionally grubs (Smythies et al., 2000).

7. Black-and-Yellow Broadbill - *Eurylaimus ochromalus* (Raffles, 1822)

One of the most common Broadbill found in the region and exists in most habitats up to 1,200m a.s.l (Myers, 2009; Phillips & Phillips, 2016). This species flocks in groups and feed on smaller prey especially insects from the middle-storey to upper-storey and crowns. Smythies (1999) noted the behaviour of this species occupying a look-out perch before sallying for insects in the foliage. Calls are distinguishable from other birds but may be mistaken to the call of its sister species, the Banded Broadbill (*Eurylaimus javanicus*).

8. Green Broadbill - *Calyptomena viridis* (Raffles, 1822)

A common resident throughout Borneo up to 1,200m a.s.l where it exists in mixed dipterocarp forest and overgrown plantations (Myers, 2009). This species is more common among the other green broadbills that is endemic in Borneo including Hose's Broadbill (*Calyptomena hosii*) and Whitehead's Broadbill (*Calyptomena whiteheadi*) (Phillips & Phillips, 2016). They prefer the lower strata and primarily consume fruits, palms and figs (Smythies et al., 2000).

9. Streaked Bulbul - *Ixos malaccensis* (Blyth, 1845)

An uncommon resident throughout Borneo, this species exists in primary and secondary dipterocarp and lower montane forests up to 1,300m a.s.l (Myers, 2009). It is an arboreal frugivore and insectivore, and flocks at fruiting trees with other species of birds (Phillips & Phillips, 2016).

10. Short-tailed Babbler - *Trichastoma malaccense* (Hartlaub, 1844)

A common resident throughout Borneo that inhabits the primary and secondary lowland dipterocarp, peatswamp, plantations and hill forests up to 1,600m a.s.l (Myers, 2009; Phillips & Phillips, 2016). This species is a terrestrial babbler, where it prefers the understorey and forages invertebrates including insects such as ants, black beetles and grasshoppers among litters at the forest floor- (Smythies et al., 2000).

11. Rail-Babbler - *Eupetes macrocerus* (Temminck, 1831)

This is a rare resident, occurring throughout Borneo at very low density in lowland dipterocarp and hill forests up to 1,100m a.s.l (Myers, 2009). Both Smythies (1999) and Phillips and Phillips (2016) concur this species prefers hilly slopes and submontane habitat of their range. They are mostly confined to the ground level, and prefer to walk rather than fly when disturbed. As a terrestrial bird, this species hunts invertebrates on the forest floor with great speed. This species is also known for its shyness and skittish behaviour and hence is easily overlooked by birders and researchers (Myers, 2009). Calls are almost similar to the Garnett Pitta (which does not occur in Sabah), Blue-banded Pitta and Black-and-crimson Pitta.

As this species was recorded through vocalisation, we used a playback call for the pitta species listed above which has similar call to the Rail-babbler. The calling bird in the thickets only responded to the specific call of the Rail-Babbler while ignoring the other playback calls, which further confirmed its presence in this area. The area where the bird was recorded is also at a slope of more than 50° which is known to be the preferred habitat for a rail-babbler.

## Conclusion

This assessment resulted in a checklist of sciurids and avifauna in Kadamaian - Kinabalu Park which can be further developed into an ecotourism attraction for this area. Continuous studies on the diversity, occurrence and behaviour of these wildlife are needed to capitalise on potentials of ecotourism in Kadamaian - Kinabalu Park.

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## **Short Notes**

# **Dragonflies and Damselflies (Odonata) of Kadamaian, Kinabalu Park, Sabah**

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## **Abstract**

The Odonata fauna of Kadamaian was surveyed from 15<sup>th</sup> to 19<sup>th</sup> October, 2019 during the Borneo Geographic Expedition 2019 Kadamaian. The altitude of the survey area ranged from 400 m to 850 m above sea level, representing the lower part of Kinabalu Park. A total of 23 species in nine families were recorded - 10 species in Libellulidae, three species in Platynemididae, two species each in Platystictidae, Calopterygidae and Coenagrionidae, and one species each in Chlorocyphidae, Devadattidae, Euphaeidae and Synthemistidae. Of these, only one species is a new record for Kinabalu Park - *Pericnemis dowi*. The published Odonata records were compiled to produce a species list known from Kinabalu Park. The total number of species known to Kinabalu Park is now 71. Many more parts in Kinabalu Park need to be explored for a more comprehensive Odonata fauna of the park.

**Keywords:** Biodiversity, Borneo, expedition, new records, species list

## **Introduction**

Dragonflies and damselflies are conspicuous insects in the order Odonata. They are well distributed in the tropical and subtropical regions. The nymphs are aquatic but the adult insects are terrestrial. They are an important biological component of freshwater ecosystems. According to the IUCN, Odonata are one of the four major taxonomic groups (fish, aquatic plants, molluscs and Odonata) for biodiversity assessment of wetlands (Springate-Baginski et al. 2009). Odonata are a good indicator for water quality and aquatic ecosystem health. Close to 6,000 Odonata species are distributed throughout the world (Dijkstra et al., 2013). In Malaysia, more than 400 species have been recorded (Choong et al., 2017), and Borneo has at least 300 species (Orr, 2003).

Kinabalu Park is located at the northern tip of Borneo Island in the state of Sabah, Malaysia. It is Malaysia's first UNESCO World Heritage Site for its significant biota. The park covers an area 754 square kilometres (Hämäläinen, 1994). Much of Mount Kinabalu is located inside the park with its highest peak at 4,095 m - the highest mountain in Southeast Asia. The Odonata of Kinabalu Park have been well studied. Hämäläinen (1994) compiled most Odonata records of Kinabalu Park from literature, and he listed 38 species. However, Hämäläinen (1994) missed a few published records (Laidlaw, 1931; Asahina & Kitagawa, 1992; Tsuda & Kitagawa, 1989). Later on, Odonata records for Kinabalu Park are found in various publications (Orr, 2003; Malkmus, 2005; Dow, 2010, 2017; Dow et al., 2015, 2016, 2017, 2020; Dow & Orr, 2010; Orr & Hämäläinen, 2013). Apart from all these published records, a few additional Odonata species were recorded by Rory Dow at Poring Hot Springs and Sayap during the 2012 Dutch/Malaysian Expedition to Kinabalu Park and Crocker Range National Park - *Drepanosticta cf crenitis* Lieftinck, 1933, *Vestalis amaryllis* Lieftinck, 1965 and *Coelliccia borneensis* (Selys, 1886) (Dow personal communication, 2021). Rory Dow and Graham Reels spotted and photographed *Orolestes wallacei* (Kirby, 1889) at Poring Hot Springs during their visit to the Kinabalu Park headquarters in 2005 (Dow personal communication, 2021). These records have not been published, but are added to the Kinabalu Park list with permission. Kadamaian is located at the northwest of the park (N6° 12'14.2" E116° 30' 34.6"). To our knowledge, no Odonata record has been available from this part of Kinabalu Park. In this paper we report the Odonata species found at Kadamaian. At the same time, we produce an updated Odonata list for Kinabalu Park.

## Methodology

The Odonata of Kadamaian was surveyed from 15<sup>th</sup> to 19<sup>th</sup> October, 2019 during the Borneo Geographic Expedition 2019 Kadamaian. The altitude of the survey locations ranged from 400 m to 850 m above sea level (Table 1), and this represents the lower part of Kinabalu Park. The survey was carried out at various aquatic habitats found at the locations - streams, rivers, waterfalls and water pools. Adult insects were caught using an aerial net. The collected specimens were treated with acetone and then dried in silica gel for preservation. Identification to species level was done based on references and comparison with other specimens in collection. The specimens are kept in BORNEENSIS (Universiti Malaysia Sabah).



Table 1. Sampling locations in Kadamaian

Location	Description of aquatic habitat
A	Kadamaian campsite; rivers and small streams; N6° 12'14.2" E116° 30' 34.6"
B	waterfall, rivers and small streams; N6° 10'47.1" E116° 29' 11.1"
C	waterfalls, rivers and small streams; N6° 15'19.5" E116° 29' 38.6"
D	Wasai Waterfall; waterfall and river; N6° 13'24.2" E116° 30' 18.2"

## Results

For a period of five days of survey (15<sup>th</sup> to 19<sup>th</sup> October, 2019), a total of 23 Odonata species in nine families were recorded (Table 2; columns A-D). Family Libellulidae dominated the list with 10 species, and this was followed by family Platynemididae with three species. Families Platystictidae, Calopterygidae and Coenagrionidae had two species each while one species each was recorded for families Chlorocyphidae, Devadattidae, Euphaeidae and Synthemistidae. Of these, only one species is a new record for Kinabalu Park, i.e. *Pericnemis dowi* Orr & Hämäläinen, 2013. The previously known records (70 species) are listed in Table 2 (column E). The total number of species known from Kinabalu Park is now 71. Some of the species photographed at Kadamaian are shown in Figure 1.

**Table 2.** Odonata species recorded at Kinabalu Park. Columns A–D: sampling locations in Kadamaian (see Table 1), and column E: previously known records for Kinabalu Park.

\* indicates new record for Kinabalu Park.

No.	Family/Species	A	B	C	D	E	IUCN status
Family Lestidae							
1	<i>Orolestes wallacei</i> (Kirby, 1889)					/	LC
Family Platystictidae							
2	<i>Drepanosticta actaeon</i> Laidlaw, 1934					/	LC
3	<i>Drepanosticta barbatula</i> Lieftinck, 1940					/	DD
4	<i>Drepanosticta cf crenitis</i> Lieftinck, 1933					/	-
5	<i>Drepanosticta rufostigma</i> (Selys, 1886)	/				/	LC
6	<i>Protosticta joeponi</i> Dow, Phan & Choong, 2020	/				/	NA
7	<i>Protosticta kinabaluensis</i> Laidlaw, 1915					/	VU
8	<i>Telosticta fugispinosa</i> Dow, Afendy & Rahman, 2016					/	NT

<hr/>				
	Family Calopterygidae			
9	<i>Matronoides cyaneipennis</i> (Förster, 1897)		/	LC
10	<i>Neurobasis longipes</i> Hagen, 1887		/	LC
11	<i>Vestalis amaryllis</i> Lieftinck, 1965		/	LC
12	<i>Vestalis amnicola</i> Lieftinck, 1965	/	/	LC
13	<i>Vestalis anacolosa</i> Lieftinck, 1965		/	LC
14	<i>Vestalis beryllae</i> Laidlaw, 1915	/	/	LC
<hr/>				
	Family Chlorocyphidae			
15	<i>Heliocypha biseriata</i> (Selys, 1859)	/	/	LC
16	<i>Rhinocypha aurofulgens</i> Laidlaw, 1931		/	LC
17	<i>Rhinocypha humeralis</i> Selys, 1873		/	LC
18	<i>Rhinocypha moultoni</i> Laidlaw, 1915		/	DD
19	<i>Rhinocypha stygia</i> Förster, 1897		/	NT
20	<i>Rhinoneura villosipes</i> Laidlaw, 1915		/	VU
<hr/>				
	Family Devadattidae			
21	<i>Devadatta aran</i> Dow, Hämäläinen & Stokvis, 2015		/	LC
22	<i>Devadatta tanduk</i> Dow, Hämäläinen & Stokvis, 2015	/	/	DD
<hr/>				
	Family Euphaeidae			
23	<i>Euphaea basalis</i> (Laidlaw, 1915)		/	NT
24	<i>Euphaea subcostalis</i> Selys, 1873	/	/	LC
25	<i>Euphaea subnodalis</i> (Laidlaw, 1915)		/	LC
<hr/>				
	Family Platycnemididae			
26	<i>Coelliccia arcuata</i> Lieftinck, 1940	/	/	LC
27	<i>Coelliccia borneensis</i> (Selys, 1886)		/	LC
28	<i>Coelliccia cyaneothorax</i> Kimmins, 1936		/	LC
29	<i>Coelliccia cf nemoricola</i> Laidlaw, 1912	/	/	-
#30	<i>Copera vittata</i> (Selys, 1863)		/	LC
31	<i>Prodasineura hyperythra</i> (Selys, 1886)	/	/	LC
<hr/>				
	Family Coenagrionidae			
32	<i>Agriocnemis femina</i> (Brauer, 1868)		/	LC
33	<i>Ceriagrion bellona</i> Laidlaw, 1915		/	LC
34	<i>Ceriagrion cerinorubellum</i> (Brauer, 1865)		/	LC
35	<i>Ischnura senegalensis</i> (Rambur, 1842)		/	LC
*36	<i>Pericnemis dowi</i> Orr & Hämäläinen, 2013	/		NT
37	<i>Pericnemis kiautarum</i> Orr & Hämäläinen, 2013		/	VU
38	<i>Pseudagrion perfucatum</i> Lieftinck, 1937		/	NA
39	<i>Stenagrion dubium</i> (Laidlaw, 1912)	/	/	LC
<hr/>				
	Family Aeshnidae			
40	<i>Gynacantha basiguttata</i> Selys, 1882		/	LC
41	<i>Indaeschna grubaueri</i> (Förster, 1904)		/	LC

42	<i>Tetracanthagyna degorsi</i> Martin, 1895		/	NA		
Family Gomphidae						
43	<i>Leptogomphus coomansi</i> Laidlaw, 1936		/	LC		
44	<i>Leptogomphus pasia</i> van Tol, 1990		/	DD		
45	<i>Leptogomphus pendleburyi</i> Laidlaw, 1934		/	LC		
46	<i>Leptogomphus williamsoni</i> Laidlaw, 1912		/	LC		
47	<i>Megalogomphus buddi</i> Dow & Price, 2020		/	NA		
48	<i>Sieboldius japonicus</i> Selys, 1854		/	LC		
Family Chlorogomphus						
49	<i>Chlorogomphus dyak</i> (Laidlaw, 1911)		/	DD		
Family Macromiidae						
50	<i>Macromia euterpe</i> Laidlaw, 1915		/	DD		
Family Synthemistidae						
51	<i>Macromidia fulva</i> Laidlaw, 1915	/	/	LC		
Family Corduliidae						
52	<i>Procordulia fusiformis</i> Lieftinck, 1977		/	LC		
Family Libellulidae						
53	<i>Agrionoptera insignis</i> (Rambur, 1842)		/	LC		
54	<i>Cratilla lineata</i> (Brauer, 1878)		/	LC		
55	<i>Cratilla metallica</i> (Brauer, 1878)	/	/	LC		
56	<i>Diplacodes trivialis</i> (Rambur, 1842)	/	/	LC		
57	<i>Hylaeothemis clementia</i> Ris, 1909	/	/	LC		
58	<i>Lyriothemis cleis</i> Brauer, 1868		/	LC		
59	<i>Lyriothemis magnificata</i> (Selys, 1878)		/	LC		
60	<i>Neurothemis ramburii</i> (Brauer, 1866)		/	LC		
61	<i>Neurothemis terminata</i> Ris, 1911		/	LC		
62	<i>Orthetrum chrysis</i> (Selys, 1891)	/	/	LC		
63	<i>Orthetrum glaucum</i> (Brauer, 1865)	/	/	LC		
64	<i>Orthetrum pruinosum schneideri</i> Förster, 1903		/	LC		
65	<i>Orthetrum sabina</i> (Drury, 1770)	/	/	LC		
66	<i>Orthetrum testaceum</i> (Burmeister, 1839)	/	/	LC		
67	<i>Pantala flavescens</i> (Fabricius, 1798)	/	/	LC		
68	<i>Trithemis aurora</i> (Burmeister, 1839)		/	LC		
69	<i>Trithemis festiva</i> (Rambur, 1842)	/	/	LC		
70	<i>Zygonyx iris errans</i> Lieftinck, 1953		/	LC		
71	<i>Zyxomma obtustum</i> (Albarda, 1881)		/	LC		
Total number		12	10	4	3	70

# Härmäläinen (1994) states “teneral whitish specimen of *Copera*, most likely *C. marginipes*”, but he failed to collect the specimen. The description of the teneral matches teneral *Copera vittata*. Therefore, it is listed here as *Copera vittata*.



**Figure 1.** Some of the Odonata species photographed at Kadamaian. A. *Hylaeothemis clementia*, B. *Trithemis festiva*, C. *Pantala flavescens*, D. *Vestalis amnicola*, E. *Euphaea subcostalis*, F. *Coeliccia arcuata*, G. *Protosticta joepani* and F. *Pericnemis dowi*.

## Discussion

The number of species recorded from the expedition was not high. This was mainly due to the limited type of aquatic habitat in the survey locations - rivers, streams and waterfalls. Of course, another contributing factor to the low number of species is the short period of survey (only five days). However, we still managed to add one species to the species list of Kinabalu Park (Table 2, marked with \*). This might have indicated that the species list of Kinabalu Park (Table 2) is far from complete. We believe that many more common species have yet to be recorded for Kinabalu Park. It is also worthwhile to note that Kinabalu Park is a refuge for a good number of uncommon species, and quite a few species were described from there. Hämäläinen (1994) provided information on 13 species with type locality from Kinabalu Park. Later on, a few more species were described from the park - Orr & Hämäläinen (2013) described *Pericnemis kiautarum* from Poring Hot Springs, and Dow et al. (2015) described *Devadatta tanduk* from Poring Hot Springs.

The most interesting species recorded from Kadamaian were *Pericnemis dowi* and *Protosticta joepani* Dow, Phan & Choong, 2020. A male of *P. dowi* was spotted on vegetation next to a forest stream at location A, and it was photographed (Figure 1H). This species was described from Brunei, and it is also found in various parts of Sarawak such as Gunung Mulu National Park, Lanjak Entimau Wildlife Sanctuary, the Kelilngklang Range etc. (Orr & Hämäläinen, 2013; Dow et al., 2018). Kadamaian is the first location of this species in Sabah, and therefore it is a new record for Sabah. Sabah now has three *Pericnemis* species - *P. dowi*, *P. kiautarum* and *Pericnemis triangularis* (Laidlaw, 1931), and the first two species are found in Kinabalu Park. *Protosticta joepani* was recorded at forest path by a stream in location A. It is a recently described species from Borneo (Dow et al., 2020). In Sabah, this species has been recorded in the Crocker Ranger (Afendy et al., 2017), the Imbak Canyon (Choong & Chung, 2019), as well as other parts of Kinabalu Park - Poring Hot Springs and Sayap (Dow et al., 2020).

As it is now, 71 species in 15 families are known from Kinabalu Park. This number of species is not high for an area claimed to have high level of biota. Therefore, more effort is needed to explore many other parts of the park.

## Conclusion

Kinabalu Park is a refuge for some rare and uncommon Odonata species endemic to Borneo. At present the Odonata list of Kinabalu Park has 71

species. It must be noted that this species list is far from complete as many parts of the park have yet to be explored for Odonata. Nevertheless, the species list may provide a reference for the study of Odonata diversity in Sabah and Borneo.

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## Short Notes

# Butterfly Fauna in Kadamaian Area, Kota Belud, Sabah: A Survey during Borneo Geographic Expedition 2019

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## Abstract

During the Borneo Geographic Expedition 2019 in Kadamaian area in Kota Belud, a survey on butterfly fauna was conducted for four days from 21<sup>st</sup> to 24<sup>th</sup> October, 2019. Three sites selected for the butterfly sampling were Site 1, Site 2 and Site 4. The methods applied were fruit and carrion baited traps, and aerial netting. A total of 56 individuals were sampled and belonged to 25 species from four families (Nymphalidae, Pieridae, Lycaenidae and Papilionidae). Nymphalidae was the dominant family with *Ragadia makuta* recorded as the most abundant species. About 60% of the butterflies sampled in the area are forest species, while 40% of the overall species have narrow geographical distribution restricted to Sundaland. The butterfly fauna in Kadamaian area is comparable to other forest types in Sabah in terms of their diversity and species richness. The findings reflected the potential of Kadamaian area as a nature tourism site, and the area could also serve as a corridor for the conservation of flora and fauna as it is located adjacent to Kinabalu Park.

**Keywords:** Butterflies, diversity, forest species, Kadamaian area, conservation

## Introduction

The Malaysian State of Sabah, as part of Borneo, is well-known as one of the world's biodiversity hotspots, including diverse insect species. Many beautiful and endemic species of butterflies can be found in Sabah. There are about 950 species of butterfly species that have been recorded on Borneo (Otsuka, 2001), of which 81 species (8.5%) are endemic (Gohun et al., 2021). Butterflies are classified under the order Lepidoptera and suborder Rhopalocera. They play an important role as a pollinator by transferring pollens to the stigma of flowers (Webb, 2008). Butterflies are also widely accepted as a bioindicator of habitat quality as they are very sensitive to subtle changes in their surrounding (Mobeen et al., 2016).

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The Borneo Geographic Expedition 2019 was carried out in Kadamaian in Kota Belud, Sabah. This expedition was co-organised by Universiti Malaysia Sabah and Sabah Parks and conducted from 14<sup>th</sup> to 25<sup>th</sup> October, 2019. Among the objectives of the expedition was to survey the biodiversity of flora and fauna in the area, including the butterfly faunal composition which consists of five families.

## Methodology

### a. Background of the study area

Kadamaian is located at the north-west of Sayap substation (Kinabalu Park) (Figure 1). Three sites were surveyed for this study namely Podos-Nopungguk Trail (Site 1), Melangkap Noriou (Site 2), and Kampung Tinata (Site 4). A basecamp site was set up in Kampung Podos at Podos Heritage Homestay (N06°12'44.2" E116°30'31.2") at about 417 m.a.s.l (Figure 2).

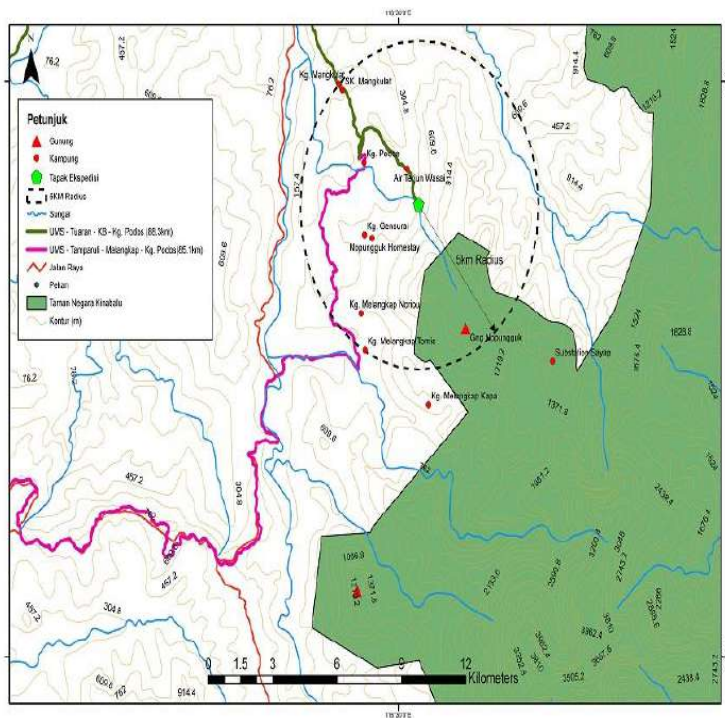


Figure 1. Map of the expedition area



Figure 2. Basecamp site at Podos Heritage Homestay, Kg. Podos

*b. Sampling methods*

The butterfly samplings were conducted from 21<sup>st</sup> to 24<sup>th</sup> October, 2019. The sampling was done from 8.00 am until 6.00 pm. Baited-traps and aerial netting techniques were used to sample the butterflies. Both techniques were used simultaneously in this survey to increase the sample size.

*i. Baited-trap technique*

This technique is widely used to sample butterflies that belong to the fruit feeding guild, most are members of Nymphalidae (De Vries et al., 1997) and also butterflies that are attracted to carrion (Hamer et al., 2006). In this survey, ten baited traps were set up only at Site 1. The traps were hung on the tree branches and placed at 50 m intervals along a 500 m transect. Ripe bananas and shrimp paste were used as baits. Five traps were baited with ripe bananas while the other five traps were baited with shrimp paste, placed alternately. All traps were checked twice daily (in the morning and afternoon).

*ii. Aerial netting technique*

The aerial netting technique is usually used to sample butterflies that feed on nectar, which cannot be sampled by using the baited-trap technique. At Site 1, the sampling of butterflies using aerial netting was conducted for 10 minutes at each of the trapping stations. Butterflies were captured within a 5 m radius at each station (Figure 3). At Site 2 and Site 4, the butterflies were captured randomly by using this method.

### *iii. Catch, mark and release*

In order to reduce the number of butterflies killed, individuals that could be identified in the field were marked and released. The butterflies were marked with a marker pen at the underside of their wings before they were released (Figure 3). For each of the species sampled and identified in the field, three or fewer individuals were taken as specimens.



Figure 3. Catch, mark and release technique

### *c. Preservation and Identification*

All the individuals that could not be identified in the field were taken as specimens and brought back to the laboratory at the Institute for Tropical Biology and Conservation (ITBC), Universiti Malaysia Sabah (UMS) for the preservation and identification processes. The specimens were identified based on Otsuka (1988) and the collections in BORNEENSIS at ITBC, UMS.

### *d. Data Analysis*

The data were analyzed using descriptive and statistical analyses. The descriptive analysis was done in Microsoft Excel 2010, while the statistical analysis was performed by using Paleontological Statistics Software (PAST) version 4.03. The butterfly diversity was measured using the Shannon-Wiener diversity index ( $H'$ ), Simpson's Index ( $D$ ) and Margalef's Index ( $D_{Mg}$ ).

i. *Shannon-Wiener Diversity Index (H')*

This index is a measure of diversity that combines species richness and their relative abundance. The diversity in a community increases as H' increases, and the values usually range between 1.5 and 3.5, and rarely exceeds 4 (Magurran, 2004).

$$H' = - \sum_{i=1}^S (p_i \ln p_i)$$

$p_i$  = proportion of species  $i$  in population

$S$  = total number of species

ii. *Simpson's Diversity Index (D)*

Simpson's Diversity Index is a measure of diversity which takes into account the number of species present, as well as the relative abundance of each species. The value of Simpson's  $D$  ranges from 0 to 1. As species richness and evenness increase, diversity increases. With this index, the greater value of  $D$ , the greater diversity of the sample (Moore, 2013)

$$D = 1 - \left( \frac{\sum n(n-1)}{N(N-1)} \right)$$

$n$  = the total number of organisms of a particular species

$N$  = the total number of organism of all species

iii. *Margalef's Index ( $D_{Mg}$ )*

Margalef's Index is a measure of species richness (Magurran, 2004)

$$D_{Mg} = S - 1 / \ln(N)$$

$S$  = the number of species,

$N$  = the total number of individuals in the sample

## Results and Discussion

A total of 56 individuals comprising of 25 species from four families (Nymphalidae, Pieridae, Lycaenidae and Papilionidae) were sampled during the expedition. Nymphalidae was the dominant family with 18 species and 43 individuals (Figure 4). Many members of this family are attracted to ripe fruits, carrion and animal excretion (Corbet & Pendlebury, 1992; De Vries, 1988). Nymphalinae and Satyrinae were the dominant subfamilies with seven species

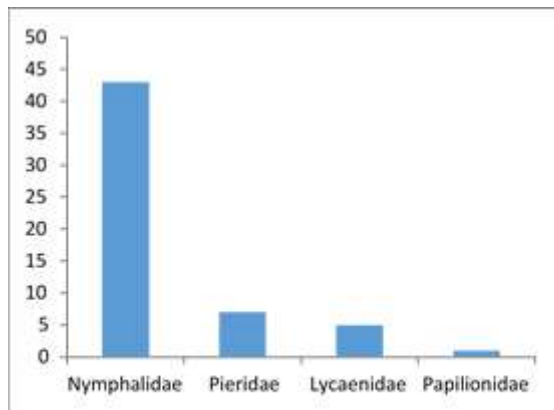


Figure 4. The number of species based on the families

recorded, respectively. In terms of abundance, Satyrinae recorded more individuals (22 individuals) compared to Nymphalinae (15 individuals).

Satyrinae is generally a small to medium-sized butterfly. Its wings are usually dull brown in colour with submarginal eye-spots. Most of the species in this subfamily are weak in flight and prefer shady conditions. The members in this subfamily are frequently found among low herbage, and some species are confined to the forest habitat. The hostplants of this subfamily are mainly Gramineae (grasses) and Palmae (palms) (Corbet & Pendlebury, 1992). Nymphalinae consists of butterflies of medium to large size. Many species have bright and beautiful wing colour patterns. The subfamily Nymphalinae are usually sun-loving butterflies and often can be seen at flowering plants in gardens and forests. This subfamily is known as strong and rapid fliers (Corbet & Pendlebury, 1992). *Ragadia makuta* was recorded as the most abundant species in the survey. The result could be explained by the abundance of the hostplant *Selaginella* spp. in the area (Figure 5). *Ragadia makuta* distribution is highly dependent on the presence of *Selaginella* spp. as the adult and larval hostplant (Hill et al., 2003).

An interesting finding from this survey is more than half (60%) of the total species sampled during this expedition are considered forest species (Table 1). The result may reflect the study area as a suitable forest habitat for many forest species. No species endemic to Borneo were found in this area. However, about 40% of the species sampled have a narrow geographical distribution (restricted to Sundaland) (based on Corbet & Pendlebury (1992), Otsuka (1988) Otsuka (2001). This includes an individual of *Trogonoptera brookiana* which is listed as

protected species under CITES (CITES, 2013) or least concern by IUCN (Malaysia Biodiversity Information System, 2020). This remarkable species is also known as Rajah Brooke's Birdwing and frequents habitats with streams. Phon et al. (2018) reported this species is facing heavy commercial exploitation and habitat loss: between 2001 and 2010, the total reported export trade of wild specimens was 5,060 individuals.



Figure 5. *Selaginella* spp. at the study area

In terms of species diversity, Kadamaian ( $H' = 3.04$ ) is comparable to Kangkawat ( $H' = 3.64$ ), as reported by Norradhihah et al. (2000) where 43 species had been sampled during four days of sampling in this area, which is part of Imbak Canyon Conservation Area, Class 1 (Protection) Forest Reserve. As for species richness, Chung et al. (2019) reported that 17 species of butterflies were recorded in five days during a survey at Sg. Rawog Conservation Area in Segaliud Lokan Forest Reserve, which is a logged-over forest, as compared to 25 species recorded in Kadamaian area.

**Table 1.** The list of butterfly species and the number of individuals recorded in the study area

Family	Subfamily	Species	No.individual	
Nymphalidae	Nymphalinae	<i>Bassarona dunya</i> <sup>a,b</sup>	5	
		<i>Discophora necho</i> <sup>a,b</sup>	2	
		<i>Dophla evelina</i> <sup>a</sup>	2	
		<i>Euthalia iapis</i> <sup>a,b</sup>	1	
		<i>Junonia atlites</i>	2	
		<i>Kallima limborgii</i> <sup>a,b</sup>	2	
		<i>Lassipa heliodore</i>	1	
		<i>Mycalesis anapita</i> <sup>a,b</sup>	1	
	Satyrinae	<i>Mycalesis fusca</i> <sup>a,b</sup>	2	
		<i>Mycalesis orseis</i> <sup>a</sup>	4	
		<i>Ragadia makuta</i> <sup>a,b</sup>	7	
		<i>Xanthotaenia busiris</i> <sup>a</sup>	3	
		<i>Ypthima baldus</i>	1	
		<i>Ypthima pandocus</i>	4	
		Morphinae	<i>Faunis canens</i> <sup>a</sup>	1
			<i>Faunis kirata</i> <sup>a,b</sup>	2
	<i>Melanocyma faunala</i> <sup>a</sup>		1	
	Pieridae	Coliadinae	<i>Taenaris horsfieldii</i> <sup>a,b</sup>	2
			<i>Eurema hecabe</i>	2
<i>Eurema sari</i>			2	
<i>Eurema simulatrix</i>			1	
Lycaenidae	Lycaeninae	<i>Gandaca harina</i>	2	
		<i>Neopithecops zalmora</i> <sup>a</sup>	4	
Papilionidae	Papilioninae	<i>Caleta elna</i>	1	
		<i>Trogonoptera brookiana</i> <sup>a,b</sup>	1	
Total	25 species		56 individuals	

**Note:** Forest species are indicated with 'a' and species with narrow geographical distribution are denoted by 'b' (based on Corbet & Pendlebury, 1992; Otsuka, 1988; Otsuka, 2001)

## Conclusion

The findings from this survey highlighted the value of the Kadamaian area for the conservation of butterfly fauna with the occurrence of many forest species as well as species of narrow geographical distribution and also a protected species under CITES. Moreover, the Kadamaian area is located adjacent to Kinabalu Park, and could therefore serve as a corridor for the persistence of many fauna and flora species. Kadamaian also has great potential to be promoted as a nature tourism site. An effective approach needs to be put forward to enhance conservation awareness among villagers and the authorities.



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**Research Article**

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**Chemical Composition of Essential Oil from *Etlingera coccinea* (Blume) S. Sakai & Nagam in Kadamaian, Kota Belud, Sabah**

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**Abstract**

*Etlingera coccinea* (Blume) S. Sakai & Nagam is a member of *Zingiberaceae* family. It is commonly known as ‘Tuhau’ in Sabah, Malaysia and consumed as a local delicacy and used as a traditional remedy for stomachache, food poisoning, and gastric problems. The plant has been reported to have bioactive properties such as anticancer, antioxidant and antibacterial. Due to the high demand for this bioactive compound in national and international markets, chemical profiling of leaves, stems and rhizomes from *E. coccinea* was carried out. Eight germplasms were collected from Trail 1 (Kg. Gensurai) and Trail 2 (Kg. Melangkap Noriou) and submitted to the hydrodistillation process to obtain the essential oil before analysing with GC-MS. From the result obtained, a total of 85 compounds were found and 26 of these were terpenoid compounds. There are several classes of compound present in different parts of *E. coccinea*, such as, monoterpene, sesquiterpene, alcohol, aldehyde, alkane, alkene, ketones, fatty acids derivatives, esters, amines as well as norterpene. Most of the compounds found in *E. coccinea* are monoterpenes at 18 compounds, followed by alcohols (14 compounds), alkanes and alkenes (12 compounds), sesquiterpenes (8 compounds), aldehydes (7 compounds), ketones, fatty acid derivatives, esters (4 compounds), and lastly, amine and norterpene, one compound each. However, total terpenoids from all plant parts from both sites were less than 50% of total abundance. Only the rhizome part from site 2 showed the highest terpenoid abundance (43.34%). Hence, the identified compounds from the study could be expended for large-scale profiling to obtain higher yields of important constituents.

**Keywords:** *Etlingera coccinea*, Essential oil, Chemical composition, GC-MS

**Introduction**

The genus *Etlingera* belongs to the *Zingiberaceae* family (Tachai et al., 2014) and consists of many edible ginger plants (Daniel-Jambun et al., 2018). One of the most commonly known species of *Etlingera* is *Etlingera coccinea* or commonly known as “Tuhau” according to the KadazanDusun. The leafy shoots with stout rhizome produce a stingy smell when it is crushed (Jualang et al.,

2015). As stated by Naive et al. (2018), it is a terrestrial herb and primarily grows in the secondary forest at bright-lit to deeply shaded locations along streams with moist to wet soil. It also grows at an elevation between 300 to 1400 metres above sea level. *E. coccinea* is native to Java, Sumatra, Thailand, Malay Peninsula and throughout Borneo, it is also discovered in the Philippines (Naive et al., 2018).

In Sabah, Malaysia, *E. coccinea* is used by indigenous ethnic groups especially the KadazanDusun to flavour local dishes (Jualang et al., 2015). According to Naive et al. (2018), the pith of the leafy shoot is used as a condiment in Borneo and Java and is also eaten as a vegetable. The fruits are edible, and the seed oil has a characteristic aroma. It is also consumed as a pickle and utilized as a traditional remedy for stomach ache, food poisoning and gastric problems (Mahdavi et al., 2017). Apart from its ability to treat these ailments and being a source of food, it was also discovered to be used in the skincare industry as a face scrub and is commercialized (Geraldine, 2017).

To date, there are only two studies on the essential oil of *E. coccinea* and these previous reports were only focused on the rhizome part of the ginger. Vairappan et al. (2012) reported they successfully extracted essential oils from five *Etlingera* species namely, *E. pyramidosphaera*, *E. megalocheilos*, *E. Elatior*, *E. brevilabrum* and *E. coccinea*. The chemical composition of each essential oil was then screened using GC-MS analysis and tested for their cytotoxic and antibacterial activities. From the study, they found that there are 39 volatile compounds in five species of *Etlingera* and only nine of the volatile compounds found in *E. coccinea* which are 3-Thujanone, Borneol, Camphor, Cedr-9-ene, L-Calamenene, Carophyllene oxide,  $\alpha$ -Bisabolol,  $\alpha$ -Epi-murolol and Cycloartanyl acetate. The most abundant volatiles in *E. coccinea* is Borneol (25.8%).

Meanwhile, a study was conducted by Nagappan et al. (2017) on the essential oils from the rhizomes of *Etlingera* spp. including *E. coccinea*, the barks of *Cinnamomum* spp. and culms of *Schizostachyum* spp. which were extracted by hydrodistillation using Clevenger-type apparatus. The aim of this study was to investigate the diversity of volatile compounds in these extracted plants and test them for their antibacterial activity. From the result obtained, they divided the volatile compounds into two groups which are major and minor groups. They found that Borneol (28.2%) is the most abundant volatile compound in *E. coccinea* followed by Aromadendrene oxide (10.9%), Elemicin (9.7%), Lauryl aldehyde (5.9%), 1-dodecanol (3%) and 2 compounds in the minor group; Camphor (2.8%) and 5-Decen-1-ol (1.3%).

As estimated by the World Health Organisation (WHO), 65 - 80% of the primary health care needs of the world population are only achieved through plant-based traditional medicine. According to Saxena et al. (2007), "Natural and therefore safe and effective", is the cause why there is phenomenal growth in the plant-based medicinal sector. People trust plant-based medicines over synthetic ones. Despite the toxicology of natural and synthetic chemicals being quite similar as mentioned by Ames et al. (1990), there are trade-offs between natural and synthetic chemicals effects on living organisms.

Due to the fact that synthetic chemicals give a lot of negative effects towards the environment and living organisms, it raises awareness regarding the extensive use of synthetic chemicals to inhibit any illness and disease spread in humans, animals as well as plants that can cause carcinogens, clastogens (agents that break chromosomes), teratogens and mutations. It has led researchers to further study the natural chemical constituents and continue exploring the potential bioactive compound that can be useful to mankind. Since *E. coccinea* plays an important role in the plant species community, it is very crucial for the species to be fully explored in terms of its chemical content in hope to find new or existing bioactive compounds. Therefore, the purpose of this study was to investigate the chemical composition of essential oil from different plant tissues of *E. coccinea* by using Gas Chromatography-Mass Spectrometry (GC-MS) analysis.

## Methodology

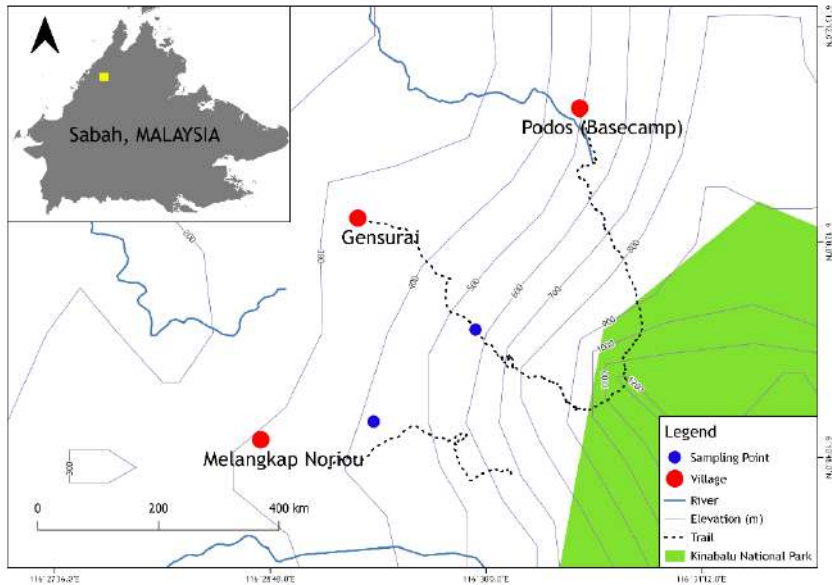
### *Sampling and Study Area*

The plant samples were collected from Trail 1 (Kg. Gensurai) and Trail 2 (Kg. Melangkap Noriou), Kota Belud, Sabah (Figure 1) on 15<sup>th</sup> to 19<sup>th</sup> October, 2019 (5 days). The whole part of *Etlingera coccinea* was collected, starting from the rhizome, stem and leaves. Only disease-free and fresh samples were collected.

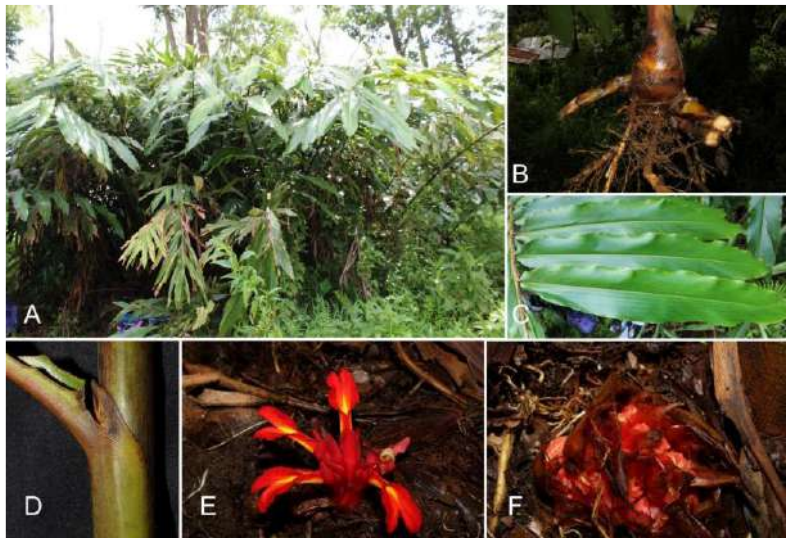
Figure 2 shows the habit, rhizome, leaves, detail of ligule, inflorescence and infructescence of *E. coccinea* (Naive et al., 2018). This species can be identified from the leaves, which are narrowly obovate lamina, sessile, ciliate margin and acuminate apex (Naive et al., 2018). *E. coccinea* also has a unique, distinct pungent smell that can be easily identified by crushing the leaves by hand.

Fresh plant samples were collected and put in plastic bags with GPS coordinate labels and the date of collection. Ropes were used to secure the plants, which

were then transported to Universiti Malaysia Sabah (UMS) for preparation of plant samples.



**Figure 1.** Sampling points during Borneo Geographic Expedition in Kadaamaian, Kota Belud. Inset: Yellow box shows Kadaamaian area in the north of Sabah.



**Figure 2.** *Etlingera coccinea* : A. Habit B. Rhizome C. Leaves D. Detail of ligule E. Inflorescence F. Infructescence (Naive et al., 2018)

### *Plant Sample Preparation*

The samples were divided into leaves, stems and rhizomes. Each sample was cut into smaller pieces and stored immediately in -80°C before further analysis.

### *Extraction of Essential Oil from Plant*

The extraction of the essential oil from *E. coccinea* was conducted following Mahdavi et al. (2017) with slight modification. 250g-300g of leaves were chopped into smaller pieces and ground together with distilled water using a blender to increase the surface area before being subjected to hydrodistillation. One litre of distilled water that mixed well with the leaves was then added and placed into a 2L round bottom flask. The 2mL of 99% (v/v) n-pentane (BHD, Germany) was added to trap the condensed oil, through the top of the condenser into the round bottom flask containing samples. The extraction was carried out at 100°C for 7 to 8 hours. The light-yellow coloured mixture of pentane, water and essential oil then dried over anhydrous sodium sulfate ( $\text{Na}_2\text{SO}_4$ ) overnight, and then filtered. Finally, the essential oil was concentrated by blowing the pentane using nitrogen gas, leaving only the essential oil kept in airtight vials at 4°C, and dark conditioned before further analysis.

### *Identification of Volatile Compound of Essential Oil*

The GC-MS analysis was based on previous experiments by Vairappan et al. (2012) with several changes. The analysis of the essential oils was performed using a Shimadzu QP-2010 gas chromatograph attached to a Shimadzu GCMSQP-2010 plus detector (Shimadzu Corp., Japan) with a SGE BPX-5 (30.0 m X 0.25  $\mu\text{m}$  i.d., film thickness 0.25  $\mu\text{m}$ ) fused silica capillary column. High purity helium was used as the carrier gas at a constant flow rate of 1.0 mL min<sup>-1</sup>. One  $\mu\text{L}$  of sample was injected in splitless mode into the GCMS using an AOC5000 auto-injector. The initial temperature was set at 40°C, then heated at a rate of 2°C min<sup>-1</sup> to 280°C and held isothermally for 5 minutes. The ion source temperature was set at 200°C and the interface temperature at 280°C. The mass spectrometer was set to operate in EI mode with ionizing energy of 70 eV and an acquisition mass range from 35 a.m.u. to 550 a.m.u. at 0.25 scan s<sup>-1</sup>. Solvent delay was set for 5 minutes.

## **Results and Discussion**

### *Sampling and study sites*

Five samples of *Etlingera coccinea* were collected along Trail 2 in three different GPS points and elevation (N 06°10'58.9", E 116°29'29.5" at 514m elevation, N 06°10'59.9", E 116°29.22.5" at 509m elevation and N 06°11'02.2", E

116°29'27.2" at 520m elevation) and three samples were collected from Trail 1 (N 06°19'18.5", E 116°49'90.4") at elevation 558m.

#### GC-MS Chromatograms

The GC-MS chromatograms of the leaves, stems and rhizomes essential oil showed the presence of a total of 85 compounds. The compounds were identified through comparison of the fragmentation patterns in the resulting mass spectra with those published in literature and using NIST mass spectral database. So far, there have been no reports on the chemical profiling of the essential oil of *E. coccinea* leaves and stems, only rhizome. The GC-MS analysis on the rhizome of *E. coccinea* have been used in the past by Vairappan et al. (2012) and Nagappan et al. (2017). Therefore, these are the preliminary data for the chemical compositions of leaves, stems and rhizomes of *E. coccinea*.

In previous studies by Vairappan et al. (2012) and Nagappan et al. (2017), the rhizome part of *E. coccinea* were extracted using hydrodistillation and essential oil obtained on GC-MS. There were nine volatile compounds detected in *E. coccinea* rhizome which are 3-Thujanone, Borneol, Camphor, Cedr-9-ene, L-Calamenene, Carophyllene oxide,  $\alpha$ -Bisabolol,  $\alpha$ -Epi-murolol and Cycloartanyl acetate (Vairappan et al., 2012). Another seven compounds found in the rhizome of *E. coccinea* by Nagappan et al. (2017) are Borneol, Aromadendrene oxide, Elemicin, Lauryl aldehyde, 1-dodecanol, Camphor and 5-Decen-1-ol. Borneol and Camphor were the only compounds found in both papers. However, in this report, Cyclododecane and Geranyl formate are the most abundant in the rhizome part of both trails and no compound in these two previous reports were found in this paper. The same happened in the case of *Etlingera brevilabrum*, no similar chemical compound of rhizome essential oil was found in both papers reported by Vairappan et al. (2012) and Mahdavi et al. (2016), even the major chemical compositions are different. According to Boaro et al. (2019), this difference in chemical composition can be influenced by abiotic factors (mineral nutrition, water, light, temperature, and soil types), and biotic factors, such as attacks of pathogens, pests, and herbivores.

Table 1 shows the volatile profiling of leaves, stems and rhizomes essential oil from *E. coccinea* by hydro-distillation technique analysed using GC-MS from both trails in Kadamaian, Kota Belud. There are several classes of compound present in different parts of *E. coccinea*, such as, monoterpene, sesquiterpene, alcohol, aldehyde, alkane, alkene, ketones, fatty acids derivatives, esters, amines and norterpene.



**Table 1.** Volatile profiling of leaves, stems and rhizomes essential oil from *Etilingera coccinea* by hydro-distillation technique analysed using Gas Chromatography-Mass Spectrometry (GC-MS)

No.	Molecular formula	Compound name	<i>Etilingera coccinea</i> (%)							
			Trail 1				Trail 2			
			(Kg. Gensurai)				(Kg. Melangkap Noriou)			
			Leaves	Stems	Rhizomes	Leaves	Stems	Rhizomes		
<b>Sesquiterpenes</b>										
1	C <sub>15</sub> H <sub>24</sub> O	10-epi-italicene ether	-	-	7.01	-	-	-	-	-
2	C <sub>15</sub> H <sub>24</sub> O	Caryophyllane 4, 8-α-epoxy	-	-	3.23	-	-	-	-	-
3	C <sub>15</sub> H <sub>22</sub> O	Dehydrofukinone	-	-	0.59	-	-	-	-	-
4	C <sub>15</sub> H <sub>26</sub> O	neo-Intermedeol	-	-	1.21	-	-	-	-	-
5	C <sub>17</sub> H <sub>28</sub> O <sub>2</sub>	Nerolidyl acetate	-	-	0.43	-	-	-	-	-
6	C <sub>16</sub> H <sub>26</sub> O <sub>2</sub>	Trans-Nerolidyl formate	-	-	0.44	-	-	-	-	-
7	C <sub>15</sub> H <sub>24</sub>	8-Guaiene	-	-	0.36	-	-	-	-	-
8	C <sub>15</sub> H <sub>24</sub>	8-Patchoulene	-	-	0.41	-	-	-	-	-
<b>Norterpene</b>										
9	C <sub>19</sub> H <sub>40</sub>	Pristane	2.11	-	-	0.83	0.89	-	-	-
<b>Monoterpenes</b>										
10	C <sub>10</sub> H <sub>18</sub> O	Isotujol	19.34	9.93	30.48	23.75	6.61	43.34	-	-
11	C <sub>10</sub> H <sub>16</sub>	8-Pinene	-	-	-	0.05	-	-	-	-
12	C <sub>10</sub> H <sub>16</sub>	3-Carene	0.14	-	-	4.03	-	-	-	-
13	C <sub>10</sub> H <sub>18</sub> O	4-Terpeneol	9.86	-	0.58	-	-	-	-	-
14	C <sub>10</sub> H <sub>16</sub> O	cis-Limonene oxide	-	-	-	0.06	0.04	-	-	-
15	C <sub>10</sub> H <sub>16</sub> O	cis-Pinocampnone	-	-	-	0.34	-	-	-	-
16	C <sub>10</sub> H <sub>16</sub>	D-Limonene	0.25	-	-	0.60	-	-	-	-
17	C <sub>10</sub> H <sub>18</sub> O	Eucalyptol	-	-	-	0.08	-	-	-	-
18	C <sub>11</sub> H <sub>18</sub> O <sub>2</sub>	Geranyl Formate	-	9.93	29.90	-	-	41.24	-	-
19	C <sub>10</sub> H <sub>18</sub> O	Isocineole	0.36	-	-	-	0.19	-	-	-
20	C <sub>10</sub> H <sub>18</sub> O	Pinocampnoneol	-	-	-	0.10	0.09	-	-	-
21	C <sub>10</sub> H <sub>16</sub>	Sabinene	-	-	-	0.15	-	-	-	-
22	C <sub>10</sub> H <sub>16</sub>	α-Phellandrene	-	-	-	0.08	-	-	-	-

(Continued on next page)

Table 1. (continued)

23	C <sub>10</sub> H <sub>16</sub>	α-Thujene	8.43	-	-	2.92	-	-
24	C <sub>10</sub> H <sub>16</sub>	B-Myrcene	0.30	-	-	14.83	5.73	1.87
25	C <sub>10</sub> H <sub>16</sub>	B-Phellandrene	-	-	-	0.07	-	-
26	C <sub>10</sub> H <sub>16</sub> O	B-Thujone	-	-	-	0.45	-	-
27	C <sub>10</sub> H <sub>16</sub>	δ-Carene	-	-	-	-	0.56	0.22
<b>Ketones</b>								
28	C <sub>6</sub> H <sub>12</sub> O	2-Hexanone	24.50	0.26	1.52	21.44	42.90	0.08
29	C <sub>10</sub> H <sub>16</sub> O <sub>2</sub>	cis-piperitone epoxide	-	-	-	-	-	0.08
30	C <sub>13</sub> H <sub>24</sub> O	Citronellyl acetone	24.50	0.26	0.39	20.42	42.90	-
31	C <sub>9</sub> H <sub>14</sub> O	Nopinone	-	-	1.13	0.12	-	-
			-	-	1.33	0.90	-	-
<b>Fatty acids derivatives</b>								
32	C <sub>10</sub> H <sub>20</sub> O <sub>2</sub>	4-Methylnonanoic acid	-	5.19	1.85	-	0.44	-
33	C <sub>12</sub> H <sub>22</sub> O <sub>2</sub>	Ethyl trans-2-decenoate	-	3.22	0.52	-	0.12	-
34	C <sub>9</sub> H <sub>18</sub> O <sub>2</sub>	Nonanoic acid / Pelargonic acid	-	-	-	-	0.32	-
35	C <sub>8</sub> H <sub>16</sub> O <sub>2</sub>	Octanoic acid	-	0.30	-	-	-	-
			-	1.67	1.33	-	-	-
<b>Esters</b>								
36	C <sub>12</sub> H <sub>22</sub> O <sub>2</sub>	(E)-2-Decenyl acetate	0.48	1.37	2.31	0.82	0.33	0.10
37	C <sub>8</sub> H <sub>12</sub> N <sub>2</sub> O <sub>2</sub>	N-tert-Butoxycarbonylimidazole	-	0.27	-	-	-	-
38	C <sub>9</sub> H <sub>13</sub> F <sub>7</sub> O <sub>2</sub>	Pentadecyl heptafluorobutyrate	-	0.86	1.94	-	-	0.10
39	C <sub>6</sub> H <sub>10</sub> O <sub>2</sub>	Vinyl butyrate	0.48	-	-	0.82	0.33	-
			-	0.24	0.37	-	-	-
<b>Amines</b>								
40	C <sub>6</sub> H <sub>14</sub> N <sub>2</sub>	1-Aminohomopiperidine	0.41	0.30	0.39	0.24	-	0.03
<b>Alkenes</b>								
41	C <sub>13</sub> H <sub>26</sub>	(2Z)-4,5-Dimethyl-2-undecene	2.55	2.09	1.60	5.85	2.12	1.61
42	C <sub>16</sub> H <sub>32</sub>	(Z)-7-Hexadecene	-	0.67	1.21	-	-	0.41
43	C <sub>12</sub> H <sub>24</sub>	1-Dodecene	-	0.24	-	-	-	-
44	C <sub>15</sub> H <sub>30</sub>	1-Pentadecene	0.91	-	-	0.53	-	-
45	C <sub>13</sub> H <sub>26</sub>	1-Tridecene	-	0.20	0.19	-	-	-
46	C <sub>13</sub> H <sub>26</sub>	1-Tridecene	-	0.16	-	-	-	-
			-	0.30	0.20	-	-	-

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Table 1. (continued)

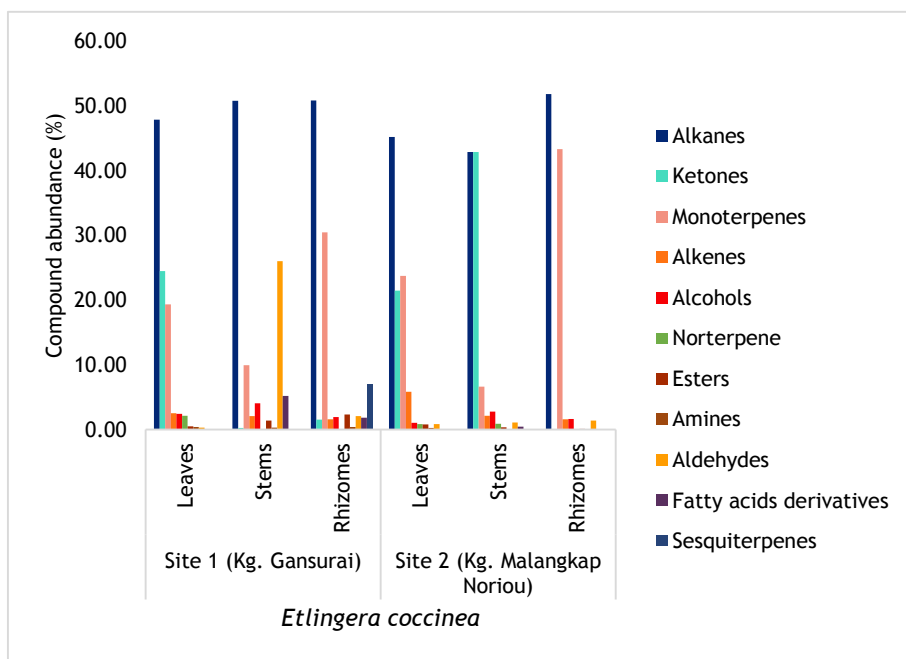
47	C <sub>14</sub> H <sub>26</sub>	2,6,10-Trimethylundeca-1,3-diene	-	-	-	-	-	-	0.40
48	C <sub>10</sub> H <sub>20</sub>	3,7-Dimethyl-1-octene	0.14	-	-	-	0.05	-	-
49	C <sub>12</sub> H <sub>24</sub>	3-Dodecene	0.56	-	-	-	0.62	1.02	0.00
50	C <sub>18</sub> H <sub>36</sub>	5-Octadecene	-	0.52	-	-	-	-	-
51	C <sub>14</sub> H <sub>28</sub>	5-Tetradecene	0.94	-	-	-	1.35	1.10	0.80
52	C <sub>10</sub> H <sub>16</sub>	Bornylene	-	-	-	-	3.29	-	-
<b>Alkanes</b>									
53	C <sub>19</sub> H <sub>40</sub>	2-Methyloctadecane	47.88	50.80	50.84	45.18	42.86	51.83	-
54	C <sub>12</sub> H <sub>24</sub>	1-Ethyl-2-heptylcyclopropane	-	0.28	-	-	-	-	0.29
55	C <sub>9</sub> H <sub>18</sub>	1-Methylpentylcyclopropane	-	0.07	0.14	-	-	-	-
56	C <sub>9</sub> H <sub>20</sub>	2,3,3-Trimethylhexane	0.19	0.41	0.62	0.13	0.16	0.24	0.24
57	C <sub>17</sub> H <sub>36</sub>	2-Methylhexadecane	-	18.03	10.25	-	-	-	3.51
58	C <sub>15</sub> H <sub>32</sub>	2-Methyltetradecane	-	12.75	7.24	-	-	-	2.44
59	C <sub>6</sub> H <sub>13</sub> NO <sub>2</sub>	2-Nitrohexane	-	1.05	1.15	-	-	-	0.25
60	C <sub>17</sub> H <sub>34</sub> F <sub>3</sub> O <sub>2</sub>	2-Trifluoroacetoxypentadecane	-	-	0.52	-	-	-	-
61	C <sub>15</sub> H <sub>32</sub>	3-Methyltetradecane	2.87	-	-	0.93	0.52	-	-
62	C <sub>10</sub> H <sub>16</sub>	5,5-Dimethyl-1-vinylbicyclo[2.1.1]hexane	-	-	-	-	-	0.30	-
63	C <sub>12</sub> H <sub>24</sub>	Cyclododecane	44.83	17.46	30.93	44.11	42.18	44.79	-
64	C <sub>13</sub> H <sub>24</sub> O <sub>2</sub>	Hexyl cyclohexanecarboxylate	-	0.76	-	-	-	-	-
<b>Aldehydes</b>									
65	C <sub>15</sub> H <sub>30</sub> O	13-Methyltetradecanal	0.31	25.99	2.07	0.85	1.09	1.37	-
66	C <sub>8</sub> H <sub>14</sub> O	2-Octenal	-	0.54	-	-	-	0.28	-
67	C <sub>12</sub> H <sub>24</sub> O	Dodecanal	0.14	-	0.31	0.13	0.58	0.37	-
68	C <sub>11</sub> H <sub>22</sub> O	Methyl Octyl Acetaldehyde	-	17.57	1.76	-	-	0.08	-
69	C <sub>14</sub> H <sub>28</sub> O	Tetradecanal	-	7.88	-	-	-	0.15	-
70	C <sub>9</sub> H <sub>14</sub> O	Trivertal	0.09	-	-	-	0.19	0.15	-
71	C <sub>11</sub> H <sub>20</sub> O	Undec-(8Z)-enal	0.08	-	-	0.72	0.31	0.35	-
<b>Alcohols</b>									
72	C <sub>10</sub> H <sub>22</sub> O	1-Decanol	2.42	4.08	1.91	1.04	2.76	1.65	-
73	C <sub>12</sub> H <sub>22</sub> O	1-Dodecyn-4-ol	-	-	-	-	0.27	-	-
74	C <sub>8</sub> H <sub>18</sub> O	1-Octanol	-	1.18	-	-	-	-	-
			-	0.23	0.20	-	-	0.03	-

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Most of the compounds found in *E. coccinea* are monoterpenes at 18 compounds, followed by alcohols (fourteen compounds), alkanes and alkenes (twelve compounds), sesquiterpenes (eight compounds), aldehydes (seven compounds), ketones, fatty acid derivatives, esters (four compounds), and lastly, amine and norterpene, one compound each.

Despite the number of compounds present in each class, the relative abundance for each compound differed. Referring to Figure 3, the major compound class are monoterpenes, ketones, aldehydes and alkanes. Alkanes are the most abundant in all parts of *E. coccinea*, ranging from 42.86% to 51.83%. In the previous study by Cui et al. (2008), the alkanes in defoliated leaves were higher compared to fresh leaves. The plant sample collection was completed one week earlier before it could be stored and extracted to get essential oil, a lot of samples were already defoliated, hence, supporting the abundance of alkanes in the result obtained.



**Figure 3.** The relative abundance of several classes of the compound found in *Etlingera coccinea* essential oil

As shown in Table 1, the ranks from highest to the lowest abundance of monoterpenes found in different parts of the plant are in the order of rhizomes > leaves > stems. According to Gershenzon et al. (2000), in most plants monoterpenes are easily released to the atmosphere. The biosynthesis and accumulation of monoterpenes are also easily influenced by the external environment and improper cultivation (Zhang et al., 2017). Therefore, further assessment needs to be conducted before stating that the abundance of monoterpenes are only concentrated in some parts of a plant. Ketones, on the other hand, mostly dominate the leaves part of *E. coccinea* and sesquiterpenes were only found in rhizomes of *E. coccinea* that had been collected from Trail 1. Plants have always been known for their ability to produce a natural product that has an essential role in their defence mechanism against herbivores and any pathogens, these natural products or metabolites are synthesized in specific organs, specific stages of development, environmental effect or induced by getting attacked by other organisms (Gershenzon et al., 2000). Therefore, the presence of a chemical compound in each plant part is not necessarily the same or in the same concentration. For example, in a study of sesquiterpenes, researchers found that high emission of sesquiterpenes was detected in maize leaves that were damaged by the herbivores compared to undisturbed maize leaves (Köllner et al., 2013). Another study by Liu et al. (2020), has reported that any intact and uninjured tea plant does not emit any sesquiterpene but those that have been damaged mechanically produce one sesquiterpene ((E, E)- $\alpha$ -farnesene) and insect-damaged ones emit two sesquiterpenes ((E, E)- $\alpha$ -farnesene and (E)-nerolidol). Though our data does not reflect similar observation, this could be one of the reasons why sesquiterpenes were only found in Trail 1. Apart from this, the environmental condition where samples were collected were not really the same, Trail 2 was nearer to urban areas and Trail 1 was in the forest.

### *Terpenoid content*

Terpenoids are terpenes that have different functional groups and are an oxidized methyl group attached or detached from the compound at numerous positions. It is divided into several classes, which are monoterpenes ( $C_{10}H_{16}$ ), sesquiterpenes ( $C_{15}H_{24}$ ), diterpenes ( $C_{20}H_{32}$ ), sesterpenes ( $C_{25}H_{40}$ ), and triterpenes ( $C_{30}H_{48}$ ) varying on its carbon units. The most reliant terpenoids that varied in their formations are biologically active and extensively used all over the world as a source of cure for ailments (Perveen, 2018). From the result obtained in Table 1, the terpenoids found in *E. coccinea* are divided into two, monoterpenes and sesquiterpenes. The terpenoid content in *E. coccinea* was

highly abundant in the rhizomes, then followed by leaves and stems parts for both Trail 1 and Trail 2.

There are 26 terpenoids found in *E. coccinea* (Table 1). However, the terpenoids abundance in all plant parts for both trails are less than 50%. The rhizome part collected from Trail 2 recorded the highest terpenoid abundance (43.34%). Terpenoids were known to exhibit various beneficial properties. Based on previous studies, B-Pinene that can be found in the leaves of *E. coccinea* was found to be a good anti-inflammatory (Rivas da Silva et al., 2012) and anti-depressant agent (Guzmán-Gutiérrez et al., 2012). While Pinocampeol compound that can be found both in leaves and stems of *E. coccinea* was reported to have promising insecticidal activity (Kalechits & Kozlov, 2008). Other terpenoid that can be found in the leaves of *E. coccinea* are known as D-limonene, a major compound in citrus peel. It has a major role in breast cancer prevention and treatment according to Miller et al. (2011). Moreover, geranyl formate, a monoterpene compound that can be found mostly in the floral-rosy scent of plants, such as citronella, geranium, rose and verbenas. As stated by Jirovetz et al., (2006), geranyl formate showed antimicrobial activity when tested against yeast and gram (+) and gram (-) bacteria.

## Conclusion

The result of this study has successfully shown that the composition of chemical compounds from different parts of tissues is very diverse. Potential biological activities such as anti-oxidants and anti-microbial of *E. coccinea* essential oil as reported previously, has increased the value of *E. coccinea* itself on the market. Further studies, such as the environmental factors of *E. coccinea*, need to be continued to determine a unique potential for this species.

## Acknowledgements

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## Short Notes

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# A preliminary survey of Araceae of Kadamaian-Kinabalu Park, Kota Belud, Sabah, Malaysia.

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## Abstract

A preliminary survey on Araceae in Kadamaian - Kinabalu Park, Kota Belud, Sabah was conducted between 14<sup>th</sup> to 20<sup>th</sup> October, 2019. Four trails were surveyed namely Malangkap-Nariou Trail (425-670 m asl), Gansurai-Nopunggok Trail (~600m asl), Wasai Waterfall Trail (~425m asl) and Kampung Pinolobu Trail (~220m asl). A total of 25 species in nine genera of Araceae were recorded with at least two undescribed species. This finding shows that Kadamaian - Kinabalu Park indeed possesses unique forest ecology and should be gazetted as a protected area.

**Keywords:** Aroids, Araceae, Borneo, Kinabalu Park, Kota Belud

## Introduction

Araceae family comprises about 144 genera with 6,000 estimated species of which currently 4,000 were formally described (Mayo et al., 1997, Boyce and Croat, 2011). This family can be found mainly at tropical areas but is also distributed worldwide (Cusimano et al., 2011, Croat 1979, 1994). The family is defined by having minute sessile flowers on spadix and covered by a spathe. The spadix may bear either unisexual or bisexual flowers. Most of the climbers have bisexual type of flowers while others have unisexual flowers. Ecologically, aroids can be found in streams, ponds and canals, terrestrial habitats, tidal mud, swamps and wasteland, forest floor, climbers, epiphytes and rheophytes (Mashhor et al., 2012).

The Araceae of Borneo currently stand at 575 species, of which 433 are formally described (Wong, 2016). Most Bornean terrestrial Araceae show marked local endemism, often to a very high degree, and are frequently associated with geological obligation. Fieldtrips in Sabah have resulted in numerous undeterminable species of aroids which on subsequent flowering in cultivation have proved to be taxonomic novelties (Kartini et al., 2017).

**Methodology**  
*Sampling and Study Area*

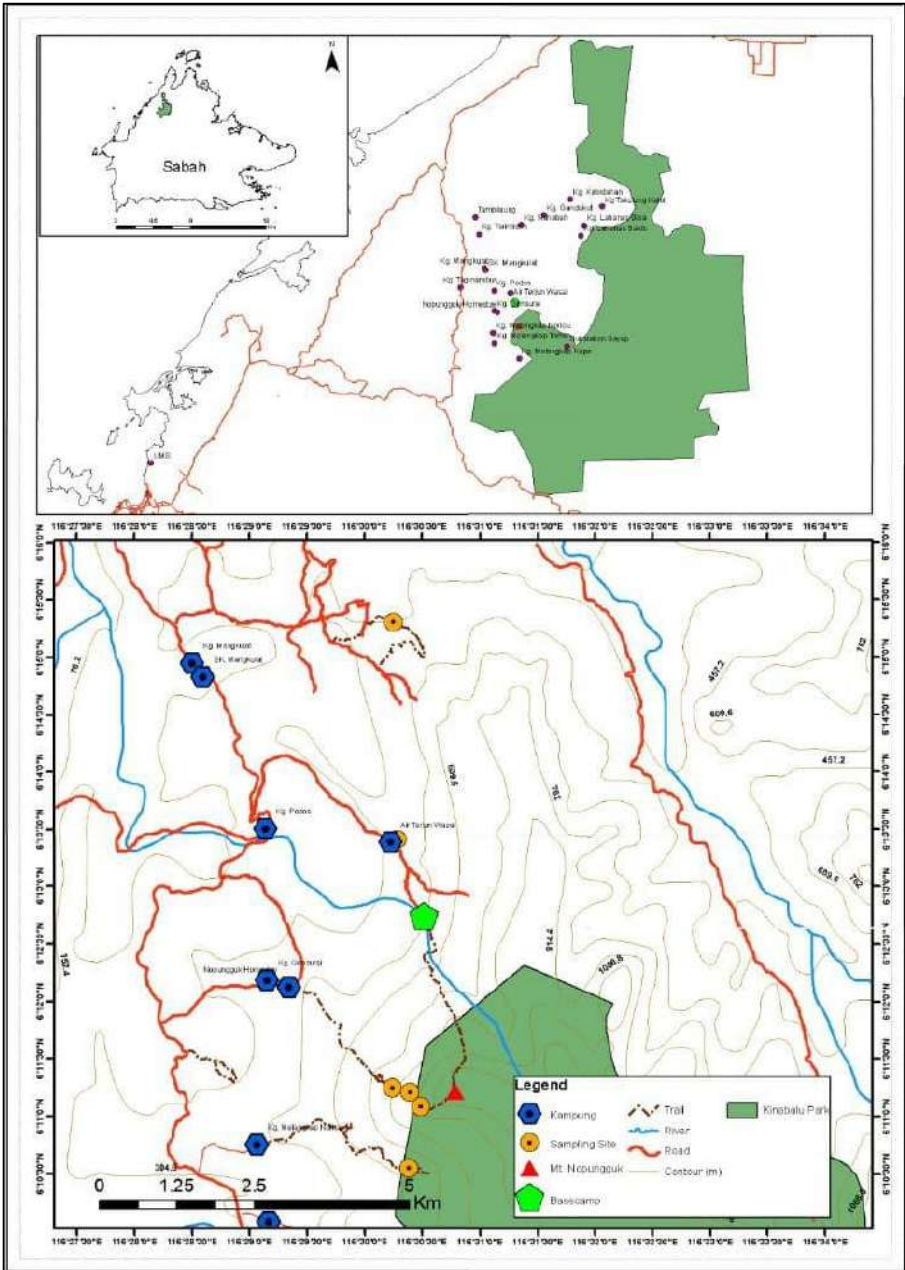


Figure 1. The location of the study area.

A preliminary survey of Araceae of Kadamaian - Kinabalu Park was conducted between 14<sup>th</sup> to 20<sup>th</sup> October, 2019 along four existing trails (Figure 1) namely Malangkap-Nariou Trail (425-670m asl; N 06°10.552' E116°30.380'), Gansurai-Nopunggok Trail (~600m asl; N 06°12.544' E116°30.275'), Wasai Waterfall Trail (~425m asl; N 06°13.414' E116°30.284') and Kampung Pinolobu Trail (~220m asl; N 06°15.303' E116°30.249'). A total of 50 plant individuals of Araceae family (with or without flowers) have been collected. The plant samples were brought back to the Institute for Tropical Biology and Conservation (ITBC), Universiti Malaysia Sabah (UMS) for specimen preparation and identification. The specimens are in the process of depositing in BORNEENSIS (BORH).

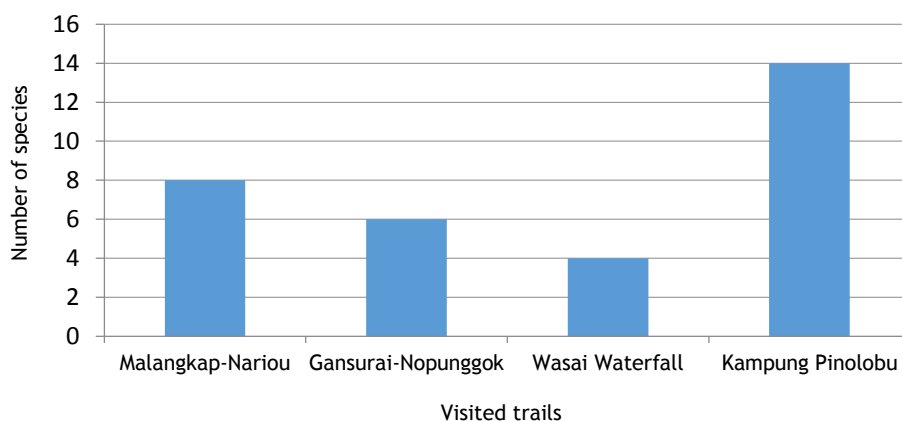
### *Morphological identification*

Living plant samples were collected and grown at the nursery of ITBC, UMS awaiting flowering for further identification. Identifications are based on Boyce (2004), Boyce et al. (2001), Hay & Yuzammi (2000), Low et al. (2018) and Wong (2016).

## Results

Three subfamilies and nine genera comprising 25 species were found in the sites surveyed (Table 1 and Figure 2). Pothoideae which is mostly a climber is represented by two species -- *Pothos scandens* and *Pothos* sp.

Most of the Monsteroideae and Aroideae species were found along riverbanks while *Nabalu corneri* was found abundant at rocky areas. Some of the *Homalomena* sp. was found abundant on mossy stones.



**Figure 2.** Total number of species in different trails.

Table 1. Araceae species in Kadamaian - Kinabalu Park.

Subfamily	Genus and Species	Malangkap-Nario Trail (425-670m asl) N 06°10.552' E116°30.380'	Gansurai-Nupungok Trail (~600m asl) N 06°12.544' E116°30.275'	Wasai Waterfall Trail (425m asl) N 06°13.414' E116°30.284'	Kampung Pinolobu Trail (220m asl) N 06°15.303' E116°30.249'
Pothoideae	<i>Pothos scandens</i> L.	-	-	-	+
	<i>Photos</i> sp.	-	+	-	+
Monsteroideae	<i>Amydrium medium</i> (Zoll. & Moritz) Nicolson	+	+	-	-
	<i>Rhaphidophora korthalsii</i> Schott	+	+	-	-
	<i>Rhaphidophora latevaginata</i> M. Hotta	-	-	-	+
	<i>Scindapsus curranii</i> Engl. & K. Krause	-	-	-	+
	<i>Scindapsus</i> sp. 1	+	-	-	+
	<i>Scindapsus</i> sp. 2	-	-	-	+
		-	-	+	-
Aroideae	<i>Aglaonema nitidum</i> (Jack) Kunth	-	-	-	+
	<i>Alocasia</i> cf. <i>longiloba</i>	+	-	-	-
	<i>Alocasia</i> cf. <i>scabriuscula</i>	-	-	-	-
	<i>Homalomena marasmiella</i> Kartini, P. C. Boyce & S.Y. Wong	+	-	-	+
	<i>Homalomena</i> sp.1 - Chamaecladon Clade	-	+	-	+
	<i>Homalomena</i> sp.2	-	+	-	-
	<i>Homalomena</i> sp.3	-	-	+	-
	<i>Homalomena</i> sp.4	+	-	-	+
	<i>Nabalu corneri</i> (A. Hay) S.Y. Wong & P. C. Boyce	+	-	+	-
	<i>Schismatoglottis ahmadii</i> A. Hay	-	-	-	+
	<i>Schismatoglottis</i> sp.1	+	-	-	-
	<i>Schismatoglottis</i> sp.2 - Patentinervia Clade	+	-	-	-
	<i>Schismatoglottis</i> sp.3 - Trifasciata Complex	-	+	-	-
	<i>Schismatoglottis</i> sp.4 - Antu Clade	-	+	-	-
	<i>Schismatoglottis</i> sp.5	-	-	-	+
	<i>Schismatoglottis</i> sp.6 - Asperata Clade	-	-	-	+
	<i>Schismatoglottis unifolia</i> A. Hay & P. C Boyce	-	-	+	+
		-	-	-	-

Throughout the sampling period, Kampung Pinolobu Trail recorded the most number of species with only four species formally described, followed by Malangkap-Nariou Trail with five out of eight species recorded having been formally described. Wasai Waterfall Trail and Gansurai-Nopunggok Trail only recorded four and six species of Araceae respectively.

## Discussion

The preliminary survey of Kadamaian revealed Araceae comprised largely of rather widespread species associated with disturbed habitats. The climbers *Pothos scandens*, *Rhaphidophora korthalsii*, *R. latevaginata* are the most widespread species of the genus in Sabah. *Scindapsus* sp. 1 is a widespread species that occurs throughout northern Borneo but has yet to be formally described.

The taxonomically intractable species complexes, *Alocasia* cf. *longiloba* is common and widespread but has yet to be described (Hay, 1998).

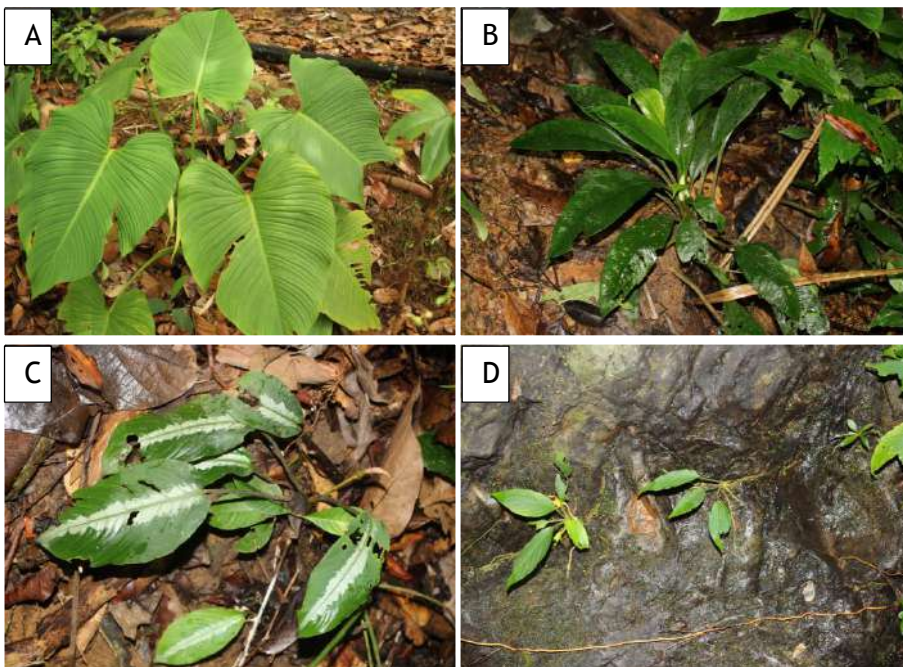


Figure 3. (A) *Nabalua corneri* (A. Hay) S.Y. Wong & P.C. Boyce; (B) *Schismatoglottis* sp. 2 (Patentinervia Clade); (C) *Schismatoglottis* sp. 3 (Trifasciata Complex); (D) *Schismatoglottis* sp. 4 (Antu Clade).

*Homalomena* is usually an aromatic herb distinguished by its persisting spathe and is mostly an undescribed species in Sabah (Wong, 2016). The *Homalomena* Chamaecladon clade (*Homalomena* sp.1) is the least well known four clades in the genus (Wong et al. 2020a & 2020b).

*Nabalu corneri* (A. Hay) S.Y. Wong & P.C. Boyce (Figure 3A) is a massive arborescent species frequenting forest gap-phases where it often becomes almost weedy (Low et al. 2018).

*Schismatoglottis* sp.2 (Figure 3B) in Patentinervia Clade resembles *Schismatoglottis retinervia* Furtado (Wong et al. 2017).

*Schismatoglottis* sp. 3 (Figure 3C) in the Trifasciata Complex and *Schismatoglottis* sp. 4 (Figure 3D) in the Antu Clade are unquestionably undescribed (Kartini et al. 2017; Wong & Boyce 2015).

## Conclusion

Kadamaian - Kinabalu Park proved to be an interesting site for Araceae and should be gazette as a protected area due to the presence of two species of Sabah's endemics namely *Homalomena marasmiella* and *Schismatoglottis unifolia*.

## Acknowledgements

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## Research Article

# Community preparedness in ecotourism development and their role in maintaining the natural resources in Kadamaian area, Sabah

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## Abstract

Community preparedness in ecotourism development and their role in maintaining natural resources is necessary. Yet, many examples throughout the world suggest that local communities are unable to participate in development of ecotourism and maintaining natural resources. A preliminary study was carried out to identify the preparedness of local communities in ecotourism, as well as to determine the Strength, Weakness, Opportunities, and Threat (SWOT) that effect the preparedness of local communities. Sociological interviews was a recommend networking as the determining the factor. This paper aims to assess the potential of ecotourism in the Kadamaian area and the preparedness in development of ecotourism by the local community. We focused on evaluating the relationship between environment impact and ecotourism by the local community in terms of sustainable tourism development, which can provide an insight in future management for government, stakeholders and managers. This preliminary study shows that local communities are highly prepared in ecotourism development and inmaintaining natural resources, and the result could assist them in conserving natural resources.

**Keywords:** Ecotourism, SWOT, Conservation, Local Network, Tourism Development.

## Introduction

Ecotourism is a type of niche market in the tourism industry. Researchers including from the United National Educational Scientific and Cultural Organization (UNESCO)(2008) state that niche markets in the tourism industry can be classified as Dark tourism, Cultural & Heritage tourism, Adventure

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tourism, Agricultural tourism, 3S tourism, Food tourism, Medical tourism, Cycling tourism, Sports tourism, Health tourism, Green tourism, Pink tourism, Ecotourism and others. Ecotourism is a niche market of the tourism industry, as stated above. According to Buckley (2009), the word “ecotourism” has been debated about in terms of its definition, for at least two decades. Ecotourism is defined as a promise that each tourist has responsibility during travel to natural areas; which means tourists will make a positive contribution towards the conservation of the natural environment and enhance the well-being and life style of local communities (Angelica, Eben & William, 2009). Cheia (2013) states ecotourism was and is a human activity with an extensive search and development model of a country, especially in the 21st century. Furthermore, Ikonen (2012) also indicated that ecotourism can be defined as a form of tourism that sees tourists’ main motivation being observation and appreciation of nature and trying their best to conserve and minimize the negative impacts on nature and the socio-cultural environment in ecotourism destination. The data collected from the survey was analysed using the Statistical Package for Science and Social Sciences (SPSS). In addition, SPSS is used to investigate the community preparedness and to test the reliability and the correlation of the study.

Strengths, Weaknesses, Opportunities and Threats (SWOT) analysis has been widely used in business and management assessment, but it can also be usefully applied in other fields (Rizzo, 2005). SWOT can directly describe and measure factors in a study, interpret the strength and weakness of the project as well as its opportunities and threats. In this research, the development of ecotourism in Kadamaian area was considered after doing a survey on the perception of the local community and a SWOT analysis to measure the potential attraction of the area. This paper aims to assess the potential of ecotourism in Kadamaian area and preparedness for the local community to develop ecotourism. We focus on evaluating the relationship between environment impact and ecotourism by the local community in terms of sustainable tourism development, which can bring insights for future management for the government, stakeholders and managers. There was no information on community ecotourism development from Kadamaian at the time the current survey was done. As a result, this research is critical in terms of providing significant baseline data for conservation reasons via inventory, as well as information on community preparedness and their involvement in maintaining natural resources.

*Literature review*

Ecotourism can be illustrated as nature tourism that intentionally seeks to provide a net of positive contributions towards environmental conservation and to sustain this for local communities development (Weaver & Lawton, 2007). Moreover, ecotourism has been a preferable option in utilizing resources within a protected area and it is considered as a bridge between nature conservation and rural economic development.

Neopolist theory suggests that the local community should be central to tourism development and management, and encourage them to achieve or to formalize the system at local level planning. Local involvement is a critical element of preparedness of ecotourism development and the role of communities to maintain natural resources. The preparedness of local communities can be seen by their participation. Some scholars have created a typology of participation. However, not all of it deals directly with tourism development (Leksakundilok, 2006).

SWOT analysis is a useful method for the comprehensive assessment of sustainable tourism development (Reihanian, 2012). The analysis identified two main factors for evaluation development: Internal factors determine the advantages and disadvantages of the region; external environmental factors include opportunities and threats (Miandehi, 2013). The SWOT framework has been used as a good research tool to not only evaluate business, but to also conduct a comprehensive assessment for ecotourism development, and it is an effective way to identify the future directions of tourism development (Collins-Kreiner & Wall, 2007; Miandehi & Masrouri, 2013). SWOT can be applied for management processes that highlight important factors that interact with each other (Pickton & Wright, 1998), to overcome the weaknesses and threats, also to enhance strengths and opportunities in development. It plays a significant role in evaluating the potential of ecotourism in the local community, which provides a simple and clear strategy for managers and governments.

Table 1 shows community preparedness in ecotourism when they are actively involved in the development at community level. By examining community attitude and preparedness for ecotourism, this research encourages the local community to maintain available resources and to understand current issues that impact nature and enhance or limit participation in controlling development.

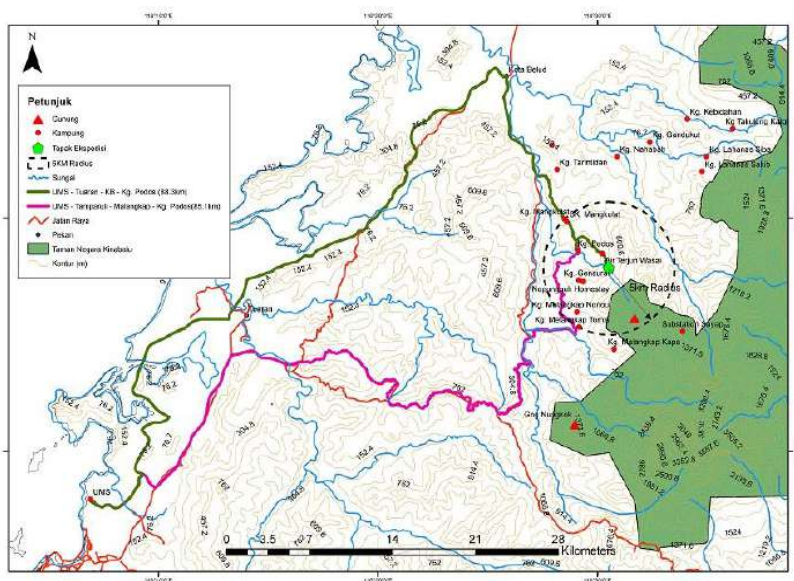
**Table 1.** Typologies of Participation

Levels	Types	Characteristic
Genuine participant (active)	Empowerment	Local people may directly contact explorer tourist and develop tourism by themselves: Local people have a control over all development without any external forces or influence.
Symbolic participant (towards active)	Partnership	There are some degrees of local influence in the tourism development process.
	Interaction	People have greater involvement at this level. The rights of the local people are recognized and accepted in practice at the local level (Pretty, 1995a).
	Consultation	People are consulted in several ways through meetings, seminars etc.. Developers may accept some contribution from locals (Arnstein, 1969).
Non-participant (passive)	Informing	People are told about tourism development programmes that have been already decided by the community. The developers, run the projects without getting any feedback from local communities.
	Manipulation	Tourism development is generally developed by powerful individuals, government, or outsiders without any discussion with the local communities (Arnstein, 1969).

*Note.* Source: Adapted from Leksakundilok (2006)

## Methodology

### Research Site

**Figure 1.** Map of Kadamaian Area

This research was conducted in the Kadamaian area of Kota Belud, from 20<sup>th</sup> 25<sup>th</sup> October, 2019 (6 days). Kadamaian is located to the north west of Kinabalu Park in the west coast of Sabah. The basecamp site is proposed to be set up in Kampung Podos at Podos Heritage Homestay compound (N06°12'44.2" E116°30'31.2") about 30 minutes from Kota Belud town, or about 1 hour 30 minutes from Kota Kinabalu (88.3 km).

A trail leading to the summit of Mount Nopunggok (1,430 m asl) will pass through different forest types, from lowland mixed dipterocarp forest, upland mixed dipterocarp forest to lower montane forest. Pristine forest can be seen inside the Kinabalu Park area.

#### *Research Techniques and Procedure*

The process of the investigation of the community's preparedness can be shades understanding in factors that is limiting the contribution of local communities in ecotourism and the support of conservation.

In conducting the preparedness of local communities and their role in maintaining natural resources, two types of data were required. Primary data was obtained through a survey from the site in the form of a sociological survey as a guideline since this study does not have control over event. As for secondary data, it was obtained through various journals and reports on the preparedness of local communities. In addition, any changes in development and their role in maintaining natural resources will be investigated through available reports. Because of the time constraints in completing this study, there are fewer samples and observations. In order to obtain fresh information in future, this study will serve as a guide for future researchers, ensuring that they have accurate information about the study's topic. It may not be much, but it should be useful.

#### *Questionnaire Design*

The research was conducted using quantitative data. Quantitative data consisted of a survey to describe and to measure the relationship of the variables (PenState University, 2017). Questionnaires were distributed without the respondent's name to keep the data private and to encourage critical opinion. Respondents for the study came from four villages in Kadamaian, (1) Kg. Melangkap Noriou, (2) Kg. Mengkulat, (3) Kg. Gensurai, and (4) Kg. Podos. Questionnaires used a Likert Scale of six as it has higher reliability when using Cronbach's Alpha Coefficient tool and will provide higher value (Chomeya, 2010). The questionnaire was were bilingual, in English and Bahasa Malaysia. In

general, the questionnaire consisted of three (3) sections, i.e (1) demographic which is the respondent's profile, (2) independent variable that includes the demographic variable, government policy, and benefit of ecotourism, and (3) perception of the local community in ecotourism and conservation.

#### *Cronbach's Alpha*

The reliability involved when developing variables from the accumulative scale is used as the predictive component of the target model. Santos (1999) mentioned that since accumulative scales are a collection of items designed to measure the interconnection of infrastructure, it is significant to know whether the same set of items elicit the same response if the same questions are redesigned and readministered to the same respondents. Variables derived from test instrumentation are confessed as reliable only if they provide a test, which is stable and a reliable response to repeated administrations.

Cronbach's alpha is an indicator in reliability test, which is associated with the variation accounted for by the real score of the "underlying construct." Construct is the hypothetical variable that will be measured (Santos, 1999).

Santos (1999) stated that the reliability of factors draw from questions with two possible answers or multi-point formatted questionnaires or scales (i.e., rating scale: 1 = poor, 5 = excellent) are described by Alpha coefficient ranges in value from 0 to 1. The higher the score, the more trustworthy the generated scale is. It is also indicated for 0.7 to be a fit and good reliability coefficient; however lower thresholds are sometimes used in the literature.

#### *SWOT Analysis*

SWOT analysis was conducted in order to investigate the potential of ecotourism development and management in the future in Kadamaian. This analysis tested the internal factors including strengths and weaknesses as well as external factors which are opportunities and threats in this region in terms of community ecotourism and strategies. Values of factors were identified by the environment, local community and economy in regards to successful factors of ecotourism (Parker & Khare, 2005).



## Results and Discussion

### *Cronbach's Alpha*

**Table 2.** Results of reliability Cronbach's Alpha for the variables

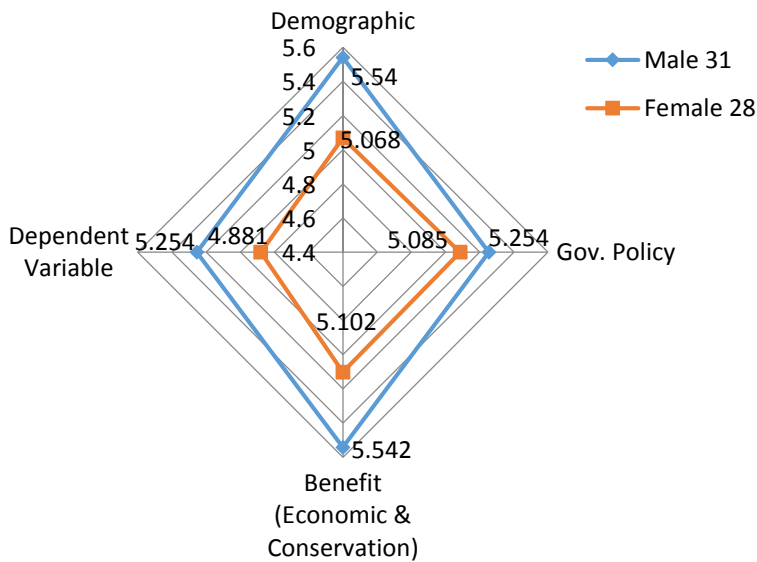
Variables	No. of Items	Cronbach's Alpha
Perception of Local Community on Ecotourism and Conservation	9	.865
Demographic Variable	5	.853
Government Policy	5	.764
Benefits of Ecotourism (Economic & Environmental Conservation)	7	.831

The Cronbach's Alpha values of the questionnaire were above the acceptable level, at a value range of 0.764 - 0.865. The second independent variable, government policy, was found to be the lowest in terms of reliability among other variable with 5 items:  $\alpha = 0.764$ . The third independent variable, that is the benefit of ecotourism (economic and environmental conservation), showed an average reliability with 7 items:  $\alpha = 0.831$ . The first independent variable, the demographic variable, was found to be the highest and with good reliability among independent variables with 5 items:  $\alpha = 0.853$ . Meanwhile, the dependent variable, perception of the local community on ecotourism and conservation, which consisted of 9 items, showed excellent reliability among all variables, with a reliability value of 0.865.

### *Demographic*

The relationship of gender on the preparedness of the local community for ecotourism and conservation is very important in order for them to understand and to be more aware of the environment instead of self-development. Figure 2 shows the awareness of both genders on the four items of the perception of ecotourism and conservation. The spider chart shows that males have higher information and understanding about ecotourism and conservation with a mean of 5.54 on the demographic variable, 5.245 on government policy, 5.542 on benefit of ecotourism and conservation (economic and environmental conservation) and the independent variable 5.254 which is the perception of local community on ecotourism and conservation. However, females have an average understanding and information with the mean for each dependent variable for demographic variable at 5.068; 5.085 for government policy, and 5.102 for benefit of ecotourism (economic and environmental conservation) and

the dependent variable with a mean of 5.254 for the perception of the local community on ecotourism and conservation.



**Figure 2.** The relationship of gender on the perception of local community on ecotourism and conservation.

Establishing relationships between the same variables in two populations or between two or more variables in the same population is known as correlational research (Curtis, Comiskey & Dempsey, 2016). Correlation can refer to a single digit that explains the degree of relationship between two variables. In order to identify the relationship between two variables, coefficient is used and described by a unitary value.

There is a statistical technique to identify the connection between two quantitative and continuous variables. It shows how strongly pairs of variables are related. The result of a correlation is called correlation coefficient, with ranges from -1.0 to +1.0.

Table 3 shows that demographic variables and perception of local community on ecotourism and conservation were statistically significantly correlated,  $r = 0.635$ ,  $p < 0.01$ . The correlation coefficient indicated a moderate association between the demographic variables and local community perception.

There was a statistically significant correlation between government policy and perception of local community on ecotourism and conservation with  $r = 0.501$ ,  $p < 0.01$ . The correlation coefficient indicated a moderate association between government policy and the perception of local tourists.

**Table 3.** Result of Pearson Correlation Analysis

	Perception of Local Community on Ecotourism and Conservation	Demographic Variables	Government Policy	Benefits of Ecotourism and Conservation
Perception of Local Community on Ecotourism and Conservation	1	0.635**	0.501**	0.758**
Demographic Variables	0.635**	1	0.619**	0.651**
Government Policy	0.501**	0.619**	1	0.626**
Benefits of Ecotourism and Conservation	0.758**	0.651**	0.626**	1

\*\*Correlation is significant at the 0.01 level (2-tailed).

The benefits of ecotourism and the perception of local tourists towards ecotourism destinations in Malaysia were statistically significantly correlated,  $r = 0.758$ ,  $p < 0.01$ . The correlation coefficient indicated a moderate association between benefits of ecotourism and local tourist's perception.

In a nutshell, demographic variables, government policy and benefits of ecotourism were significantly correlated with the perception of local community on ecotourism and conservation with moderate relationships.

#### *Internal Factor Estimate Matrix (IFEM)*

There were 15 factors to strength this study; the effectiveness score ranged between 2 and 4. Geographical features, biodiversity of forests, traditional and cultural experience activities, community motivation and the organization of ecotourism in the community had the highest weight. In regards to weakness,

the highest weight was no disposal plans and the lowest was lack of travel and ecotourism agencies. The final score was 3.44, which is larger than 2.5; SWOT analysis shows that if the value is less than 2.5, it means that there are fewer opportunities than threats. If the value is greater than 2.5, the opportunity is greater than the threat, which indicates that the value has a high potential for the internal development of the tourism industry and can be explained in the external development. (Ganjali, 2014; Ghorbani, 2015).

Ecotourism sustainable development strategies using SWOT and QSPM model: A case study of Kaji Namakzar Wetland, South Khorasan Province, Iran.

**Table 4.** Internal Factor Estimate Matrix

No.	Strength	Weight	Effectiveness score	Final Score
1.	Well maintained road to access, not far from city; i.e. 1 hour to Kota Belud & 2 hours to KK city centre. Tourists do not need to travel far to experience ecotourism.	0.05	3	0.15
2.	Near Mount Kinabalu, the forest ecosystem is very well protected, cool temperature weather. Abundant topography, river, waterfall, forest, mountain.	0.06	4	0.24
3.	Various wild flora and fauna (large ferns, tuhau, butterflies etc.) including protected species (Pangolin & Rafflesia spp).	0.06	4	0.24
4.	Special landscape featuring, many different types of waterfall and river, different scenery in each trail.	0.03	2	0.06
5.	Big camping sites are ready for tourist usage; on flat high land close to the river. Well built restrooms available with proper sewage system.	0.03	2	0.06
6.	Clean, piped water available at tourist centre as well as houses within the villiage. Power supply is sufficient for tourist usage in the camping area.	0.04	4	0.16
7.	Attraction signs have settled along the trail based on its different natural geography feature. Trails with proper resting place for tourists.	0.03	2	0.06
8.	Developed homestay, advertisement on social media, with experienced guide, detailed map and trail route.	0.04	3	0.12
9.	There are four (4) main hiking trails determined by locals themselves. Local guides will explain the flora and fauna to tourists along the trail.	0.05	3	0.15
10.	Traditional and cultural experience activities that enables tourists to learn from local community; including making handcraft, traditional food and cultural night.	0.06	3	0.18

11.	Other tourism packages provided to enrich activity, such as visiting bee farm, buffalo riding, rafting and karaoke and BBQ.	0.03	2	0.06
12.	Local communities are actively involved in the tourism activities & are trained regarding environmental education. They are also known as Citizen Scientist.	0.04	3	0.12
13.	High level of community awareness & compliance regarding biodiversity conservation.	0.05	4	0.2
14.	Community has high motivation to improve their economic situation through ecotourism; as well as the drive to implement their plans for the benefit of the environment and the peoples' socio-economic condition.	0.06	4	0.24
15.	Well organized community towards building ecotourism in their locality, willingness to learn and share experiences with community's vicinity kampungs.	0.06	4	0.24
No.	Weaknesses	Weight	Effectiveness score	Final Score
1.	Trails are not user friendly to beginners and poor strength people, as the routes are difficult for trekking.	0.05	4	0.20
2.	Unpredictable weather easily interrupts activity.	0.05	4	0.20
3.	Shortage of shelter for resting and rain along the trail.	0.04	4	0.16
4.	Facilities are not well maintained along the trail, i.e. stairs are too old and broken.	0.05	4	0.20
5.	Lack of travel and ecotourism agencies.	0.02	2	0.04
6.	No disposal plans, may cause waste pollution if increase tourism activities.	0.06	4	0.24
7.	Limited knowledge of fauna and flora in terms of sustainable ecotourism.	0.04	3	0.12
<b>Total</b>		<b>1</b>		<b>3.44</b>

#### *External Factor Estimate Matrix (EFEM)*

Seven factors were identified to opportunities and 6 factors to threats. Research and educational potential and increase conservation awareness of the ecosystem to local community had the highest weight. Promotion of sustainable tourism to the public had the lowest weight. Threat to local ecosystem conservation and habitat disturbance and risk of local resources destruction and land use issues regards of tourism development had the highest weight in terms of threats. The lowest weights of threat were inconvenience for foreign tourists such as language barriers and the local community losing its traditional way of life. The value of external factor was 3.27 interpreting that opportunities were more than threats. Hence, Kadamaian area seems to have significant potential for ecotourism development, however, there are still many facilities and conservation management that need to be improved.

**Table 5.** External Factor Estimate Matrix

No.	Opportunities	Weight	Effectiveness Score	Final Score
1.	More potential attraction can be explored in various landscape.	0.07	3	0.21
2.	Research and educational potential according to sustainable ecotourism.	0.10	4	0.40
3.	Increase conservation awareness of ecosystem to local community through tourism.	0.10	4	0.40
4.	Job opportunity and economic development for local community.	0.09	4	0.36
5.	Example of ecotourism management and market for neighbor community around Mt. Kinabalu Park.	0.06	3	0.18
6.	Promotion of sustainable tourism to public.	0.05	2	0.10
7.	Enhance culture and traditional context conservation.	0.07	2	0.14
No.	Threats	Weight	Effectiveness Score	Final Score
1.	Threat to local ecosystem conservation and habitat disturbance.	0.10	4	0.40
2.	Risk of local resources destruction and land use issues regards of tourism development.	0.10	4	0.40
3.	Lack of environmental impact assessment for ecotourism facilities.	0.08	3	0.24
4.	Shortage of pollution treatment for waste disposal, i.e. hotel, food, vehicle.	0.08	3	0.24
5.	Inconvenience for foreign tourists such as language barrier.	0.05	2	0.10
6.	Local community lose their traditional way of life.	0.05	2	0.10
<b>Total</b>		<b>0.93</b>		<b>3.27</b>

Internal and external factors in the matrix of SO, ST, WO and WT strategies were done for the Kadamaian community. Table 6 below shows sustainable tourism development strategies in Kadamaian.

**Table 6.** Strategies of internal and external factors

<b>SO strategies</b>
Consider the area climate and weather which affects the potential activities when planning for ecotourism tours.
More conservation research and education should be conducted in this area increasing local community knowledge and awareness.
Consider particular ecosystem environment of Kadamaian, special geographical features and biodiversity should be developed for this.
Consider developing ecotourism management in future in order to enhance the tourism industry for the local community.
Education of teaching and training tourism skills provide more opportunities for the local community.

**ST strategies**

Government making management plan and strategies to avoid negative impact of tourism on the local ecosystem and biodiversity.  
 Improving infrastructure to have better service and facilities to attract tourists.  
 Increasing conservation activities to improve conservation capacity and awareness.  
 Conducting Environmental Impact Assessment to prevent landscape exploitation and habitat disturbance.  
 Develop more traditional activities to keep local culture and marketing.

**WO strategies**

Design proper trails for safety and for tourists who are not strong enough.  
 Establish shelter and maintaining facilities along the trails for resting and visiting.  
 Environmental education and culture building through public media, academic conferences, and NGOs.  
 Cooperate with tour agencies to promote ecotourism package tours for the local community.  
 Communicate and introduce ecosystem attractions by using well-designed education packages.  
 Consider the plan to deal with waste disposal for mass tourism.

**WT strategies**

Plan alternative tourism packages in case of unexpected weather.  
 Getting funds from stakeholders to continue maintaining facilities.  
 More conservation knowledge education taught by professional experts to local guides.  
 Government makes regulations and laws preventing negative impacts by tourism.  
 Environmental impact management formulated for increasing number of tourists in the future.  
 Waste treatment system planned for accommodation and other activities.

## Conclusion

The community preparedness in ecotourism and their role in maintaining the natural resources are still under study, and more studies are needed. After considering the result from the study, it has contributed preparedness of the local community on ecotourism. The main purpose of this study is to ensure natural resources are well managed and aware about environmental issues. Subsequently, Kadamain has significant potential for ecotourism development. The natural geographic features and biodiversity lay the foundation for the tourism industry. However, there is still lack of infrastructure and promotion of this area. The local community has potential to do ecotourism, the perception for conservation and their awareness show they are prepared to do this. In addition, the government and organizations need to support policy and other aspects for better management and development. It is of utmost importance to conserve and maintain the ecotourism industry in Sabah, not only for sightseeing, but to also maintain the existence of the natural resources which is has value not only for tourists but also for the local community.

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## Research Article

# A Species Checklist of Wild Orchids in Selected Sites in Kadamaian, Kota Belud, Sabah

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## Abstract

A brief orchid diversity study was conducted from 14<sup>th</sup> to 19<sup>th</sup> October 2019 in Kadamaian, located northwest of Kinabalu area, through a scientific geographic expedition. A convenience sampling method was employed with only the flowering individuals collected and preserved as herbarium specimens. A total of 58 species and 32 genera were identified during the field visits to the Mount Nopungguk, Melangkap Noriou, Ulu Sungai Melawa, and basecamp site. The finding comprises of 43 epiphytes, 13 terrestrials, and two mycoheterotrophs. *Appendicula congesta*, *Bulbophyllum disjunctum*, *Dendrobium kiauense*, and *Goodyera rostellata* are species endemic to Borneo found during the expedition along with *Crepidium multiflorum*, a hyper-endemic species to Kinabalu area. Several rare species which narrowly distributed to primary forest habitat were discovered, including jewel orchids; *Cystorchis variegata* var. *variegata* and *Anoectochilus geniculatus*, and two mycoheterotrophs; *Aphyllorchis pallida* and *Lecanorchis multiflora* var. *multiflora*.

**Keywords:** Borneo, Kinabalu area, Orchidaceae

## Introduction

Kadamaian is a township or *mukim*, situated immediately to the northwest of Kinabalu Park in the west coast of Sabah. It is located within the Kota Belud district, and 88.3km from Kota Kinabalu, the capital of Sabah. The name 'Kadamaian' is taken from river found within the area vicinity; Kadamaian river. The highest peak in Kadamaian is Mount Nopungguk, with a height of 1,430m above sea level. Falls within the Kinabalu Park, a World Heritage Site,

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Kadamaian is blessed with different forest types, which are lowland mixed dipterocarp forest, upland mixed dipterocarp forest, and lower montane forest. Due to its richness and abundance of biodiversity, a lot of recreation activities were initiated by locals such as river rafting, homestays, and hiking spots. The prominent Kinabalu Park has been set aside for the conservation and protection of the richest yet endangered plant community (Phillips, 1988; van der Ent, 2013).

Orchidaceae is one of the largest and most diverse families of flowering plants, comprising of 736 genera worldwide with around 27,000 species (Chase et al., 2015). The family is widely distributed in a variety of terrestrial ecosystems especially in the tropics, although it is absent from polar and desert areas (Romero, 1996). Borneo alone, is indeed rich with orchids, having 2,500 to 3,000 species, which is equivalent to 10% of the world's orchids and 75% of the Malesian orchid flora (Lamb, 1991). Of these, 30%-40% are thought to be endemic to Borneo (Chan et al., 1994). The forests of Kinabalu alone have over 1,200 species of wild orchids (Phillips, 1988).

To the best of our knowledge, the first published list of Bornean orchids was by Ridley in 1896 (Wood & Cribb, 1994). The author described 49 new species and recorded 224 species in 62 genera. The list was then updated in 1942 by Masamune (Wood & Cribb, 1994), in which the number was increased to 1,203 species in 99 genera. After that, an account by Wood & Cribb (1994) lists over 1,400 species in 149 genera in Borneo. Following publication of the 1993 orchid volume, Wood et al. (1993), an extensive compilation of the orchids of Mount Kinabalu became available, Wood et al. (2011), lists 866 taxa in 134 genera of orchids.

Since then, a number of studies were conducted to further profile the orchid diversity around Sabah. Most notably within the Crocker Range National Park (Majit et al., 2014), Kinabalu Park (Majit et al., 2014), and gazetted conservation areas such as Danum Valley (O'Malley, 2009), Imbak Canyon (Tsukaya et al., 2013), and Maliau Basin (Suetsugu et al., 2018). One recent study by Juiling et al. (2020) conducted an IUCN Red List assessment on 136 endemic species to Sabah, including in the Kinabalu and Crocker Range parks. So far, there is no known study on wild orchids documented for the Kadamaian area. Nevertheless, we hypothesized a high abundance of orchids could be present in the Kadamaian area given that the area is made of more than one type of forest and located within the biodiverse-mountain range. Moreover, the Bornean highland area possesses a wealth of wild orchids (Wood & Cribb, 1994; Beaman et al., 2001;

Majit et al., 2014; Besi et al., 2020). Orchids habitat specificity (Fay & Chase, 2009; Schödelbauerová et al., 2009) combined with the small populations exhibited by most species and narrow-pattern distribution (Chung et al., 2005; Rodrigoues & Kumar, 2009) makes them vulnerable to the threats of habitat loss and degradation (Coates & Dixon, 2007; Swarts & Dixon, 2009). In the last four decades, Sabah has lost 39.5% of its forest cover (Gaveau et al., 2014). Moreover, over-collection for ornamental and medicinal usage has become another significant threat to the survival of Orchidaceae (Hágsater & Dumont, 1996; Koopowitz et al., 2003).

As a part of conservation efforts to preserve Sabah's nature heritage, a scientific expedition was conducted, and aimed to record the diversity, composition and distribution of wild orchids in selected sites in the Kadamaian area. Here we report the preliminary finding.

## Materials and Methods

### *Study site*

A botanical convenience sampling was carried out from 14<sup>th</sup> to 19<sup>th</sup> October, 2019 in lowland to lower montane dipterocarp forests of Kadamaian, Sabah, Malaysia. Due to the limited time available, only four selected sites were studied including the basecamp area (Table 1, Figure 1 and Figure 2).

**Table 1.** Selected study sites

Sites	Vegetation types	Coordinates		Elev. (m)	Total distance (km)
		Labels in Fig. 1B	Latitudes and longitudes		
Nopungguk	Lower montane forest	1	N 06° 9' 7.8114", E 116° 18' 13.752"	245	ca. 6
		2	N 06° 11' 13.3", E 116° 30' 47.8"	1,419	
		3	N 06° 11' 46.7", E 116° 30' 48.8"	885	
		4	N 06° 11' 55.1", E 116° 30' 45.0"	849	
		5	N 06° 12' 25.6", E 116° 30' 34.1"	471	
Melangkap Noriou	Riparian forest	6	N 06° 11' 13.3", E 116° 29' 30.6"	513	ca. 2
		7	N 05° 50' 80.7", E 118° 07' 57.6"	631	
Hill near basecamp	Forest ridge	8	N 06° 12' 30.3", E 116° 31' 14.4"	875	ca. 1
Ulu Sungai Melawa	Mixed hill dipterocarp forest	9	N 06° 12' 15.0", E 116° 31' 20.9"	933	ca. 1

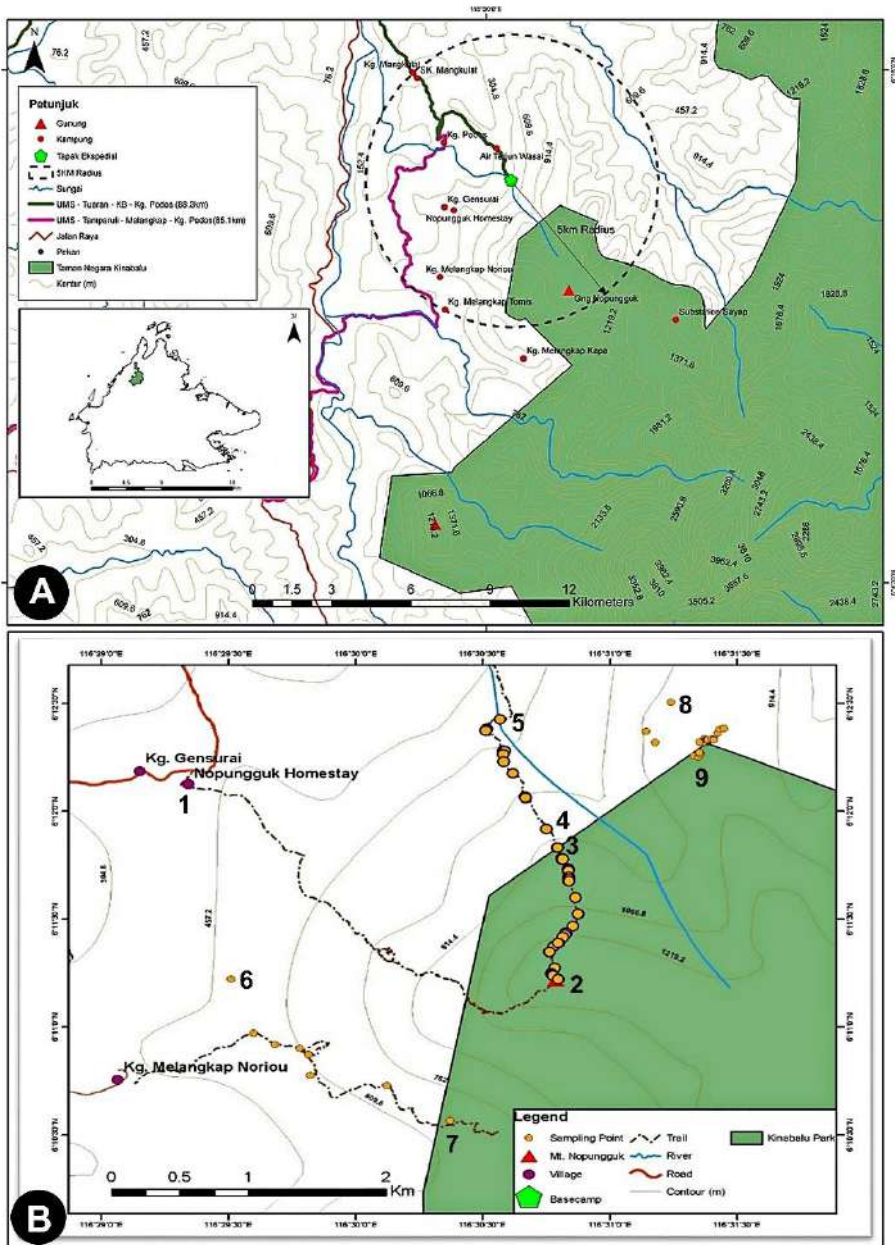


Figure 1. Kada maian, Kota Belud District in Sabah (A) and locations of the study plots (B).

The sites were selected based on the vegetation type. The sampling was done along the existing and off-road four-wheel trails within a width of 10m to the left and 10m to right. Nopungguk is a relatively undisturbed lower montane forest. It is characterized by a cool and dim environment. There are two major trails, Podos-Nopungguk and Nopungguk-Gensurai. We climbed to the peak via Podos-Nopungguk trail and headed back via Nopungguk-Gensurai trail. As we travelled to the higher elevation area, the tree crowns were thicker and the surrounding atmosphere was dimmer and cooler. The ground was covered with leaf litter, protecting the substrate from disturbance. The upper trail was steep and quite narrow banked by cliffs at both sides. The Melangkap Noriou trail is majorly made of a riverine (riparian) forest. The area is a commonly visited recreation site. As we walked through the trail, we passed a secondary forest and rubber plantation, and then we entered a deeply shaded riparian forest that led us to the cascading waterfall of Kuminangkad. The Ulu Sungai Melawa is a hill forest connected to a four-wheel off-road track and forest ridge situated just above the basecamp.



**Figure 2.** Study sites in Kadamaian: A) an emergent tree in Nopungguk trail; B) inland vegetation at the upper area of Nopungguk trail; C) Mirolian Stone (Batu Besar Bertingkat) in the Nopungguk trail; D) riverine trail in Melangkap Noriou; E) Kuminangkad waterfall at the end of Melangkap Noriou trail; F) forest ridge in Ulu Sungai Melawa; G) disturbed hill forest near basecamp; H) a dying tree accumulates epiphytic orchids in the hill forest of Ulu Sungai Melawa.

### *Sampling and specimen processing*

Each site was only visited once during this preliminary study. One individual of the flowering specimens were collected and ripped or dug out properly by ensuring that the whole plant was extracted, including its root. The specimens were preserved using standard herbarium technique after Bridson & Forman (2000) and deposited in the herbarium of Institute for Tropical Biology and Conservation (ITBC), Universiti Malaysia Sabah (UMS). The non-flowering ones were not collected during the expedition following the restriction employed by the managing committee. All orchids encountered, either flowering or non-flowering, were photographed and important notes on their habit and morphology were recorded in a notebook.

The collected orchids were identified into their respective taxa based on their morphological characters and habits. Some identifications were done in the field and made possible based on photographs. Reliable references were used in the identification and classification processes such as Seidenfaden & Wood (1992), Wood (1997), Beaman et al. (2001), Comber (2001), and Wood (2003).

We also studied some of the digitalized herbarium specimens deposited in the international databases such as Harvard University Herbaria & Libraries (AMES) ([https://kiki.huh.harvard.edu/databases/specimen\\_index.html](https://kiki.huh.harvard.edu/databases/specimen_index.html)), Swiss Orchid Foundation (SOF) (<https://orchid.unibas.ch/index.php/en/>), National Herbarium of the Netherlands (NHN) accessed through Browse Dutch Natural History Collections: BioPortal (Naturalis) (<http://bioportal.naturalis.nl/>), and also Kew World Checklist of Selected Plant Families (WCSP) (Govaerts et al., 2020). The information on the current distribution status was retrieved from published checklists by Wood et al. (1993), Wood & Cribb (1994), and Beaman et al. (2001); and the online databases as mentioned above.

## **Results and Discussion**

A total of 58 species and 32 genera were identified during a brief visit to the Mount Nopungguk, Melangkap Noriou, Ulu Sungai Melawa, and basecamp sites of the 2019 Kadamaian Scientific expedition. Only 23 orchids were identified into their respective taxa, with the remaining only identified to their genera level or closest affinity as the floral structure was lacking upon assisting the identification. From the total orchids collected during the expedition, only 30% were flowering and fruiting, presumably due to the non-flowering period for orchids in the area. Some orchids were found with seed pods or dehiscent seed pods signalling that we missed the flowering season during the expedition. Our



finding consists of 43 epiphytes, 13 terrestrials, and two mycoheterotrophs (Figure 3), and of these, 52 species are Epidendroideae, four are Orchidoideae, and one each for Apostasioideae and Vanilloideae (Figure 4). *Bulbophyllum*, *Coelogyne* and *Dendrobium* are the most abundant genera (Figure 5).

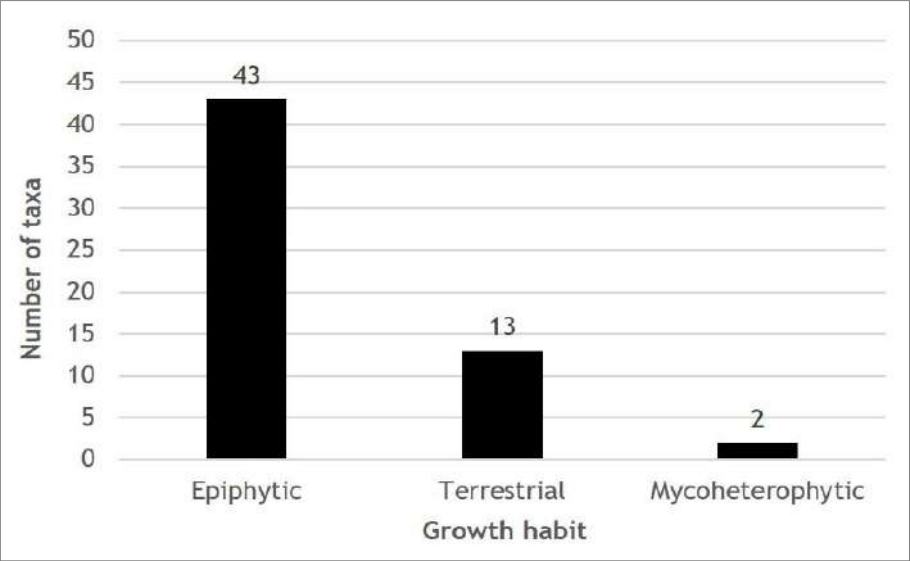


Figure 3. Number of orchid species with different types of growth habits in Kadamaian.

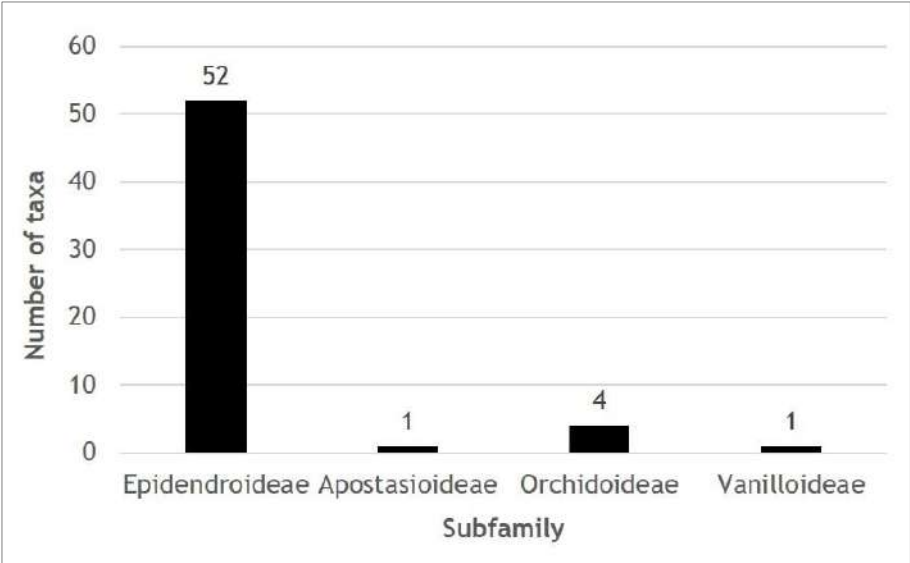


Figure 4. Number of orchid species from different subfamilies found in Kadamaian.

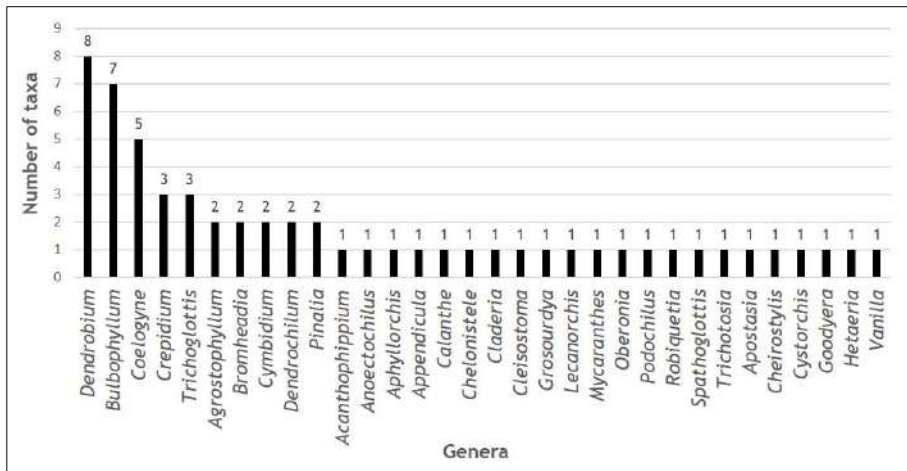


Figure 5. Number of orchid genera found in Kadamaian.

The inland lower montane forest of Nopungguk harboured the most abundant orchid species (Figure 6), as it is roofed by emergent trees, shaded and moist. Also, the trail is undisturbed and longer than the other visited sites, which provided more opportunity to encounter orchids. The soil in this area is typically that of humus and peat forming the upper layer. This is likely due to a high soil water content, reduced sun radiation, and low rates of decomposition. Epiphytic orchids were in great abundance, notably the necklace orchids, *Coelogyne* and its allied genera such as *Chelonistele*, *Dendrochilum*, and *Pholidota*. Also, in great abundance are species of *Bulbophyllum* and *Dendrobium*. However, terrestrial orchids also showed a high dominance as the shady and moist forest floor was ideal for their growth.

The riverine forest should have accumulated an abundant orchids species, particular favourites of epiphytes that prefer microclimates where it is often cooler, with swift air currents (Wood, 2008). However, the Melangkap Noriou trail was deeply shaded and dark even during the daytime where sunlight was perching above the canopy. Presumably, the thick canopy roofed the lower canopy area, limiting exposure to sunlight. For orchids, lower light levels and too low temperatures are limiting factors and here they are less abundant (Wood, 2008). Also, 100m from the trail's entrance is a secondary forest and rubber plantation. The secondary forest is occupied by the pioneers, such as *Macaranga*. Invasive species from family Poaceae and Cyperaceae were also very prominent in this area. Such forests are poor in orchid species diversity (Wood,

2013). Only a few *Dendrobiums* found a refuge in the plantations, including *D. acerosum*.

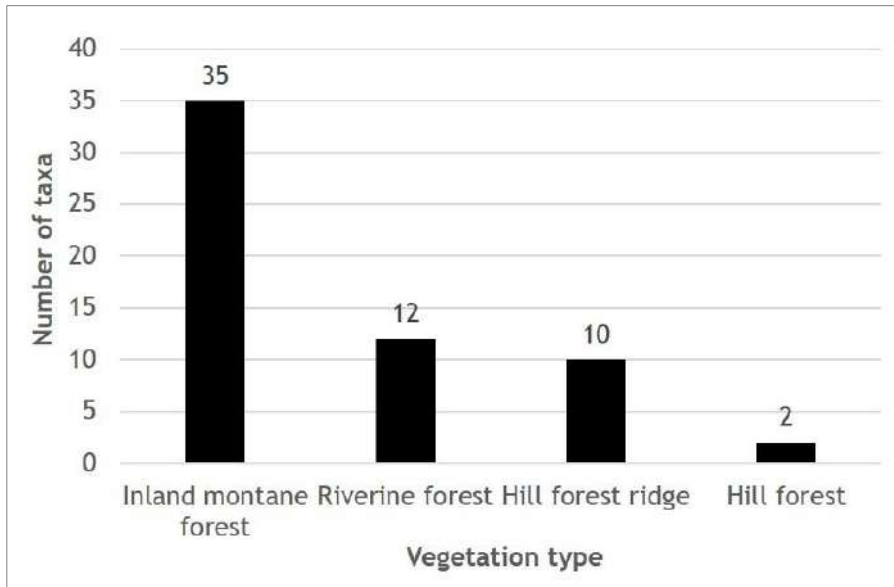


Figure 6. Number of orchid species from different vegetation types found in Kadamaian.

Four species were endemic to Borneo; *Appendicula congesta*, *Bulbophyllum disjunctum*, *Dendrobium kiauense*, and *Goodyera rostellata*, with *Crepidium multiflorum*, a hyper-endemic species, endemic to Mount Kinabalu area. Three of the endemic species, *A. congesta*, *B. disjunctum*, and *D. kiauense*, were found in the Nopungguk trail, a relatively undisturbed inland montane forest, dominated by tall trees with thick interlocking crown, allowing very minimal sunlight exposure. Meanwhile, *G. rostellata* was found in the Melangkap Noriou trail, a riverine forest connected to a secondary forest and a rubber plantation. Several species narrowly distributed to the primary forest habitat were discovered, including a jewel orchid; *Cystorchis variegata* var. *variegata* and *Anoectochilus geniculatus* (Figure 7). Notably, a rare jewel orchid, or probably a new one, was found during the expedition. We have not managed to accurately identify the specimen to its respective taxa nor the genus. In this paper, it is conferred to a *Cheirostylis* species referring to the leaf pattern (Figure 7H). None of the known jewel orchids genera in Borneo resembles our specimen. A further study could not be carried out as no other specimen was collected.



**Figure. 7.** Rare and endemic orchid species found in Kadamaian: A) *Anoectochilus geniculatus* (plant); B) *Anoectochilus geniculatus* (flower); C) *Appendicula congesta*; D) *Bulbophyllum disjunctum*; E) *Cystorchis variegata* var. *variegata*; F) *Dendrobium kiauense*; G) *Goodyera rostellata*; H) Unidentified plant (cf. *Cheirostylis*).

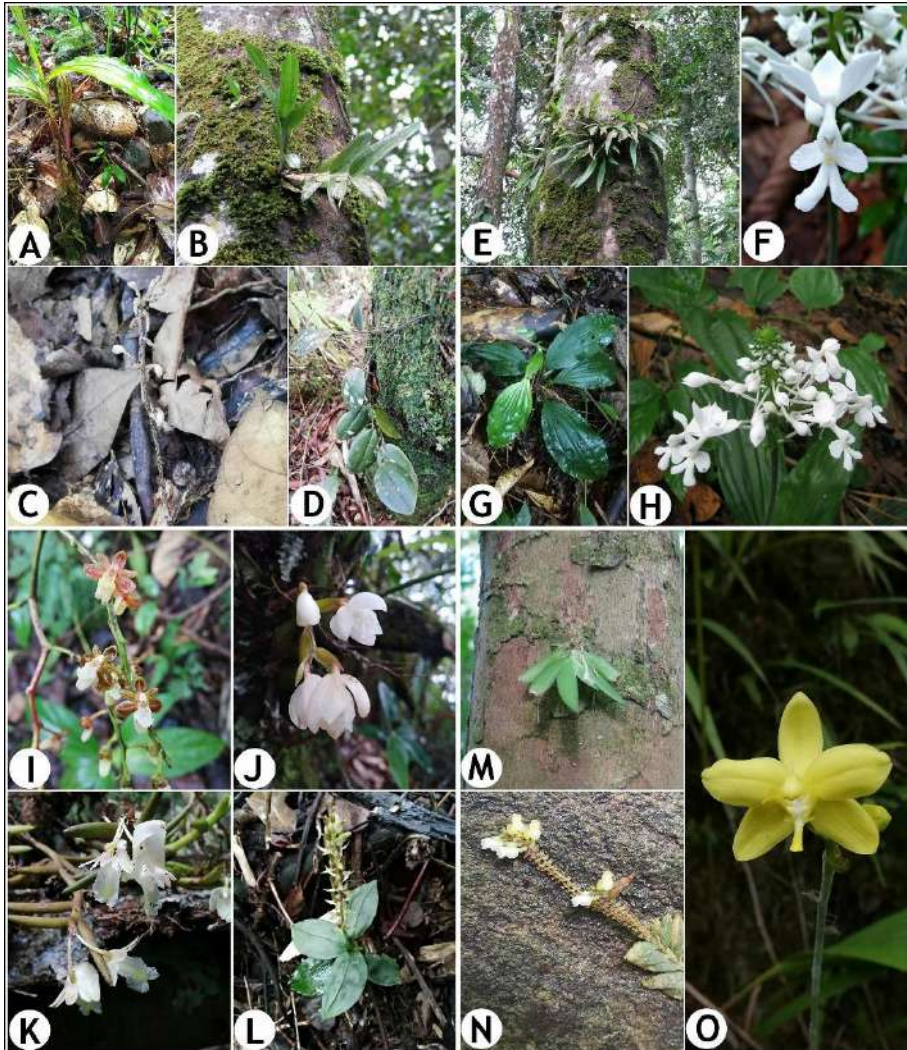
*Crepidium multiflorum* is an erect plant with flowers small, non-resupinate, the front edge of the lip simple or more or less 3-lobed, and the apical part often emarginated or bilobulate (Figure 8). Also, the plant that we discovered is very similar to the holotype of *Clemens* AMES 16941 (AMES-photo!), similar in the size of the plant, the spreading ovate to wide elliptic leaves, morphology of the flowers based on the illustrations on the herbarium sheets, except the flowers of *C. multiflorum* described in the holotype are orange whereas our plants have flowers that are greenish-yellow. The *C. multiflorum* is almost identical to *C. balabacense* and *C. moluccanum* in both vegetative and floral morphologies. However, the latter two species have not been recorded in Borneo until now. Further revision of this complex is recommended to evaluate the legitimate status. It commonly occurs in hill forest at elevation 900-1,200m above sea level (Wood et al., 1993). In the current study, the plant was found growing along with *Spathoglottis aurea* on a disturbed ground along the margin of a four-wheel off-road track situated just above our basecamp.



Figure 8. *Crepidium multiflorum*: A) Inflorescence; B) Plant.

Importantly, we discovered two mycoheterotrophs; *Aphyllorchis pallida* and *Lecanorchis multiflora* var. *multiflora*. The occurrence of these flagship species along the trails were rare with 2-3 individuals growing along the mentioned trails. The findings were listed in Table 2. The checklist includes brief information on each species growth habits, sites, and vegetation types. Some of the collected orchid species are shown in the Colour Plate (Figure 9). Among the selected sites, Melangkap Noriou trail was the most disturbed and occupied invasive species and these were seen to affect the abundance of wild orchids in the area. If the disturbance is not minimized and appropriate conservative measures, especially habitat restoration, are not taken, the effect of habitat destruction and ecological alteration will be detrimental and eventually destroy the vulnerable terrestrial orchids in the area. The Nopungguk trail was relatively undisturbed and there was no sign of destruction in the area. Presence of the jewel and terrestrial orchid species in a forest area serve as an indicator of a pristine environment. The selected sites are all within the Kinabalu Park area and put under the care of Sabah Parks, which is considerably well-protected under their jurisdiction. To quote Juiling et al. (2020), a collaboration with any ex-situ conservatories in Sabah, especially the Poring Orchid Garden, as well as local community for wild orchids ex-situ conservation is highly recommended.





**Figure 9.** Wild orchids of Kadamaian and their growth habit: A) *Acanthophippium javanicum*; B) *Agrostophyllum* sp.; C) *Aphyllorchis pallida*; D) *Bulbophyllum* sp.; E) *Bulbophyllum* sp.; F) *Calanthe triplicata* (flower); G) *Calanthe triplicata* (plant); H) *Calanthe triplicata* (flowers); I) *Cleisostoma racemiferum*; J) *Coelogyne clemensii*; K) *Dendrobium acerosum*; L) *Hetaeria* sp.; M) *Oberonia* sp.; N) *Podochilus lucescens*; O) *Spathoglottis aurea*.

Table 2. Preliminary list of orchid taxa found in the selected sites in Kadamaian, Sabah.

No	Subfamilies	No	Genera	No	Taxa	Growth habits	Localities
1	Epidendroideae	1	<i>Acanthophippium</i>	1	<i>A. javanicum</i> Blume	TR	MN
		2	<i>Agrostophyllum</i>	2	<i>A. sp</i> (1)	EP	NP
				3	<i>A. sp</i> (2)	EP	NP
		3	<i>Anoectochilus</i>	4	<i>A. geniculatus</i> Ridl.	TR	NP
		4	<i>Aphyllorchis</i>	5	<i>A. pallida</i> Blume	MH	NP
		5	<i>Appendicula</i>	6	<i>A. congesta</i> Ridl.	EP	NP
		6	<i>Bromheadia</i>	7	<i>B. brevifolia</i> Ridl.	EP	NP
				8	<i>B. sp.</i>	TR	NP
		7	<i>Bulbophyllum</i>	9	<i>B. disjunctum</i> Ames & C.Schweinf. in O.Ames	EP	NP
				10	<i>B. membranaceum</i> Teijsm. & Binn.	EP	SM
				11	<i>B. sp</i> (1)	EP	NP
				12	<i>B. sp</i> (2)	EP	NP
				13	<i>B. sp</i> (3)	EP	NP
				14	<i>B. sp</i> (4)	EP	NP
				15	<i>B. sp</i> (5)	EP	SM
		8	<i>Calanthe</i>	16	<i>C. triplicata</i> (Willemet) Ames	TR	NP & MN
		9	<i>Chelonistele</i>	17	<i>C. cf. sulphurea</i>	EP	NP
		10	<i>Claderia</i>	18	<i>C. viridiflora</i> Hook.f.	EP	NP
		11	<i>Cleisostoma</i>	19	<i>C. racemiferum</i> (Lindl.) Garay	EP	MN
		12	<i>Coelogyne</i>	20	<i>C. clemensii</i> Ames & C.Schweinf. in O.Ames	EP	NP
				21	<i>C. cuprea</i> H.Wendl. & Kraenzl.	EP	NP
				22	<i>C. sp</i> (1)	EP	NP
				23	<i>C. sp</i> (2)	EP	NP
				24	<i>C. sp</i> (3)	EP	SM
		13	<i>Crepidium</i>	25	<i>C. multiflorum</i> (Ames & C.Schweinf.) Szlach.	TR	B
				26	<i>C. sp</i> (1)	TR	MN
				27	<i>C. sp</i> (2)	TR	MN
		14	<i>Cymbidium</i>	28	<i>C. sp</i> (1)	EP	NP
				29	<i>C. sp</i> (2)	EP	MN
		15	<i>Dendrobium</i>	30	<i>D. acerosum</i> Lindl.	EP	MN
				31	<i>D. crumenatum</i> Sw.	EP	NP
				32	<i>D. kiauense</i> Ames & C.Schweinf. in O.Ames	EP	NP
				33	<i>D. sp</i> (1)	EP	NP
				34	<i>D. sp</i> (2)	EP	NP
				35	<i>D. sp</i> (3)	EP	SM
				36	<i>D. sp</i> (4)	EP	SM
				37	<i>D. villosulum</i> Wall. ex Lindl.	EP	NP
		16	<i>Dendrochilum</i>	38	<i>D. sp</i> (1)	EP	NP
				39	<i>D. sp</i> (2)	EP	NP
		17	<i>Grosourdya</i>	40	<i>G. sp.</i>	EP	SM

	18	<i>Lecanorchis</i>	41	<i>L. multiflora</i> J.J.Sm. var. <i>multiflora</i>	MH	NP	
	19	<i>Mycaranthes</i>	42	<i>M. sp.</i>	EP	NP	
	20	<i>Oberonia</i>	43	<i>O. sp.</i>	EP	MN	
	21	<i>Pinalia</i>	44	<i>P. sp</i> (1)	EP	NP	
			45	<i>P. sp</i> (2)	EP	MN	
	22	<i>Podochilus</i>	46	<i>P. lucescens</i> Blume	EP	SM	
	23	<i>Robiquetia</i>	47	<i>R. sp.</i>	EP	SM	
	24	<i>Spathoglottis</i>	48	<i>S. aurea</i> Lindl.	TR	B	
	25	<i>Trichoglottis</i>	49	<i>T. sp</i> (1)	EP	NP	
			50	<i>T. sp</i> (2)	EP	NP	
			51	<i>T. sp</i> (3)	EP	SM	
	26	<i>Trichotosia</i>	52	<i>T. sp.</i>	EP	NP	
2	Apostasioideae	27	<i>Apostasia</i>	53	<i>A. nuda</i> R.Br. in N.Wallich	TR	NP
3	Orchidoideae	28	<i>Cheirostylis</i>	54	<i>C. sp.</i>	TR	NP
		29	<i>Cystorchis</i>	55	<i>C. variegata</i> Blume var. <i>variegata</i>	TR	SM
		30	<i>Goodyera</i>	56	<i>G. rostellata</i> Ames & C.Schweinf. in O.Ames	TR	MN
		31	<i>Hetaeria</i>	57	<i>H. sp.</i>	TR	MN
4	Vanilloideae	32	<i>Vanilla</i>	58	<i>V. sp.</i>	EP	MN

Notes: EP = Epiphytic, TR = Terrestrial, MH = Mycoheterotrophic, NP = Nopunguk, MN Melangkap Noriou, SM = Ulu Sungai Melawa, B = Hill near basecamp area.

## Conclusions and Recommendations

Our collection of 58 orchid species belonging to four subfamilies is considerably high despite the brief visit and limited time spent in the selected sites. Here, we emphasize the importance of undisturbed vegetation type on the species abundance, where the highest abundance was found in the inland montane forests. No species is listed as a new record as most of the specimens have not been completely identified to the respective taxa due to lack of floral structures. One limitation of our current study is insufficient samples. Nevertheless, there could be more to be discovered in the highland area due to the undisturbed environment. Protection of the undisturbed forest area, especially the Mount Nopunguk and Ulu Sungai Melawa areas, is imperative to ensure the survival and richness of these precious wild orchids are maintained. Mount Nopunguk has great potential as a hiking spot where tourists would enjoy the magnificent view of Mount Kinabalu up-close. To reverse the threats, an integrated diversity and population study, and ecology and conservation assessments in the Kadamaian area must first be conducted.



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**Research Article**

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**Soil Physico-Chemistry in the Habitat of *Rafflesia* in Kinabalu Park, Sabah, Malaysia.**

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**Abstract**

This study was conducted to identify the type of soil texture, and its relationship with *Tetrastigma* sp., a host of the *Rafflesia* sp. in Kinabalu Park, Sabah, Malaysia. The soil samples were collected from five study areas: Losou Podi, Losou Minunsud, Sayap Substation, Langanan and Gansurai. The plot was selected when the host exhibited traits of being infected by *Rafflesia*, either by the presence of buds, flowers or residual scar marks found on the host. The result reveals that the soil in the habitat of *Rafflesia* sp. and their host is sandy loam type, with a high volume of sand compared to silt and clay, between 65.40-79.25%. The soil moisture in the area is low, ranging from 14.89% to 27.96%. The soil in the plots was less fertile due to low value of soil organic matters (1.12-1.40%), with slightly acidic soil pH value (4.08-4.73). The most abundant elements contained in the soil were Fe, Al and Mg. The different *Rafflesia* habitats were observed to have a relationship with different soil factors: either physical, chemical, or both to promote the growth of *Rafflesia*. There was relationship between Sayap Substation with some chemical elements in the soil, rather than the soil's physical characteristic. Both Langanan and Losou Podi were only influenced by the physical characteristics of the soil. In comparisons, the Gansurai and Losou Minunsud have a relationship influenced by a combination of physical properties and chemical elements in the soil. From this study, it can be concluded the presence of *Tetrastigma* sp. in the different *Rafflesia* habitats has its own relationship with the soil and is not influenced by one factor.

**Keywords:** Kinabalu Park, *Rafflesia*, Soil physico-chemistry, *Tetrastigma*.

## Introduction

Kinabalu Park is located on the West Coast of Sabah, Malaysian Borneo, specifically in the Crocker Formation, underpinned by the Temburong Formation consisting of three types of rock units: thick sandstone, sandstone and shale interval unit, and thin sandstone and shale interval unit (Tracy et al., 2018). The parent soil material consists of sandstones, which explains the exceptionally high sand content in this area (Keng et al., 2020). Most of the soils in the Crocker Formation area are characterized by textured loam types (Nor Azlan et al., 2017) and clayey loams found at an altitude of 921 meters a.s.l. (Keng et al., 2020).

Both *Tetrastigma* and *Rafflesia* are often found in slightly rocky and sandy habitats (Balete et al., 2010; Barcelona et al., 2007). Several studies have recorded that the soils in their habitats were slightly acidic (Nasihah, 2016; Nur Hayati et al., 2021) to almost neutral (Ali et al., 2015; Laksana et al., 2018; Lianah, 2014). The high acidity level of the soil significantly affects the types and amount of chemicals in the soil. The increase in the number of certain chemicals present in the soil is due to decrease in soil pH value (Widowati & Sukristyonubowo, 2012). Excess trace elements in the soil will increase the soil toxicity (Purwanti et al., 2018) and soil pollution (Sellan et al., 2019). Severe soil toxicity will cause death to organisms, thus disrupting the habitat's ecosystem balance.

The *Rafflesia* sp. (Rafflesiaceae) which is famous for its spectacular large flower (Abang Hashim & Hans, 2000) can be found in Sabah (Nais, 2001). Three species have been recorded in Sabah namely *Rafflesia pricei*, *R. keithii* and *R. tengku-adlinii* (Mat-Salleh, 1991; Nais, 2001). *Rafflesia* is a holoparasitic plant (Nikolov et al., 2014) that lives on its host, the *Tetrastigma* that belongs to family Vitaceae (Mat-Salleh et al., 2011; Nasihah et al., 2016; Takhtajan, 2009). To date, there are no records reporting *Rafflesia* inhabiting a host from another genus besides *Tetrastigma*.

*Rafflesia* faces the threat of habitat destruction and extinction (Yeo et al., 2012). Their flowers have been used as an ingredient in traditional medicine (Fu et al., 2011; Lianah, 2014) and for multi-uses in daily life (Chettri & Barik, 2013; Kar et al., 2013). In addition, logging activities and natural disasters also contribute to their extinction (Latiff & Mat-Salleh, 1991; Yahya et al., 2010). In Sabah, both *Rafflesia* and *Tetrastigma* are fully protected plants under the Wildlife Conservation Enactment 1997, Schedule I (Part II, Section 54 (1) (a)) to prevent the public from consuming these two plants (Sabah Wildlife Department, 1997). In addition, Sabah Parks has introduced a scheme to

encourage the conservation of these two plants through the *Rafflesia* Conservation Intensive Scheme (RCIS) involving private land owned by villagers around Kinabalu Park (Nais & Wilcock, 1998).

*Rafflesia* is extremely sensitive to changes in its surroundings, especially when the host suffers from any damage (Nais, 2001). With efforts to preserve and conserve the host, these flowers are also indirectly protected. Therefore, it is crucial to understand the ecology of *Tetrastigma* to ensure that *Rafflesia* can grow well. This study aims to evaluate the physical and chemical characteristics of soil in *Rafflesia*'s habitat at Kinabalu Park, to gain a better understanding of the soil characteristics in the habitat of *Rafflesia*. This information is crucial for in-situ conservation of *Rafflesia* sp. in Kinabalu Park and Sabah state. In-situ conservation of this plant is very important because its natural habitat is decreasing rapidly. This research will improve our knowledge of this plant, which is important for conservation.

## Methods and Materials

### *Study location*

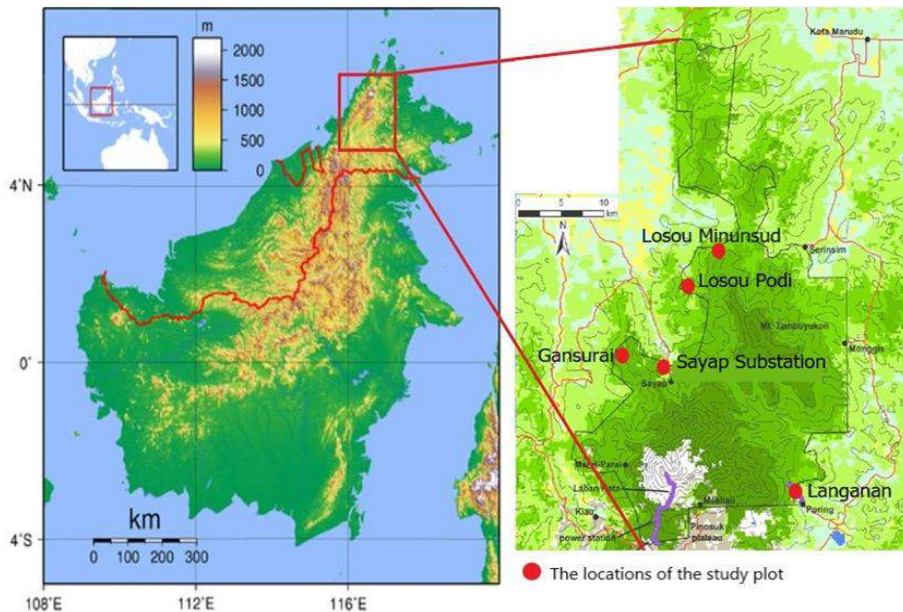
The study was conducted in Kinabalu Park, located about 20km from Ranau Town, Sabah and it covers an area of 754 km<sup>2</sup>. The geographical position is at latitude N 6° 5' and longitude E 160° 33', with the study area's average elevation ranging from 634 to 994 metres a.s.l., and the vegetation type varying according to altitude (Rafiqpoor & Nieder, 2006). All the five study areas surveyed in the present study comprised of hill dipterocarp forest.

### *Soil Sampling*

Overall, there were five circular-shaped plots established within the Kinabalu Park area. There were two districts involved -- in Kota Belud: namely Losou Podi (LP) at 666 m a.s.l. (N 06° 19' 13.9", E 116° 38' 32.8"), Losou Minunsud (LM) at 634 m a.s.l. (N 06° 20' 54.7", E 116° 37' 47.1"), Sayap Substation (SS) at 909 m a.s.l. (N 06° 10' 02.8", E 116° 33' 51.6"), and Gansurai (GA) at 744 m a.s.l. (N 06° 11' 25.0", E 116° 29' 57.0"); and in Ranau: Langanan (LA) at 994 m a.s.l. (N 06° 03' 49.9", E 116° 41' 14.8"). The plot selection was made based on the presence of a *Rafflesia*'s host, (*Tetrastigma* sp.) at the study locations, and *Tetrastigma* sp. represent the centre point for the plot. The selection of hosts was based upon traits exhibited when the *Tetrastigma* sp. was infected by a *Rafflesia* (either the presence of buds, flowers or residual scar marks) (Suwartini et al., 2008).

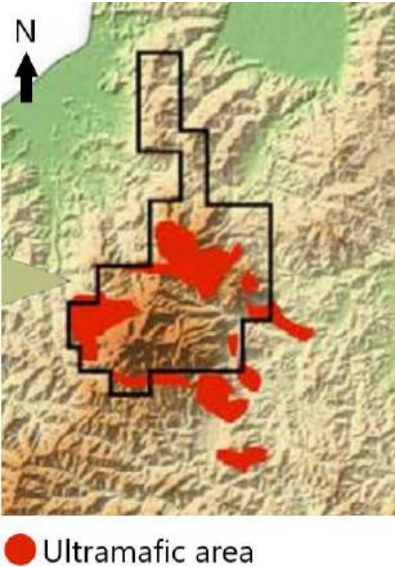
According to van der Ent et al. (2018), the land in Kinabalu Park consists of ultramafic and non-ultramafic types. The map provided by the author indicates that none of the study plots were located within the ultramafic soil type area. Figure 1 shows the location of each study plot around Kinabalu Park, Sabah.

The plots were circular, with a radius of 20 metres (Figure 3), and they were established by applying the placement of a *Rafflesia*'s host in the plots as point zero (Nur Hayati et al., 2021). The total area of all surveyed plots was 0.6285 ha<sup>-1</sup> (0.1257 hectares). An auger was used to collect soil samples at a depth of 20cm, at random, with a total of nine replications per plot (Nur Hayati et al., 2021). The soil samples were stored in plastic bags (Sarker et al., 2018) and were processed in the Faculty of Science and Natural Resources, Universiti Malaysia Sabah for soil analyses.

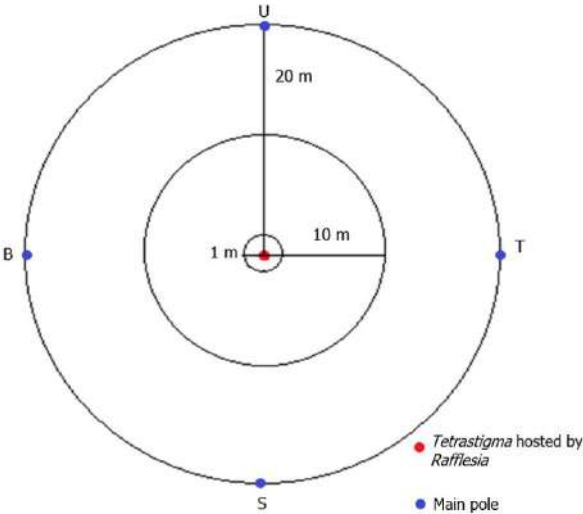


**Figure 1:** The red dot in the map shows the study location around Kinabalu Park, Sabah. Source: Modified from Harris et al. (2012).





**Figure 2.** Shaded relief map of Kinabalu Park with ultramafic occurrences (marked in red).  
Source: Modified from van der Ent et al. (2018).



**Figure 3.** Plot design of the study.

The soil samples were dried by applying the air-drying method at a temperature of around 24-30°C (Anderson & Ingram, 1993; Sarker et al., 2018; Tangketasik et al., 2012) before being processed. Some of the completely dried soil samples were carefully crushed to separate the roots and large rocks. Next, the soil was sieved using a 2mm filter (Alhameid et al., 2017; Anderson & Ingram, 1993; Bottinelli et al., 2017), before it was used to determine minerals in the soil. The soil texture was determined by using the closed beaker sedimentation method (Whiting et al., 2011) and the soil texture triangle (USDA, 1960). Fresh soil samples (without being dried) were used to read the soil pH value, soil moisture content and soil organic matter (SOM).

#### *Determination of the Soil pH, Soil Moisture & Soil Organic Matter*

A total of 10 grams of fresh soil sample was used to read the soil pH value. The soil was dissolved in 25ml distilled water in a beaker and was stirred for 10 minutes before being left for 30 minutes before taking the pH reading by using a pH meter. This procedure was repeated 3 times to obtain the mean value (Anderson & Ingram, 1993).

To measure the soil moisture, the soil and crucible samples were weighed and recorded as M1. The samples were then heated overnight at 105°C. The sample was left to cool in a desiccator before being reweighed (M2). The water content present in the soil and the dry weight of the soil was calculated using the following formula (Anderson & Ingram, 1993):

$$\text{Percentage of soil moisture} = \frac{M1 - M2}{M1} \times 100$$

Where;

M1 = Initial weight of the soil

M2 = Final weight of the soil

The same dried soil samples were used to calculate the total soil organic matter (SOM). Soil samples were measured and labelled as M1 before being placed overnight in a furnace up to a temperature of 400°C. The sample was then reweighed and recorded as M2 after the soil sample had cooled. The formula to calculate the SOM is the same as soil moisture formula.

#### *Determination of the Soil Texture*

Roots and large rocks were removed from the soil samples after undergoing a drying process. Clumped soils were crushed to avoid any errors during the sedimentation process which may result in errors in the readings.

A long, tapered clear beaker was filled with soil and water in a 1:2 ratio. The lid of the beaker was tightly closed and shaken for 10 minutes to break up and separate the mineral particles in the soil. After 1 minute, the depth of sand was measured. The sample was left uninterrupted for 2 hours before measuring the silt depth. The sample was left again uninterrupted until the water became completely clear before measuring the depth of the clay soil. Figure 4 shows the condition of a fully completed soil layer (Whiting et al., 2011). This procedure was repeated for all samples using different beakers.

From the sediment thickness of the soil layers formed, the percentage of each soil particle was obtained using the following formula;

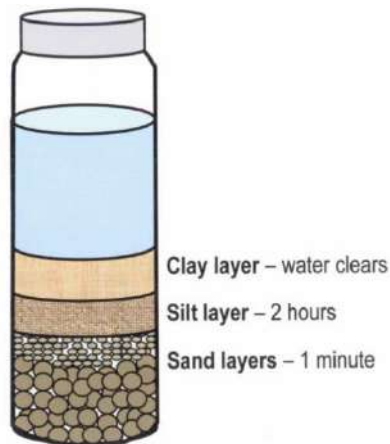
$$\text{Percentage of particle types in the soil} = \frac{h}{H} \times 100$$

Which is;

$h$  = Particle layer thickness

$H$  = The overall thickness of the sediment

The percentage of each layer of these particles was then applied by referring to the soil texture triangle (Figure 5) to obtain the cross point between the percentage of particles to determine the soil type.



**Figure 4.** Measuring soil texture (Whiting et al., 2011).

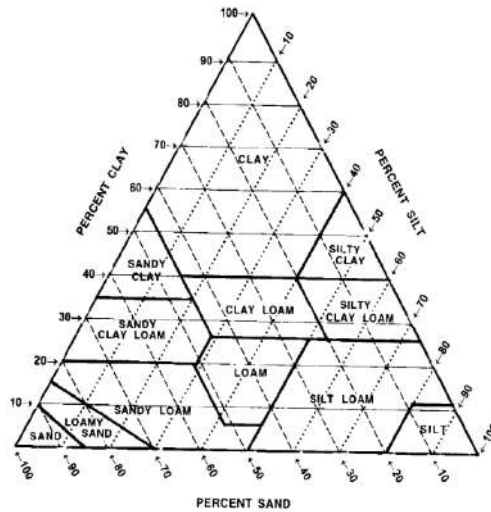


Figure 5. Soil texture triangle (USDA, 1960).

#### *Determination of Minerals in Soil*

The dried and filtered soil sample in powder was mixed with aqua-regia (a mixture of nitric acid and hydrochloric acid) and was left overnight to allow the mineralization process to complete. The sample solution was filtered and diluted to 50ml before being analysed using Inductively Coupled Plasma ICP-OES equipment Perkin Elmer Optima model 5300DV to measure the content of elements, which are Aluminium (Al), Iron (Fe), Potassium (K), Calcium (Ca), Magnesium (Mg), Manganese (Mn), Sodium (Na) and Vanadium (V) (Owens & Cornwell, 1995; Santoro et al., 2017; ).

#### *Data Analysis*

The collected samples were analysed for physical and chemical soil parameters such as soil pH, soil texture, and chemical in the soil. By using one-way ANOVA, significant difference in physical and chemical characteristics in each study plot was tested. To investigate the relationship between soil factors and *Rafflesia* hosts, Principal Component Analysis (PCA) was conducted. All statistics were analysed using Paleontological Statistic (PAST) version 3.26.

## **Results and Discussions**

#### *Physical properties of soil*

Table 1 shows that the soil has significant difference for physical characteristics between surveyed plots with  $p < 0.05$ . All plots have a higher content of sand

(65.40±8.39 - 79.25±9.84%) when compared to the content of clay (10.76±7.4 - 11.93±7.75%) and silt (9.84±8.06 - 23.84±9.80%). According to the soil texture triangle by USDA (1960), all soil textures in the study area are sandy loam type. The soil moisture content and SOM were low with the range from 14.89% to 27.96% and 1.12% to 1.40%, respectively. Only soil moisture showed a significant difference between the plots ( $p<0.001$ ). Meanwhile, soil texture that comprised of sand, clay and silt did not show any significant differences between the plots ( $p>0.05$ ).

Table 1. Physical content of soil at the study site.

Plot	Soil Moisture (%)	SOM (%)	Sand (%)	Silt (%)	Clay (%)	Soil Type
LP	20.38±1.65 <sup>bc</sup>	1.21±0.45 <sup>a</sup>	65.40±8.39 <sup>ab</sup>	23.84±9.80 <sup>a</sup>	10.76±7.4 <sup>a</sup>	Sandy loam
LM	19.42±5.49 <sup>bc</sup>	1.26±0.43 <sup>a</sup>	73.66±8.08 <sup>ab</sup>	14.94±9.44 <sup>ab</sup>	11.40±4.91 <sup>a</sup>	Sandy loam
SS	14.89±5.41 <sup>c</sup>	1.12±0.54 <sup>a</sup>	79.25±9.84 <sup>a</sup>	9.84±8.06 <sup>b</sup>	10.91±6.98 <sup>a</sup>	Sandy loam
LA	27.96±6.66 <sup>a</sup>	1.40±0.59 <sup>a</sup>	72.98±12.19 <sup>ab</sup>	15.95±7.88 <sup>ab</sup>	11.07±5.78 <sup>a</sup>	Sandy loam
GA	25.82±2.62 <sup>ab</sup>	1.25±0.89 <sup>a</sup>	71.89±9.97 <sup>ab</sup>	16.17±11.48 <sup>ab</sup>	11.93±7.75 <sup>a</sup>	Sandy loam
<i>p</i> -value	0.000	0.915	0.078	0.054	0.994	-

Values are mean ± SD; Values with similar alphabets were not significantly different with  $p>0.05$  based on Tukey HSD test. Note: LP=Losou Podi; LM=Losou Minunsud; SS=Sayap Substation; LA=Langanan; GA=Gansurai; SOM=Soil organic matter.

The soil moisture in the study area is slightly higher when compared to the lower soil moisture in Royal Belum State Park (RBSP) (3.81-7.13%) (Nur Hayati et al., 2021). The soil in the study area was found to be less fertile due to low SOM value (Subowo, 2010; Tangketasik et al., 2012) when compared with the findings of Nur Hayati et al., (2021), with 2.30-8.17% and Nasihah (2016) with 2.36-4.19% in Lojing Highlands, Kelantan. A high level of SOM can help improve soil aggregates to reduce soil permeability to water (Zulfahmi et al., 2007).

Sandy loam soil type can accommodate a sufficient amount of water (Noborio & Kubo, 2017) for the use of both *Rafflesia* sp. and *Tetrastigma* sp. without damaging the host's roots and the decay of *Rafflesia* due to excess water. The soil with higher percentage of sand content can provide sufficient amount of water during the dry season, which allows the soils in *Rafflesia* and *Tetrastigma* habitats to have a good underground drainage system (Balet et al., 2010; Barcelona et al., 2007).

As stated in Nur Hayati et al. (2021), *Rafflesia* and *Tetrastigma* habitat soils in RBSP were more characterized by loamy soils with higher sand content, compared to silt and clay. However, the percentage of silt and clay content in RBSP were similar. The total percentage of sand content in RBSP was lower when

compared to that from this study. However, Ali et al. (2015) stated that the *Rafflesia* habitat in West Java is characterized as silty clay loam habitat type with high clay content, followed by silt and the lowest was sand. The percentage of soil content between these areas were significantly different.

#### *Chemical properties of soil*

The soil in the study area was slightly acidic with a pH range of 4.08-4.73. GA has the highest Fe, Al and V concentration compared to the other habitats, 131.01±95.76 ppm, 75.65±57.67 ppm and 0.42±0.34 ppm, respectively. SS has the highest concentrations of Mg, K, Ca and Na with readings of 20.86±16.90 ppm, 11.26±8.81 ppm, 9.75±6.77 ppm and 0.76±0.60 ppm respectively. LM had the highest concentration of Mn elements with a value of 4.05±2.70 ppm (Table 2). From the one-way ANOVA analysis, only Al and Na did not show any significant difference between the plots ( $p > 0.05$ ).

The pH values obtained from this study were similar to van der Ent et al. (2018), where the pH ranged between 4.6-6.0 in Kinabalu Park. However, Quintela-Sabaris et al. (2020) stated that the pH value of ultramafic soils in Northern Sabah is almost neutral, at around pH 7. Several *Tetrastigma* habitats in Peninsular Malaysia also recorded pH values between 3.51 to 5.80 (Mohd Afiq Aizat, 2018; Nasihah, 2016; Nur Hayati et al., 2021; Syamsurina et al., 2018). However, the pH value of soils in *Tetrastigma* habitat in Indonesia was close to neutral with pH of 5.7-7.0 (Ali et al., 2015; Laksana et al., 2018; Lianah, 2014).

Based on Kitayama et al. (1998), the serpentinite rocks around Mount Kinabalu located in the Kinabalu Park area have a higher content of Fe, Mg, Ni, Cr and Co. Meanwhile, Ca, K and P were found in lower concentrations. This study recorded similar results where the habitat shows higher amounts of Fe, Al and Mg compared to the other elements: Ca, K, V, Mn and Na (Table 2). Additionally, van der Ent et al. (2018) also noted higher concentrations of Ca, Fe, Mg, and Mn in the ultramafic region; whereas the elements K, Na, P and Si had higher concentrations in the non-ultramafic region.

**Table 2.** Chemical composition of soil at the study site (ppm).

Plot	LP	LM	SS	LA	GA	P-value
pH	4.25±0.1 <sup>4b</sup>	4.71±0.27 <sup>a</sup>	4.73±0.40 <sup>a</sup>	4.08±0.21 <sup>b</sup>	3.8±0.24 <sup>b</sup>	0.000
Al	16.09±9.53 <sup>b</sup>	48.54±38.75 <sup>ab</sup>	48.69±53.33 <sup>ab</sup>	33.35±28.79 <sup>ab</sup>	75.65±57.67 <sup>a</sup>	0.055
Ca	1.99±0.96 <sup>bc</sup>	7.63±5.31 <sup>ab</sup>	9.75±6.77 <sup>a</sup>	1.08±0.90 <sup>c</sup>	4.56±5.74 <sup>abc</sup>	0.001
Fe	42.70±26.96 <sup>b</sup>	83.96±68.21 <sup>ab</sup>	59.32±50.34 <sup>ab</sup>	56.56±49.67 <sup>ab</sup>	131.01±95.76 <sup>a</sup>	0.039
K	1.68±0.87 <sup>a</sup>	4.75±4.07 <sup>a</sup>	11.26±8.81 <sup>a</sup>	5.72±8.12 <sup>a</sup>	2.57±1.92 <sup>a</sup>	0.009
V	0.04±0.03 <sup>b</sup>	0.09±0.07 <sup>b</sup>	0.18±0.16 <sup>b</sup>	0.07±0.07 <sup>b</sup>	0.42±0.34 <sup>a</sup>	0.000

<b>Mg</b>	1.30± 1.02 <sup>b</sup>	15.09±14.84 <sup>ab</sup>	20.86±16.90 <sup>a</sup>	9.08± 11.01 <sup>ab</sup>	16.85±13.99 <sup>ab</sup>	0.023
<b>Mn</b>	1.47±1.14 <sup>bc</sup>	4.05±2.70 <sup>a</sup>	0.87±0.74 <sup>c</sup>	0.88±0.83 <sup>c</sup>	3.62±2.78 <sup>ab</sup>	0.001
<b>Na</b>	0.35±0.20 <sup>a</sup>	0.68±0.52 <sup>a</sup>	0.76±0.60 <sup>a</sup>	0.33±0.23 <sup>a</sup>	0.51±0.34 <sup>a</sup>	0.115

Values are mean ± SD; Values with similar alphabets were not significantly different, with  $p > 0.05$  based on Tukey HSD test. Note: LP=Losou Podi; LM=Losou Minunsud; SS=Sayap Substation; LA=Langanan; GA=Gansurai; AL=Aluminium; Ca=Calcium; Fe=Ferum; K=Potassium; V=Vanadium; Mg=Magnesium; Mn=Manganese; Na=Sodium.

### *Relationship between soil physical characteristics and Rafflesia's host (Tetrastigma)*

The principal components, PC-1 and PC-2 contributed about 84.29% of the total variance in the data (Table 3). The first principal component, as given in Table 3, had variance (Eigenvalue) of 2.55 and accounts for 51.08% of the total variance. PC1 was contributed from soil moisture, sand, silt and SOM. The second principal component had variance of 1.66 and accounted for 33.21% of the data variability. PC2 is influenced by all five variables. However, clay is the stronger contributor to this PC2 (Table 4).

**Table 3.** Summary of the Eigenvalues for soil physical.

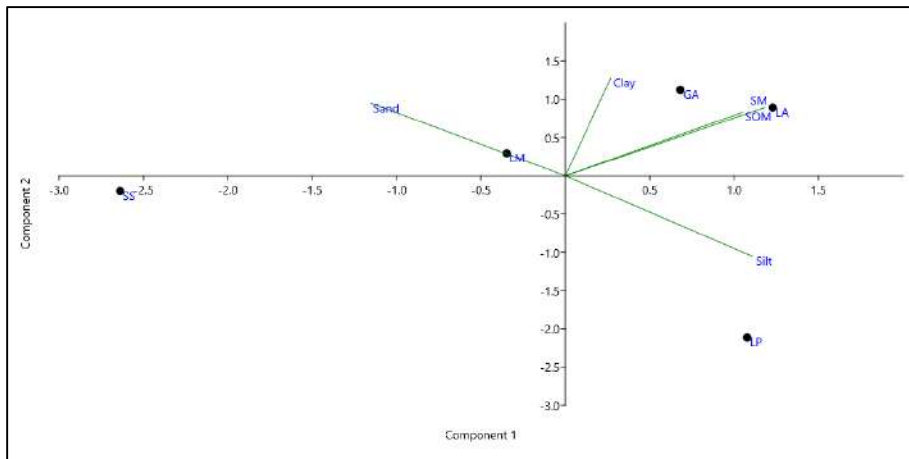
PC	Eigenvalue	% variance	Cum. % Var.
1	2.55	51.08	51.08
2	1.66	33.21	84.29
3	0.72	14.33	98.61
4	0.07	1.39	100.00

**Table 4.** Correlation matrix of PCA for soil physical.

	PC 1	PC 2	PC 3	PC 4
Soil Moisture (%)	0.52	0.39	-0.13	-0.75
Sand (%)	-0.51	0.42	-0.26	-0.09
Silt (%)	0.49	-0.47	0.18	0.07
Clay (%)	0.12	0.56	0.78	0.25
Soil organic matter (%)	0.46	0.37	-0.53	0.61

In Figure 6, it was observed that the habitat of *Rafflesia* was not affected by a single factor; with three main groups observable from the PCA analysis. Both Langanan and Gansurai habitats which were located near each other, were strongly influenced by soil organic matter and soil. Losou Podi exhibited a strong interrelation with silt. The third group showed that Losou Minunsud was strongly interrelated with sand in the habitat. The results obtained in this study were similar in Nasihah (2016) and (Nur Hayati et al., 2021) by having sand and silt in the same group. However, Sayap Substation was observed to have no relation

with any soil physical characteristics. Soil moisture in *Rafflesia*'s habitat has a strong positive correlation with SOM values, similar to Sumarno et al. (2009) and Tangketasik et al. (2012), whereby the percentage of sand, clay, and silt, and the amount of SOM affects the soil moisture in a habitat.



**Figure 6:** PCA of soil physical characteristics in *Rafflesia* habitat at Kinabalu Park, Sabah. (LP=Losou Podi; LM=Losou Minunsud; SS=Sayap Substation; LA=Langanan; GA=Gansurai; SM=Soil Moisture; SOM=Soil Organic Matter).

#### *Relation between pH and chemistry in soil and Rafflesia's host (Tetrastigma)*

The principal components, PC-1 and PC-2 contributed approximately 86.88% of the total variance in the data (Table 5). Table 5 shows the first principal component had a variance (Eigenvalue) of 4.94 and accounted for 54.86% of the total variance. PC1 was contributed by pH, Al, Ca, Mg and Na. The variance of the second principal component was 2.88, accounting for 32.02% of the data variability. PC2 was contributed by pH, Al, Fe, K, V and Mn (Table 6).

**Table 5:** Summary of the Eigenvalues for soil chemicals.

PC	Eigenvalue	% variance	Sum. % Var.
1	4.94	54.86	54.86
2	2.88	32.02	86.88
3	1.02	11.36	98.24
4	0.16	1.76	100.00

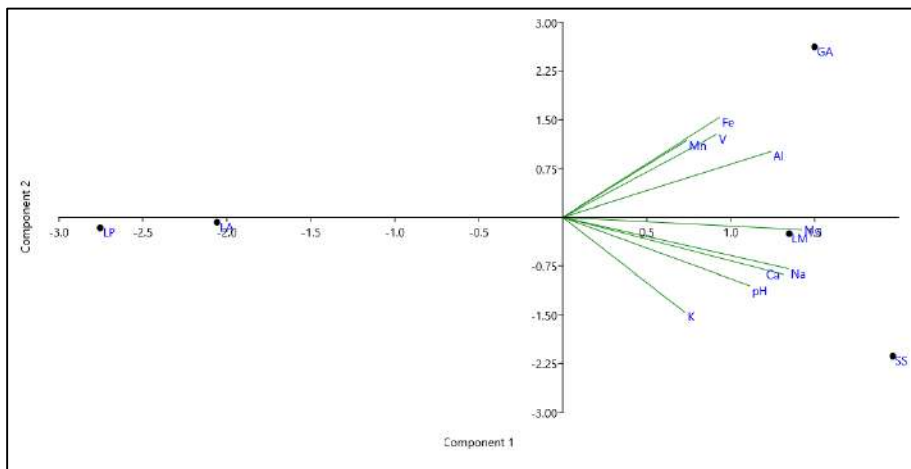
The results of this analysis indicated two main groups. Langanan and Losou Podi were observed to have no interrelation with any minerals found in the soil of the study area. Meanwhile, Gansurai had high positive interrelation with the elements Fe, V, Mn and Al. Sayap Substation did not exhibit any interrelation to



the physical characteristics of the soil, but had strong interrelations with minerals (K, Ca, Na, and Mg) and pH was observed in Figure 7. This second group also showed that Losou Minunsud had a direct contact with Mg. Sellan et al. (2019) explained that the different mineral content and pH found in soil can influence the growth of a species in its habitat. However, it was clear that the habitat of *Rafflesia* was not influenced by single specific factor, as observed in Nur Hayati et al. (2021).

**Table 6.** Correlation matrix of PCA for soil physical.

	PC 1	PC 2	PC 3	PC 4
pH	0.33	-0.32	0.38	-0.22
Al	0.37	0.30	-0.21	0.20
Ca	0.39	-0.26	0.14	-0.30
Fe	0.28	0.46	-0.03	0.13
K	0.22	-0.44	-0.43	0.36
V	0.27	0.38	-0.37	-0.64
Mg	0.43	-0.06	-0.26	0.36
Mn	0.22	0.36	0.61	0.35
Na	0.40	-0.24	0.18	-0.13



**Figure 7:** PCA of soil chemical elements in *Rafflesia* habitat at Kinabalu Park, Sabah. (LP=Losou Podi; LM=Losou Minunsud; SS=Sayap Substation; LA=Langanan; GA=Gansurai).

## Conclusion

This study revealed that the habitat of *Rafflesia* host i.e., *Tetrastigma* in Kinabalu Park, Sabah is interrelated to soil conditions with a high percentage of sand content compared to silt and clay elements. The soil type is sandy loam

with a high soil moisture content. In comparison, the SOM readings in the study area were very low, indicating that the soil in this habitat is less fertile.

We also discovered that soils in the *Rafflesia* habitat in Kinabalu Park have high levels of Al, Fe and Mg content. Only Gansurai, Sayap Substation and Losou Minunsud have an interrelation with the chemical factor; especially the elements Fe, Mn, V, Mg and soil pH.

The soil characteristics of different habitats of *Rafflesia* sp. and *Tetrastigma* sp. are influenced by different factors such as the percentage of sand content in the soil, soil moisture, and the mineral content of the element Mg. This proves that the habitat for these flowers can be complex and not affected by a single common soil factor.

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## Short Notes

# **A note on the Diversity of Rare and Wild Fruits Species in Sungai Kangkawat RS, Imbak Canyon Conservation Area (ICCA), Tongod, Sabah, Malaysia**

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## **Abstract**

Rare and wild fruits diversity of Sungai Kangkawat Research Station, Imbak Canyon Conservation Area (ICCA), Tongod District, Sabah Malaysia was inventorised for flora composition assessment. A total of 34 species of rare and wild fruits species were identified along three trails, namely Kawang, Nepenthes and Pelajau. During this inventory trip, only a few species were fruiting due to unsynchronized fruiting season. The inventorised species consist of 21 important genera namely *Artocarpus*, *Alstonia*, *Aralidium*, *Baccaurea*, *Cissus*, *Cnestis*, *Durio*, *Diospyros*, *Dacryodes*, *Ficus*, *Garcinia*, *Girardinia*, *Goniothalamus*, *Horsfieldia*, *Lasianthus*, *Lepisanthes*, *Lithocarpus*, *Mangifera*, *Osmosium*, *Rhodomyrtus* and *Uvaria*. A total of 61 herbarium specimens from various wild species were also collected and prepared from the trails for safe deposition at the respective herbaria.

**Keywords:** Rare fruits, Wild fruits, Sungai Kangkawat, Imbak Canyon Conservation Area, Tongod, Sabah, Malaysia

## **Introduction**

Rare and wild fruits species diversity is an important component of Malaysia's biodiversity. Our warm and humid tropical climate stimulates and harbours vast diversity of these species in our tropical rainforests and it is estimated 370 species of rare and wild fruits species are still thriving in the forest fringes, homegardens and orchards with most of them in semi domesticated state without any significant attention given (Ong 2004; Rukayah 2004). In view of this situation, it is important to record and document the diversity and uses of these wild fruits species that can be a useful source of the genetic pool for

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horticultural fruits species as well for nutritional and phytochemistry analysis. One of the important sites that can be considered a natural Gene-bank for these wild fruits species is located in Sabah, Borneo - Imbak Canyon Conservation Area (ICCA), Tongod District. This site spans a newly gazetted Class I (Protection) Forest Reserve of pristine rainforests and is considered rich in plant biodiversity as well as home for many rare and endangered animal species (Chua & Suleiman 2015; Latiff & Sinun 2011). A plant inventory survey was carried out in Sungai Kangkawat Research Station from 29<sup>th</sup> September to 2<sup>nd</sup> October 2018, aiming to document the diversity of rare and wild fruits species and to enrich herbarium collections.

## Methodology

Sungai Kangkawat Research Station is one of the main conservation plots in Imbak Canyon Conservation Area (ICCA), Tongod District, Sabah, Borneo Malaysia. During this inventory, trails provided in Sungai Kangkawat Research Station were botanised and the diversity of rare and wild fruits species that are available in this plot were recorded. Three trails provided for research purpose in this inventory were tagged as Nepenthes, Kawang and Pelajau trail. The main objective of this inventory is to provide a checklist of the fruits diversity using Rapid Plant Assessment Technique in which the plant listing was recorded on the surveyed trails and important plant parts (morphological and fertile specimens) were collected for further evaluation. This technique covers 10m radius on the the right and left sides of the trails to ensure the full plant compositions of these trails were fully inventorised. Plant description were referred to Corner (1952); Soepadmo et al. (2014) & Turner (1995). Specimens are deposited in the MDI and BORH Herbarium and recorded in the Agrobiodiversity Information System (AgroBIS) database. The checklist contained vouchers, herbarium records and also sighted record.

## Results and Discussion

This inventory reported 34 species that are available along the provided trails in Sungai Kangkawat RS (Imbak Canyon Conservation Area), with frequency of two to four individuals of every species existing along trails. From this observation, ecology of those rare and wild fruits species were dominated by seedlings to medium-sized trees, that occurred with the mature fruit trees species. Table 1 shows the list of species observed, with the type of plant form. Systematic diversity of this survey showed the composition of the important rare and wild fruits species in this site consists of 21 genera in 20 families.

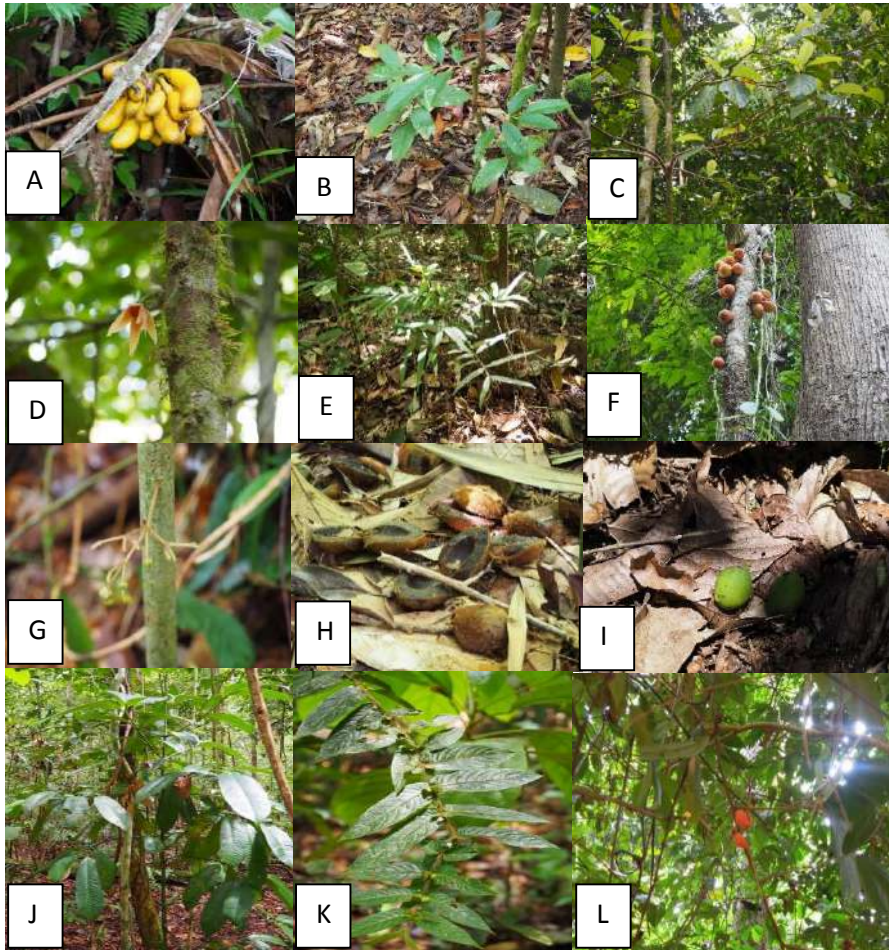


**Table 1.** List of species inventorised in three trails in Sungai Kangkawat Research Station.

Family	Species	Plant type
Actinidiaceae	<i>Actinidia</i> sp.	Tree
Anacardiaceae	<i>Mangifera caesia</i>	Tree
	<i>Mangifera</i> sp. 3	Tree
	<i>Mangifera</i> sp. 4	Tree
Annonaceae	<i>Uvaria borneensis</i>	Woody climber
	<i>Goniothalamus</i> sp.	Tree
Apocynaceae	<i>Alstonia</i> sp.	Tree
Torreelliaceae	<i>Aralidium pinnatifidum</i>	Tree
Malvaceae	<i>Durio griffithii</i>	Tree
	<i>Durio grandiflorus</i>	Tree
	<i>Durio graveolens</i>	Tree
	<i>Durio oxleyanus</i>	Tree
Burseraceae	<i>Dacryodes</i> sp.	Tree
Connaraceae	<i>Cnestis</i> sp.	Tree
Ebenaceae	<i>Diospyros lanceifolia</i>	Tree
Phyllanthaceae	<i>Baccaurea tetrandra</i>	Shrub - Tree
	<i>B. parviflora</i>	Tree
	<i>Baccaurea</i> sp. 2	Tree
Fagaceae	<i>Lithocarpus</i> sp.	Tree
Guttiferae	<i>Garcinia griffithii</i>	Tree
	<i>Garcinia</i> sp.	Tree
Leguminosae	<i>Osmosia</i> sp.	Tree
Meliaceae	<i>Aglia borneensis</i>	Tree
Moraceae	<i>Ficus punctata</i>	Tree
	<i>Artocarpus elasticus</i>	Tree
	<i>Artocarpus lowii</i>	Tree
	<i>Artocarpus</i> sp.	Tree
Myrtaceae	<i>Rhodomyrtus</i> sp.	Tree
Myristicaceae	<i>Horsefieldia</i> sp.	Tree
Rubiaceae	<i>Lasianthus borneensis</i>	Tree
Sapindaceae	<i>Lepisanthes alata</i>	Tree
	<i>L. tetraphylla</i>	Tree
	<i>L. amoena</i>	Tree
Ulmaceae	<i>Gironniera nervosa</i>	Tree
Vitaceae	<i>Cissus</i> sp.	Climber

Several interesting species that bear fruits such as *Uvaria borneensis* (Annonaceae) were also encountered during our survey. This *Uvaria borneensis* is locally known as pisang - pisang group is categorised as a woody climber is unique as out of 24 species and 9 genera recorded in Malaysia, 17 species (4 genera) are categorised as native to Sabah and Sarawak (Soepadmo et al. 2014). Other than that, it is clearly indicated from our records and sighted species that

the Anacardiaceae, Malvaceae, Rubiaceae and Moraceae formed the majority plant coverage of wild and rare fruits family represented by 34 species in 21 genera.



**Figure 1.** Plate of rare and wild fruits species. A) *Uvaria borneensis*, B) *Durio griffithii*, C) *Baccaurea tetrandra* D) *Goniiothalamus* sp., E) *Lepisanthes alata*, F) *Ficus punctata*, G) *Actinidia* sp., H) *Horsfieldia* sp., I) *Mangifera* sp., J) *Garcinia griffithii*, K) *Lasianthus* sp., L) *Aglaia borneensis*

Important rare and wild fruits genera observed along the provided trails are *Artocarpus*, *Alstonia*, *Aralidium*, *Baccaurea*, *Cissus*, *Cnestis*, *Durio*, *Diospyros*, *Dacryodes*, *Ficus*, *Garcinia*, *Gironniera*, *Goniiothalamus*, *Horsfieldia*, *Lasianthus*, *Lepisanthes*, *Lithocarpus*, *Mangifera*, *Osmosium*, *Rhodomirtus* and *Uvaria*, this

is agreeable with the rich diversity of Borneo's wild fruits species that can be found on this island (Agriculture and Agrifood Department Brunei, 2017).

In conclusion, the diversity of rare and wild fruits of Imbak Canyon Conservation Area is diverse in genera and species for Bornean taxa. However, our studies have only covered the accessible trails in this conservation area, and it is suggested that further inventorisations should be conducted, critically during the forest's fruiting season to attain complete specimens and identification of its diversity.

## Conclusion

High diversity of the wild and rare fruits species was expected as 34 species of rare and wild fruits species were identified in the observed trails from the total area. Thus, further studies about the diversity in this area should be conducted. It is suggested the site's present condition is preserved with more emphasis and constructive efforts for conservation of Imbak Canyon's ecological habitats to ensure the diversity of these rare and wild fruits species can be conserved with their natural elements.

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## Research Article

# The Non-Volant Terrestrial Small Mammals at Ulu Muda Forest Reserve, Kedah, Malaysia

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## Abstract

An eight days survey of non-volant small mammals was conducted at the Ulu Muda Forest Reserve in the state of Kedah, Malaysia. To sample the small mammals, we placed 100 cage traps and 50 bucket pitfall traps randomly along the existing man-made trails in four sampling sites within the study area. Total trapping effort for the cage traps was 796 trap nights, while the total trapping effort for the bucket pitfall traps was 400 trap nights. Overall, 24 non-volant small mammal individuals represented by seven species from the Family Muridae were captured. The Red spiny rat, *Maxomys surifer*, was the most dominant species accounting for 42% of the total individuals captured. We also caught the Chestnut white-bellied rat (*Niviventer fulvescens*) which is a new record for the study area. Although sampling was conducted only briefly, our study has demonstrated that Ulu Muda Forest Reserve still holds a high diversity of forest rat species, some of which are of international or regional conservation concern. Increasing the sampling effort, i.e., by surveying more areas over a longer period, would likely increase the possibility of capturing more small mammal species in this area.

**Keywords:** Rats, Ulu Muda Forest Reserve, species diversity, *Niviventer fulvescens*.

## Introduction

Ulu Muda is recognized as the largest lowland dipterocarp rainforest in the northern part of Peninsular Malaysia (Bashir Ali, 2014). The geology of Ulu Muda is composed of hills of 300 meters (m) with moderately high peaks and lowland area of less than 200m above sea level, and this area covers 162,931 hectares (Suksuwan, 2008). In addition to providing important habitats for many wildlife species, the Ulu Muda area plays an important role as a water catchment for Muda Lake, Peru Lake and Ahning Lake. In terms of forest types, Ulu Muda is covered by hill dipterocarp, and limestone vegetation (Marden et al., 2013). This location is also unique for having many natural saltlicks that are an important

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source of essential minerals for large mammal species (Stevens, 1968; Matsubayashi et al., 2007; Bashir Ali, 2014). Based on camera trapping methods, the World Wide Fund for Nature recorded 31 species of mammals at saltlick sites in Ulu Muda (Bashir Ali, 2014).

Since over 10 years ago,, the Ulu Muda Forest Reserve has recorded approximately 112 mammalian species which account for 50% of mammalian species found throughout Peninsular Malaysia (DWNP, 1993; WWF-Malaysia, 2002; Mariana et al., 2005; Sharma et al., 2005; Shukor et al., 2005; DWNP, 2010). The animals reported here include charismatic species such as the Asian elephant (*Elephas maximus*), Malayan tapir (*Tapirus indicus*) and Sambar deer (*Rusa unicolor*). Additionally, the Hairy-nosed otter (*Lutra sumatrana*), which is an endangered otter species usually found in a peat swamp forest, has also been recorded in Ulu Muda (Hubback, 1932; Bashir Ali, 2014; Salahshour, 2016).

Although efforts to gazette Ulu Muda as a protected area started since 1930, the official gazettement of this area has yet to be made to date (Elagupillay, 1987; Stevens, 1968; DWNP, 1989; MOCAT, 1996; Bernama, 2009). While Ulu Muda's large mammalian faunal has been extensively investigated in the past, few studies concerning small mammals have been conducted (Shukor et al., 2005; Sharma et al., 2005; Bashir Ali, 2014). This has created a gap in the latest information about Ulu Muda's overall faunal diversity.

In this study, we conducted a survey focusing on the non-volant small mammal fauna of Ulu Muda Forest Reserve. Non-volant small mammals refer to any non-flying mammals whose adult live weight in the wild do not exceed five kilogrammes (kg; Hayward & Phillipson, 1979; Barnett & Dutton, 1995) such as rats, shrews, gymnures and treeshrews. The latest small mammal survey using camera trapping in Ulu Muda by (Bashir Ali, 2014) recorded only two non-volant species of rodent, i.e., the Long-tailed giant rat (*Leopoldamys sabanus*) and Malayan porcupine (*Hystrix brachyura*). Camera trapping method is not ideal for studying small mammals because species identity is not easily known from the photographs captured (Bernard et al., 2013). Thus, we conducted a survey of the small mammals using live-cage traps and bucket pitfall traps with the main aim to provide an updated checklist of the non-volant small mammal community in Ulu Muda Forest Reserve. We also gathered information concerning their distribution, regional or local conservation status and morphological characteristics to enrich the scientific data for this animal group.

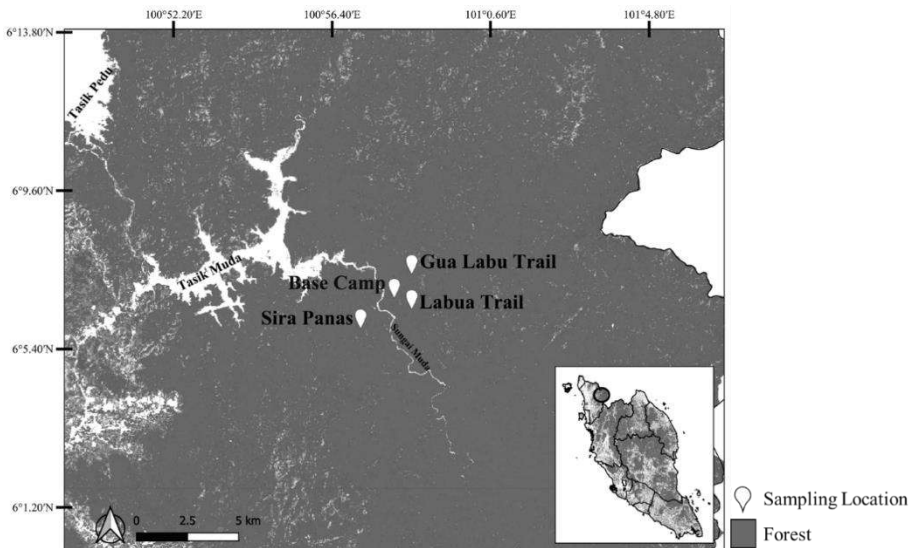
## Methodology

### *Study area*

Ulu Muda consists of seven permanent reserve forests (PRF) namely Bukit Keramat Forest Reserve (FR), Padang Terap FR, Pedu FR, Chebar Besar FR, Chebar Kecil FR, Bukit Saiong FR and Ulu Muda FR (Bashir Ali, 2014). In this study, we surveyed the distribution of non-volant small mammals in Ulu Muda Forest Reserve (UMFR) with GPS coordinate N 6° 07.536 E 100° 58.146 (Figure 1). Four sampling sites were selected in this study (1) base camp (N 06° 06.888' E 100° 57.787'), (2) Labua (N 06° 06.833' E 100° 57.784'), (3) Gua Labu (N 06° 07.026' E 100° 58.114') and (4) Sira Panas (N 06° 06.482' E 100° 57.509'). These sites are located near the Muda River and covered by secondary forests with an elevation range between 100m to 150m. Sampling was conducted from 6<sup>th</sup> to 14<sup>th</sup> December, 2019. Sira Panas is one of the many natural saltlicks in Ulu Muda Forest Reserve. Other natural saltlicks around this forest reserve that are known to us are Sira Bongor and Sira Air Hangat.

### *Taxon sampling*

One hundred cage traps with size of 28cm x 15cm x 12cm each were set up at all sampling sites (25 cage traps at each site) along the existing man-made trails. All cage traps were baited with either cut ripe banana coated with peanut butter, oil palm kernels, salted or fresh fishes. In addition, 50 buckets (height = 25cm, top diameter = 21cm with 10 small holes at the bottom to allow water



**Figure 1.** Map showing the four sampling sites, namely Gua Labu trail, Labua trail, base camp, and Sira Panas in the Ulu Muda Forest Reserve.

drainage) were buried without bait where their openings were the same level as the forest floor. Both cage traps and buckets were deployed between 10 to 50m apart from each other. All traps were active for eight consecutive days and nights and were checked for animals trapped once a day in the morning between 7:00 hrs - 11:00 hrs. All traps were re-baited with new baits during the daily check. The identity of the trapped animals was determined using several field guide books i.e., Francis (2008), Aplin et al. (2003) and Herbreteau et al. (2011). All trapped animals were euthanized using Zoletil® 50 following the standards widely used by field researchers (Rivas et al., 2015; Ishak et al., 2018). All tissue samples from the liver were collected and preserved in 90% ethanol for DNA analysis. All samples were labeled and preserved in 70% ethanol as wet specimens (Tingga et al., 2012) and were stored in the Museum of Zoology, Universiti Malaya.

#### *Data analyses*

We analyzed the diversity indices of rats using Paleontological Statistics software (PAST) version 3.26 (Hammer et al., 2001). The diversity indices include Simpson diversity index ( $1-D$ ), Shannon-Wiener diversity index ( $H$ ), Buzas and Gibson's evenness ( $E$ ) and Margalef's richness index ( $D_{Mg}$ ) (Mohd-Taib et al., 2019; Tu et al., 2020). We compared the results of our survey with earlier records based on studies conducted in 2008 by Mariana et al. (2008) in the same locality (see Table 1).

To construct the species accumulation curve, the cumulative number of species from all the sampling sites were calculated using PAST software. The cumulative number of species was used against the sampling efforts which is given by the sampling days, to draw the accumulation curve. The rarefaction curve of the total individuals non-volant small mammals captured from Ulu Muda Forest Reserve, Kedah was also constructed using the same software, to access the species richness from the sampling results.

## **Results and Discussion**

A total of seven species of murid rodents have been recorded in Ulu Muda Forest Reserve during our survey based on an overall of 24 small mammal individuals caught. We did not capture other groups of rodents as recorded by Bashir Ali (2014) using the same sampling methods as our survey. This is most likely due to the brief period of our survey (8 days only). Nevertheless, we have sighted several arboreal species from the genus Tupaiidae or Scuriidae, although we could not confirm the species identity. The most frequently caught species in



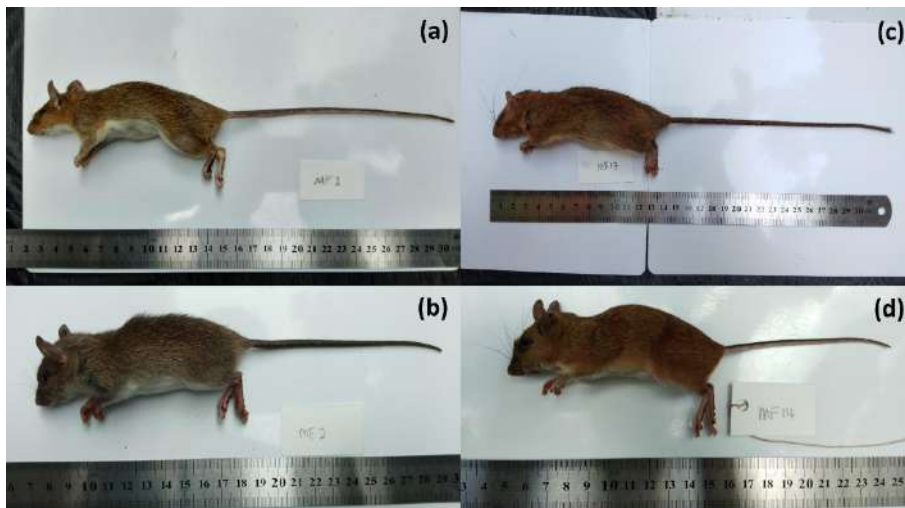
this survey was the Red spiny rat (*Maxomys surifer*) followed by Whitehead's spiny rat (*Maxomys whiteheadi*) with 10 and 6 individuals caught, respectively (Table 1). *M. surifer* is one of the common murids found in the lowland dipterocarp forest (Francis, 2008). This species is quite similar to *M. whiteheadi* (Figure 2) based on its morphology, especially the bicolored tail. *M. whiteheadi* is also the only captured species listed as Vulnerable by the IUCN Red List (IUCN, 2020; refer Appendix 1 for other species status). During this survey, we were unsuccessful in trapping Rajah spiny rat (*Maxomys rajah*). This might be due to its habitat, which possibly occurs in a drier area than the *M. surifer*. These two species are rarely found in the same locality (Francis, 2008). Two singleton species were represented by Muller's giant Sunda rat (*Sundamys muelleri*) and Indomalayan Pencil-tailed tree mouse (*Chiropodomys gliroides*). Two individuals of *Niviventer* species were also captured, which are Dark-tailed tree rat (*Niviventer cremoriventer*) and Chestnut white-bellied rat (*N. fulvescens*). The latter is a new record in Ulu Muda Forest Reserve.

**Table 1.** Taxonomic list and comparison with previous survey of non-volant small mammals collected from a survey at Ulu Muda Forest Reserve in Kedah state.

Family Species	In this study	Mariana et al. (2008)
Muridae		
<i>Maxomys surifer</i>	10	1
<i>Maxomys whiteheadi</i>	6	7
<i>Maxomys rajah</i>	0	2
<i>Niviventer cremoriventer</i>	2	0
<i>Niviventer fulvescens</i>	2	0
<i>Rattus tiomanicus</i>	2	0
<i>Sundamys muelleri</i>	1	6
<i>Chiropodomys gliroides</i>	1	0
<i>Leopoldamys sabanus</i>	0	4
Tupaiaidae		
<i>Tupaia glis</i>	0	1
<b>Total individuals</b>	<b>24</b>	<b>21</b>
<b>Total families</b>	<b>1</b>	<b>2</b>
<b>Total species</b>	<b>7</b>	<b>6</b>

There are three species of *Niviventer* that can be found in Peninsular Malaysia consisting of *N. cremoriventer*, *N. cameroni* and *N. fulvescens* (Figure 2). Out of three, only *N. cameroni* has a distinct distribution where it lived at an elevation of more than 1000m and was recorded in Cameron Highlands, Pahang (Francis, 2008). The other two species of *Niviventer* are distributed in forests throughout Peninsular Malaysia (Francis, 2008).

Based on our observation, except the body sizes and length of tails, their morphologies are quite similar to the *M. whiteheadi* and by coincidence they shared the same habitat. This might lead to species misidentification, especially with the juvenile *N. fulvescens*.



**Figure 2.** Photos of rats collected: a) *Niviventer fulvescens*, b) *Rattus tiomanicus*, c) *Niviventer cremoriventer* d) *Maxomys whiteheadi*

As compared with the previous study by Mariana et al. (2008), our results have shown slight changes in murid abundance as in Table 1. For example, Mariana et al. (2008) managed to trap six individuals of *S. muelleri* while we only found a single individual of this species which was commonly present near water sources such as streams and rivers. Same goes with the Long-tailed giant rat (*Leopoldamys sabanus*) and Common treeshrew (*Tupaia glis*) which are known to be common and widely distributed species. We did not expect to get the same result as Mariana et al. (2008), but to see the changes in diversity at the same locality. The absence of some species may be due to disturbance in the sampling sites that chase away most of the small mammals. The assumption is based on scattered animal dung especially in Labua and Sira Panas indicating the presence of medium to large mammals. Besides that, our sampling efforts are quite low as we divided the cage and pitfall traps to each sampling site. The disturbance and possible food scarcity for small mammals may force the animals to go deeper into the forests and our coverage is not enough to cover them.

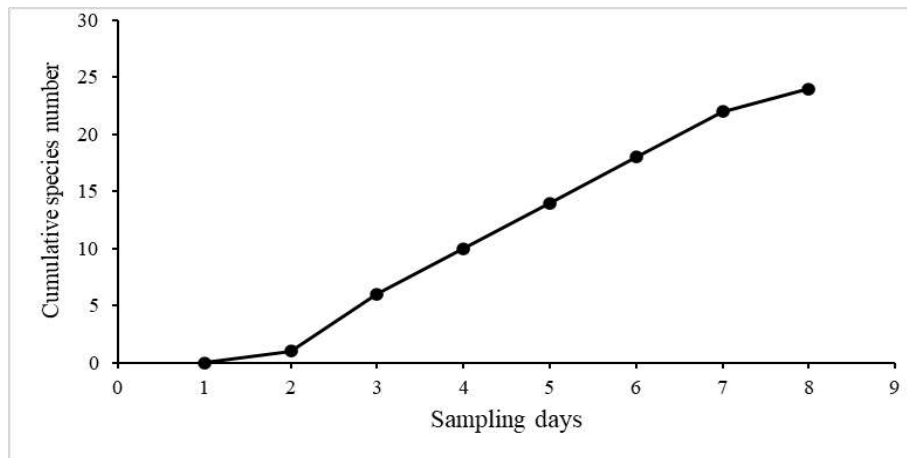
Our results showed rather moderate (Simpson's diversity index) and low values of diversity indices (Table 2), indicating a moderate species richness of small

mammals in the sampling sites and the number of individuals is not distributed evenly between the captured species. This could be seen based on the highest and lowest number of captured species. Based on our observation, there is no deforestation or new development in our study sites. The nearby nature lodge (Earth Lodge) is the only eco-tourism provider in the forest (Bashir Ali, 2014) and during our trip, no tourists were spotted there, except for our research team and MNS staff.

**Table 2.** The species diversity of non-volant small mammals analyzed in this study.

Sample analyses	This study
No. of species	7
No. of individuals	24
Simpson diversity index ( $1-D$ )	0.740
Shannon-Wiener diversity index ( $H$ )	1.597
Buzas and Gibson's evenness ( $E$ )	0.706
Margalef's richness index ( $D_{Mg}$ )	1.888

The species accumulation curve was constructed to depict the dependence of the cumulative number of species captured from the sampling sites on a variable representing sampling effort which is sampling days (Figure 3).

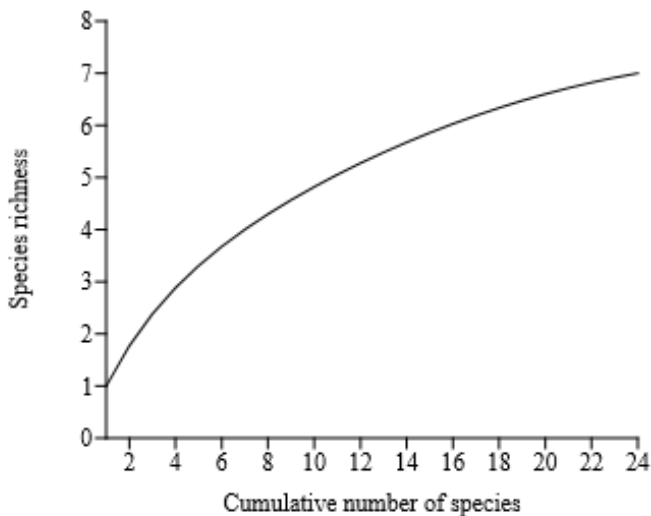


**Figure 3.** Species accumulation curve showing the cumulative species number against the sampling days (8 days of sampling period) from Ulu Muda Forest Reserve, Kedah.

The figure also shows that the curve almost achieves the horizontal asymptote of the species richness. Extended period of samplings at Ulu Muda Forest Reserve, Kedah would increase the species richness of the non-volant small

mammals captured and the species accumulation curve will probably achieve asymptote.

According to Colwell (2009), rarefaction curves are useful for comparing species richness among populations in the inventoried or not inventoried sampling sites with inadequate effort. Therefore, the rarefaction curve was constructed based on the species richness against the cumulative number of species (Figure 4). The figure shows the accumulation curve has almost reached asymptote.



**Figure 4.** Rarefaction curve of the total individuals' non-volant small mammals captured from Ulu Muda Forest Reserve, Kedah.

Based on Colwell et al. (2012), the rarefaction curve is usually used to estimate the complete richness of the samples by visualizing it as the asymptote of the accumulation curve. In this study, it can be said that the sampling efforts are suitable and enough to estimate the total richness of the collected samples. However, any additional sampling efforts are still needed to increase species richness of non-volant small mammals captured from the sampling sites so that the accumulation curve can reach asymptote.

## Conclusion

The few numbers of individuals and low number of non-volant small mammal species caught were most likely due to our short sampling duration, sampling efforts and less abundance of them in our sampling sites. However, this survey provides an updated checklist on the species occurrence at Ulu Muda Forest Reserve. Future surveys should be conducted over a longer period using a larger number of traps (increase sampling efforts) to get better results.

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**Appendix 1.** Taxonomic list and descriptive statistical measurements of non-volant small mammals collected from a survey at Ulu Muda Forest Reserve in Kedah state. Sample size is noted as (n) and its mean  $\pm$  standard deviation and range (in parenthesis) were displayed too. All measurements are in millimeters (mm) and weight in grams (g). Sexes and ages, ranging from juvenile to adult are combined.

Family	Species	Common name	n/sex	Head and body length (HB)	Tail length (TL)	Hind foot length without claw (HF)	Ear length (E)	Weight (W)	IUC N Stat us
Muridae	<i>Maxomys surifer</i>	Red spiny rat	5M, 5F	164.0 $\pm$ 19.0 (140 - 199)	173.4 $\pm$ 19.0 (149 - 206)	37.6 $\pm$ 1.6 (36 - 41)	23.5 $\pm$ 2.6 (21 - 28)	107.6 $\pm$ 41.0 (50 - 182)	LC
	<i>Maxomys whiteheadi</i>	Whitehead's spiny rat	5M, 1F	110.5 $\pm$ 6.6 (101 - 119)	105.5 $\pm$ 6.0 (100 - 116)	26.4 $\pm$ 1.9 (25 - 30)	16.4 $\pm$ 0.9 (15 - 18)	37.5 $\pm$ 7.2 (29 - 50)	VU
	<i>Niviventer cremoriventer</i>	Dark-tailed tree rat	2M	124.0 $\pm$ 24.0 (107 - 141)	176.5 $\pm$ 312 (154 - 199)	25.5 $\pm$ 0.7 (25 - 26)	18.3 $\pm$ 1.1 (18 - 19)	51.0 $\pm$ 24.0 (34 - 68)	LC
	<i>Niviventer fulvescens</i>	Chestnut white-bellied rat	1M, 1F	140	166.0 $\pm$ 12.7 (157 - 175)	23.5 $\pm$ 3.5 (21 - 26)	19.0 $\pm$ 1.4 (18 - 20)	67.0 $\pm$ 18.4 (54 - 80)	LC
	<i>Rattus tiomanicus</i>	Malayan field rat	2M	138.5 $\pm$ 34.6 (114 - 163)	136.0 $\pm$ 32.5 (113 - 159)	31.0 $\pm$ 2.8 (29 - 33)	17.5 $\pm$ 2.1 (16 - 19)	67.5 $\pm$ 37.5 (41 - 94)	LC
	<i>Sundamys muelleri</i>	Muller's giant Sunda rat	1F	212	267	46	23	307	LC
	<i>Chiropodomys gliroides</i>	Indomalayan Pencil-tailed tree mouse	1N/A	N/A	160	20	N/A	N/A	LC
		<b>Total individuals</b>	<b>24</b>						

\*M=male, F=female, N/A=Not Available, VU=Vulnerable, LC=Least Concern



## Research Article

# The Distribution and Abundance of Long-Tailed Macaques in the Main Campus of Universiti Malaysia Sabah and its Vicinity

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## Abstract

Long-tailed macaques (*Macaca fascicularis*) inhabit a wide range of natural and human-modified environments in Southeast Asia. Because of their ability to occupy the same space and utilise the same food resources as humans, long-tailed macaques have in some cases been regarded as nuisance and pest. As part of an effort to assess the status of human-macaque interactions in the main campus of Universiti Malaysia Sabah (UMS), we conducted a survey to determine the distribution range and estimate the abundance of the macaque population. We performed the survey monthly from April 2019 to March 2020 at 13 localities classified into three different habitat types i.e. secondary forest, forest edge and urban areas identified in the campus and its vicinity. The survey covered an overall area of 6.02 km<sup>2</sup>. We recorded the group or individual locations and the number of macaques with each individual's age/sex information whenever possible. The macaques consisted of three different multimale-multifemale groups and a solitary individual. We recorded a minimum population size of 54 macaque individuals with an average group size of 17.7 individuals and a population density of 8.97 individuals/km<sup>2</sup> in the surveyed areas. Macaques were encountered every month and in all habitat types defined in this study. However, they were mainly distributed at two habitat types i.e. urban areas and forest edges near Bukit UMS and the student residential colleges where anthropogenic influence appeared to be high. We speculate that the monthly availability of food resources, including anthropogenic food waste, may partly explain this distribution pattern. We suggest that further studies is conducted in this regard.

**Keywords:** *Macaca fascicularis*, distribution, abundance, management implications, human-macaque interactions, Universiti Malaysia Sabah.

## Introduction

Human-wildlife interactions are a growing concern worldwide (Dickman, 2010; Distefano, 2005; Soulsbury & White, 2016). To effectively manage the interactions, accurate knowledge on the distribution range and abundance of the animals is crucial (Dickman, 2010; Greggor et al., 2016; Redpath et al., 2013; Swan et al., 2017).

Across Southeast Asia's mainland and islands, the long-tailed macaque, *Macaca fascicularis*, is considered one of the most successful species of primates as they are widely distributed (Abegg & Thierry, 2002). Based on Mittermeier et al. (2013) and an updated checklist published in Roos et al. (2014), the species can be found in present-day Bangladesh, Myanmar, Laos, Thailand, Vietnam, Cambodia, Philippines, Malaysia, Singapore, Brunei, Indonesia and Nicobar Islands of India. There are currently at least 10 recognised subspecies, including *M. f. aureus* (Geoffroy, 1831), *M. f. philippinensis* (Geoffroy, 1843), *M. f. tua* (Kellogg, 1944), *M. f. atriceps* (Kloss, 1919), *M. f. condorensis* (Kloss, 1926), *M. f. lasiae* (Lyon, 1916), *M. f. umbrosus* (Miller, 1902), *M. f. fuscus* (Miller, 1903), *M. f. fascicularis* (Raffles, 1821) and *M. f. karimondjawa* (Sody, 1949).

In Malaysia, long-tailed macaques have been reported in Peninsular Malaysia and the states of Sabah and Sarawak on Borneo Island (Roos et al., 2014). Similar to the distribution reported in India, Indonesia, Mauritius, Singapore and Thailand, they are commonly found in riparian zones, coastal forests, mangrove forests, low elevation secondary forests, forest periphery near villages, agricultural plantations and urban/suburban areas, including recreational parks and other tourist attractions (Eudey, 2008; Fooden, 1995; Fuentes et al., 2011; Gumert, 2011; Gumert et al., 2011; Gumert et al., 2012; Hambali et al., 2012; Hansen et al., 2019; Jamhuri et al., 2018; Kurland, 1973; Malaivijitnond & Hamada, 2008; Otani et al., 2020; PERHILITAN, 2018; Riley et al., 2015; Saaban et al., 2014; Sha et al., 2009; Southwick & Cadigan, 1972; Sussman & Tattersall, 1986; Tee et al., 2019; Umapathy et al., 2003). This is most likely due to the availability and easy access to resources, especially food provisioning by humans, whether directly or indirectly, that leads to adaptation and adjustment of their behaviour to live near anthropogenic influences and, hence, interaction with humans. Indeed, on a larger scale in Peninsular Malaysia, 56,786 complaints or 66% of all reported human-wildlife conflict complaints from 2006 to 2015 involved long-tailed macaques (Saaban et al., 2016). Similarly, in 2016, the long-tailed macaque was the most

reported species, with 4,237 complaints or 63% of 6,769 complaints recorded (DWNP, 2016).

Small scale studies have been conducted on some populations in urban areas, particularly university campuses and nature parks in Peninsular Malaysia. Findings reported nuisance behaviour such as foraging at waste areas, littering, entering buildings and displaying aggression towards humans (Hambali et al., 2012; Md-Zain et al., 2011). To manage these issues, the Department of Wildlife and National Parks (DWNP) in Peninsular Malaysia have used various approaches, including culling and trapping of individual macaques (DWNP, 2016; Lappan & Ruppert, 2019; Saaban et al., 2016). Campaigns and awareness programs were also launched, resulting in the setting up of educational signages to improve public education about macaque behaviour, the introduction of macaque-proof bins and the development of manuals and action plans for managing human-macaque conflicts (Saaban et al., 2016). However, these actions' effectiveness, impacts and sustainability on managing human-macaque conflicts, in the long run, are unknown and will urgently require further assessments. Given the increasing proximity of the long-tailed macaque populations to anthropogenic activities and environments, this species is expected to remain among the top most reported species in human-wildlife conflicts in Malaysia in the future.

Long-tailed macaques inhabiting the Universiti Malaysia Sabah's (UMS) main campus are no exception; students and staff have complained reported to the student councils and the UMS management, particularly the management of student residence, regarding the nuisance behaviour of macaques. To date, however, there are no comprehensive studies conducted on the long-tailed macaque population in the campus and its vicinity. Therefore, the present study was conducted to determine the distribution range and estimate the abundance of the macaque population as an initial effort to assess the status of human-macaque interactions in and around the campus area. In this article, we report the long-tailed macaque distribution and abundance.

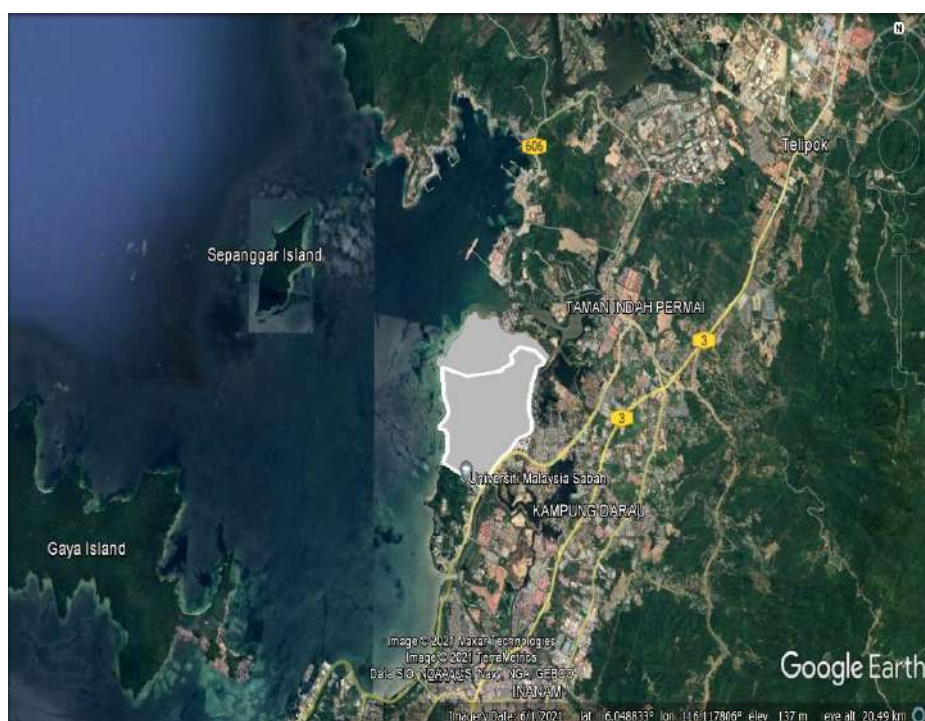
## **Methodology**

### *Study Area*

Our study area is located at Sepanggar Bay in Kota Kinabalu, Sabah, Malaysia (Figure 1). For nearly three decades until the present time, the bay has been developed as part of an urban expansion of the metropolitan area of Kota Kinabalu. Some of the most notable landmarks include the main campus of

Universiti Malaysia Sabah (UMS), Sabah Federal Government Administrative Complex, Sabah State Legislative Assembly building, residential areas and various supporting infrastructure.

Our main survey sites were located at the centre of Sepanggar Bay, where the main campus of UMS ( $6.036276^{\circ}$ ,  $116.115700^{\circ}$ ) was established in 1999. The study covered a total area of  $6.02\text{km}^2$ , with  $4.22\text{ km}^2$  inside the border of UMS campus, whereas the remaining  $1.80\text{ km}^2$  is outside of the campus area (Figure 1). Secondary forest, coastal forest, forest edge, residential and commercial buildings, roads and other urban structures make up the general landscape of the study site (Figure 2). Most of the infrastructural development took place in the southern and eastern parts of the campus. In contrast, the northern part consists of an isolated secondary forest fragment occupying the Outdoor Development Centre (ODEC) ( $6.042845^{\circ}$ ,  $116.112109^{\circ}$ ) near the coast and the highest peak, Bukit UMS ( $6.042426^{\circ}$ ,  $116.119485^{\circ}$ ) at 205m a.s.l.



**Figure 1.** Map of study area and study site in the main campus of UMS and its vicinity, Sepanggar Bay, Kota Kinabalu, where  $4.22\text{km}^2$  of the study site is in the shaded part bordered with the white line while  $1.80\text{km}^2$  is the shaded part next to it (Google Earth, 2020).

This study defined secondary forest as a continuous area of secondary vegetation, including some planted tree species. We defined forest edges as strips of vegetation that border the secondary forest between 50 - 100m in width, whereas urban areas were defined as built environments contiguous with the forest edges for human settlements and other activities. Based on secondary information on localities where macaques were frequently encountered in UMS, these areas were further divided into 13 localities to be surveyed for long-tailed macaques (Figure 2). These localities were Forest Zone 1 (FZ1), Forest Zone 2 (FZ2) and Forest Zone 3 (FZ3), representing the secondary forest habitat type. Edge Zone 1 (EZ1), Edge Zone 2 (EZ2) and Edge Zone 3 (EZ3) represented the forest edge habitat type, while Kolej Kediaman Excellent (KKE), Kolej Kediaman Tun Fuad (KKTF), Kolej Kediaman Tun Mustapha (KKTM), Institute for Tropical Biology and Conservation (ITBC), Chancellery, Jalan Samudera and non-UMS areas represented the urban area habitat type.

#### *Data Collection*

We used the general physical characteristics and age-sex classification of long-tailed macaque described by Fooden (1995; 1997) and Poirier & Smith (1974) to identify the individual macaques and distinguish the different macaque groups in this study (Table 1).

Systematic surveys were conducted monthly from April 2019 to March 2020 by walking along existing trails and other pathways at the 13 localities in the secondary forest, forest edge and urban areas. Long-tailed macaques were detected using direct sighting, vocalisation, movement sound or the presence of their faeces or food waste on the ground. When a group or an individual macaque was sighted, we (YYZ and assistant) recorded the location coordinates using a handheld GPS receiver. The number of visible individuals were counted, aided by a pair of binoculars and followed as much as possible to identify the group-type or solitary individual. To assist in group recognition, the key individuals were noted for their unique features. Other distinguishing characteristics of the group, such as the number of mothers with an infant, were also noted. The age-sex class of each individual was identified if possible; otherwise, they were marked as unknown. The group type was recorded as either multimale-multifemale group or all-male group. Observations of a single adult male individual were categorised as solitary male. In addition, other opportunistic observations of macaque groups or individuals outside of the monthly systematic surveys were also recorded, including secondary sightings by UMS students and staff.

**Table 1.** Morphological traits of each age-sex class for the identification of *Macaca fascicularis*.

Age-sex class	Morphological traits for identification
Adult male	Adult male has long and narrow rostrum, large canine, fully developed scrotum and descended testicles. Compared to adult female, length of pelage is longer and body size is bigger. Length of head, body and skull are also greater.
Adult female	Adult female has shorter pelage and smaller body size. Length of head, body and skull are shorter. The sexual skin of pregnant female may swell or redden and adult female that had previously given birth has elongated nipples.
Subadult	Body size of subadult is smaller than that of adult.
Juvenile	Body size of juvenile is smaller than that of subadult and adult. It also has greater relative tail length, hind foot and ear length.
Infant	Infants less than 3 months old have blackish dorsal pelage and bare, pinkish, unpigmented facial skin. Until it is approximately 3 months old, the dorsal pelage turns grayish or brownish in colour. Deciduous canines and first molars will start to grow as it learns to obtain food independently.

### Data Analysis

The group size of each macaque group was estimated from the maximum number of visible individuals recorded during any one of the monthly surveys, while the overall population size was estimated from the sum of the maximum group size of each identified group and the solitary individuals. The age-sex composition was estimated based on the number of adult male (AM), adult female (AF), subadult (SA), juvenile (Juv) and infant (Inf).

Monthly macaque encounter rates were computed based on the following formulas:

$$X = \frac{\sum (n_x + n_y + n_z + \dots n_i)}{N}$$

Where,  $X$  is the mean frequency of encounters at all localities in month  $y$  i.e.

$\left[ \frac{\left( \frac{\text{encounter}}{\text{sample day}} \right)}{\text{surveyed locality}} \right]$ ,  $n_x$  is the mean frequency of encounters at locality  $x$  in month  $y$

i.e.  $\left( \frac{\text{encounter}}{\text{sample day}} \right)$ , and  $N$  is the number of surveyed localities in month  $y$ .

$n_x$  was calculated based on the formula:

$$n_x = \frac{\sum (n_1 + n_2 + n_3 + \dots n_i)}{N}$$

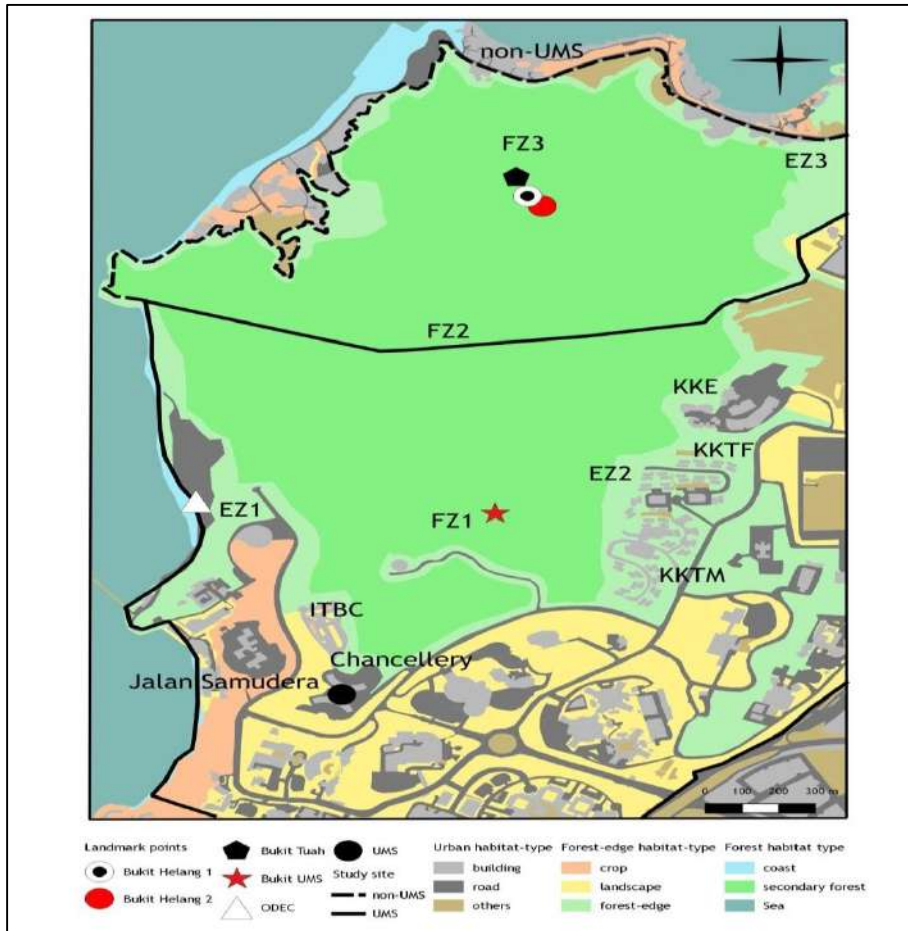
Where,  $n_i$  is the frequency of encounter in sample day  $i$ , and  $N$  is the number of sample days in month  $y$ .

Macaque encounter rates were also expressed by localities and were calculated based on the following formula:

$$X = \frac{\sum (n_x + n_y + n_z + \dots n_i)}{N}$$

Where,  $X$  is the mean frequency of encounters during all months at locality  $y$

i.e.  $\left[ \frac{(\text{encounter})}{(\text{sample day})} \right]$ ,  $n_x$  is the mean frequency of encounters during month  $x$  at locality  $y$  i.e.  $\left( \frac{\text{encounter}}{\text{sample day}} \right)$ ,  $N$  is the number of surveyed months in locality  $y$ .



**Figure 2.** Map of our landmark points, study site borders and the localities and features of each habitat type in the main campus of UMS and its vicinity (QGIS, 2018).

To generate the density distribution and heatmap of macaque in UMS campus and its vicinity, we used the Quantum GIS 2.18.24 software (QGIS Development Team, 2018) based on location coordinates data collected from the monthly population surveys, personal sightings and other secondary sightings. We used

the same software to show the overall distribution range of the identified macaque groups and solitary individuals, based on the location coordinates of each identified group and individual collected from the monthly population surveys.

## Results and Discussion

### *Number of macaque groups, number of solitary individuals, group size, population size, age-sex composition*

Overall, we sampled 75 days of population surveys over the 12 months survey periods (i.e. 1-12 days of survey per month; monthly average surveys of 6.25 days). The monthly number of detected macaques ranged from 0 to 38 individuals. The monthly population surveys and other personal observations and secondary sightings yielded 103 encounters of a group or individual macaque.

Three macaque groups, namely Group A, Group B, Group C and a solitary individual, were identified. Group A and B were identified based on the number of infants they had, i.e. Group A had two infants, while Group B had four infants. Both groups and the solitary male macaque were sighted at different localities in the campus during a survey conducted in January 2020. Group C was noted as a separate group based on personal observations and the localities where the individual macaques in this group were sighted, which were different from the usual localities where individuals of Group A and Group B were sighted. The group size of Group A was 22 individuals, Group B (21 individuals) and Group C (10 individuals). The estimated population size was 54 individuals. The overall age-sex composition consisted of 8 adult males, 9 adult females, 12 sub-adults, 11 juveniles and 6 infants; 8 individuals were unidentified (Table 2).

**Table 2.** The age-sex composition of the long-tailed macaque population where age-sex is represented by adult male (AM), adult female (AF), sub-adult (SA), juvenile (Juv), infant (Inf) and unidentified (UF).

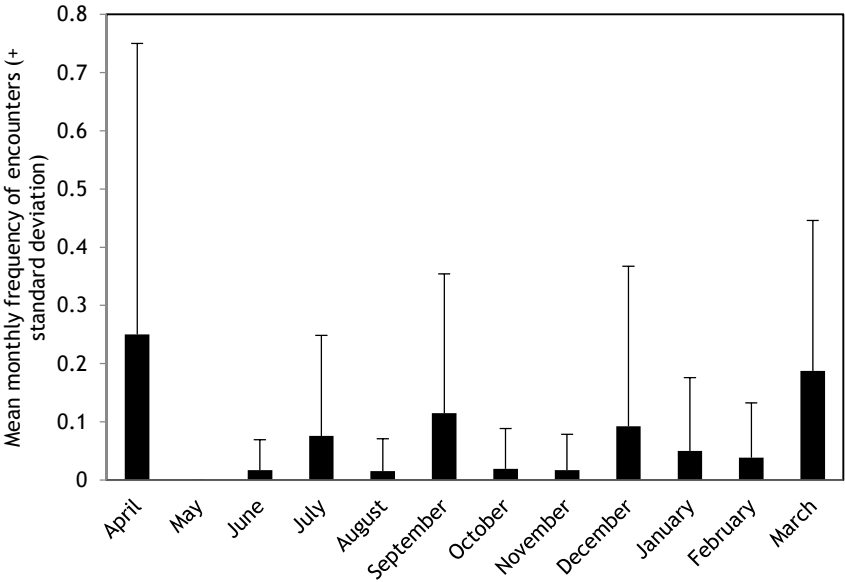
Group/Individual	Total individuals	AM	AF	SA	Juv	Inf	UF
Group A	22	3	5	5	6	2	1
Group B	21	3	4	7	2	4	1
Group C	10	1	0	0	3	0	6
Solitary	1	1	0	0	0	0	0
Total individuals	54	8	9	12	11	6	8



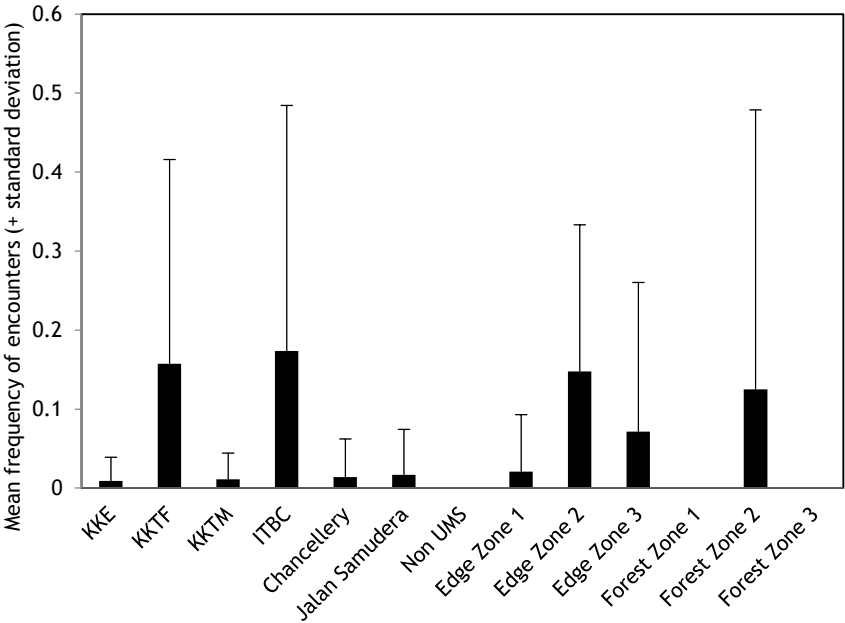
Compared to the group size of long-tailed macaques reported in other university campuses in Malaysia (Universiti Kebangsaan Malaysia: UKM; Universiti Malaya: UM), the group size in our study was much smaller; the mean group size in our study was 17.7 individuals ( $n = 3$ ), whereas it was  $\leq 45.3$  individuals ( $n = 7$ ) in UKM and 49 individuals ( $n = 4$ ) in UM (Md-Zain et al., 2011; Osman, 1998). On the other hand, the group size range found in our study i.e. 10-22 individuals was rather similar to the ones reported in non-provisioned free-range long-tailed macaque groups that generally consume natural food e.g. 19-25 individuals in Singapore (Sha and Hanya, 2013);  $\leq 25$  individuals in Kalimantan, Indonesia (Yeager, 1996); and 10-30 individuals in the wild (Fooden, 1995). This would probably be due to food resources and seasonal provisioning. This information is essential for the future assessment of the status of human-macaque interactions in UMS campus because direct (e.g. being fed by humans) and indirect (e.g. anthropogenic food waste-eating) provisioning both can typically affect primate demography. Access to energy-rich artificial food generally leads to an increase in primate birth rate, possibly inducing larger group sizes and greater dependence on anthropogenic food (Rothman and Bryer, 2019), for example, in UKM (Ruslin et al., 2019) and Bukit Melawati, Kuala Selangor (Mohd-Daut et al., 2021) of Peninsular Malaysia. Therefore, even though macaques in UMS may not be highly dependent on anthropogenic food, further evaluation of their dietary choices with detailed food habits is necessary in future studies.

#### *Macaque density, encounter rate and distribution range*

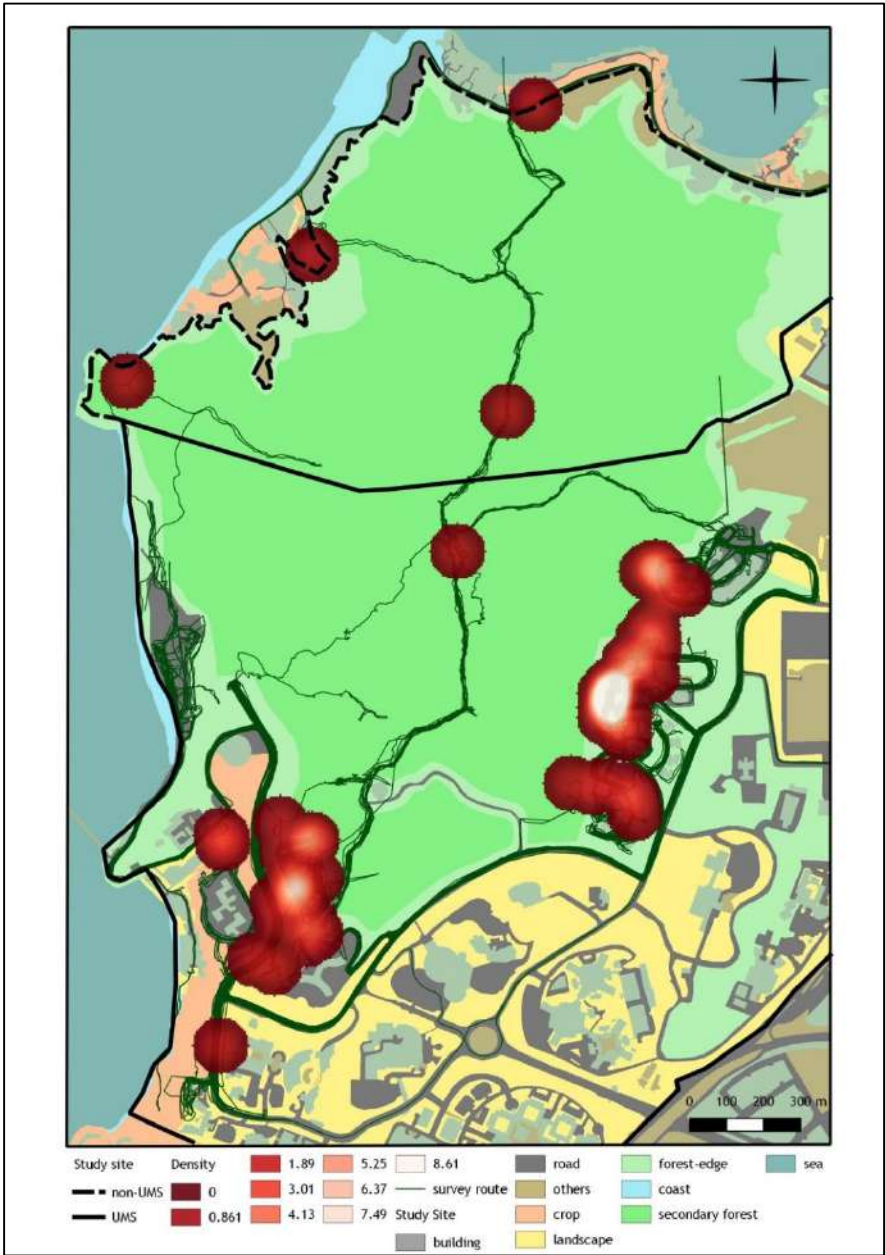
Based on the estimated population size of 54 individuals in UMS campus and its vicinity, the density of the macaque population estimated over an area of 6.02 km<sup>2</sup> was 8.97 individuals/km<sup>2</sup>. Macaques were sighted every month during the study, except in May 2019 due to low sampling effort during that month (Figure 3). Although macaques were encountered in all habitat types as defined in our study i.e. the secondary forest, forest edge and urban areas, the mean frequency of encounters at the 13 surveyed localities varied markedly. Some localities recorded no encounters of macaque, such as at non-UMS area, Forest Zone 1 and Forest Zone 3. Macaques were most frequently encountered at ITBC, followed by KKTF and Edge Zone 2 (Figure 4). In general, the macaques were found to be mostly distributed at two habitat types i.e. the forest edges consisting of Edge Zone 1 near Bukit UMS and Edge Zone 2 near the student residential colleges, as well as the urban areas consisting of the student residential colleges i.e. KKE, KKTF, KKTM, ITBC and Chancellery (Figure 5, Figure 6).



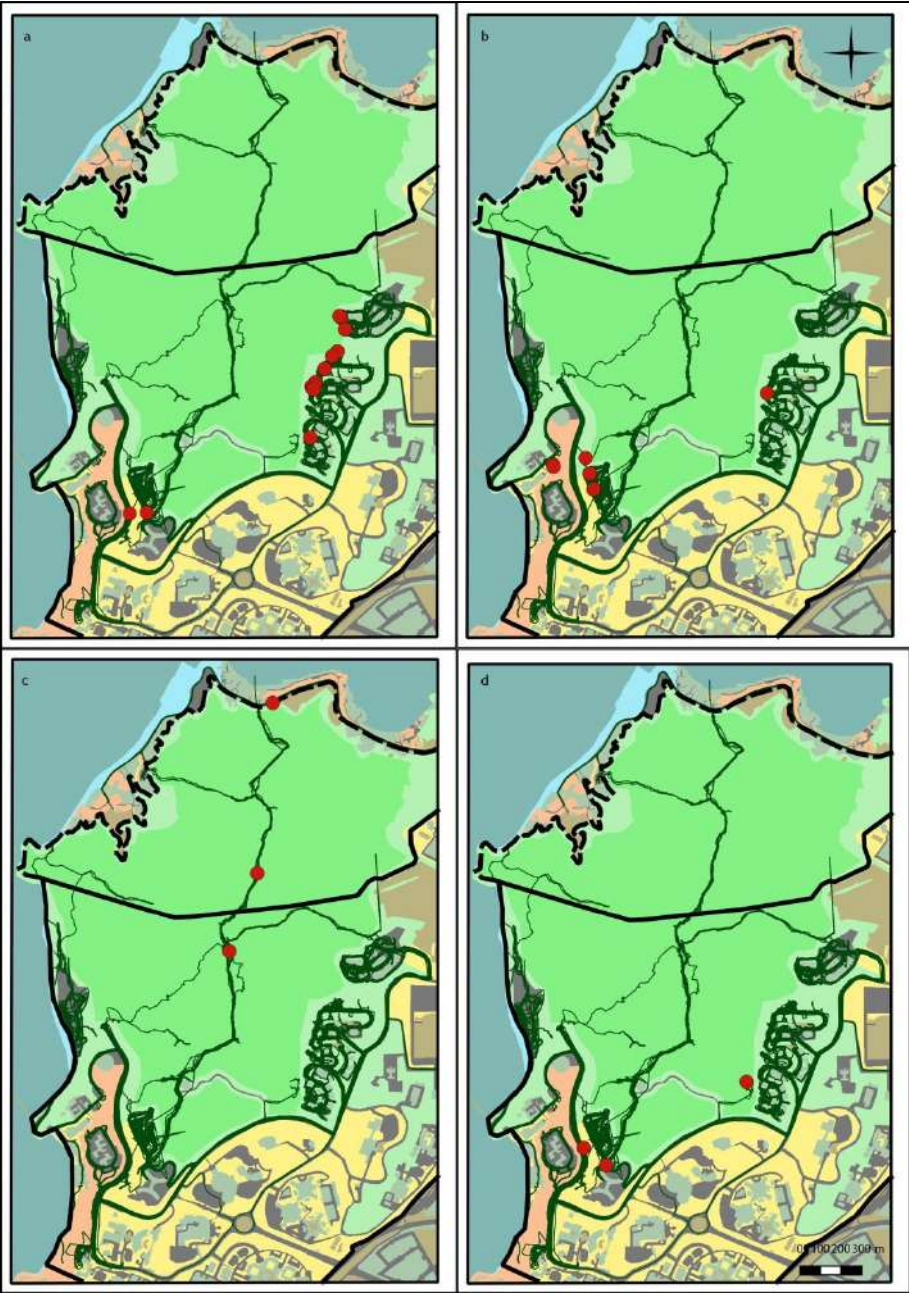
**Figure 3.** Macaque encounter rate i.e. mean monthly frequency of encounters (+ standard deviation) with macaque groups or solitary macaque from April 2019 until March 2020.



**Figure 4.** Mean frequency of encounters (+ standard deviation) with macaque groups or solitary macaque at the 13 surveyed localities.



**Figure 5.** Heatmap of macaque distribution in the main campus of UMS and its vicinity from April 2019 to March 2020, with hotspots represented based on the density distribution of the data points i.e. location coordinates collected from the population survey, personal observations and secondary sightings.



**Figure 6.** Map of macaque distribution range (red dots) in the main campus of UMS and its vicinity from April 2019 to March 2020, with (a), (b), (c) and (d) represented based on location coordinates of Group A, Group B, Group C and solitary individual collected from population survey.

The population density of macaques in our study was lower than that recorded in Singapore i.e. 28.2 individuals/km<sup>2</sup> (Sha et al., 2009). As discussed in Sha et al. (2009), even the macaque density in Singapore was much lower than that in other areas like Hong Kong, Bali and Pulau Penutjang off Java, suggesting that the population of long-tailed macaques in UMS campus may not be beyond its carrying capacity. However, since the macaques in UMS were mainly distributed near areas of high human activities (i.e. in the forest edge and urban areas), rather than in the secondary forest, the macaques may simply be more visible in the campus, thus giving a false impression that macaques in the campus area are abundant. Although further studies are needed to elucidate the different types of interactions between humans and macaques on UMS campus, it is essential to monitor the changes in the population density of the macaques. This is to ensure that potential increase in human-macaque interactions resulting from increased macaque population density could be minimised, preventing a more severe human-macaque conflict in the future.

The fact that the encounter rates of macaques varied widely temporally and spatially, as found in this study, suggests that the macaques in UMS prefer to utilise different localities in different months during the study period. The availability of anthropogenic food waste, due to indiscriminate disposal in the campus may potentially affect the distribution patterns of the macaques, especially during certain months in urban areas and forest edge localities near student residential colleges, where the amount of anthropogenic food waste is expected to increase or decrease significantly, depending on the presence or absence of students during the academic semester or off semester periods.

### **Conclusion and management implications**

The present study has provided some basic information about the distribution and abundance of the long-tailed macaque population that would be instrumental in assessing the status of human-macaque interactions in UMS, so that any negative impacts brought about by such interactions can be ameliorated. In summary, results from the present study showed that long-tailed macaques were found in UMS campus area and its vicinity with an estimated population size of 54 animals and a density of 8.97 individuals/km<sup>2</sup>. Three groups and a solitary male were present during the study, with group size ranging between 10-22 individuals. Based on our comparison with other studies on macaque populations elsewhere, the population density of long-tailed macaques in UMS campus was generally low and likely to be below its carrying capacity. Furthermore, the macaques were encountered more often in

the forest edge and urban areas where anthropogenic activities are high during certain periods, especially near residential colleges during the academic semesters.

In conclusion, while it is imperative to do more detailed studies, especially on the factors influencing long-tailed macaques monthly distribution patterns in relation to the availability of anthropogenic food waste and natural food in the forest, we suggest the following initial actions should be implemented: firstly, we recommend that the authorities in UMS take appropriate action to make students and staff aware that they should not feed the macaques, directly or indirectly; secondly, educational programmes about the negative impacts of provisioning on the ecology of long-tailed macaques should be endeavoured (e.g. Sha et al., 2009); thirdly, proper management and disposal of anthropogenic food waste including using macaque-proof bins or bin with latches should be implemented at macaque hotspots (Animal Neighbours Project, pers. comms.; Hambali et al., 2019).

### **Acknowledgement**

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## Research Article

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# Habitat's Effects on Biomass Content of Gelam (*Melaleuca cajuputi* Maton & Sm. Ex R. Powell subsp. *Cumingiana* (Turcz.) Barlow; Myrtaceae)

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## Abstract

Gelam commonly grows in clumps in peat swamp forests of South and Central Kalimantan. This study aims to determine the effects of growing sites on the biomass content of gelam stands. The study was carried out by analyzing the soil content of growing sites toward the growth of gelam (i.e. the number of individuals and biomass per hectare). The study was conducted at two sites, with the following habitat types: site A - shallow peat with a depth of 51-100cm and only inundated on a high tide, and site B - a moderate peat with a depth of 101-200cm, not flooded but with a groundwater depth of less than 50cm at high tide. A forest fire at site B resulted in higher soil fertility (especially the element K) compared to site A, due to the presence of a pile of ash and charcoal. Also, the fire at site B created more open space, controlled weeds, and at the same time, killed some seedlings and saplings, thinning the site out. The growth rate of gelam at site B was higher than that of site A; at site B, the average height was 10.9m and the average diameter was 10.9cm while at site A, the average height was 9.97m and the average diameter was 10.3cm. The presence of ash, charcoal and more open space after the forest-fire resulted in a higher biomass content at site B (147.223 tons/ha) compared to site A (131.578 tons/ha).

**Keywords:** Biomass, *cajuputi*, peat, Gelam, habitat, Kalimantan

## Introduction

In 1973, intensive use of peatlands for transmigration projects began. In 1995, the Indonesian government executed a massive new rice field programme on peatland in the province of Central Kalimantan, the programme itself was known as the million-hectare Peatland Project (PLG), but it ultimately failed. The land

from the PLG project became neglected, and gelam grew in the area (Poniman et al., 2006).

Gelam (*Melaleuca cajuputi* Maton & Sm. Ex R. Powell subsp. *Cumingiana* (Turcz.) Barlow) grows predominantly in tidal swamps, and freshwater flooded peatlands (Craven & Barlow, 1997; Colton & Murtagh, 1999; Craven, 1999). Gelam tolerates high acidity, which is found in Kalimantan, especially in Central and South Kalimantan (Craven & Barlow, 1997; Rachmanady et al., 2003). Gelam is frequently found growing in areas with high acidity in freshwater, flooded peatlands (Colton & Murtagh, 1999; Craven, 1999; Supriyati et al., 2015). Gelam is a sub-species of Cajuputi (Craven & Barlow, 1997) with a distribution from mainland Indochina to the western part of Flora Malesiana, with the largest population in Kalimantan. This species also produces essential oil, which is similar to its relative in eastern Indonesia, *M. leucadendra*. In international trade, it is also called cajuput oil with Indonesia and Malaysia being the world's main producers, and the main harvesting area being the island of Borneo (Doran, 1999).

Kinnon et al. (1996) described gelam as a pioneer species ecologically. This was confirmed by Suyanto et al. (2001) who noted that gelam is a fast-growing species. Daryono (2009) and Supriyati et al. (2016) also suggested that gelam can grow in habitats that contain pyrite, with good natural regeneration, especially in shallow and moderate peats with acid sulfate soils. A previous study showed that gelam grows predominantly in the area formerly set aside for the PLG; the species grows in clumps as forest composition of the area (BPDAS Kahayan, 2007). The aim of this study is to determine the effects of growing sites on the biomass content of gelam stands (i.e. number of individuals and biomass per hectare).

## Material and Methods

### Study area

The study was conducted in the area of the former PLG (Figure 1) (BPDAS Kahayan 2007). The study area was divided into two sites, (1) site A with shallow peat thickness (51-100cm) located at S 02°50'355'' - S 02°50'520''; E 114°20'383'' - E 114°20'544'', and (2) site B with medium thickness of peat (101-200cm) lying at S 02°49'369'' - S 02°49'627''; E 114°17'462'' - E 114°18'109''. Both sites were selected during a preliminary survey, which found dominant gelam stands and met the parameters of the study plan.

### *Data collection*

The study was carried out in several steps, as follows:

Fieldwork: An inventory of total individuals of gelam (seedlings, saplings, poles and trees) at site A and B was made using a one-hectare sampling-plot for each location. The diameter of each specimen and total number of specimens was measured.

Soil Sampling: Soil samples were collected from both sites to determine soil fertility. The samples were collected from three levels of soil depth: 25cm, 75cm and 125cm, and put into a sealed plastic container to maintain their original condition.

Sample selection: Seedlings (four individuals) with height <1.5cm were selected along with saplings (five individuals) with height of > 1.5cm, and diameter of <10cm, poles (five individuals) with a diameter of between 10cm and 20cm, and trees (five individuals) with a diameter of > 20 cm (Soerianegara and Indrawan 2005). A total of 38 samples were selected from both sites (19 each).

Sample collection: Seedlings and saplings were collected by pulling, while poles and trees were laid down first and then pulled out, in other words, samples were collected by a destructive sampling method. Roots, stems, fruit, flowers, branches and leaves, in various stages of growth (seedlings, saplings, poles, and trees) were taken, and weighed to determine the biomass in wet, air-dry, and oven-dry conditions, so that the moisture content and biomass content could be calculated.

### *Data analysis*

Soil samples from each site were analyzed using the following parameters: pH (H<sub>2</sub>O), carbon (%), total N (%), P<sub>2</sub>O<sub>5</sub> (ppm), K (me/100g), and CEC (me/100g), and followed the procedures provided by Yuwono (2003) and Sulaeman et al. (2009). The biomass was estimated under three conditions (wet, air-dry, and oven-dry), using the allometric equation:

$$Y = a X^b$$

Where: Y = biomass; X = diameter at breast height (dbh) for saplings, poles, and trees; while for seedlings as high as 30cm from the ground; a, b = coefficient. The next step was to make an allometric equation model based on the diameter and number of individuals to calculate the biomass content in tons per hectare.



## Results and Discussion

**Table 1.** Total individuals of gelam at different growth stages

Growth stages	avg. number of individuals per sampling plot	avg. diameter per sampling plot (cm)	total area of sampling plot (m <sup>2</sup> )	conversion factor to hectare	avg. number of individuals per hectare
<b>Site A</b>					
Seedlings	144.0	0.5	100	100	14,400
Saplings	442.6	4.1	625	16	7,082
Poles	257.2	12.6	2,500	4	1,029
Trees	65.0	23.9	10,000	1	65
Total	-	-	-	-	22,575
Average		10.3			
<b>Site B</b>					
Seedlings	239.0	0.4	100	100	23,900
Saplings	114.8	6.0	625	16	1,837
Poles	236.0	13.4	2,500	4	944
Trees	70.0	23.7	10,000	1	70
Total	-	-	-	-	26,751
Average		10.9			

**Table 2.** Parameters of selected samples

<b>Site A</b>							
Growth stages	C (cm)	Ø (cm)	H (cm)	C-h (cm)	ØC (cm)	Rd (cm)	ØR (cm)
Seedlings	0.63	0.20	70.00	32.83	9.85	9.97	9.37
	1.26	0.40	112.00	38.57	12.67	12.90	11.58
	1.89	0.60	129.00	39.67	13.62	14.80	14.23
	2.51	0.80	146.00	49.53	16.47	19.23	18.13
Saplings	4.50	1.43	288.63	134.50	59.50	42.50	65.00
	9.50	3.02	715.00	250.00	100.00	40.00	80.00
	13.00	4.14	872.00	300.00	150.00	55.00	90.00
	19.00	6.05	1020.00	400.00	170.00	75.00	110.00
Poles	26.00	8.28	1050.00	400.00	180.00	80.00	150.00
	32.00	10.19	1071.00	406.00	238.00	85.00	180.00
	38.00	12.10	1173.00	553.00	243.00	90.00	200.00
	44.00	14.01	1289.00	570.00	250.00	95.00	215.00
Trees	50.00	15.92	1341.00	575.00	332.00	98.00	240.00
	56.00	17.83	1343.00	600.00	358.00	99.00	248.00
	63.00	20.05	1347.00	634.00	379.00	100.00	270.00
	75.00	23.87	1490.00	800.00	386.00	115.00	275.00
Total	83.00	26.42	1750.00	820.00	570.00	118.00	304.00
	94.00	29.92	1850.00	850.00	600.00	133.00	327.00
	100.00	31.83	1880.00	800.00	600.00	139.00	353.00
	713.29	227.06	18,936.63	8,253.10	4,668.11	1,421.40	3,160.31
Average	37.54	11.95	996.67	434.37	245.69	74.81	166.33

Site B							
Growth stages	C (cm)	Ø (cm)	H (cm)	C-h (cm)	ØC (cm)	Rd (cm)	ØR (cm)
Seedling	0.63	0.20	62.00	29.08	8.07	9.05	7.97
	1.26	0.40	106.00	43.33	12.17	15.50	11.83
	1.89	0.60	131.00	51.30	15.37	19.33	14.52
	2.51	0.80	144.00	53.67	18.90	21.73	18.90
Poles	4.50	1.43	331.75	136.00	62.00	54.00	61.25
	9.50	3.02	622.00	250.00	90.00	70.00	100.00
	13.00	4.14	780.00	270.00	100.00	80.00	110.00
	19.00	6.05	1100.00	300.00	120.00	80.00	120.00
Sapling	26.00	8.28	1190.00	350.00	155.00	80.00	130.00
	32.00	10.19	1370.00	530.00	220.00	90.00	170.00
	38.00	12.10	1430.00	550.00	250.00	90.00	200.00
	44.00	14.01	1450.00	560.00	300.00	100.00	220.00
Tree	50.00	15.92	1460.00	580.00	330.00	100.00	230.00
	56.00	17.83	1620.00	660.00	340.00	110.00	235.00
	63.00	20.05	1650.00	670.00	350.00	110.00	250.00
	75.00	23.87	1720.00	690.00	400.00	130.00	250.00
Total	83.00	26.42	1740.00	720.00	430.00	135.00	260.00
	94.00	29.92	1850.00	770.00	450.00	140.00	300.00
	100.00	31.83	1945.00	795.00	450.00	140.00	320.00
	713.29	227.06	20,701.75	8,008.38	4,101.51	1,574.61	3,009.47
Average	37.54	11.95	1,089.57	421.49	215.87	82.87	158.39

Notes: C: Circular; Ø: Diameter at breast height (dbh) for saplings, poles and trees, and 30 cm from ground level for seedlings; H: Height; C-h: Canopy height; ØC: Canopy diameter; Rd: Root depth; ØR: Root diameter.

**Table 3.** Biomass Weight in Wet, Air-dry, and Oven-dry

Growth stages	dbh (cm)	Site A			Site B		
		W-w (kg/idv)	Ad-w (kg/idv)	Od-w (kg/idv)	W-w (kg/idv)	Ad-w (kg/idv)	Od-w (kg/idv)
Seedlings	0.2	0.016	0.006	0.006	0.007	0.003	0.003
	0.4	0.034	0.014	0.012	0.025	0.010	0.009
	0.6	0.060	0.025	0.022	0.051	0.021	0.019
	0.8	0.105	0.044	0.038	0.093	0.040	0.035
Saplings	1.4	0.892	0.394	0.346	0.980	0.462	0.406
	3.0	6.250	2.830	2.482	5.600	2.834	2.475
	4.1	9.930	4.814	4.218	10.890	5.738	5.010
	6.1	25.210	12.294	10.733	30.660	17.407	15.045
Poles	8.3	48.980	24.332	21.174	61.920	35.408	30.562
	10.2	70.940	35.373	30.764	93.060	53.321	46.189
	12.1	128.900	64.351	55.723	143.670	80.886	69.956
	14.0	132.110	67.899	58.889	200.360	114.428	98.955
Trees	15.9	236.590	121.735	105.351	320.040	188.334	163.257
	17.8	269.450	140.188	121.212	332.460	197.840	170.199
	20.0	321.160	167.995	145.955	412.710	247.882	214.562
	23.9	631.350	340.959	295.821	616.080	375.923	324.977
Total	26.4	912.950	508.988	440.388	920.180	575.799	497.731
	29.9	1,116.020	620.458	539.036	1,122.050	705.389	611.691
	31.8	1,089.760	622.972	540.138	1,265.820	825.646	708.424

Notes: dbh= Diameter at breast height for saplings, poles and trees, and 30 cm from ground level for seedlings; W-w= Wet weight; Ad-w= Air-dry weight; Od-w= Oven-dry weight

Table 1 shows seedlings that grew at site B after the fire were more numerous and younger with a smaller diameter. Table 2 shows that the gelam samples at



site B had an average height of 10.9m, an average bole height of 8.22m, and an average diameter of 10.9cm while site A samples had an average height of 9.97m, average bole height of 7.08m, and average diameter of 10.3cm.

**Table 4.** Allometric equations for estimating biomass content

Correlation between	Sites	Coefficient of determination	Allometric Equations
Diameter at 30 cm from the ground to biomass at seedling stage	A	$R^2 = 0.979$	$B=0.045Dp^{1.31}$
Diameter at breast height to biomass at sapling, pole, tree growth stages		$R^2 = 0.996$	$B=0.153D^{2.36}$
Diameter at 30 cm from the ground to biomass at seedling stage	B	$R^2 = 0.996$	$B=0.048Dp^{1.76}$
Diameter at breast height to biomass at sapling, pole, tree growth stages		$R^2 = 0.999$	$B=0.183D^{2.39}$

Notes: Dp= Diameter at 30 cm from ground level for seedlings; D= Diameter at breast height (dbh) for saplings, poles and trees;  $R^2$ = Coefficient of determination; B= biomass.

The biomass data (oven-dry weight) for each growth stage in Table 3 were used to perform the allometric equation presented in Table 4. Krisnawati et al. (2012) stated that allometric models of biomass estimation - which have commonly been used in Indonesia - are presented in the form of rank functions, namely  $Y = aX^b$  where Y is the dependent variable (biomass), X is the independent variable (dbh or a combination of dbh and height), a is the coefficient, and b is the exponent of the allometric model.

**Table 5.** Biomass content at each growth stage

Growth stages	Biomass ton/ha	Percentage (%)
<b>Site A</b>		
Seedlings	0.280	0.213
Saplings	49.780	37.833
Poles	63.344	48.142
Trees	18.173	13.812
Total	131.578	100.000
<b>Site B</b>		
Seedlings	0.252	0.171
Saplings	31.931	21.689
Poles	89.031	60.473
Trees	26.010	17.667
Total	147.223	100.000

Allometric equations were applied to the collected data to calculate the biomass content of each stage of growth (seedlings, saplings, poles and trees) in

tons/hectare. The calculation results of biomass content are presented in Table 5, which is the basic data for calculating the total biomass in tons/hectare unit presented in Table 6.

**Table 6.** Total biomass content

Sites	Biomass ton/ha
A	131.578
B	147.223

**Table 7.** Parameters of soil fertility

Soil depth (cm)	Sam ple	pH (H <sub>2</sub> O)	Carbon (%)	Organic material (%)	N Total (%)	P <sub>2</sub> O <sub>5</sub> (ppm)	K (me/100 g)	CEC (me/100 g)
<b>Site A</b>								
25	1	3.88	3.43	5.92	0.19	2.08	0.34	12.54
25	2	3.81	3.45	5.94	0.19	2.12	0.35	11.98
75	1	3.81	3.16	5.45	0.21	1.99	0.37	11.71
75	2	3.78	3.48	6.00	0.19	2.00	0.36	11.83
125	1	3.91	4.26	7.34	0.20	2.17	0.40	8.65
125	2	3.80	4.25	7.33	0.20	2.17	0.37	9.00
Average		3.83	3.67	6.33	0.20	2.09	0.37	10.95
Criteria *		Very sour	High		Low	Low	Medium	Low
<b>Site B</b>								
25	1	3.63	3.40	5.86	0.17	1.49	0.48	9.87
25	2	3.75	3.20	5.52	0.16	1.57	0.48	10.01
75	1	3.32	2.99	5.16	0.14	1.36	0.65	15.09
75	2	3.34	2.97	5.13	0.15	1.32	0.68	14.87
125	1	3.40	3.80	6.70	0.16	1.59	0.57	13.25
125	2	3.36	3.86	6.66	0.17	1.59	0.59	13.22
Average		3.47	3.37	5.84	0.16	1.49	0.58	12.72
Criteria *		Very sour	High		Low	Low	High	Low

Table 6 shows that the total biomass at site B was higher than site A. The result of soil analysis between the two sites are relatively similar based on the parameters of pH, carbon, organic matter, N total, P<sub>2</sub>O<sub>5</sub>, and CEC, while K content at site B was greater than site A (Table 7). A fire at site B resulted in gelam growing well in terms of height and diameter, this is supposedly due to a pile of ash and charcoal formed by the forest fire. Ash contains silicate which had a positive effect on the growth of gelam, especially at site B. This is confirmed by Najiyati et al. (2005) who stated that ash is the residue of burning organic materials such as wood, litter and weeds. Excess ash can increase nutrients, both micro and macro, pH (8.5-10), the soil is not easily leached, and contains base cations such as K, Ca, Mg, and Na which are relatively high. This is evident from a soil analysis that showed site B contained higher K element than site A due to the ash, formed after the fire. This finding is also confirmed

by Komarayati et al. (2004) who state that the ash content of litter charcoal is quite high, at 13.7%, which explains why litter charcoal is often used as fertilizer for both perennial and annual crops. Forest fires result in the formation of charcoal from burning biomass which is supposed to fertilize the soil. Gusmailina et al. (2003) explained that charcoal has an important role in fertilizing the soil, including raising the soil pH level, improving soil structure and texture, creating ideal conditions for soil microorganisms, and increasing the CEC. Ogawa (1989) states that charcoal has plenty of pores which can improve water and air circulation in the soil, and in turn expand the root system of plants.

The fires that occurred at site B caused the death of saplings and seedlings and cleared out weeds. This resulted in improved growth of gelam at site B compared to site A. After the fire, the growing area became more open, weeds were absent, and at the same time, a thinning process occurred as some seedlings and saplings had died. Fires do not kill poles and trees, so after the fire there was more growth of the poles and trees due to the increased open space. Trees and poles at site A were not easily burnt due to a high tide that floods the site, keeping the area wet. The growth of gelam at site A was slower due to a denser groundcover, which covered the stand of gelam, such as Kalakai or Lemidi (*Stenochlaena palustris*; Blechnaceae) (Figure 1.), also the dense stand caused the growth of gelam to be slower. This is in line with the study of Nilsen and Strand (2008) which revealed that the spruce, when thinned out gradually from 2070 trees/ha to 1100 trees/hectare, and finally 820 trees/ha could increase the average diameter from 6.4cm (n = 2070), to 6.5cm (n = 1100), and 7.5cm (n = 820), respectively. This is also in line with the results of Susila's study (2010) which revealed that the duabanga (*Duabanga moluccana*; Lythraceae) stand before being thinned out had an average diameter of 20.30cm (n = 508 trees/ha), an average height of 12.64m, and volume of 181.36 m<sup>3</sup>/ha. After thinning, 172 trees per hectare were left, resulting in an average diameter of 32.88cm, an average height of 19.69m, and a volume of 250 m<sup>3</sup>/ha.

Forest fires can accelerate the process of peat maturation. The more mature peat will increase soil fertility. This was confirmed by Kurnain (2005) who explained that fires in peatlands could increase the recast of peat material, so that it would quickly mature. Water levels that inundated both sites did not directly affect the growth of gelam, but Yamanoshita et al. (2001), who

examined the growth response of *Melaleuca cajuputi* to flooding, found that the higher the water level, the higher the height of *M.cajuputi*.

The aforementioned is the result of analysis and facts in the field of a naturally growing gelam stands. Forest fires certainly cause many losses and must be avoided for forest sustainability. The results of this study suggest gelam can grow well if the habitat is suitable, i.e there is some open space, weeds are controlled and there is fertile soil.

## Conclusions

Forest-fires resulted in better soil fertility and higher levels of the element K. Fires also resulted in more open space, less weeds and at the same time, a thinning process had occurred as some seedlings and saplings had died. There was better growth of gelam at site B compared to site A, and this positively correlated with the biomass content. Forest-fires certainly cause many losses and must be prevented for forest sustainability. However, the results of this study can be a reference that gelam can grow well if the area has some open space, is weed-free and has fertile soil.

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## Research Article

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# Avifaunal survey of Mengilan Forest Reserve and its surrounding areas in Pensiangan, Sabah, Malaysia

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## Abstract

An avifaunal survey was carried out at Mengilan Forest Reserve. The MacKinnon List method was used to assess species diversity. The three-day survey recorded a total of 18 lists with 486 individuals detected. Seventy-five species from 33 families were recorded, with  $H=3.86$  and  $E_H=0.62$ . True species richness was estimated (using SuperDuplicates® online calculator) to be approximately 91 species, with approximately 16 species not detected. The seven Bornean endemics detected were: Black-crowned Pitta, Bornean Leafbird, Bornean Black Magpie, Bornean Necklaced Partridge, Dusky Munia, White-crowned Shama and Yellow-rumped Flowerpecker. Pellorneidae (jungle babblers) and Pycnonotidae (bulbuls) were the most speciose families with six species. Nectariniidae (sunbirds and spiderhunters) and Cuculidae (cuckoos) each had five species. Timaliidae had four species. Apodidae (swifts), Pycnonotidae and Cisticolidae (warblers) had the highest percentages of individuals detected with 10.7%, 9.7% and 8.4%, respectively. The five species with the highest relative abundance index were Silver-rumped Spinetail (0.084), Dusky Munia (0.058), Yellow-bellied Prinia (0.045), Blue-crowned Hanging Parrot and Green Iora (both 0.043) and the Eurasian Tree Sparrow (0.039). The majority of the species detected (68) were forest-dependent species. Of these, 55 species were strictly forest birds. Insectivores made up the most dominant dietary guild, i.e., a total of 46 species (from 21 families) with 41 species in 18 families being strict insectivores.

**Keywords:** avifaunal survey, MacKinnon List method, Mengilan Forest Reserve, Pensiangan district, feeding guilds.

## Introduction

Birds are important indicators of forest ecosystem health and have only recently been included when surveying biodiversity in the forest reserves of Sabah, Malaysia. Realising this, the Sabah Forestry Department has begun rapid assessment of avifaunal communities to help it determine forest ecosystem health. This paper documents the outcomes of a brief bird survey conducted during the Mengilan Forest Reserve (MFR) Scientific Expedition from 10<sup>th</sup> to 15<sup>th</sup> August, 2020. The expedition was organised by the Forest Research Centre,

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Sabah Forestry Department, under the auspices of the Heart of Borneo Initiative. The main objective of this survey is to provide information for future forest management initiatives. Surveys using the MacKinnon List (ML) method (MacKinnon & Phillipps, 1993) were conducted at two sites within the forest reserve and one site close to the expedition base camp.

The Forest Research Centre of the Sabah Forestry Department aims to develop a rapid assessment methodology based on a modified ML method specifically for the department's researchers and field staff with limited time (three to four days) for field work. The Mengilan survey was part of a series of on-going field trials.

## **Methods**

### *Site description*

The Mengilan Forest Reserve (MFR), a Class I Protection Forest Reserve gazetted in 2012, lies approximately within latitudes 4.333-4.45 N and longitudes 116.9-117.017 E, or about 100km west of Tawau town. The forest reserve comprised two parts with the northern portion being larger than the one in the south. Both parts are separated by a logging road and surrounding secondary forests. Its southernmost boundary is also the international border between Sabah and Kalimantan, Indonesia. With an area of approximately 6,684 ha, MFR is located in the Pensiangan district and is jointly administered by the Serudong and Kalabakan district forestry offices. Approximately 60% of its soils are of the Maliau Association while the remainder are of the Serudong Association. The natural vegetation comprised the upland mixed dipterocarp forest (MDF), upland kerangas and the lower montane kerangas forest types. The forest reserve is extremely hilly with many slopes above 70°. The elevation ranged from 500 to 1,500m a.s.l. Due to accessibility and safety concerns, the bird surveys were conducted mainly where the main logging road intersected the larger portion of the forest reserve in its southwest, and northwards from there into the reserve until we faced the sheer cliffs.



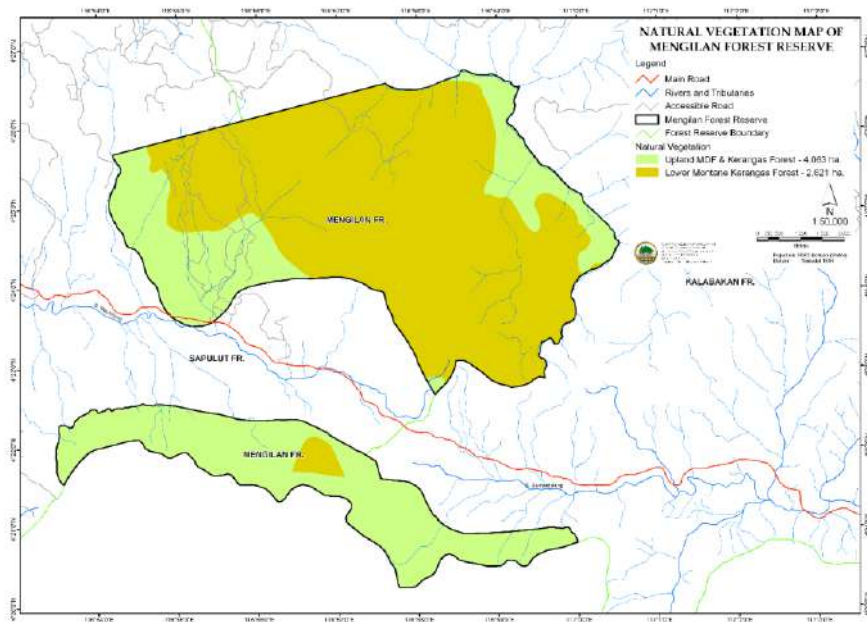


Figure 1: Natural vegetation map of Mengilan Forest Reserve.

### Survey methods

The MacKinnon Lists (ML) method is a time-efficient and cost-effective sampling approach developed for studying avian tropical biodiversity, in which a series of lists of species recorded are collected from a single survey (MacKinnon & Phillipps, 1993). It was designed for those who have limited time, resources and personnel to carry out surveys, such as government agencies, non-governmental organisations, citizen scientists and forest concessionaires. It also accounts for differences in effort, observer experience and knowledge, and weather (Poulsen et al., 1997). As the method relates species richness to the number of observations rather than to time, area or walking speed, it allows for comparison of data obtained by different observers or under varying field conditions (Herzog et al., 2002). The ML method has been gaining popularity since the 1990s, not just in avian surveys but also biodiversity assessments of mammal and fish species (Bach et al., 2020).

To apply the ML method, we compiled lists of consecutive bird species recorded aurally and visually. Each list consisted of 15 species. A species accumulation curve was generated from the addition of those species not recorded on any of the previous lists to the total species number, which was then plotted as a function of the list number. However, in contrast to the traditional ML method,

the number of individuals in each species observed within each list was also recorded. This was to provide more accurate species abundance ranks and to decrease the chances of double-counting of individuals.

### *Observation methods*

Every observer had a pair of Nikon binoculars (8 x 42s). The reference field guide of choice was the 'Phillipps' *Field Guide to the Birds of Borneo*, 3<sup>rd</sup> Ed., 2014. The latest taxonomic changes were determined from online sources and published papers. For example, the Brown Fulvetta was recently placed in the new family Alcippeidae (Cai et al., 2019), and the species has been treated accordingly in this paper. Audio identification was verified using pre-recorded bird songs.

The initial four-day survey was reduced to three days (11<sup>th</sup> to 13<sup>th</sup> August, 2020) due to a team member participating in a search-and-rescue operation. For the first two days, the survey was conducted for 4-5 hours beginning at about 8am. The delayed starts were due to rainy conditions experienced during the expedition, which in turn exacerbated the slippery road conditions heading towards the sites. Surveys were conducted along the main logging road that intersects the forest reserve, along its boundary and up to 1.8km into the forest reserve. On the third day, the survey began at 6am at the base camp and its nearby forests and road network. Due to the slippery and muddy road conditions during the expedition, surveys to detect nocturnal birds were carried out close to the expedition base camp.

All observations were recorded by a designated person. Care was taken to prevent intra-list and inter-list double-counts of individuals. As about half of the individuals were detected by their calls/vocalizations, individuals were listed only when and if the observers were certain that they were different individuals, especially when inputting abundance data within the same 15-species list. Criteria for determining difference in individuals were: a) when the calls originated from a different direction; b) there were two or more calls heard subsequently from a similar direction of a previously recorded individual of the same species; c) the distance from the previously recorded individual was deemed far enough for a call to be considered a different individual. For species in flocks, such as the Chestnut-crested Yuhina, photographs were taken using a handphone and then immediately viewed to estimate the number of individuals. Care was taken not to double-count the same flock. When the trails were not looped, only bird species not recorded earlier were recorded on the return leg

of the trails. Evening surveys were conducted at the expedition base camp, located about 8km from the north-westernmost boundary of MFR.

### *Analyses*

From the acquired data, basic diversity information was extracted, such as species richness, a diversity index (H), relative abundance (EH), most common families, most speciose families and Bornean endemics. A species accumulation curve was generated from the addition of those species not recorded on any of the previous lists to the total species number, which was then plotted as a function of the list number. To estimate true species richness of the area, we used the SuperDuplicates® online calculator developed by Chao et al. (2017), which requires only the total number of species observed and the number of species observed only once (uniques/singletons). The relative abundance indices of species observed were calculated. The most common families and species, and Bornean endemics, were also determined.

Analyses of feeding guilds provided information on how communities of species utilize certain forest resources (for example fruits, insects, arthropods and seeds) and may indicate the condition or health of the forest ecosystem. Thus, the species were categorised according to six feeding guilds based on their preferred diet; carnivores (Car), frugivores (Fru), insectivores (Ins), nectarivores (Nec), granivores (Gra) and omnivores (Omn). Species were considered as omnivores if they are known to consume roughly similar amounts of animal- and plant-based food resources, such as Ins/Gra, Fru/Ins and Nec/Fru/Ins. Guild information was determined mainly from Phillipps (2014) and Wells (1999 & 2007). The feeding guilds were then described according to habitat types (for example, forest, forest edge and open areas) to examine the importance of habitats to different guilds.

## **Results and Discussions**

### *Avifaunal Composition and Species Richness*

The three survey days yielded 18 lists and 486 detected individuals, of which 283 (58.2%) individuals were detected by their calls/vocalisations. A total of 75 species belonging to 33 families were recorded (see Appendix I for the complete species list). The Shannon Diversity Index (H) value was 3.86 with Evenness Index ( $E_H$ ) of 0.62. Compared to the lowland mixed dipterocarp forest of the Kabili-Sepilok Virgin Jungle Reserve (Class 6) with 308 species and 59 families (Petol & Ong, 2013), the lower value for the total number of species may be, amongst others, due to the MFR having been logged in the past. It has to be stated that

the avifaunal data of Sepilok had been collected since the 1970s and thus, provided a more accurate checklist compared to the rapid four-day survey of MFR.

The survey also yielded seven Bornean endemics (Table 1). Besides Bornean Black Magpie, Black-crowned Pitta and Bornean Necklaced Partridge, the other four species were categorised as Least Concern (LC) in the IUCN Red List of Threatened Species. The Bornean Necklaced Partridge was the sole species listed as Vulnerable (VU).

**Table 1.** Species endemic to Borneo and their respective categories in The IUCN Red List of Threatened Species.

No.	Species	Category
1	Black-crowned Pitta	NT
2	Bornean Black Magpie	NT
3	Bornean Leafbird	LC
4	Bornean Necklaced Partridge	VU
5	Dusky Munia	LC
6	White-crowned Shama	LC
7	Yellow-rumped Flowerpecker	LC

Table 2 lists species that were listed as NT and VU in the IUCN Red List of Threatened Species. The sole species listed as Critically Endangered was the Helmeted Hornbill. A large majority of those in the NT category were common lowland MDF species, with the exception of the Great Argus and the Chestnut-naped Forktail. Besides the Bornean Necklaced Partridge, the Rhinoceros and Asian Black Hornbills were also listed as VU.

**Table 2.** Species listed as Near Threatened (NT) and Vulnerable (VU) in the IUCN Red List of Threatened Species.

No.	Species	Category	No.	Species	Category
1	Black-crowned Pitta	NT	14	Great Argus	NT
2	Bornean Black Magpie	NT	15	Green Iora	NT
3	Black-and-yellow Broadbill	NT	16	Lesser Green Leafbird	NT
4	Black-throated Babbler	NT	17	Moustached Hawk-cuckoo	NT
5	Black-throated Wren-babbler	NT	18	Puff-backed Bulbul	NT
6	Brown Fulvetta	NT	19	Rufous-crowned Babbler	NT
7	Buff-necked Woodpecker	NT	20	Short-tailed Babbler	NT
8	Buff-vented Bulbul	NT	21	Sooty-capped Babbler	NT
9	Chestnut-naped Forktail	NT	22	Yellow-crowned Barbet	NT
10	Crested Jay	NT	23	Bornean Necklaced Partridge	VU
11	Dark-throated Oriole	NT	24	Asian Black Hornbill	VU

12	Diard's Trogon	NT	25	Rhinoceros Hornbill	VU
13	Fluffy-backed Tit-babbler	NT			

Table 3 shows that Pellorneidae (ground babblers) and Pycnonotidae (bulbuls) had six species respectively. This was followed by other common families, many of which had similar numbers of species. All 33 families had a mean number of species of 2.27 with a standard deviation of 1.5.

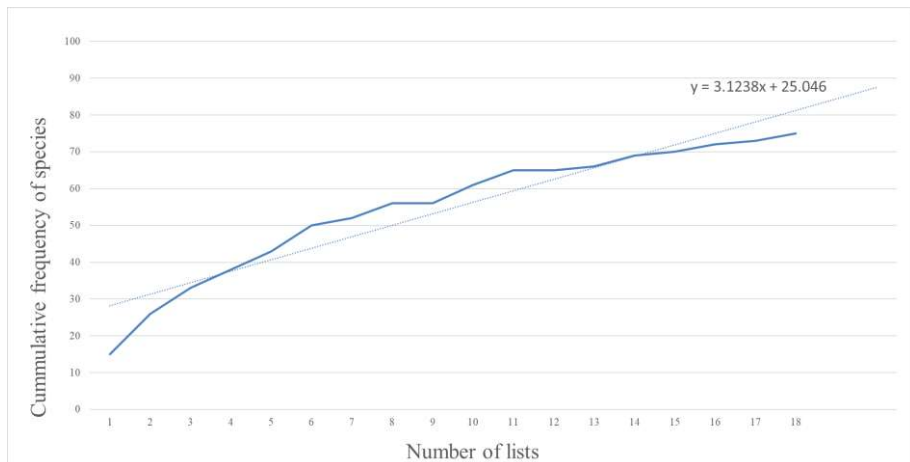
**Table 3.** Top four most speciose families (with shared rankings).

Rank	Family	No. of species
1	Pellorneidae	6
1	Pycnonotidae	6
2	Nectariniidae	5
2	Cuculidae	5
3	Timaliidae	4
4	Apodidae	3
4	Cisticolidae	3
4	Corvidae	3
4	Megalaimidae	3
4	Rhipiduridae	3
4	Muscicapidae	3
4	Bucerotidae	3

As shown in Table 4, individuals from Apodidae (swifts) were the most commonly detected with 52 (10.7%), with the Silver-rumped Spinetail accounting for 41 individuals. With a total of 6 species, the Pycnonotidae (bulbuls) ranked second with 47 individuals detected, approximately 57% of which were the Spectacled and the Yellow-vented Bulbuls. The Cisticolidae, ranked third, was represented by Red-headed Tailorbird, Rufous-tailed Tailorbird and Yellow-bellied Prinia. Being highly vocal, 88.8% of these individuals were detected by their calls. The fourth ranked munia family (Estrilidae) was solely represented by the Dusky Munias. Sharing the same rank was Timaliidae (Old World babblers), represented by four species with the Bold-striped Tit-babblers accounting for 50% of the individuals detected. Both the Psittaculidae (Old World parrots) and Nectariniidae (sunbirds/spiderhunters) ranked fifth with the former represented by Blue-crowned Hanging Parrots. From the five species of Nectariniidae, about 57% of the individuals comprised Little Spiderhunters and Purple-naped Sunbirds. From the list, both Aegithinidae and Alcippeidae, ranked seventh and tenth respectively, were represented by one species each, i.e. Green lora and Brown Fulvetta, with 19 and 14 individuals detected respectively.

**Table 4.** Ten families with the highest percentage of individuals detected (note similar rankings).

Rank	Family	No. of individuals	% of individuals detected
1	Apodidae	52	10.70
2	Pycnonotidae	47	9.67
3	Cisticolidae	41	8.44
4	Estrilidae	28	5.76
4	Timaliidae	28	5.76
5	Psittaculidae	21	4.32
5	Nectariidae	21	4.32
6	Cuculidae	20	4.12
7	Aegithinidae	19	3.91
7	Passeridae	19	3.91
8	Pellorneidae	18	3.70
9	Rhipiduridae	16	3.26
9	Eurylaimidae	16	3.29
9	Megalaimidae	16	3.29
10	Alcippeidae	14	2.88



**Figure 2.** Species accumulation curve and linear regression line of bird species in MFR.

As expected for the ML rapid assessment method, and with a 3-day duration of the survey, the species accumulation curve (Figure 2) had not achieved asymptote. To estimate the true species richness, the SuperDuplicates® online calculator was used (Chao et al., 2017). Only the total number of species detected and the number of singletons (species detected only once) were

needed to input into the calculator. Table 5 is a summary of the results. It estimated Chao1 (species richness using abundance data) to be approximately 91 species, with an upper and lower threshold of approximately 108 and 83 species respectively, at 95% confidence interval. The number of doubletons was estimated to be about 10 while the actual number detected during the survey was 13 species. The calculator also estimated that approximately 16 species were undetected, i.e. the survey managed to detect approximately 84% of the total species in the area. Based on the linear regression line in Figure 2, it estimated that another three lists, or an extra survey day, were needed to detect the estimated 91 species of birds.

**Table 5.** Results from SuperDuplicates®.

Estimated number of doubletons	Estimated species richness	Standard error	95% C.I. lower	95% C.I. upper	Number of undetected species	Undetected percentage (%)
9.53	90.87	6.1	109.91	133.29	16.96	14.38

#### *Relative abundance index*

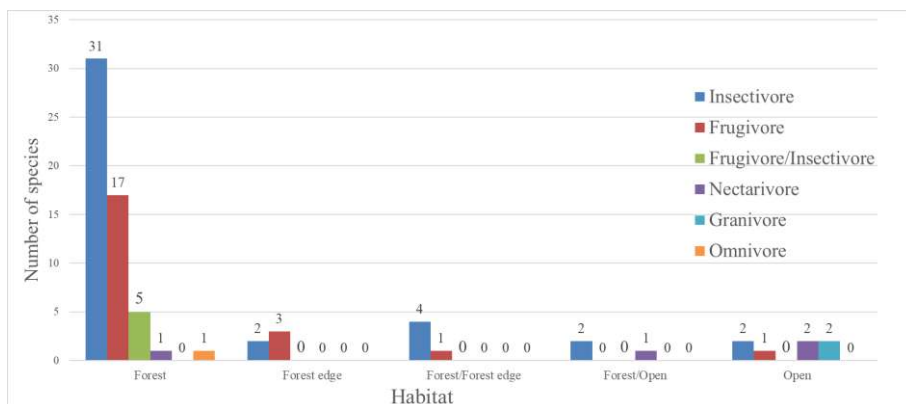
During the survey, 283 individuals (58.2%) were detected aurally. Table 6 shows the top five species with the highest relative abundance index. With the exception of the Green lora, all the other five species were detected along the logging road that intersected the MFR in the south, and in open areas close the expedition base camp. By nature, Silver-rumped Spinetail, Dusky Munia and Eurasian Tree Sparrow were highly visible and were often seen flying in flocks. However, Yellow-bellied Prinia and Blue-crowned Hanging Parrot were mainly detected by their calls; the former being amongst the long grasses and the latter heard from high above the canopy.

**Table 6.** Top 5 species with the highest relative abundance index (note similar rankings).

Rank	Species	Family	Total individuals	Relative abundance index
1	Silver-rumped Spinetail	Apodidae	41	0.0844
2	Dusky Munia	Estrildidae	28	0.0576
3	Yellow-bellied Prinia	Cisticolidae	22	0.0453
4	Blue-crowned Hanging Parrot	Psittaculidae	21	0.0432
5	Green lora	Aegithinidae	19	0.0391
5	Eurasian Tree-sparrow	Passeridae	19	0.0391

### *Habitat types and feeding guilds*

The species were categorised according to their preferred habitats (e.g. forests, forest edges, open area) and respective feeding guilds (Figure 3). The majority of the species (63 species) detected were forest-dependent species. Of these, 55 species from 27 families were strictly forest birds. The high number of forest-dependent species—and the low number of open area specialists—reflected the relatively intact forest ecological functions of the MFR. The presence of eight individuals from three species of hornbills may support this assumption as they are highly dependent on tall forest trees with crevices to breed. Additionally, a pair of the nomadic endemic Bornean Leafbirds were seen. The presence of families such as Campephagidae (minivets), Dicruridae (drongos), Monarchidae (monarchs), Phasianidae (pheasants), Pittidae (pittas), Rhipiduridae (fantails) and Trogonidae (trogons) further reflects the relatively intact ecological functions of the forest. However, the absence of certain families was also bewildering and we lack any explanation for this phenomenon. For example, not a single bird-of-prey or kingfisher was detected aurally nor visually during the survey. Additionally, there was a lack of woodpeckers (Picidae) with only five Buff-necked Woodpeckers and a single Rufous Piculet detected.



**Figure 3.** Number of species according to habitat types and feeding guilds in MFR.

As expected, insectivores made up the most dominant dietary guild, with a total of 45 species in 21 families. Of these, 41 species in 19 families were strict insectivores and the rest were mixed-diet insectivores. The dominance of insectivorous (strict and otherwise) bird species in MFR indicated the presence of plentiful food resources. The second most dominant guild was the frugivores with 21 species, 5 of which were mixed-diet frugivorous species. All 4 species of nectarivores were from the spiderhunter/sunbird family, Nectariniidae. The sole



granivore was the Bornean endemic and very common Dusky Munia. The sole, predominantly omnivorous species was the Hill Myna although it may be more frugivorous in forests.

## Summary

The survey team managed to obtain a preliminary insight on the avian diversity and ecology in MFR. Its avian diversity (75 species from 33 families) is not representative of the forest that included three forest types, namely the upland MDF and kerangas forest, and the lower montane kerangas forest. However, this was mainly attributed to our team's inability to access these forest types due to the lack of roads and the extreme steepness of the terrain. Thus, the inaccessible areas of the lower montane kerangas forest (2,621 ha) in the north, central and southeast portions of the forest reserve were not surveyed, resulting in the lower diversity numbers. However, taking into account the low observation hours of approximately 20.5 hours, the results of this survey is sufficient to describe the current forest ecology when including information from other fields of ecological research that were carried out during the scientific expedition. This forest reserve deserves priority in future ecological research and monitoring activities as and when road access to the said areas improves in the future.

## Acknowledgements

We thank the then Chief Conservator for Forests, Datuk Mashor bin Mohd. Jaini, for supporting the expedition. Our appreciation also goes to the research officers and staff of the Forest Research Centre who organised the Mengilan Forest Reserve Scientific Expedition.

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## Appendix

List of species detected in Mengilan Forest Reserve

No.	Common name	Species	Family
1	Green lora	<i>Aegithina viridissima</i>	Aegithinidae
2	Brown Fulvetta	<i>Alcippe brunneicauda</i>	Alcippeidae
3	Mossy-nest Swiftlet	<i>Aerodramus salangana</i>	Apodidae
4	Plume-toed Swiftlet	<i>Collocalia affinis</i>	Apodidae
5	Silver-rumped Spinetail	<i>Rhaphidura leucopygialis</i>	Apodidae
6	Asian Black Hornbill	<i>Anthracoceros malayanus</i>	Bucerotidae
7	Helmeted Hornbill	<i>Rhinoplax vigil</i>	Bucerotidae
8	Rhinoceros Hornbill	<i>Buceros rhinoceros</i>	Bucerotidae
9	Scarlet Minivet	<i>Pericrocotus speciosus</i>	Campephagidae
10	Lesser Green Leafbird	<i>Chloropsis cyanopogon</i>	Chloropseidae
11	Red-headed Tailorbird	<i>Orthotomus ruficeps</i>	Cisticolidae
12	Rufous-tailed Tailorbird	<i>Orthotomus sericeus</i>	Cisticolidae
13	Yellow-bellied Prinia	<i>Prinia flaviventris</i>	Cisticolidae
14	Green Imperial Pigeon	<i>Ducula aenea</i>	Columbidae
15	Bornean Black Magpie	<i>Platysmurus leucopterus aterrimus*</i>	Corvidae
16	Crested Jay	<i>Platylophus galericulatus</i>	Corvidae
17	Slender-billed Crow	<i>Corvus enca</i>	Corvidae
18	Banded Bay Cuckoo	<i>Cacomantis sonneratii</i>	Cuculidae
19	Greater Coucal	<i>Centropus sinensis</i>	Cuculidae
20	Moustached Hawk-cuckoo	<i>Hierococcyx vagans</i>	Cuculidae
21	Plaintive Cuckoo	<i>Cacomantis merulinus</i>	Cuculidae
22	Raffles's Malkoha	<i>Rhinortha chlorophaea</i>	Cuculidae
23	Orange-bellied Flowerpecker	<i>Dicaeum trigonostigma</i>	Dicaeidae
24	Yellow-rumped Flowerpecker	<i>Prionochilus xanthopygius*</i>	Dicaeidae
25	Bronzed Drongo	<i>Dicrurus aeneus</i>	Dicruridae
26	Greater Racquet-tailed Drongo	<i>Dicrurus paradiseus</i>	Dicruridae
27	Dusky Munia	<i>Lonchura fuscans*</i>	Estrildidae
28	Black-and-red Broadbill	<i>Cymbirhynchus macrorhynchos</i>	Eurylaimidae
29	Black-and-yellow Broadbill	<i>Eurylaimus ochromalus</i>	Eurylaimidae
30	Asian Fairy-bluebird	<i>Irena puella</i>	Irenidae
31	Bornean Leafbird	<i>Chloropsis kinabaluensis*</i>	Irenidae
32	Blue-eared Barbet	<i>Psilopogon cyanotis</i>	Megalaimidae
33	Gold-whiskered Barbet	<i>Psilopogon chrysopogon</i>	Megalaimidae
34	Yellow-crowned Barbet	<i>Psilopogon henrici</i>	Megalaimidae
35	Red-bearded Bee-eater	<i>Nyctyornis amictus</i>	Meropidae
36	Black-naped Monarch	<i>Hypothymis azurea</i>	Monarchidae
37	Chestnut-naped Forktail	<i>Enicurus ruficapillus</i>	Muscicapidae
38	Verditer Flycatcher	<i>Eumyias thalassinus</i>	Muscicapidae
39	White-crowned Shama	<i>Copsychus stricklandii*</i>	Muscicapidae
40	Brown-throated Sunbird	<i>Anthreptes malacensis</i>	Nectariniidae
41	Little Spiderhunter	<i>Arachnothera longirostra</i>	Nectariniidae
42	Olive-backed Sunbird	<i>Cinnyris jugularis</i>	Nectariniidae
43	Plain Sunbird	<i>Anthreptes simplex</i>	Nectariniidae
44	Purple-naped Sunbird	<i>Kurochkinogramma hypogrammicum</i>	Nectariniidae
45	Dark-throated Oriole	<i>Oriolus xanthonotus</i>	Oriolidae
46	Eurasian Tree-sparrow	<i>Passer montanus</i>	Passeridae
47	Black-throated Wren-babbler	<i>Napothera atrigularis</i>	Pellorneidae
48	Ferruginous Babbler	<i>Napothera atrigularis</i>	Pellorneidae
49	Rufous-crowned Babbler	<i>Malacopteron magnum</i>	Pellorneidae
50	Short-tailed Babbler	<i>Pellorneum malaccense</i>	Pellorneidae
51	Sooty-capped Babbler	<i>Malacopteron affine</i>	Pellorneidae
52	Bornean Necklaced Partridge	<i>Tropicoperdix charltonii*</i>	Phasianidae

53	Great Argus	<i>Argusianus argus</i>	Phasianidae
54	Buff-necked Woodpecker	<i>Meiglyptes tukki</i>	Picidae
55	Rufous Piculet	<i>Sasia abnormis</i>	Picidae
56	Black-crowned Pitta	<i>Erythropitta ussheri*</i>	Pittidae
57	Blue-headed Pitta	<i>Hydrornis baudii</i>	Pittidae
58	Blue-crowned Hanging Parrot	<i>Loriculus galgulus</i>	Psittaculidae
59	Buff-vented Bulbul	<i>Iole crypta</i>	Pycnonotidae
60	Hairy-backed Bulbul	<i>Tricholestes criniger</i>	Pycnonotidae
61	Olive-winged Bulbul	<i>Pycnonotus plumosus</i>	Pycnonotidae
62	Puff-backed Bulbul	<i>Euptilotus eutilotus</i>	Pycnonotidae
63	Red-eyed Bulbul	<i>Pycnonotus brunneus</i>	Pycnonotidae
64	Spectacled Bulbul	<i>Ixodia erythrophthalmos</i>	Pycnonotidae
65	Yellow-vented Bulbul	<i>Pycnonotus goiavier</i>	Pycnonotidae
66	Pied Fantail	<i>Rhipidura javanica</i>	Rhipiduridae
67	Spotted Fantail	<i>Rhipidura perlata</i>	Rhipiduridae
68	White-throated Fantail	<i>Rhipidura perlata</i>	Rhipiduridae
69	Hill Myna	<i>Gracula religiosa</i>	Sturnidae
70	Black-throated Babbler	<i>Stachyris nigricollis</i>	Timaliidae
71	Bold-striped Tit-babbler	<i>Macronus bornensis</i>	Timaliidae
72	Chestnut-winged Babbler	<i>Cyanoderma erythropteron</i>	Timaliidae
73	Fluffy-backed Tit-babbler	<i>Macronus ptilosus</i>	Timaliidae
74	Diard's Trogon	<i>Harpactes diardii</i>	Trogonidae
75	Rufous-winged Philentoma	<i>Philentoma pyrhoptera</i>	Vangidae

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\*Bornean endemics

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**Research Article**

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**Avian Diversity, Feeding Guilds and Conservation Status in Mt. Pantaron, San Fernando, Bukidnon, Philippines**Mary Cor S. Salolog<sup>1</sup>, Heidi C. Porquis<sup>1</sup>, Arturo G. Gracia, Jr<sup>2\*</sup><sup>1</sup>*Department of Biology, Central Mindanao University, Musuan, Bukidnon, Philippines*<sup>2</sup>*Department of Natural Sciences and Mathematics, Surigao del Sur State University-Main Campus, Surigao del Sur, Philippines***\*Corresponding author:** artzgracia@gmail.com**Abstract**

Bird conservation is one of the main concerns of conservation societies due to its crucial role in maintaining ecosystem balance. However, for this to be realized, there is a need for continuous updating on ecological information necessary for conservation planning, especially for less explored habitats. Hence, this study was conducted to evaluate the diversity and status of birds in one of the understudied areas in Bukidnon, Philippines. Mist-netting for a total of 200 net-days, transect-line, and point-counts were carried out to record the birds. A total of 31 species with 386 individuals in 23 families were noted. Overall diversity value was relatively higher with  $H'$  index of 2.87, equitability value of 0.84, and a level of dominance ( $D$ ) of only 0.08. Thus indicating that the habitat is diverse. However, concerning the uniformity of the population within the community, the birds' assemblage was noted to be moderately even with  $J'$  value of 0.56. The assemblage of birds was a mixture of insectivores, frugivores, carnivores, nectarivores, granivores, and omnivores which are essential for seed dispersal, pollination, pest control, and ecosystem re-constructing. Seven endemic species were noted. Among the birds, *Padda oryzavora* was the threatened species observed, while nine species were assessed to have a declining population. The threats observed in the area include hunting traps called "pulot," slingshot, slash and burn (*Kaingin*), and mist-netting. Based on the results, despite the limitations on the actual observation and documentation, Mt. Pantaron Range was found to house various species of avian fauna, which is worth saving from the different threats like habitat destruction and unregulated exploitation.

**Keywords:** Assemblage, Birds, Feeding Guild, Species Richness, and Threats**Introduction**

Biodiversity conservation has been one of the pressing global issues in the last few decades. The avian fauna particularly has drawn much public attention because of their fascinating appeal and the various unique roles they play in the

web of life. Both enthusiasts and researchers alike have intensified activities towards generating public awareness and support for this taxon. National and global databases are being developed. But these data need continual updating, and here is where local inventories become relevant.

Barangay Kawayan is one of the 24 barangays in the municipality of San Fernando, Bukidnon. This 3,402.21-hectare community is nested in the Pantaron Range, which is a proposed protected area in the province of Bukidnon (Barangay Kawayan Development Plan, 2008). Information obtained from the elderly residents of this barangay indicates that back in the 1960s, the area was still considerably forested, with large tall trees such as *Shorea contorta* White Lawaan, *Shorea negrosensis* Red Lawaan, *Shorea polysperma* Tanguile, *Dacrydium* sp. Tiger tree, and *Pterocarpus indicus* Narra being relatively common. The same report also indicates the presence of different animals like hornbills, owls, doves (*balud*, *punay*, and *manatad*), parrots, *kuruwakwak* (black Pigeon), hawk, falcon, eagles, flying lemur, tarsier, and herptiles.

However, logging operations began in 1971 (Gallardo, 2008). The first concessioner, Balor Ansans, opened the first road to the Barangay. Other logging operators followed. These include the Alejandro Almendras Group, the NAREDICO, El Labrador, the Balderama, some Koreans, and Japanese. Based on this information, the latter was interested in the local fauna and treasure hunting. But opposition against these logging and other activities soon began and intensified. Local and indigenous people, such as Tigwahonon, Manobo and the migrants, started rallies and pickets from the barangay up to the capital town, now the City of Malaybalay. As a result, logging operations were terminated.

What had existed and what is left of Barangay Kawayan's avian resources have not been recorded formally through the scientific process. In fact, not even a single scientific report on avian diversity, guild composition, or status and other ecological information is available for public scrutiny or for future scientific investigations. This is despite the fact these ecological data are necessary for conservation planning, especially for areas that are highly susceptible to anthropogenic activities and less explored. Thus, this study is intended to generate this important database of the avian fauna in the area. This necessity is further pressed by the current project of the national government to open the Bukidnon-Davao Road via San Fernando. This means that development will be intensified around the area, and it will most likely primarily affect its avian populations.

## Materials and Methods

### *Place and duration of the study*

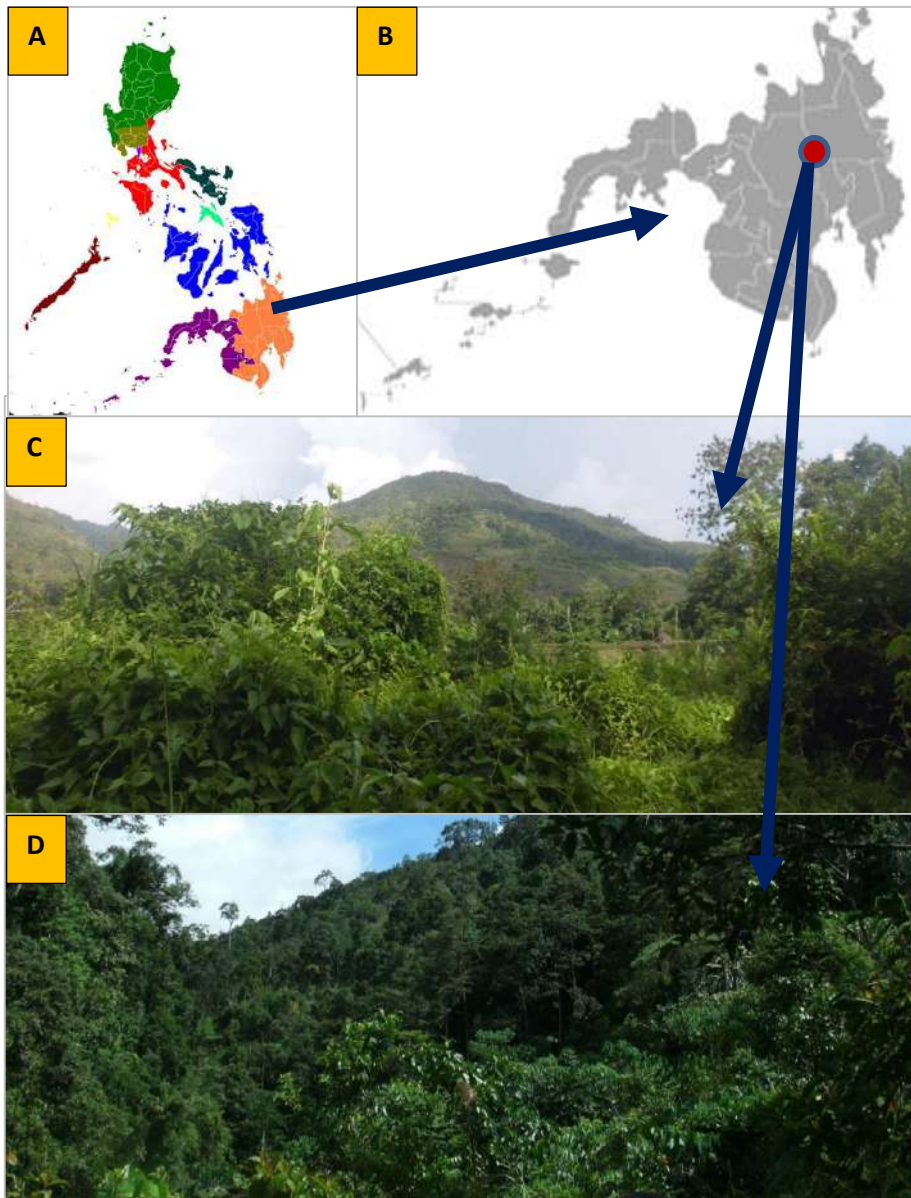
The study was conducted in two varying vegetation types of Mt. Pantaron located at Barangay Kayawan, San Fernando, Bukidnon, Philippines, positioned at 7.892737 North and 125.395830 East (Figure. 1A-B). The first station was located at the agroecosystem part of the mountain range (Figure. 1C), while the second station was situated at the lower montane part of the mountain (Figure. 1D). The main fieldwork was conducted from December 20-24 and 26-29, 2010, covering nine days of field working days. On September 8 and 9, 2014, a supplementary survey was conducted in the area. However, the approach was performed passively and was more on the perspective of opportunistic observations only.

### *Habitat Description*

The dominant plant species in the agroecosystem consist primarily of grasses, sedges, ferns, *Cocos nucifera*, and scarcely distributed trees, mostly fruit trees. The topography was generally plain. Availability of water bodies was evident for there were streams near the sampling areas at a distance of at least 20 metres to 100 metres. Some parts of the transect also by-passed areas that are just adjacent to rice fields with water irrigations. The closest human settlement was observed to be at least 500 metres away from the nearest sampling point. Canopy cover was almost absent considering the plant structure present in the area was below human height. As for the sampling station in the lower montane forest, the area was dominated by various species of trees, ferns and shrubs. The topography was quite angular for a terrain was quite elevated with a slope that ranging from 30-50 degrees. The forest canopy was around 80 to 90 percent. Occasional fallen logs were also observed inside the habitat, and leaf litter was quite thick. Presence of water bodies was also prominent in some points of the sampling area within this habitat. The distance of the nearest human settlements to the sampling area was estimated to be around 1.5 kilometres.

### *Sampling Techniques and Data Collection*

Since the study was performed by researchers who have limited knowledge in bird call identification, only the traditional mist-netting and ocular surveys were conducted. For mist-netting, three partitions were considered. The first was the ground net, established around 0 to 5 metres above the ground. The second one was the sub-canopy net, set around 10 to 15 metres; lastly, the canopy or Skynet was established 20 metres above the ground. All mist-nets had a dimension of seven metres long, five metres wide, and 30 mm mesh.



**Figure 1.** Geographic location of the study area (A and B). A portion of the sampled habitat in Brgy. Kayawan, Mt. Pantaron: Agroecosystem (C) and Lower Montane (D).



The checking and monitoring of nets were performed at intervals of one to two hours from 5:00 AM to 6:00 PM. A total of 100 mist-net days were accumulated throughout the sampling period per sampled habitat, thus, garnering an overall sampling effort of 200 net days.

For the ocular survey, the established human trail was followed and served as the transect line. This trail or transect line bypassed the two sampled stations. Along this transect, point stations were considered. All observations that were noted in the agroecosystem were pooled together. The same was done for the birds observed in the lower montane area. A total of 64 observation hours were recorded. Of these, 54 hours were contributed from the first visit and ten hours from the second visit. All bird species that were observed visually were noted. The book of Kennedy et al. (2000) entitled "A guide to the birds of the Philippines" was used as the key guide in the identification and verification of these observed birds in the area. After the identification, the birds were grouped according to their foraging types. The assignment into what foraging guild the birds belong to was based on the feeding report of Kennedy et al. (2000) and IUCN (2021). Numerical representation of the different foraging type depended on how many species fell into the respective guilds.

#### *Data Analysis*

The Biodiversity Professional Software by McCleece (1997) was used in the determination of the similarity index across the sampled habitat. The Paleontological Statistical Software by Hammer et al. (2001) was used to determine the diversity indices, descriptive statistics and species rarefaction. Visual presentations were created using Microsoft Excel 2016. Assessments were based on the current International Union for Conservation for Nature's report (IUCN). The primary diversity indices used were species evenness ( $J'$ ), Simpson Diversity Index ( $D$ ) which is a measure of species dominance, and Shanon-Weinner Diversity Index ( $H'$ ). The species evenness is a measure for species abundance uniformity within the community, wherein the more even the population of the species is, it suggests a higher diversity. This measure is represented through a numerical value of zero to one, with one as the highest value and suggesting higher diversity. The dominance on the other hand is the inverse of diversity and evenness, wherein if the value of dominance is closer to one, this suggests low diversity, and a value of zero entails the opposite. The  $H'$  index represents the totality of the whole diversity of the area which is calculated using the data set on species richness and abundance. The higher the value, the more diverse the habitat is. However, using  $H'$  index alone can be subjected, especially into the idea of what value should be considered as a

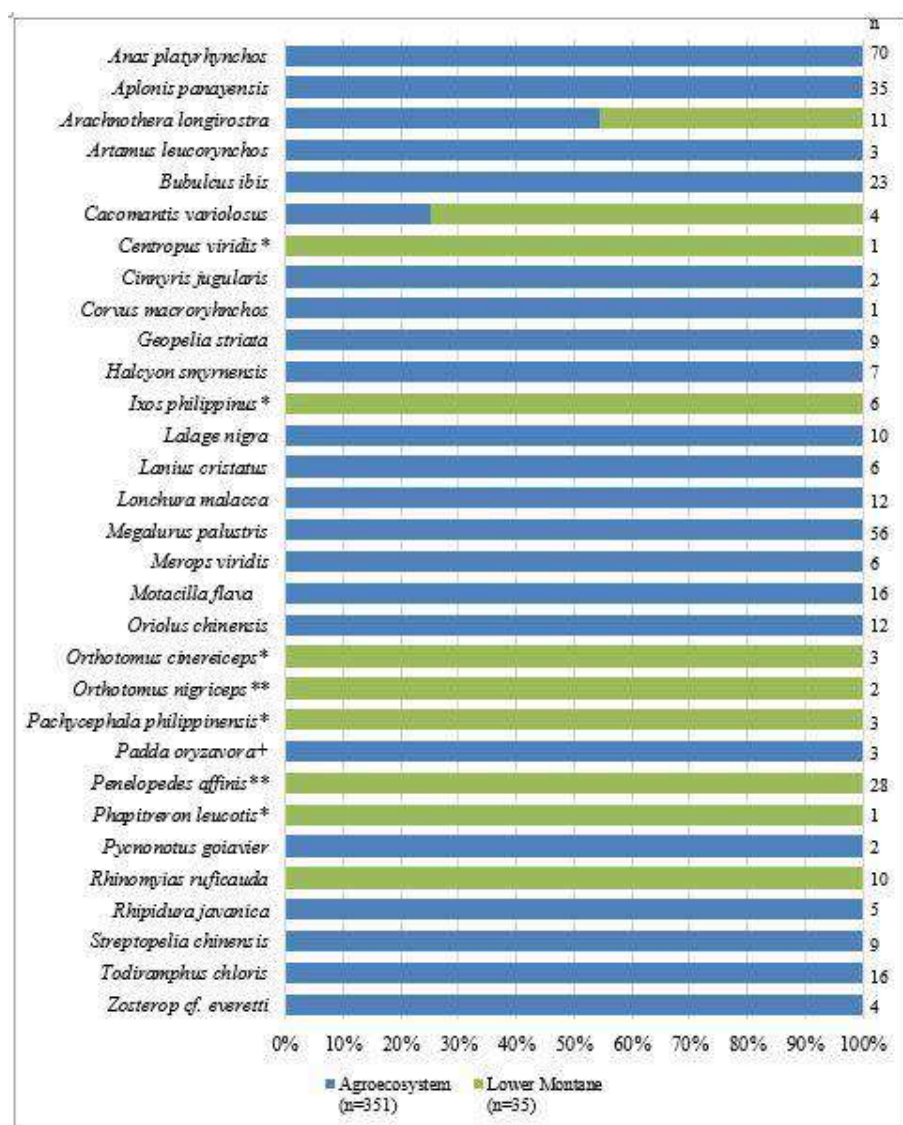
marker in saying that the area is diverse. Hence, the value of equitability was calculated to serve as reference for making a remark. The equitability was determined by calculating the ratio of the H' value the H' index theoretical value. An equitability value of one suggests the highest diversity while zero indicates the opposite.

## Results and Discussion

### *Assemblage and Diversity*

A total of 386 individuals classified into 23 families, grouped into 30 genera, and identified as 31 different species of birds were recorded. Of these, four species namely *Anas platyrhynchos* (n=70, 18%), *Lonchura malacca* (n=56, 15%), *Aplonis panayensis* (n=35, 9%), and *Padda oryzavora* (n=28, 7%) were the abundantly observed (Figure 2). These four species comprised 49% of the abundance proportion in the area. The high number of *A. platyrhynchos* is associated with its behavioural characteristics. These wild ducks are always in a group wherever and whenever they are (Kennedy et al., 2000), just like the other duck species. Meanwhile, the most prominent part in abundance is the high number of observations of the threatened species *Padda oryzavora*. The species is considered to fall under the vulnerable criterion of the IUCN assessment, yet it was among the most well-numbered bird in the area.

The overall avian faunal composition observed comprises 4.5% of the total species in the Philippines (n=691: Allen et al., 2017) and 0.3% based on the global records (n=10,000: Thayer, 2017). The totality is comparatively lower to the reports of Gracia et al. (2021) in Awasian Water Forest Reserve in Mt. Hilong-hilong (n=82); Calimpong & Nuñez (2015) in Bega Watershed, Prosperidad (n=83); Nuñez et al. (2017) in Mt. Matutum Protected Landscape (n=81); Lagat and Causareen (2019) in Upland Cavite (n=121); Amoroso et al. (2018) in Mt. Hamguitan Expansion Site (n=41); Mohagan et al. (2015) in Mt. Apo Long-Term Ecological Research site (n=38); and Alviola et al. (2010) in Malagos, Watershed, Davao del Sur (n=54). However, this low representation of birds in the area compared to the other sampled habitats across the Philippines, as mentioned above, does not necessarily mean that the site is poor in terms of bird assemblage. It is important to elucidate that as compared with other findings, just like in the case of Gracia et al. (2021), Nuñez et al. (2017), Mohagan et al. (2015), Alviola et al. (2010), and the rest of the studies, they had someone who



**Figure 2.** The proportion of species abundance across sampled habitats. Note: The species name with a single asterisk at the upper right denotes that the bird is a Mindanao Endemic. Two asterisks denote Philippine Endemic. Meanwhile, the plus sign means that the bird is a threatened species.

is actually well-versed in documenting the birds through vocalization. Others had access to more sophisticated tools that are used for birding, thus, maximizing the potential of observing more species in the area even if these are

not seen. On the contrary, the current study was highly dependent on documenting species through mist-netting, for it was the most reliable thing to do during that time. Also, although an ocular survey was also performed, but due to lack of mastery on bird identification, other observed species were counted as part of the data mortality to avoid the issue on uncertainty.

With regards to comparative ecological information between the sampled habitats, the agroecosystem was noted to hold more species ( $n=23$ ) than the lower montane forest ( $n=10$ ). Also, abundance was relatively higher in the agroecosystem comprising 91% ( $n=351$ ) of the total population than the lower montane, which covers only 9% ( $n=35$ ) of the accumulated record (Figure 2). Species diversity-wise, still, the agroecosystem had the highest values in terms of Shannon index ( $H'=2.64$ ) against the lower montane's value of  $H'=2.07$  (Table 1).

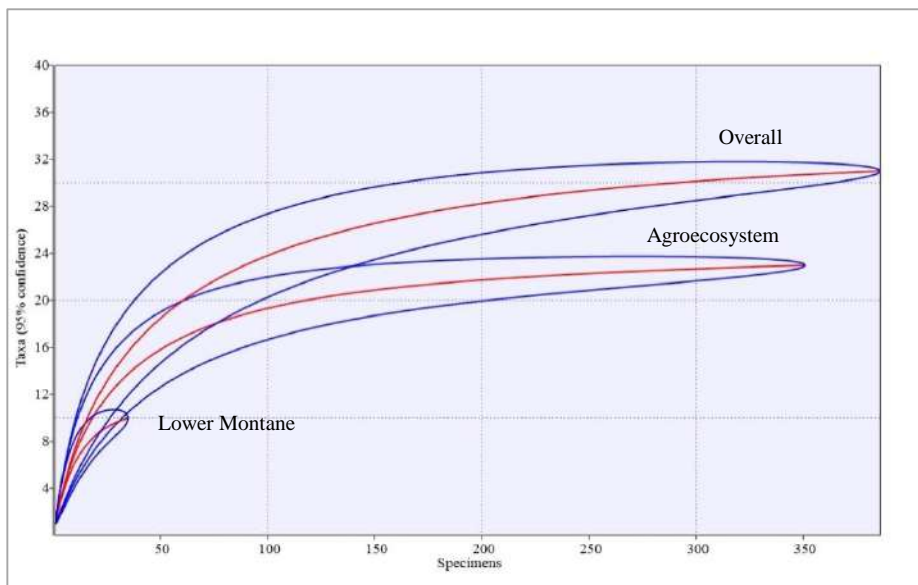
**Table 1.** Ecological information in two sampled vegetations and for the entire site of Mt. Pantaron.

Ecological Information	Site		
	Agroecosystem	Lower Montane	Overall
Shannon-Weiner Diversity Index ( $H'$ )	2.64	2.07	2.87
Species Evenness ( $J'$ )	0.61	0.79	0.56
Species Dominance ( $D$ )	0.09	0.15	0.08
Equitability value	0.84	0.90	0.84

The high bird abundance in the agroecosystem was already expected, for it is usually a given idea (Harvey & Villalobos, 2007) as species commonly found in this type of habitat are generalist (Van Der Wal et al., 2012). Also, most of the time, the species inhabiting such areas are dominant ones with a wide distribution range (Tanalgo et al., 2015). However, the rest of the results opposed the commonly reported ecological scenario, such as in the studies of Mohagan et al. (2015), Barzan et al. (2015), and Laube et al. (2007) that forested areas are more species-rich and diverse habitats. Although at some point, it does align with the findings of Mulwa et al. (2012). Still, this study could not concur that the agroecosystem harbours more species than the lower montane. The primary reason for this aligns and lies on a single concept - the sampling limitation.

This result is a pure manifestation of the barriers observed during the conduct of the study. Unlike in the lower montane, the area in the agroecosystem is more open with fewer ocular barriers. Hence, the birds that were present at that time

were much easier to track and observe. When doing the actual visual survey in the lower montane, the high density and cover of shrubs and trees made it more challenging to see the birds. This claim is further supported by the reports of Maghuyop et al. (2000), Saguindang et al. (2002), and Dumalahay (2009) that dense and closed forest cover result in low bird counts. Although some familiar bird calls and bird appearances were noted, it was still not enough to contribute to the list. This limitation is clearly manifested in the simulated species rarefaction (Figure 3). As shown, the graph projection in the lower montane is way too small and suggests an increasing pattern. This graph entails that the data does not yet reach the saturation level; thus, the probability of observing more species in the area is still high. On the contrary, the graph representation for the agroecosystem is already projecting an asymptote pattern. This result entails data saturation. The graph further suggests that the chance of observing additional species is now lower.



**Figure 3.** Species rarefaction for the two sampled habitats and for the entire Mt. Pantaron.

On the other hand, despite the limitations incurred in the lower montane, the overall diversity index was recorded to be generally higher, with a Shannon-Weinner index of 2.87, equitability value of 0.84, and a level of dominance of only 0.08 (Table 1). The result suggests that Mt. Pantaron itself is an abode for avian fauna. However, species evenness-wise ( $J'$ ), the value is observed to be moderately even with  $J'=0.56$ . The species evenness is highly influenced by the

dominant species recorded in the agroecosystem, especially by these four dominant birds, namely *A. platyrhynchos*, *L. malacca*, *A. panayensis*, and *P. oryzavora*. Ecologically, the presence of dominant species affects the evenness of the community. The higher the number of dominant species and their representation (outliers), the lower the evenness would be. In relation to diversity, the lower the evenness, the diversity value would also fall, for it is directly proportional. But it is also crucial to point out this is just one of the components that contribute to diversity. Factors such as species richness and composition are also essential contributors that influence the diversity value. Thus, despite the presence of those dominant species, the area still holds high diversity. Further, a lot more species were noted to be evenly distributed within the community. Hence, those species outliers contributed only a minor fallback to the  $H'$  index. That is clearly observed through the obtained value on dominance ( $D = 0.08$ ) - another ecological measure that is inversely proportional to the Shannon Index.

#### *Feeding guilds*

The identified species were assessed based on their diet. Results revealed that the birds belong to six different feeding guilds. The well-represented group was the Insectivores with 16 species, followed by Frugivores ( $n=5$ ), Granivores ( $n=4$ ), Carnivores ( $n=3$ ), Nectivores ( $n=2$ ), and Omnivores ( $n=2$ ) (Figure 4). The findings suggest that the habitat has good ecological support that ranges from seed dispersal, pollination, pest control, and ecosystem re-constructing, which are an important component for any ecosystem to thrive (Heine & Speir, 1989; Kati & Sekercioglu, 2006). This is because the presence of different ecological niches is vital in any community for it supports each other to maintain the balance of the ecosystem (Tabur & Ayvaz, 2010; Law, 2019). Meanwhile, the high concentration of the insectivores is highly associated to the fact that most of the species are those found in the agroecosystem going to the forest edge, where insect diversity is noted to be high - a concept which is primarily anchored on the idea of the predator-prey relationship. At some point, this also explains why there was low to no representation of the large frugivores in the site - an observation which aligns to the report of Sekercioglu (2012). As mentioned, the sampling conducted was more biased on the agroecosystem due to various limitations. Also, knowing that the habitat is an open area dominated by grasses and sedges (reasons for observing granivores) where there were only limited fruit trees, the chance of documenting fruit-eating birds or even nectar-feeding and flower-pecking bird species was also limited. This is because bird assemblage is highly associated with food availability (Bhatt & Joshi, 2011).

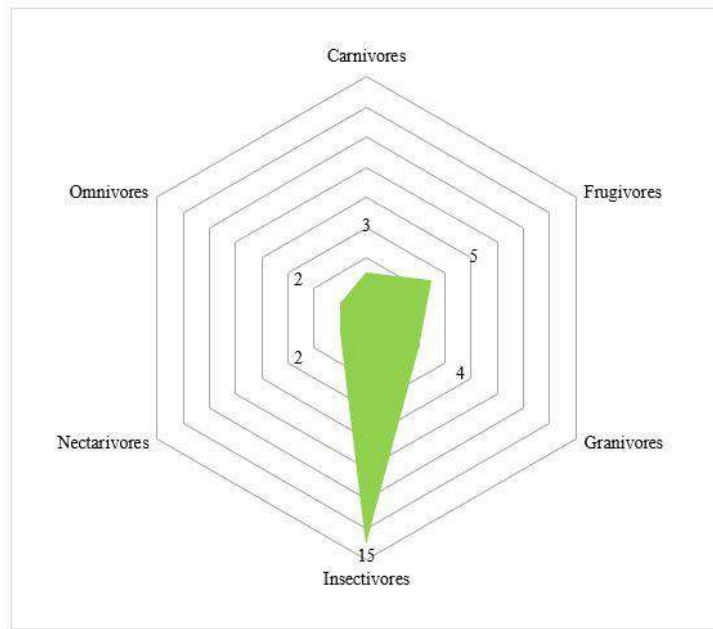
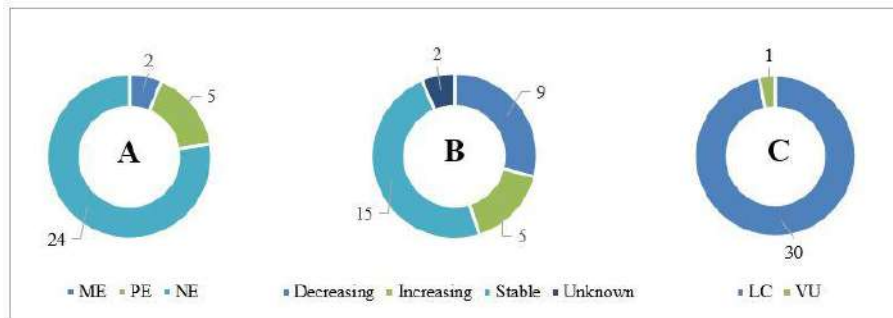


Figure 4. The distribution of the species in terms of their feeding guilds.

#### *Endemism, status, and threats*

Only seven endemic species of birds were noted, comprising 22% of the records. Five were geographically restricted in the Philippine archipelago (*Centropus viridis*, *Ixos philippinus*, *Orthotomus cinereiceps*, *Pachycephala philippinensis*, and *Phapitreron leucotis*), while two were restricted in the Mindanao faunal region (*Orthotomus nigriceps* and *Penelopedes affinis*) (Figure 5A). It was observed that despite the agroecosystem being the most species-rich habitat, not even a single endemic species was found in the area. All the endemic species, either Philippine or Mindanao endemic, were documented in the lower montane forest even with limited observation. This result suggests that highly specialized species are associated with a more intact forest and are most likely to shun areas with a high level of human disturbance (Dans & Gonzalez, 2010). The finding further suggests that a more extensive and intensive study with an increased focus on this type of vegetation is necessary because more endemic species are still waiting to be found.

In terms of conservation status, the species *P. oryzavora* is the only threatened species recorded. While for the population trend, a noteworthy remark is the documentation of the nine species with decreasing numbers based on the IUCN



**Figure 5.** Percentage distribution of species in term of Endemicity (A), Population Trend Status (B), and Conservation Status (C).

assessment (2021) (Figure 5B and 5C). These species include *Lalage nigra*, *Lanius cristatus*, *Motacilla flava*, *Oriolus chinensis*, *Pachycephala philippinensis*, *Padda oryzavora*, *Penelopides affinis*, *Rhinomyias ruficauda*, *Todiramphus chloris*, and *Zosterops cf. everetti*. The presence of these species in the area, including the endemic ones, suggests a call for conservation measures and management plans, especially that couple of threats were also noted in the site. These threats include habitat destruction through slash and burn (*Kaingin*), unregulated anthropogenic activities through bird hunting (*Pulot*), and trapping of birds with high economic demand like the threatened species *P. oryzavora* that are usually being traded in the nearby city. Also, considering the idea that the constructed road that cuts across the mountain range itself is about to open to the broader public. Thus, possible development in the area could lead to an environmental problem that will lead to species displacement if measures are not prepared in advance.

## Conclusion and recommendation

Based on the results, Mt. Pantaron is an ideal abode for various bird species. The area showed a high diversity value, diverse composition of ecological niche, and home to endemic birds, threatened, and those who have a declining population. With this, it is recommended that conservation initiatives for the area be considered, especially evident threats that could affect the area's biodiversity. Furthermore, a more rigorous study is suggested to further enhance the profiling of the birds in the area, particularly in the lower montane forest. These are highly necessary for crafting efficient and pragmatic conservation measures.



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## Short Communication

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# Silvered Langur (*Trachypithecus cristatus*) Survey in Sibuti Wildlife Sanctuary, Miri, Sarawak

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## Abstract

A rapid and passive primate survey using a scanning technique was conducted in Sibuti Wildlife Sanctuary (Sibuti WS) from 11<sup>th</sup> to 16<sup>th</sup> July, 2018. This study provided the first confirmed record of the presence and the diet of silvered langur in the wildlife sanctuary. A group of silvered langur that comprise 20 to 30 individuals were observed in the vicinity of the sanctuary. The diurnal langur feeds on fig plants (*Ficus* sp.), *simpoh air* (*Dillenia suffruticosa*) and tree from the family Leguminosae. However, this langur population appears to be isolated and is probably decreasing due to limited food sources, intra- and interspecific competition, and hunting pressure by local people.

**Keywords:** diet, ecology, scan sampling, silvered langur

## Introduction

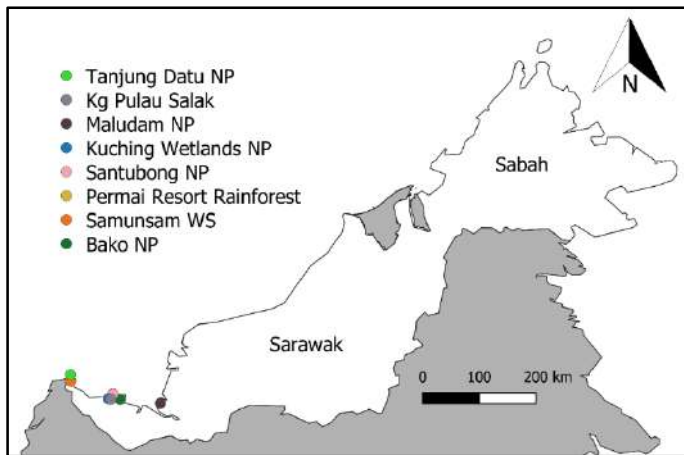
The Malaysian primate population is not an exception to the ongoing decline of the global primate population due to human activities such as land conversion for oil palm plantations (Meijaard & Nijman, 2020). Despite the anthropogenic activities, ongoing monitoring efforts are still the practical means to assess changes in wildlife population trends. This is particularly important for Colobine monkeys in determining their population size and distribution. This information will be critical in developing conservation plans of the targeted species such as silvered langur to increase their population size (Revoro et al., 2015; Matsuda et al., 2020). The IUCN Red List has listed *Trachypithecus cristatus* (silvered langur) as a rare species that is categorised as Vulnerable (VU) due to poaching activities by local people (Chivers & Davies, 1979; Meijaard, & Nijman 2020).



**Figure 1.** Silvered langur was found at Sibuti WS headquarters.

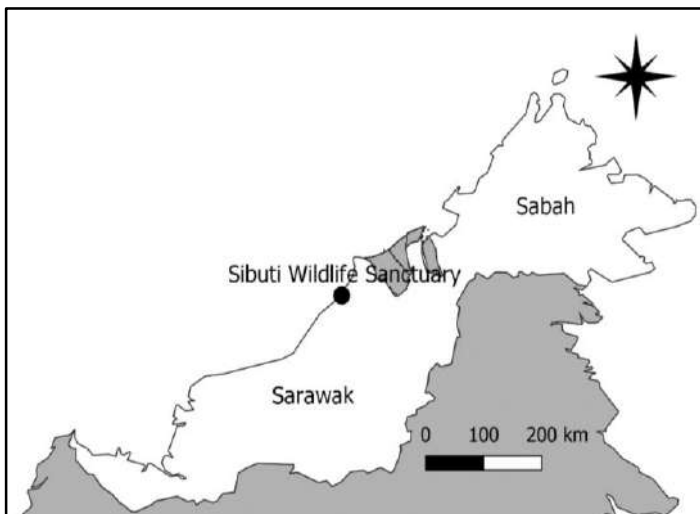
A silvered langur, also known as a silver leaf monkey (Figure 1), has unique characteristics such as a silver spike on its head and straight whiskers which differentiate them from other langur species. Silvered langur are also known as folivorous primates because their diet comprises 91% of leaves and 9% of fruits (Hock & Sasekumar, 1979). Diet studies on silvered langur in Bako National Park, Sarawak recorded at least 10 species of plants which include *Buchanania* sp., *Vitex pubescens*, *Pandanus* sp., *Nephrolepis bisserata*, *Dillenia suffruticosa*, *Cerriops tagal*, *Pongamia pinnata*, *Calophyllum inophyllum*, *Hibiscus tiliaceus*, and *Barringtonia asiatica* (Laman et al., 2007; Wan-Azman, 2017). This unique langur can be found in the riparian and mangrove forests (Meijaard & Nijman, 2020). Silvered langur can be found in Sumatra, Natuna Island and Borneo (Harding, 2010).

In Malaysian Borneo, specifically Sarawak, silvered langur has been recorded in many protected areas, as listed in Figure 2 (e.g. Laman et al., 2007; Vun et al., 2011; Shuib et al., 2012; Kombi & Abdullah, 2016; Phillipps & Phillipps, 2016; Khan et al., 2017). These include Bako National Park (Bako NP), Kuching Wetlands National Park (Kuching WNP), Kampung Pulau Salak (Salak KP), Santubong National Park (Santubong NP), Permai Resort Rainforest (Permai RR), Samunsam Wildlife Sanctuary (Samunsam WS), and Maludam National Park (Maludam NP). Although Sibuti Wildlife Sanctuary (Sibuti WS) is along a coastal



**Figure 2.** Location of Silvered Langur that has been recorded in Sarawak (e.g. Laman et al., 2007; Vun et al., 2011; Shuib et al., 2012; Kombi & Abdullah, 2016; Philipps & Philipps, 2016; Khan et al., 2017)

area where a majority of silvered langurs are recorded, information on their distribution at Sibuti WS remained unclear. The only known information is from visitors' online blogs. Herein, we present the first report on this species, along with a note on their diet from the observation recorded at Sibuti WS.



**Figure 3.** Location of survey site which covered mangrove forests at Sibuti Wildlife Sanctuary, Bekenu, Miri, Sarawak

Sibuti WS is one of the five wildlife sanctuaries in Sarawak (Samunsam WS, Lanjak Entimau WS, Pulau Tukong Ara-Banum WS and Sungai Jelangai WS) and was gazetted on 29th May 2000, covering 678 hectares. It is located in the Miri district or, to be more specific, in the Bekenu area, about 51.8 km away from Miri town (or about 1-hour drive from Miri). According to Shah et al., (2016), the sanctuary is mostly covered by a riverine mangrove forest, which is dominated by *Bakau minyak* (*Rhizophora apiculate*). Sibuti WS is a natural stopping area for migratory birds such as the stork-billed kingfisher (*Pelargopsis capensis*) and olive-winged bulbul (*Pycnonotus plumosus*). Sibuti WS personnel also highlighted that proboscis monkeys (*Nasalis larvatus*) could be occasionally spotted, although the exact microhabitat of the animal within the sanctuary remains undetermined. Besides birds and the possible proboscis monkey record, not much is known about wildlife in the sanctuary. Despite being unexplored, Sibuti WS was previously reported to have undergone anthropogenic activities (Saifullah et al., 2014; Shah et al., 2016). Such observation highlights the importance of continuous diversity assessment to document and assess the wildlife population trend in the sanctuary.

## Methodology

Surveys were conducted from 11<sup>th</sup> to 16<sup>th</sup> July, 2018 to record the population size and foraging activities. Surveys were done around the sanctuary's headquarters (N 4°2'4.023" E 113°47'19.798), and boat surveys were conducted along Sibuti River (N 4°1'18.1812" E 113°47'1.8494") and Kelulit River (N 4°3'29.466" E 113°50'39.094) for five consecutive days. For boat surveys, boats were slowed down if silvered langur was sighted, to record their population size and their diet. Surveys were conducted with the assistance of Sibuti WS staff to locate and record silvered langur's diet at Sibuti WS. Surveys were performed twice per day; 6.30 am to 9.00 am and 12.00 noon to 5.00 pm. These periods were chosen based on previous reports of the active and foraging time for silvered langur (Laman et al., 2007; Wan-Azman, 2017).

## Result

Within five days of sampling, the silvered langur was only sighted on the first and the last day of the survey at the Sibuti WS headquarters. A social troop of silvered langur was seen during the daytime on the first and the last day. Both observations suggest that they came from the same group consisting of 20 to 30 individuals. From the observations, their diet comprises of unripe fruit from *Simpoh air* (*Dillenia suffruticosa*), fig plants (*Ficus* sp.) and fruit from family

Leguminosae. The boat survey at Sibuti and Kelulit rivers only found a few individuals of non-targeted species, which is the long-tailed macaque (*Macaca fascicularis*).

## Discussion

Unlike other sites where silvered langur was recorded, in Sibuti WS the silvered langurs quickly leaped away once they detected human presence. In contrast, in Bako NP, the silvered langur was already habituated to visitors coming into the park. However, they produce a warning sound whenever they feel threatened, as they can make 13 different vocalisations such as warning, conflict, fear and others (Harding, 2010). In terms of food supplies, in Bako NP the silvered langur has various food sources such as *Ceriops tagal*, *Pongamia pinnata*, *Calophyllum innophyllum*, *Hibiscus tilaceus* and others (Laman et al., 2007; Wan-Azman, 2017). Meanwhile in Sibuti WS the silvered langur may have limited variety of food sources compared to Bako NP. As observed in Sibuti WS and compared to those reported in Bako NP, the silvered langur from both sites preferred to consume the *simpoh air* plant as their main dietary intake (Wan-Azman, 2017). Additionally, there are two types of dietary intake namely *Ficus* sp. and seeds from the family Leguminosae ingested by silvered langur in Sibuti WS, which was not reported in Bako NP (Laman et al., 2007). *Ficus* sp. from the family Moraceae has elements of flavonoids, sugar, vitamin A and vitamin C, which may boost the silvered langur's immune system (Somasekhar et al., 2013). In addition, it is a good food source as it may help prevent constipation (Chan et al., 2017). Seeds or legumes from the family Leguminosae provide a range of essential nutrients such as protein, low glycemic index carbohydrates, dietary fibre, minerals and vitamins (Cakir et al., 2019). Legumes are also composed of high protein content compared to other cultivated plants because of the nitrogen-fixation bacteria that live in the nodule of legume roots (Kouris-Blazos & Belski, 2016). The presence of derived bioactive peptides in legumes may further add to their food quality (Lopez-Barrios et al., 2014; Ortiz-Martinez et al., 2014). The young leaves and seed-based diets also provide additional nutritional advantages as young leaves provide a good source of mineral and seeds can serve as an energy source (Hanya & Bernard, 2015). This finding also highlights the vital role of primates in seed dispersal to sustain the forest ecosystem.

## Conclusion

Silver langur's dietary intake in Sibuti WS is slightly different from the populations in Bako NP due to the limited food sources. Therefore, these langurs put extra effort in their foraging activity to get enough food supplies. Their diet comprises *simpoh air*, *Ficus* sp. and seeds from the family Leguminosae. These wild nutritional foods have benefited them in surviving at Sibuti WS. An additional long term study that focuses on their population structure and trend is important to better manage them in Sibuti WS.

## Acknowledgement

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## Research Article

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# Highly Versatile, Non-Invasive Method for Collecting Buccal DNA from Free-Ranging Non-Human Primates

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## Abstract

Non-invasive techniques for collection of DNA samples of suitable quality and quantity are important for improving the efficiency of genetic wildlife research. The development of a non-invasive method for collection of DNA samples from wild stump-tailed macaques (*Macaca arctoides*) is described herein. Sterilized polyester rope was cut into 10 cm pieces, which were then soaked in a 20% sugar solution to bait individuals. Rope swabs were immediately collected and transferred to a lysis buffer solution after subjects had picked up, chewed and discarded them. DNA was later extracted from the buffer. Quantitative real-time PCR and both allelic dropout and genotype failure rates were used to compare the quantity and quality of the buccal DNA samples to those of intestinal slough cell DNA samples collected from freshly dropped feces. The buccal samples yielded significantly more DNA ( $27.1 \pm 33.8$  ng/ $\mu$ L) than did the fecal samples ( $11.4 \pm 15.4$  ng/ $\mu$ L) and exhibited lower allelic dropout and genotyping failure rates for the 10 autosomal microsatellites investigated. Buccal cell collection was also simple, inexpensive, reliable and less time-consuming compared to fecal sampling. Thus, this method should facilitate genome-wide studies of non-human primates and other wildlife species.

**Keywords:** Non-invasive DNA collection, Microsatellite markers, Quantitative real-time PCR, Allelic dropout

## Introduction

Wildlife, including non-human primates, has been subject to genetic analyses in a wide variety of research fields, such as evolutionary biology (e.g., Liu et al., 2020; Rogers et al., 2019; van der Valk et al., 2019; Williams et al., 2020),

population genetics (e.g., de Manuel et al., 2016; Liu et al., 2018; Nater et al., 2017), phylogeography (e.g., Bunlungsup et al., 2016; Yao et al., 2017), pedigree analysis (e.g., Snyder-Mackler et al., 2016), and conservation biology (e.g., Lynn et al., 2016), using a variety of DNA markers. Mitochondrial DNA (mtDNA), for example, is generally used for investigating maternal relationships and phylogeography (Liedigk et al., 2015), whereas Y-chromosome genes of mammals are used to investigate paternal relationships and male dispersal (Tosi et al., 2000; Tosi et al., 2002). Meanwhile, autosomal markers, such as microsatellite and single-nucleotide polymorphism (SNP) markers, are often used to investigate population genetics and genomic diversity (Chakraborty et al., 2015; Svardal et al., 2017).

As a result of recent advances in DNA analysis technology and growing concerns over animal welfare, genetic studies of wildlife frequently use DNA samples that have been collected by non-invasive means (Lynn et al., 2016). For example, DNA samples have been collected from egg shells (herring gull, *Larus argentatus*; Egloff et al., 2009), blood-fed mosquitos (Ejiri et al., 2011), koala feces (*Phascolarctos cinereus*; Wedrowicz et al., 2013), and bug-bite blood (Sumatran rhinoceros, *Dicerorhinus sumatrensis*; Rovie-Ryan et al., 2013). DNA has similarly been collected non-invasively in genetic studies of wild, non-human primates, for example, from trapped hairs (white-headed langur, *Trachypithecus leucocephalus*; Wang et al., 2016), semen (Japanese macaques, *Macaca fuscata*; Domingo-Roura et al., 2004), urine (Japanese macaques; Hayakawa & Takenaka, 1999), and saliva (mountain gorillas, *Gorilla beringei beringei*, and Grauer's gorillas, *Gorilla beringei graueri*; Smiley et al., 2010; Chimpanzee, *Pan troglodytes*, Inoue et al., 2007). Among these DNA resources, fecal samples have been most commonly used (Chiou & Bergey, 2018; Hernandez-Rodriguez et al., 2017; Orkin et al., 2020). However, fecal samples generally yield low quantities of low-quality DNA, and even though the markers used in some studies (e.g., mtDNA markers) can be amplified successfully owing to their high copy numbers (Bunlungsup et al., 2016), enormous efforts are required when examining nuclear markers (Navidi et al., 1992; Taberlet et al., 1996). One major problem with using fecal DNA samples for nuclear genotyping is allelic dropout, a phenomenon in which one of two autosomal alleles is not amplified by PCR, causing heterozygous genotypes to be misinterpreted as homozygous (Pompanon et al., 2005; Tebbutt & Ruan, 2008). Allelic dropout is problematic in paternity and kinship analyses using autosomal microsatellites (Vigilant et al., 2001).

As such, development of non-invasive DNA sampling methods that allow researchers to obtain large quantities of high-quality DNA samples with low levels of contamination is needed. Buccal cell collection methods have been reported previously; such as collecting sugarcane wedges or pith of terrestrial herbaceous vegetation after their chewing by wild bonobos (*Pan paniscus*, Hashimoto et al., 1996; Ishizuka et al., 2018), taking oral swabs from anesthetized mountain and Grauer's gorillas (Smiley et al., 2010), and attaching ropes to saliva-collecting devices near free-ranging Tibetan macaques (*Macaca thibetana*, Simons et al., 2012). Collecting DNA from wedges of sugar cane or other plants is a non-invasive method that does not require manipulation of animals and is thus applicable to other study sites with appropriate modification according to certain factors, such as the environment of the study and the feeding patterns of subjects. However, methods that require specialized equipment takes time and cost to produce the device. Especially in the wild, using specific devices is less flexible when collecting multiple samples from several monkeys at once due to mobilities. Such methods were inapplicable to the stump-tailed macaques at our study site in Thailand because of the difficulty in preparation and storage of the bite materials. Thus, we designed an alternative method for collecting buccal cells as reported here.

Herein, a non-invasive method for collecting buccal DNA samples using rope swabs is described as simple, reliable, inexpensive and less time-consuming than other commonly used methods. To test the effectiveness of this method, two experiments were conducted. The first was a quantitative comparative test of host DNA in 41 fecal and 41 buccal DNA samples randomly selected using real-time PCR. In addition, gel electrophoresis ("gel tests") were also used to quantitatively test DNA samples cheaply and conveniently, and their results were compared with those of costlier real-time PCR to verify their accuracy. The second experiment was a qualitative comparison based on allelic dropout and genotype failure rates in 30 fecal and 30 buccal DNA samples selected using gel tests.

## Materials and methods

### *Study site*

The present study was conducted at the Khao Krapuk Khao Taomor Non-Hunting Area, Phetchaburi Province, Thailand (12°47'59.2" N, 99°44'31.1" E), which harbours five free-ranging groups of stump-tailed macaques (*Macaca arctoides*). There are five groups: Ting group, 115 individuals; Nadam group,

91 individuals; Third group, 71 individuals; Fourth group, 75 individuals; Wngklm group, 43 individuals (Toyoda et al., 2017). The monkeys here are habituated to observer AT since 2015. This survey area is mainly a mountainous area composed of secondary forests and bamboo forests, and open areas coexist including temples and houses of local people. The moving area of monkeys was divided between north and south by large roads, and food provisioning by locals or visitors was occasionally observed along the road or at temple grounds. As for environmental conditions, the mean annual temperature and annual rainfall are 27°C and 1070 mm, respectively, based on data at the nearby national park named Keang Krachan National Park, about 30km from this study site (Wijitkosum, 2012). This site consists primarily of secondary forests, including stands of bamboo and agricultural areas.

#### *Collection and extraction of DNA samples*

Buccal cells were collected using baited ropes (hereafter *rope swabs*). Polyester ropes (6 mm in diameter; Takagi Corporation, Kagawa, Japan, JAN code: 4943 956 261 513) were cut into approximately 10 cm pieces, autoclaved, and dried to avoid contaminations (Figure 1). To bait individuals, the rope swabs were soaked in a 20 % sugar solution (70 g cane sugar dissolved in 350 mL distilled water) for at least 30 min, and then scattered on the open ground where the monkeys were found. After being chewed (Figure 2) and discarded by a monkey, the rope swab was quickly collected and transferred to a 5 mL carrying tube containing 3 mL lysis buffer (0.5 % (w/v) in SDS, 100 mM EDTA pH 8.0, 100 mM Tris-HCl pH 8.0, and 10 mM NaCl) (Hayaishi & Kawamoto, 2006). To compare the quantity and quality of the buccal DNA with that of other commonly used DNA sources, intestinal slough cells from freshly dropped fecal samples were also collected. A sterile cotton bud, which was soaked in 2 mL lysis buffer, was used to swab the surfaces of feces, following the protocol of Bunlungsup et al. (2016). To increase DNA yields, the surfaces of the feces were swabbed at least three times.

The buccal and intestinal cells that were transferred to the lysis buffer were kept at room temperature for at least five months until DNA extraction. DNA was extracted following the procedure of Kawamoto et al. (2013). Potential PCR inhibitors were removed by adding 600 mg of hydrolyzed starch (Wako, Osaka, Japan) to 1.5 mL of lysis buffer per sample. The samples were incubated at 36°C for 10 min, and then centrifuged at 1000 ×g for 15 min. Finally, 750 µL of each supernatant was processed using a commercially available DNA clean-up system (Wizard SV Gel and PCR Clean-Up System; Promega, Madison, WI, USA), and the DNA was finally eluted with 50 µL pure

water. This study including fieldwork and lab work was conducted from September 25<sup>th</sup>, 2015 to June 15<sup>th</sup>, 2017, and 74 fecal samples and 579 buccal samples were collected.

#### *DNA quantification*

The amount of host DNA was quantified by quantitative real-time PCR (Morin et al. 2001). Forty-one DNA samples extracted from buccal and 41 from fecal samples were selected randomly from all of the extracted DNA samples. The real-time PCR method was used because both the buccal and intestinal DNA samples were contaminated with other exotic DNA sources, such as bacteria, eukaryotic parasites and dietary materials (e.g., plants, insects, or small animals), which could not be differentiated using conventional spectrophotometry. The sequences of the real-time PCR primers and *c-myc* probe were 5'-GCCAGAGGAGGAACGAGCT-3' (CMYC\_E3\_F1U1), 5'-GGGCCTTTTCATTGTTTTCCA-3' (CMYC\_E3\_R1U1), and 5'-FAM-TGCCCTGCGTGACCAGATCC-TAMRA-3' (CMYC\_E3\_TMV), respectively (Morin et al., 2001). Real-time PCR was performed using a StepOnePlus real-time PCR System (Thermo Fisher Scientific, Waltham, MA, USA), and each 20  $\mu$ L reaction contained 2  $\mu$ L DNA template, 1 $\times$  TaqMan Fast Advanced Master Mix (Thermo Fisher Scientific), 250 nM probe, and 900 nM of each primer. In addition, the PCR amplification conditions included an initial denaturation step of 95 °C for 20 s, followed by 45 cycles of 95 °C for 1 s and 60 °C for 20 s. Host DNA quantity (concentration) was determined using a standard curve made by a duplicate set of DNA with known quantity. The standard set was made from DNA extracted from the blood of a northern pig-tailed macaque (*Macaca leonina*) reared in the Primate Research Unit, Chulalongkorn University (Bangkok, Thailand). The DNA was quantified using a spectrophotometer and diluted to 10 ng/ $\mu$ L, 2.5 ng/ $\mu$ L, 625 pg/ $\mu$ L, 156 pg/ $\mu$ L, 39.1 pg/ $\mu$ L, and 9.8 pg/ $\mu$ L with deionized water. The mean DNA yields obtained from the buccal and fecal samples were compared using the Wilcoxon rank sum test in R Ver. 3.4.2 (R Core Team, 2016).

Real-time PCR provides an accurate host DNA concentration for each DNA sample, and thus was appropriate for comparing the DNA yields of the buccal and fecal samples. However, real-time PCR analysis is expensive. Therefore, to select suitable samples for microsatellite genotyping, the usability of the 82 DNA samples was roughly screened using conventional PCR and agarose gel electrophoresis following the procedure of Kawamoto et al. (2013) and Ball et al. (2007) (gel electrophoresis). For the gel test, the *c-myc* gene was PCR-amplified in 12.5  $\mu$ L reactions of 1  $\mu$ L template DNA, 1 $\times$  PCR Buffer for KOD FX,

400  $\mu$ M dNTPs, 0.25 U KOD FX (Toyobo, Osaka, Japan), and 0.015 pM of both the forward and reverse real-time PCR primers, using the following conditions: initial denaturation step of 94 °C for 2 min, 45 cycles of 98 °C for 10 s, 58 °C for 30 s, and 68 °C for 30 s. The resulting amplicons were electrophoresed on 2% agarose-TAE gels, stained with SYBR Safe DNA Gel Stain (Thermo Fisher Scientific), and visualized using UV transilluminators to determine the intensity of the target band. To estimate the amount of buccal and intestinal DNA, a series of human placental DNA (Sigma-Aldrich, St. Louis, MO, USA) at concentrations of 500, 300, and 100 pg/ $\mu$ L were used as reference controls. When the luminous intensity of a PCR product was > 300 pg/ $\mu$ L of the control, the sample was considered to have sufficient yield template DNA for microsatellite genotyping and was used in the next step for microsatellite amplification. We used human placental DNA as reference following Kawamoto et al. (2013) that was different from the *Macaca leonina*'s DNA used in the real-time PCR. This was because of the difference of availability of the DNA standard in Japan and Thailand, and the difference of the species was considered not to affect the substantial results (Smith et al., 2002). The accuracy of the real-time PCR analysis and gel test screening were compared using the Wilcoxon rank sum test with continuity correction.

#### *DNA quality analysis*

To determine DNA quality, the 30 paired buccal and intestinal DNA samples that passed the gel test were randomly selected for microsatellite genotyping. Ten microsatellite loci were amplified using a modified version of the two-step multiplex method (Toyoda & Malaivijitnond, 2018). During the first step of PCR, all microsatellite loci were amplified in a single 20  $\mu$ L reaction that included 1  $\mu$ L template DNA. During the second step, the 10 loci were divided into three subsets and were amplified in 12.5  $\mu$ L multiplex PCR reactions that each included 1  $\mu$ L of non-diluted amplicon from the first multiplex PCR reaction. The PCR thermocycling conditions were the same as those from the gel test, except that 35 cycles were used for the first PCR, and 45 for the second PCR. Allelic dropout rates and false allele rates were calculated using PEDANT Ver.1 (Johnson & Haydon, 2007, available from <http://sites.google.com/site/pcdjohnson/home/pedant>). In the programme, the results of two independent PCR products per sample per locus were used to estimate the allelic dropout and false allele rates. The allelic dropout and false allele rates of the buccal and fecal sample DNA were compared using the Wilcoxon signed-rank test ( $p < 0.05$ ) in R. In addition, the genotype failure rate (a phenomenon in which the peak of an allele is detected at extremely low levels or is not detected) of each locus was calculated based on the duplicated



PCR results, and the rates of genotype failure of the buccal and fecal DNA samples were compared using the Wilcoxon signed-rank test ( $p < 0.05$ ) in R.



Figure 1. Rope swabs cut into 10cm length and 3ml of lysis buffer in 5ml tube

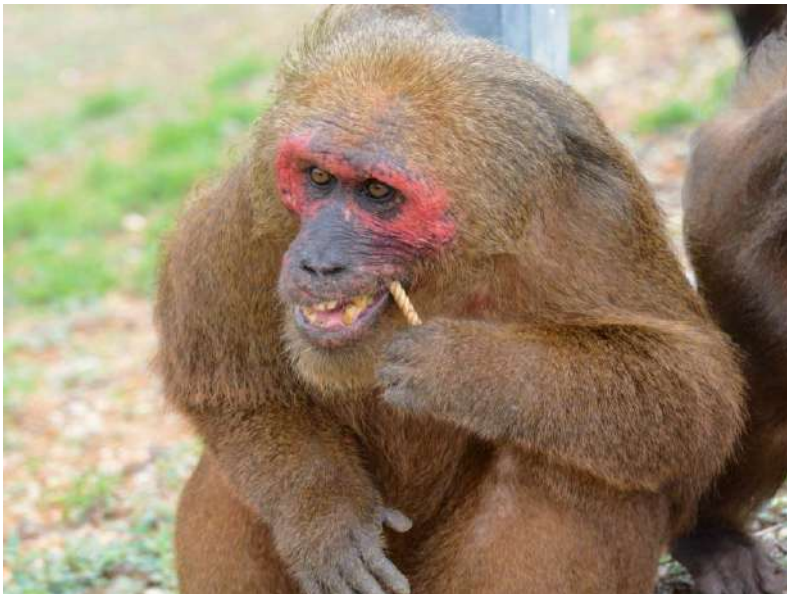
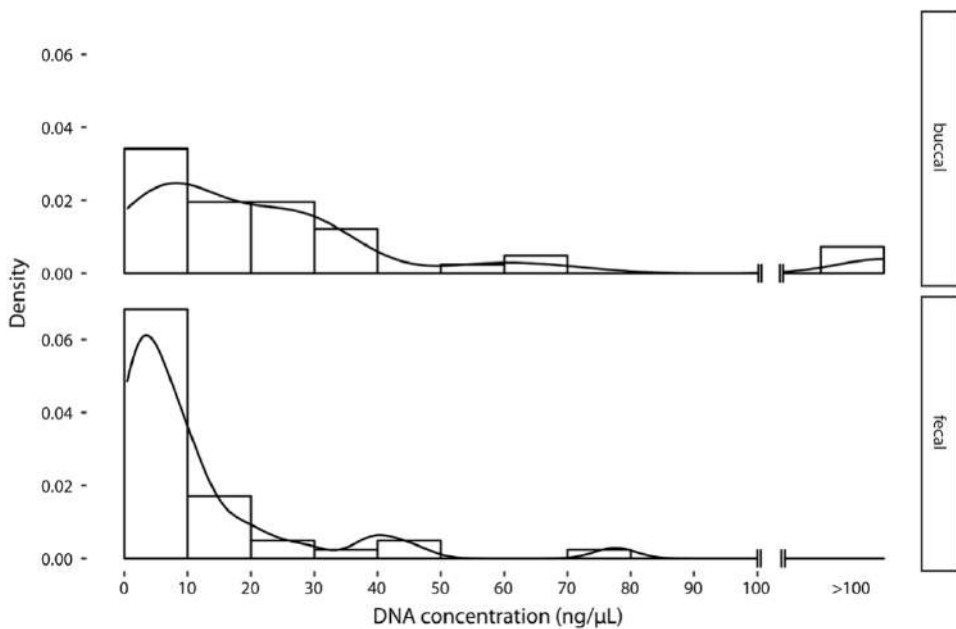


Figure 2. Monkey chewing a rope swab

## Results

### *DNA quantity*

Analysis of the 82 DNA samples (41 buccal and 41 intestinal DNA samples) revealed that the buccal samples yielded significantly more host DNA ( $27.1 \pm 33.8$  ng/ $\mu$ L) than did the fecal samples ( $11.4 \pm 15.4$  ng/ $\mu$ L;  $W = 473$ ,  $P < 0.001$ ). Although 68% (28/41) of intestinal samples yielded concentrations less than 10 ng/ $\mu$ L, only 29% of buccal samples produced such low concentrations (12/41) (Figure 3).



**Figure 3.** Frequency of buccal and fecal DNA in each DNA concentration zone. Although many fecal samples are dense in the low concentration zone, meaning that the sampling efficiency is not good, buccal samples shows a gentle peak overall, indicating that samples with high concentration can be more easily obtained.

The determination by the gel test was possibly made the presence/absence of the band. Of the 41 fecal and buccal DNA samples tested, 22 (53.7%) and 35 (85.4%) met the criterion for sufficient yield ( $\geq 300$  pg/ $\mu$ L), respectively. The concentration of host DNA that passed and failed the gel tests as measured by

real-time PCR was a significant difference ( $W = 991$ ,  $p < 0.01$ ), indicating that either real-time PCR or the gel test can be used for DNA quantification.

#### *DNA quality*

For the 30 monkeys whose samples passed the gel test, the allelic dropout rate of the 10 microsatellite loci was significantly lower for the buccal (0.00%, range: 0.00 -  $6 \times 10^{-6}$ %) than for the fecal DNA samples ( $18.12 \pm 16.12\%$ , range: 0.00-55.96%; Wilcoxon signed-rank test,  $V = 44$ ,  $p < 0.01$ ; Table 1). Estimated dropout rates were used to calculate the amount of repetition necessary for accurate results at the 99.99% certainty level (Morin et al., 2001). At least 6 repetitions were needed for fecal sample analysis to produce reliable genotype data, whereas one repetition was sufficient for buccal samples.

Similarly, the genotyping failure rate had significantly lower for buccal DNA samples ( $2.70\% \pm 3.88$ , range: 0.0-13.3 %) than for fecal DNA samples ( $35.67\% \pm 15.35$ , range: 18.3-65.0%; Wilcoxon signed-rank test,  $V = 55$ ,  $p < 0.01$ ), although the rate was variable among the loci examined (Table 1).

**Table 1.** Allelic dropout and genotype failure rates of 10 microsatellite loci for fecal and buccal DNA samples of stump-tailed macaques in Khao Krapuk Khao Taomor.

Loci	Allelic dropout rate (%)		Genotype failure rate (%)	
	Fecal	Buccal	Fecal	Buccal
D3S1768	10.54	0.00	38.33	0.00
D6S2793	25.00	0.00	58.33	2.00
D7S2204	8.80	0.00	31.67	13.33
D8S1106	13.04	0.00	45.00	0.00
D11S2002	0.00	0.00	65.00	1.67
D13S765	29.20	0.00	23.33	0.00
D14S306	0.00	0.00	18.33	1.67
D17S1290	28.45	0.00	33.33	5.00
D18S537	55.97	0.00	25.00	3.33
D19S582	10.21	0.00	18.33	0.00

## **Discussion**

### *Advantages from sampling point of view*

When fecal samples are used as genetic resources, the success in genotyping depends on various conditions; e.g. the temperature at the time of sample collection, sample desiccation (Nsubuga et al., 2004), and salt concentration (Hofreiter et al., 2001), and skill of the collectors, as most researchers experienced. Using the rope swab method in our study, the collection of high-quantity and quality DNA samples would be possible without much training, providing a more versatile option that is not dependent heavily on the level of

experience of the sample collector. Our rope swab method may also be useful for collecting samples from infants. Indeed, our method was capable of collecting samples from infants aged 2-3 weeks, even though the feces of infants were often soft, diarrhea-like or very small and often difficult or almost impossible to collect. Thus, we strongly believe that our method would be a powerful alternative to overcome the difficulty of collecting fecal samples from infants which are indispensable for genetic analysis such as paternity tests. The rope swab method is also less time consuming than fecal collection. Since the quality of DNA samples cannot be checked in-situ study, multiple fecal samples must be collected to ensure collection of an adequate sample from the target animals. On the other hand, most of buccal samples provided usable DNA, and thus, fewer specimens need to be collected from each animal. Additionally, to collect fecal samples, researchers must patiently follow the targeted animals until they defecate, which is time-consuming. Therefore, the rope swab method presented in this study has great potential to save time and mitigate these factors.

#### *Advantages from analysis point of view*

Our study showed that the rope swab method is more effective, in terms of both quantity and quality of recovered DNA, compared to extraction from fecal samples. The rope swab method yielded up to 2.4 times more host DNA than did fecal samples and exhibited much lower allelic dropout and genotype failure rates, indicating that our method possibly facilitates genotyping analyses with fewer repetitive PCR trials, which could save time, labour and money. This is because low DNA quantity increases genotyping errors that affect the reliability of genotyping in microsatellite analysis (Taberlet et al., 1999), and thus repeating experiments for each locus and extract is recommended (Goossens et al., 1998).

#### *Important notice using rope swab method*

Although our method would be useful, there are several cautionary notes while collecting samples. Firstly, in the initial phase, monkeys may not chew on the rope swabs. In this case, a habituation period using fruit juice instead of sugar water to increase the attractiveness of the swab rope is recommended. From experience, however, it seems better to switch to sugar water during the sample-collection phase. Genotyping results were not stable when using DNA samples collected with orange juice, probably due to acid or other chemical compounds present in the fruit juice.

Secondly, the collection of samples shortly after monkeys have consumed food should be avoided, especially at provisioned sites or when targeting captive animals, as fruits are the main food items given and contain acids or other chemical compounds that may inhibit PCR. Complex polysaccharides possibly originating from vegetable material in the diet are also considered potential PCR inhibitors (Monteiro et al., 1997). Thus, time of sampling may affect the quality of the sample rather than the duration for which the monkey chews the rope.

Thirdly, adjustments to the soaking time of the rope swab in the sugar-water solution and the concentration of sugar according to the condition of the subject animals or study site may be needed. Extended soaking times or high sugar concentrations could encourage monkeys to chew the rope swabs for longer periods, which may lead to greater DNA yields. However, the potential downside of a longer chewing period is that the target monkeys may move while chewing, making retrieval of the rope swabs more difficult for the researcher. Although some individuals spent significant time chewing the swabs and occasionally broke them into small fragments, no monkeys accidentally ate the rope swabs during this study period, demonstrating their safety in application.

Fourthly, the rope swabs should be well-distributed among the troop, otherwise higher-ranking males will take multiple ropes at once. When samples from subordinate individuals are needed, spreading the rope swabs over a wide area to attract high-ranking individuals, and then casting some swabs to the target individual may be an effective strategy.

Lastly, because this method requires that the rope swabs be provided to the animals, it may not be suitable for use with non-habituated, wild animals. This method also cannot be used in research sites where access to wildlife or provisioning is prohibited. Since this method involves material once contained in the mouths of animals, researchers must be aware of the possibility of touching saliva to prevent zoonosis (e.g., Kelesidis & Tsiodras, 2010). When conducting behavioural observation at the same time, the possibility of influencing the behaviour of the target animals must also be considered. Ultimately, the applicability of this method will depend on the specific needs and conditions of the research.

Furthermore, we must note the standard range of quantitative real-time PCR. In this study, the standard range of quantitative real-time PCR could not cover

the sample concentration range due to the fact that the quantity of DNA was extracted at a higher concentration than our assumption. We followed the protocol of Wizard SV Gel and PCR Clean-Up System and used 50  $\mu\text{L}$  of water for the final elution step, though 200  $\mu\text{L}$  is used in Morin et al. (2001). This difference of the final elution volume should have resulted in the higher concentration of DNA both from buccal and fecal samples in our study.

#### *Future possibility of application*

The successful DNA collection and genotyping of *M. arctoides* using our method can be further applied to different conditions as long as researchers pay attention to risks and take precautions. For example, for populations kept in captive conditions at research institutions or individuals kept in cages in laboratories are the best conditions. Also, for provisioned or well-habituated free-ranging primates such as populations living near temples which are widely seen in most Southeast Asian countries. This is a very useful method for researchers who have to obtain samples from specific individuals in a limited research period in the wild. Furthermore, with some modifications, this method can be applied for hormone and veterinary analysis (e.g., detecting a specific virus in the saliva; Musso et al., 2015; Huff et al., 2003). The non-invasive buccal cell collection method described by this study may further facilitate animal population genomic studies in both captive and field environments. Further integration of genetic information with behavioural and ecological data is expected to provide more insights into *M. arctoides*, including genetic structure and socioecological characteristics such as reproductive strategy and kinship structure.

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## Review Article

# Secondary Metabolites of the Soft Coral *Lobophytum pauciflorum*

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## Abstract

Secondary metabolites, derivatives of primary metabolites, are known for their biological activities. Marine organisms, especially marine invertebrates such as sponges, tunicates, soft corals, bryozoans, and nudibranchs are important source of secondary metabolites with diverse biological properties. Approximately 40,000 marine natural products have been identified from various marine resources. Soft corals are a group of invertebrates known for their production of a vast range of metabolites with great structural diversity. Among 39 genera of soft coral Alcyonacean, a total of eighteen different species of *Lobophytum* soft corals have been identified. Isolation of secondary metabolites from the genus *Lobophytum* is tremendously explored by researchers worldwide. This review compiles several secondary metabolites that have been isolated and published on the soft coral *L. pauciflorum*, including the compound structures and some notable bioactivity.

**Keywords:** *Lobophytum pauciflorum*, soft coral, secondary metabolite, biological activity

## Introduction

The ocean constitutes a rich source of biologically and genetically diverse marine organisms, as a result of harsh chemical and physical marine environments, such as cold temperature, high pressure, and dark conditions of the ocean (Nikapitiya, 2012). Secondary metabolites are metabolic products

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derived from primary metabolites. Although they are considered non-essential for the vegetative growth, secondary metabolites take up adaptive roles such as defence mechanism, signalling molecules, symbiosis, metal transport, competition, etc. Secondary metabolites have long been harvested for their beneficial properties such as antimicrobial, antitumor and growth promoter (Thirumurugan et al., 2018). Marine organisms produce a range of bioactive secondary metabolites which are found to be chemically and biologically distinct from terrestrial originated metabolites (Hassan et al., 2016). Secondary metabolites are produced by both macro and micro marine organisms as part of their defence strategies, signalling molecule with their environment and in response to the food chain (Hanif et al., 2019).

Soft corals are a vital component of the coral reef ecosystem, and are known to be a vast source of secondary metabolites exhibiting diverse biological properties (Amir et al., 2012). To date, eight natural products derived from marine organism have been approved as drugs, seven of which are still in use until now (Hanif et al., 2019). In recent years, soft corals have become a new source of novel marine bioactive compounds in drug development research (Changyun et al., 2008). The genus *Lobophytum*, comprising of more than 20 different species present in tropical and subtropical waters, is known to be a rich source of macrocyclic cembranoids and metabolites with significant biological activities, such as anti-viral, anti-inflammatory and anti-tumour properties (Yan et al., 2010, Putra et al., 2016). *Lobophytum pauciflorum* (Figure 1) is a common species in the shallow waters of the Indo-West Pacific, and is also one of the most abundant soft coral species in the coral reefs of southern Taiwan (Fan et al., 2005, Wessels et al., 2017). Similar to most soft corals, *L. pauciflorum* is gonochoric in nature, with dimorphic polyps (Wessels et al., 2017). The secondary metabolite profile of male and female *L. pauciflorum* have been found to be different, hence indicating a physiological difference between sexes (Fleury et al., 2006). Nonetheless, physiological difference at the secondary metabolite levels between the sexes is yet unclear (Zhao et al., 2013). Distinction in the expression of the immune system between sexes was identified as one of the factors which gives rise to physiological differences in bilateral animals (Wessels et al., 2017). The immune system may also govern the microbiome of the soft coral (Margulis and Fester, 1991). However, exposure of *L. pauciflorum* of both sexes to short-term environmental stress was found to have no significant effect on the microbial communities of males and females, thus indicating the resilient nature of the microbiome towards short-term environmental stress (Wessels et al., 2017). Hence, the

difference in secondary metabolites levels between male and female *L. pauciflorum* still remains unclear.



Figure 1. Underwater colony of *L. pauciflorum* (photo credit Kishneth Palaniveloo).

This review discusses on the secondary metabolites that have been isolated, extracted and published on the soft coral *L. pauciflorum* and structures of these compounds are presented. In addition, the biological potential of *L. pauciflorum* and the isolated secondary metabolites are also reviewed.

## Secondary Metabolites Isolated from *L. Pauciflorum*

### Diterpenes

Cembranoid diterpenes are fourteen carbocyclic ring structures with an isoprenoid skeleton (Rodrigues et al., 2019). Among the earliest investigations on *L. pauciflorum* was the investigation into the bioactivity of terpenes from the species from Japan. Isolation through repeated silica gel column chromatography revealed the presence of two cembranoid diterpenes of a 13-membered carbocyclic ring system, both with  $\alpha$ -methylene- $\gamma$ -lactone ring (Yamada et al., 1980a). Cembranolides containing a tricyclic system were also isolated by Japanese researchers. Similar to the previously isolated compounds, these compounds possessed the  $\alpha$ -methylene- $\gamma$ -lactone ring moiety as well, along with a trisubstituted epoxide and the common 14-membered carbocyclic ring. (Yamada et al., 1980a).

In 1983, Kinamoni et al. reported the isolation of cembranoids nephthenol (1) from the Red sea variety of *L. pauciflorum*. Nephthenol (1) was first isolated

and characterized in 1974 by Schmitz et al., from the soft coral *Nephthea* sp. collected from the Marshall Islands. The Australian variety of this soft coral species were discovered to produce the cembranes, 14-hydroxycembra-1,3,7,11-tetraene (2) and 15-hydroxycembra-1,3,7,11-tetraene (3) (Bowden et al., 1987). Later in the 1990s, an antipode, lobocalone (4) was reported to be isolated from *L. pauciflorum* from the Indian waters (Anjaneyulu & Rao, 1995). This compound was also isolated from the soft coral *L. caledonense* from the South China Sea by Su et al., in 1993, and from the gorgonian *Eunicea fusca* (Gopichand & Schmitz, 1978). In addition, a group of Fujisawa pharmaceutical companies in 1993 isolated four additional cembranoids (Anjaneyulu & Rao, 1995). The group also reported the isolation of six steroids from *L. pauciflorum*. Further studies on *L. pauciflorum* lead to the isolation of a 10-membered-ring diterpene, cyclolobatriene (5) of an Okinawan variety of the soft coral. Cyclolobatriene (5) was isolated together with the known compounds lobatriene (6), eunicol (7) and fuscol (8) (Govindam et al., 2012). Lobatriene (6) has been previously reported to be isolated from the soft coral *Sinularia flexibilis*, and eunicol (7) was previously reported from the same soft coral species and reported by Coll et al. in 1986. In this study, *L. pauciflorum* samples were extracted with methanol, and the methanol extract was gone through a silica-gel flash column chromatography, which uses 40 µm silica gel Hi-Flash column of size L (26 mm i.d. x 100 mm), and recycling HPLC with a LC-8A pump. The extracts of compounds 5-7 were colourless viscous oil, while compound 8 was in a colourless oil form. These compounds are of germacrene nature.

The secondary metabolites of the soft coral *L. pauciflorum* mainly comprise lobane-type compounds instead of cembranoid diterpenes. Previous records of lobanes being isolated from the genus *Lobophytum* has been reported by Raju et al. (1994) from the Andaman and Nicobar waters with the isolation of 15-nor-13-keto-8-elemene (9), 17,18-epoxyloba-8,10,13(15)-triene-16-ol (10) and loba-8,10,13(15)-triene-16,17,18-triol (11). The lobane 15-nor-13-keto-8-elemene (9) had been previously isolated from a gorgonian, *Eunicea fusca* (Raju et al., 1994) and was also reported from *L. pauciflorum* by Rao in 1990. The structures of compounds 1 - 11 are shown in Figure 2.

*L. pauciflorum* from the waters of Philippines yielded an additional four lobanes, lobatriene (6) that was oddly named as epoxylobatrienol, its acetate (12), lobatrienediol (13), its acetate (14), methoxy lobatetraene (15) and oxepin lobatrienol (16). Interestingly, the compound lobatetraene (15) was reported as fuscol (8), a compound being isolated from the gorgonian *Eunicea fusca* and re-isolated from the soft coral *L. microlobulatum* (Bonnard et al., 2010).



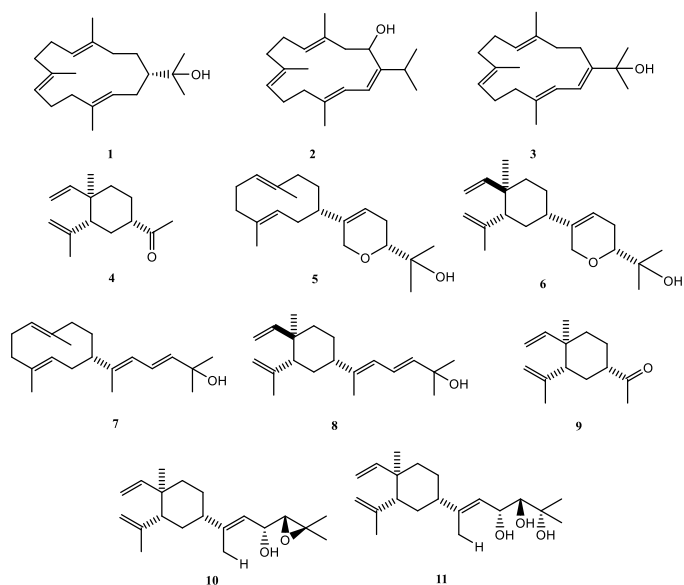


Figure 2. Lobane type diterpene and other derivatives isolated from *L. pauciflorum*.

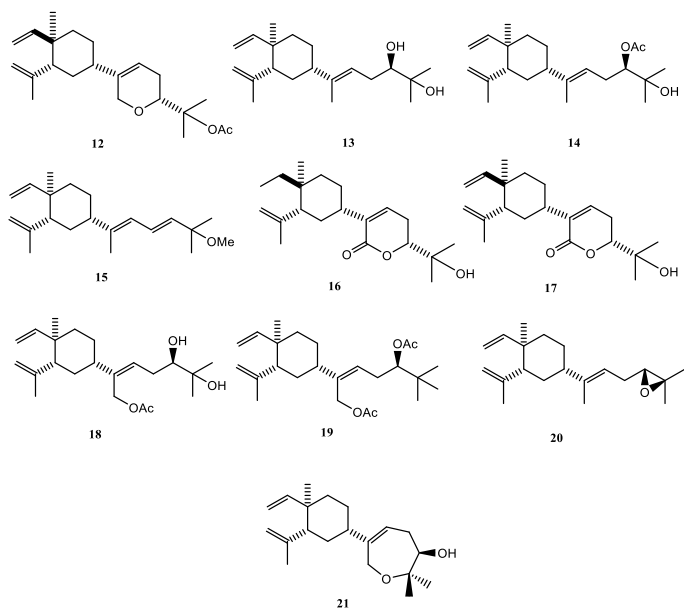


Figure 3. The structures of compounds 12 - 21.

The compounds lobatrienediol (13), lobatrienolide (17), lobatrietriol monoacetate (18), lobatrietriol diacetate (19) and lobatriene epoxide (20) were also isolated along with two compounds, lobatrienol (21) and lobatriene (6), previously reported from *Sinularia flexibilis*. These compounds were tested for their biological properties (Edrada et al., 2000). Structures of the isolated lobanes and other derivatives from *L. pauciflorum* are shown in Figure 3.

### Biscembranoids

Biscembranoids are constituents consisting of two cembranes linked together, and *L. pauciflorum* is reported to be a rich source of biscembranoids. In 2010, 27 iso-biscembranoids from *L. pauciflorum* in Chinese waters were reported. Lobophytone A (22) - Z<sub>1</sub> (48) was isolated from *L. pauciflorum* by Yan et al. between 2010 and 2011. The lobophytone were reported in four separate publications (Yan et al., 2010a; 2010b; 2010c; 2011). The first report involves lobophytone A (22) - G (28) (Figure 4), followed by lobophytone H (29) - N (35) (Figure 5), lobophytone O (36) - T (41) (Figure 6) and lobophytone U (42) - Z<sub>1</sub> (48) (Figure 3 and 4). In the final report, Yan and co-workers reported additional metabolites methyl sartortuoate (49) and nyalolide (50). The biscembranoids of this soft coral was suggested to be different than the usual tetraterpenoids due to the antipodal Diels-Alder cycloaddition between cembranoid-diene and cembranoid-dienophile. Several stereoisomers were also identified among the lobophytone. Lobophytone G (28) was suggested to be a stereoisomer of lobophytone F (27) (Yan et al., 2010a) and lobophytone L (33) is a stereoisomer of K (32). Lobophytone U (42) was similar to lobophytone A (22) and B (23), except for the presence of hydroxy moiety at C-33. Lobophytone V (43) likewise was similar to lobophytone A (22) but contained an additional oxygen atom and a methine group.

Similarly, lobophytone W (44) possessed an oxymethylene attached to C-27, while lobophytone X (45) lacked the hydroxy group in the compound. Lobophytone Z (47), on the other hand is a dehydroxylated derivative of lobophytone A (22), while lobophytone G (28) is chemically related to lobophytone W (44) with variations in rings B and C of the biscembranoid. Lobophytone Y (46) is structurally similar to methyl sartortuoate (49), while lobophytone J (31) is structurally related to the biscembrane nyalolide (50) that was reported by Yan et al. in 2011. Chemical structures of compounds 42-50 are shown in Figure 7.

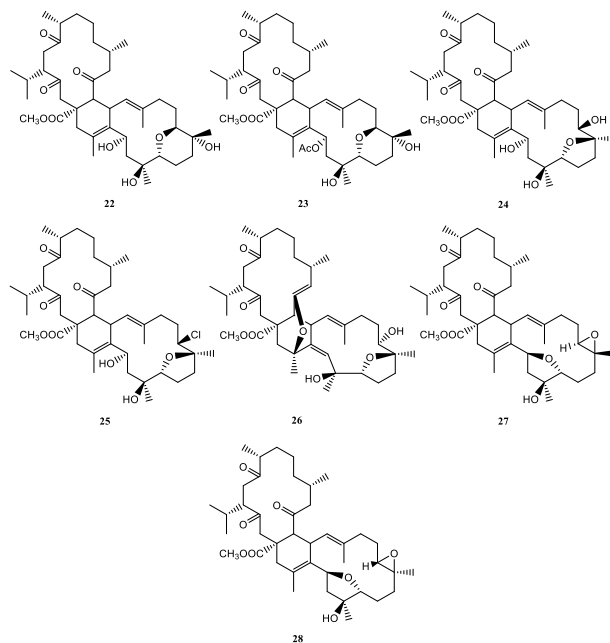


Figure 4. Biscembranoids lobophytone A (22) - G (28) from *L. pauciflorum*.

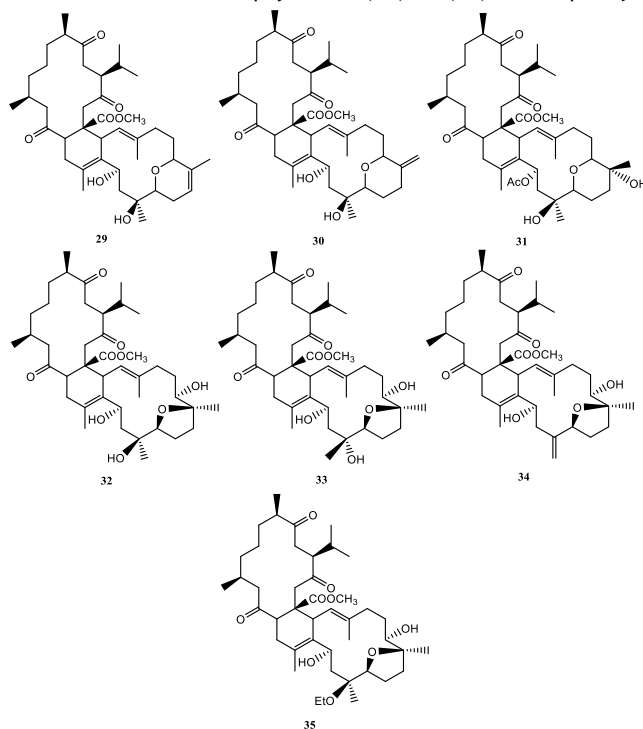


Figure 5. Biscembranoids lobophytone H (29) - N (35) from *L. pauciflorum*.

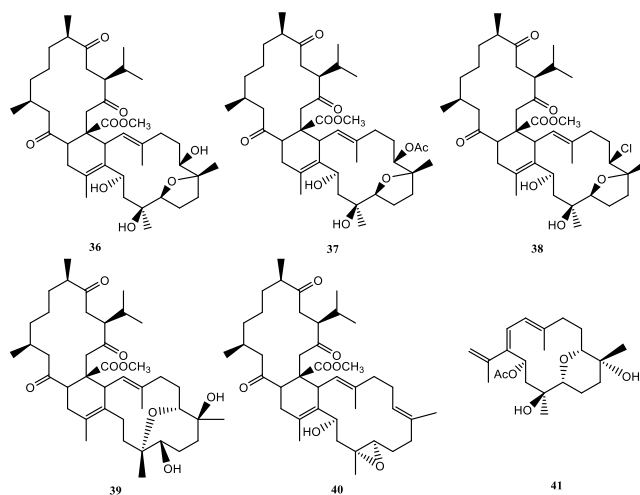


Figure 6. Biscembranoids lobophytone O (36) - T (41) from *L. pauciflorum*.

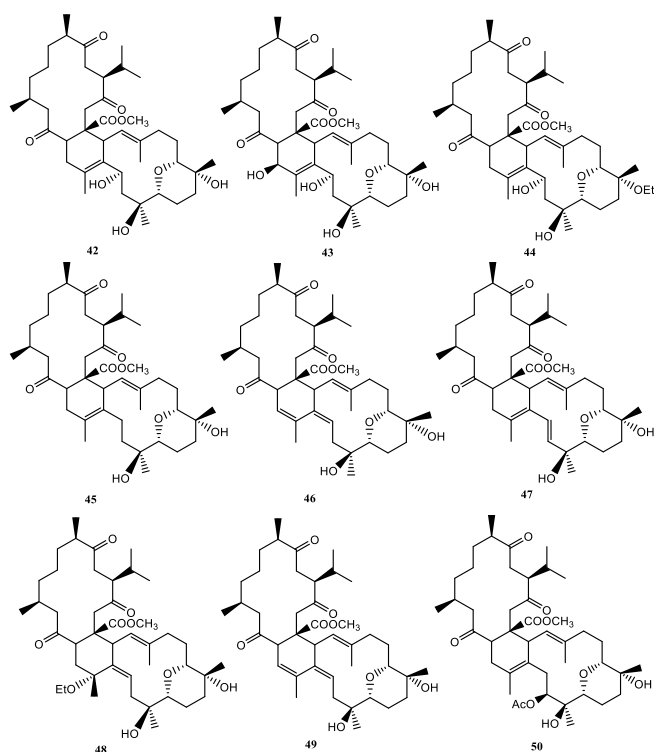


Figure 7. The structures of compounds (42-50) from *L. pauciflorum*.

### Sterol

Steroids are structures with a cyclopentane polyhydrogen phenanthrene skeleton and three side chains. Despite being widely found in animals and plants, marine steroids are found to be more diverse in their structure compared to terrestrial phytosterols (Zhang et al., 2019). Lobosterol (**51**) was the first sterol to be isolated and reported from the soft coral *L. pauciflorum*, which contains a 3 $\beta$ ,4 $\beta$ ,5 $\beta$ -trihydroxy system (Tursch, 1976). This is followed by isolation of several polyhydroxysteroids from *L. pauciflorum* in the Okinawan waters (Yamada et al., 1980b). These polyhydroxysteroids include 24 $\xi$ -methylcholastane-3 $\beta$ ,5 $\alpha$ ,6 $\beta$ ,25-tetrol (**52**), 24 $\xi$ -methylcholastane-1 $\beta$ ,3 $\beta$ ,5 $\alpha$ ,6 $\beta$ -tetrol (**53**), 24-methylencholastane-1 $\beta$ ,3 $\beta$ ,5 $\alpha$ ,6 $\beta$ -tetrol (**54**), and 24 $\xi$ -methycholestane-3 $\beta$ ,5 $\alpha$ ,6 $\beta$ ,25-tetrol 25-monoacetate (**55**). Compound **55** was previously isolated from *Sarcophyton elegans* (Moldowan et al., 1974).

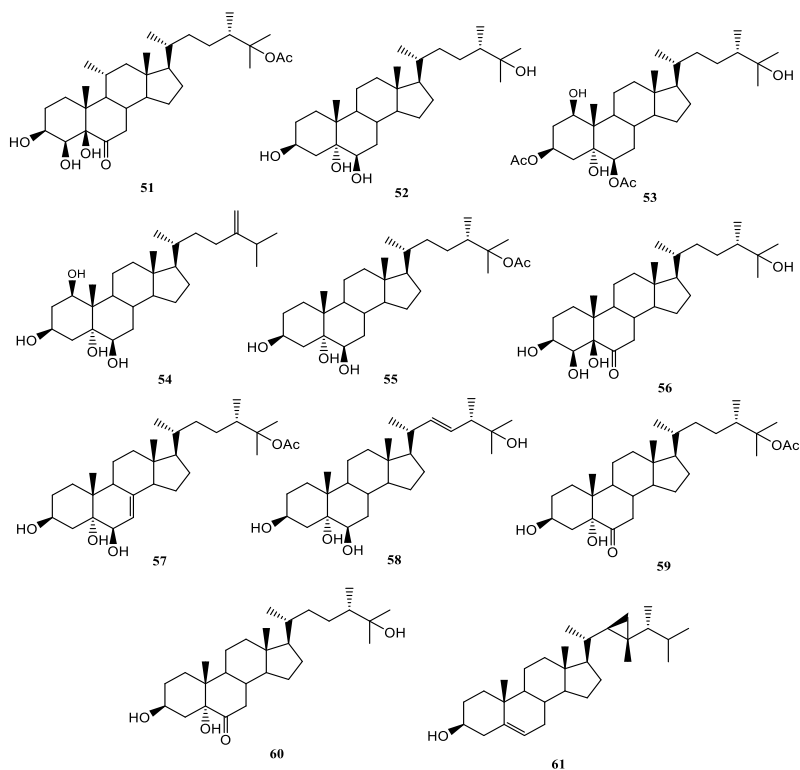


Figure 8. Polyhydroxysteroids (**51-61**) isolated from *L. pauciflorum*.

Reports on the isolation of sterols from *L. pauciflorum* continued with the isolation of additional five steroids from the soft coral variety of the Andaman and Nicobar Islands. These steroids, 25-deacetylobosterol (**56**), (24S)-24-methylcholest-7-en-3 $\beta$ ,5 $\alpha$ ,6 $\beta$ ,25-tetrol-25-monoacetate (**57**), (24S)-24-methylcholest-22E-ene-3 $\beta$ ,5 $\alpha$ ,6 $\beta$ ,25-tetrol (**58**), (24S)-24-methylcholestane-3 $\beta$ ,5 $\alpha$ ,25-triol-6-one-25-monoacetate (**59**) and its C-25 deacetoxy analogue (**60**). The steroids were isolated along with lobosterol (**51**) and two diterpene lobanes (Rao et al., 1990). In 2016, Hassan et al. isolated six known compounds from the marine soft coral *L. pauciflorum*. Among the identified compounds is the sterol gorgost-5-ene-3 $\beta$ -ol (**61**). Figure 8 represents the chemical structures of polyhydroxysteroids from *L. pauciflorum*.

### Biological Properties of *L. Pauciflorum* and Isolated Compounds

The cytotoxicity of polar and nonpolar extracts of freeze-dried *L. pauciflorum* using 50:50 ethanol-water and 50:50 ethyl acetate-methanol was investigated using brine shrimp *Artemia salina* lethality test. The polar extract of *L. pauciflorum* exhibited shrimp mortality activity of 90.7% at 1000-ppm concentration after 6 hours of exposure, and 100% mortality effect after 24-hour exposure. Moderate cytotoxicity activity of 89.1% was observed for the non-polar extract at 1000-ppm. However, it is notable that *L. pauciflorum* had the highest activity among the other three soft corals that were investigated which are *Sinularia flexibilis* (70.2%), *Sarcophyton glaucum* (69.2%), and *Lobophytum crissum* (33.3%) (Luyao et al., 2019a). Antioxidant and antimicrobial activities of polar and non-polar extracts of *L. pauciflorum* collected from the Coast of Agusan del Norte, Philippines was investigated by Luyao et al., in 2019. The total antioxidant capacity was expressed as Ascorbic Acid Equivalence (AAE) and Butylated Hydroxytoluene Equivalence (BHTE). Non-polar extract and polar of *L. pauciflorum* exhibited an AAE value of 54.58 and 53.59, respectively at 200 ppm. A BHTE value of 125.85 and 113.41 was observed for non-polar and polar extracts, respectively at 200 ppm. The DPPH radical scavenging activity (%) of non-polar and polar *L. pauciflorum* extract exhibited an activity of 1.49% and 2.88% at 500 ppm. *Lobophytum pauciflorum* displayed a minimal yet active effect towards the growth of gram-positive and gram-negative bacteria, *B. subtilis* and *E. coli*. However, no antifungal activity was observed against the tested *Aspergillus niger* and *Saccharomyces cerevisiae*.

The seven isolated compounds lobatriene (**8**), lobatrienediol (**13**), lobatrienolide (**17**), lobatrietriol monoacetate (**18**), lobatrienetriol diacetate (**19**), lobatriene epoxide (**20**) and lobatrienol (**21**) were tested for their bioactivity. Lobane was tested for its biological potential and lobatrienol (**21**) was found to possess

antibacterial and anti-fungal activities by inhibiting the gram-positive bacteria *Bacillus subtilis* and *Saccaromyces cerevisiae* as well as the phytopathogenic fungus *Candida cucumerinum* (Edrada et al., 2000). Cytotoxicity test using brine shrimp assay were also conducted. Lobatriene epoxide (**20**) displayed the highest cytotoxicity activity with IC<sub>50</sub> value of 0.64 µg/mL along with lobatetraene (**15**) at 4.18 µg/mL (Edrada et al., 1998). Bioassay-guided screening of *n*-hexane, dichloromethane, ethyl acetate and methanol fractions of *L. pauciflorum* for anti-inflammatory activity revealed its significant activity against cyclooxygenase enzymes COX-1 and COX-2. The hexane, dichloromethane showed significant anti-inflammatory activity against COX-1 compared to the positive control, and indomethacin had IC<sub>50</sub> values of 0.59, 1.37 and 1.52 mM respectively. The *n*-hexane, dichloromethane, ethyl acetate showed significant anti-inflammatory activity against COX-2 compared to positive control Celecoxib, with an IC<sub>50</sub> value of 0.12, 0.33, 0.61 and 0.43 mM respectively. Spectroscopic analysis isolated two active compounds which were identified to be nephthenol (**1**) and gorgost-5-ene-3β-ol (**61**) (Hassan et al., 2016).

Biscembranoids lobophytone Q (**38**) exhibited inhibition towards lipopolysaccharide (LPS) induced nitric oxide (NO) production in macrophages with IC<sub>50</sub> of 2.8 µM. In addition, this compound together with lobophytone T (**41**) inhibited the growth of *Staphylococcus aureus*, *S. pneumoniae* and *Saccaromyces cerevisiae* at a concentration of 20 µg/mL (Yan et al., 2010c). Similar to lobophytone Q (**38**), lobophytone Z (**47**) exhibited inhibition towards LPS induced NO production in macrophages with IC<sub>50</sub> of 2.6 µM while the other compounds displayed IC<sub>50</sub> values in the range of 3-5 µM. Lobophytone U (**42**) also was able to inhibit the growth of *S. aureus* and *S. pneumoniae* (Yan et al., 2011). The reported biological activity of *L. pauciflorum* and the active compounds is summarised in Table 1.

**Table 1.** Summary of *L. pauciflorum* biological activities and the active compounds.

Biological activity	Active compound	Reference
Antibacterial	Lobatrienol ( <b>21</b> )	Edrada et al., 2000
Antifungal	Lobatrienol ( <b>21</b> )	Edrada et al., 2000
Anti-inflammatory	Nephthenol ( <b>1</b> )	Hassan et al., 2016
	Gorgost-5-ene-3β-ol ( <b>61</b> )	
	Lobophytone Q ( <b>38</b> )	Yan et al., 2010c
Antimicrobial	Lobophytone T ( <b>41</b> )	Yan et al., 2011
	Lobophytone U ( <b>42</b> )	Luyao et al., 2019b
	Lobophytone Z ( <b>47</b> )	
Antioxidant	-	Luyao et al., 2019
Cytotoxicity	Lobatriene epoxide ( <b>20</b> )	Edrada et al., 1998,
		Luyao et al., 2019a

## Conclusion

*Lobophytum pauciflorum* is a source of a range of secondary metabolites that have diverse bioactivities. In this review, we report that three classes of secondary metabolites, namely diterpenes, biscembranoids, cembrene and sterols, have been isolated from the soft coral species *L. pauciflorum* and their bioactivity. It was observed that reports on *L. pauciflorum* focus more on the structural diversity of secondary metabolites rather than their bioactivity. Further extensive study is required to explore the biological potential of the secondary metabolites isolated from the soft coral species *L. pauciflorum*.

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## **Short Communication**

# ***Burmannia sphagnoides* (Burmanniaceae): A New Record for Kalimantan, Indonesia**

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## **Abstract**

This paper reports the first occurrence for a rare mycoheterotrophic *Burmannia sphagnoides* Becc. (Burmanniaceae) in Kalimantan (Indonesian Borneo) based on a specimen collected from Gunung Mas Regency, Kalimantan Tengah Province. A morphological description, distribution map, brief notes, and photographs are presented.

**Keywords:** Borneo, herbs, mycoheterotroph, new record, taxonomy

## **Introduction**

The family Burmanniaceae consists of eight genera with perhaps as many as 96 species accepted so far (Merckx et al., 2013). The comprehensive taxonomic study of this genus in Malesia was published over half a century ago (Jonker, 1948). According to Jonker, there are at least five genera and 39 species that have been reported from this phytogeographical region. Some new findings were also published in the recent years (e.g. Tsukaya et al., 2016; Tsuetsugu et al., 2018).

*Burmannia sphagnoides* Becc. was described for the first time by the Italian botanist Odoardo Beccarii (1877). The type specimens was collected from Kuching, Sarawak in 1865. The species is now known from Sumatra, Peninsular Malaysia, and Sarawak (Jonker, 1948; POWO, 2021). Though if collected from Malaysian Borneo, there was no record from Indonesian Borneo until this study. In 2020, a specimens was collected from Gunung Mas Regency, Kalimantan Tengah Province, in Kalimantan, Indonesian Borneo. There is no previous report of this species in Indonesian Borneo and is therefore considered a new record for the area.

## Materials and Methods

The specimen was collected during extensive High Conservation Value assessment for plants from 2019 to 2020. It was found in a remaining forest patch near an oil palm plantation and settlements. This forest is located outside of protected areas. Identification was done using comparison to published literature on mycoheterotrophic plants in Borneo (Jonker, 1948; Dancak et al., 2015; Ling et al., 2019; Tsukaya et al., 2016) and herbarium study in BO and digital specimens in FI and L (abbreviations follow Thiers, 2021-continuously updated). The morphological description of the species was prepared from the recently collected specimen.

## Results and Discussion

### Species Description

*Burmattia sphagnoides* Becc., Malesia 1: 246 (1877). Type: Malaysia, Ragiato di Sarawak, Kuching, June 1865, *Beccari* 86 (FI-image! [FI013461]) (type illustration: Beccari (1877, tab. XV, f. 8-11)). Figure 1.



**Figure 1.** *Burmattia sphagnoides* from Gunung Mas Regency, Kalimantan Tengah Province, Indonesia: A. plants, B. close-up of roots, C. stem and scales, D. close up of flowers showing bracts, E. close up of flowers.

Mycoheterotrophic plants, 4-5.5cm; glabrous. Stem unbranched, 1.5-2mm across, densely covered with scale-like leaves, imbricate in the lower portion of stem, ovate-lanceolate, up to 8mm long and  $\times$  2mm wide, narrowing to pointed, rather filiform apex, margin entire, glabrous, midrib 1. Flowers 3-5 at the apex of stem. Bracts lanceolate, c.  $7.5-8.5 \times 1.5-4$ mm, acute or subobtus. Flowers with 6 costae, 8.5-9mm. Perianth tube cylindrical, c. 4mm long, white. Outer perianth broadly ovate-triangular, c.  $2 \times 2$ mm, thick, apex acute. Inner perianth c. 1mm long. Style filiform, slightly angled, bearing 3 stigmas. Ovary broadly ellipsoid to subglobose,  $3 \times 3$ mm. Mature fruit unknown.

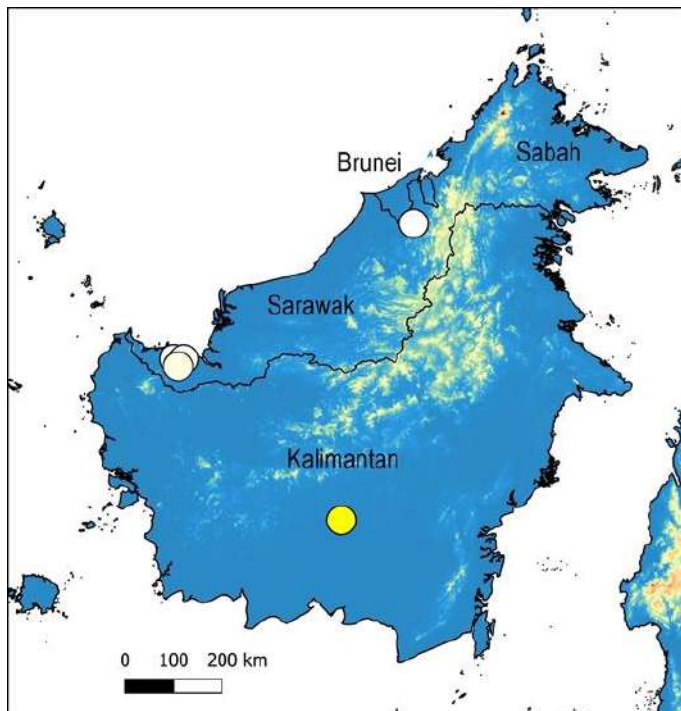
**Distribution:** This species has been recorded from Sumatra, Peninsular Malaysia, and Sarawak. The distribution in Sumatra is so far only known from the East Coast Residence, now called Sumatra Utara Province (Jonker, 1948). Distribution in Borneo including the new record is listed in Figure 2. The new record is located around 472km southeast of Kuching, Sarawak, where most Bornean specimens were collected.

**Habitat:** *Burmannia sphagnoides* was found at the lowland area around 43m asl. The habitat is a dipterocarp forest. The common species include several species from the genus *Dipterocarpus* and *Shorea* (Dipterocarpaceae), *Knema* (Myristicaceae), and *Syzygium* (Myrtaceae). Some species in this forest fragment are potentially threatened with extinction according to the IUCN Red List (2021) like *Dipterocarpus lowii* Hook.f. (NT). Endemic species like the climbing epiphyte *Bulbophyllum beccarii* Rchb.f. (Orchidaceae) also occur in the same area.

Very limited ecological data is available for this species. In the Flora Malesiana, Jonker (1948) mentions this species occurs in the forest on decaying matter. Our recent observation also shows a similar situation. The plants were found growing on an empty forest floor with a lot of organic materials like dead leaves and fallen branches or trunk. It seems this species has a very narrow type of habitat. The vegetation is of mixed forest with a small number of dipterocarp trees. More than 100 individuals were seen during the field exploration at a single site on a single occasion. In Sarawak, according to our examination of specimens, this species was collected from the forest floor on dead leaves (*Brooke 9544* [L.1487316]) or 'wet leaves' (*Brooke 9764* [L.1487317]). The only recorded type of forest is primary dipterocarp forests on sandstone substratum, with low stature and considerably thin-stemmed forest (*Jacobs 5554* [L.1487318]).

**Notes:** In Borneo, as well as Malesia, this species can be recognized from other members of the genus by the mycoheterotrophic life-style with reduced leaves, usually in the form of scales, densely covering the stem (appearing imbricate), the wingless flowers, and ovary that is equal to or longer than the perianth (Jonker, 1948).

**Specimen examined:** INDONESIA. Kalimantan: Kalimantan Tengah Province, Gunung Mas Regency, forest near Agrolestari Santosa oil palm plantation ( $1^{\circ}20'53.52''\text{S}$   $113^{\circ}31'14.88''\text{E}$ ), c. 43m asl, Mustaqim RA2020-01 (BO!). MALAYSIA. Borneo: Sarawak, 1st div., c. 25km West of Kuching, Mt Matang, 750-900m, 21 Sep 1958, *Jacobs* 5554 (K, L!-image [L.1487318]); Sarawak Division 1, 13th mile Matang, 1 Dec 1954, 1000 ft, *Brooke* 9544 [L!-image [L.1487316]]; Sarawak Division 1, Kuching, 23 Mar 1955, *Brooke* 9764 (L!-image [L.1487317]); Sarawak, Mattang, Apr 1866, *Beccari* 1502 (L!-image [L.1483719]); Sarawak Division I, Perungen, 11 May 1954, *Brooke* 8511 (L!-image [L.1487320]).



**Figure 2.** Geographic distribution of *Burmannia sphagnoides* in Borneo with new record in Kalimantan (○ = previous records; ● = new record).



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## Research Article

# Documenting Butterflies Diversity (Lepidoptera: Rhopalocera: Nymphalidae) as Potential Nature Tourism Products at Sukau Rainforest Lodge and Sukau Ecotourism Research Centre, Kinabatangan, Sabah

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## Abstract

Sukau Rainforest Lodge (SRL) and Sukau Ecotourism Research Centre (SERC) have been recognised as two ecotourism sites in Sabah. However, there were no butterfly tourism products carried out at both sites. As a result, a 5-day survey of butterfly diversity was conducted using fruit-baited traps. Butterfly diversity, richness, and abundance were evaluated using Shannon-Weiner Diversity for both sites. SWOT analysis was also carried out at both sites throughout a 3-week observation. The results showed there were 20 species from 6 subfamilies of Nymphalidae butterflies. A total of 96 individuals of each species were also tabulated, with SRL revealing a higher diversity index compared to SERC due to its large surrounding area. The butterfly specimens were collected for educational purposes as nature tourism products. Lack of research, no butterfly signage, forest fragmentation, and mass tourism activities were identified as weaknesses and threats with their strengths and opportunities as proper guidelines based on SWOT analysis. Hence, it shows that both sites are valuable for nature tourism based on their unique butterfly fauna as butterfly tourism products. This study could also provide baseline data on butterfly diversity and its potential as butterfly tourism products at both sites. Butterfly diversity data and SWOT analysis are compulsory for the creation of butterfly tourism products. Such valuable fauna can be integrated as nature tourism products in conjunction with appropriate decision-making strategies.

**Keywords:** Sukau Rainforest Lodge, Sukau Ecotourism Research Centre, Shannon-Weiner Diversity, Nymphalidae, SWOT analysis, butterfly tourism products.

## Introduction

The Kinabatangan Floodplain of north-eastern Borneo (also known as Sabah, Malaysia) has been named one of the top ecotourism destinations in Malaysia (WWFNM, 1996). Home to several tourist facilities, including the Sukau Rainforest Lodge (SRL) and the Sukau Ecotourism Research Centre (SERC), this is a unique place; one where visitors can see the rare Proboscis monkeys, gibbons, and an Asian elephant munching contentedly along the river's edge of fragmented forests (Teo & Patterson, 2005). In the last 30-40 years, much of the original dipterocarp forest has been cleared or converted for land use along the Kinabatangan River (Goossens et al., 2006; Latip et al., 2013). Oil palm plantations currently dominate the landscape along the river, and the forest is limited to isolated patches. Despite this extensive transformation of the Kinabatangan Floodplain, the forest remnants of the Kinabatangan River still contain a very diverse small mammal community (Brunke et al., 2019).

Nature-based tourism can be broadly defined as an exploration of a natural destination which may be a place for a recreational activity where interaction with animals and plants is incidental, or the object of the visit is to acquire an understanding of the natural history of the place itself (a form of ecotourism) and to interact with the animals and plants (Fennell, 2014; Wolf et al., 2019). Ecotourism is intimately associated with natural features such as scenic beauty, beaches and coastal reserves, flora and fauna, and parks and conservation areas. These prime features could be renowned for their tourist attractions (Goh, 2015). Butterflies are usually large and attractive to most people. Thus, this can provide prioritisation for nature tourism products (Takizawa et al., 2012). They are good indicators of environmental health due to their sensitivity to habitat and climate changes. As an outcome, such intervention in species richness and abundance in specific areas can be influenced by a direct impact on habitat quality and the provision of ecosystem services (Pang et al., 2016; Peters et al., 2016). A manager must have knowledge and insight to make such strategic decisions (Grant, 2008), guiding SRL and SERC management in developing nature tourism products for butterflies.

Borneo is home to 242 butterfly species belonging to the Nymphalidae family (Häuser et al., 1997), an estimated 75% of these adult species feed on rotting fruits (Hill et al., 2001). In Sabah, around 186 Nymphalidae species (76.9%) have been recorded from the Kinabalu Park area, presented in a systematic arrangement by Häuser et al. (1997). The specific purpose of this study is to document the diversity of the butterflies at the SRL and SERC. This study investigates whether the butterfly diversity is sufficient to serve as a nature

tourism product at both sites. This could also provide baseline data on butterfly diversity and its potential as a butterfly tourism product.

## Methodology

Butterfly surveys were conducted at the SRL and SERC from July 11-15, 2020, to collect butterfly diversity data.

Butterflies were collected using modified Van Someren Rydon's traps (Upton, 1991; Benedick et al., 2007a). The fruit-baited traps were implemented in this study to attract canopy and fruit-feeding butterflies to the ground level. Selected sampling sites for traps were positioned about 1-2 m above the ground and 50-100 m apart. At the SRL, nine traps were set (Figure 1), whereas, at the SERC, four traps were set (Figure 2). The trap placements at both sites were distinct as the sampling area of SRL is larger compared to the SERC. The baits were prepared early in the morning on each sampling day. Each trap was inspected 4 times a day: morning (10:00 - 12:00), afternoon (12:00 - 14:00), evening (14:00 - 16:00), and late evening (16:00 - 18:00) for five consecutive days. Similar butterfly species that were captured were released into the wild.

The list of butterfly species caught and recorded at the SRL and SERC were presented and tabulated by subfamily and species. Recent Otsuka (1988; 2001) classifications and standard reference works for taxonomy and nomenclature were followed. In addition, the butterfly diversity study was considered based on their species richness and abundance at both sites. Hence, the butterfly diversity was calculated using the Shannon-Weiner Index as instructed by Shannon & Weiner (1963), which is described by the following equation:

$$\text{Species diversity } H'(S) = -\sum_{i=1}^n p_i \log p_i$$

Where:

$p_i$	=	$n_i/N$ ,
$n_i$	=	Number of individuals of a species $i$ ,
$N$	=	Size of whole community,
$S$	=	Total number of species.

The butterfly specimens were relaxed, spread, identified, labelled and stored in the SERC collection box for environmental education purpose and for as butterfly tourism products.



Figure 1. Baited Traps at the SRL (source: [www.google.com/maps](http://www.google.com/maps)).



Figure 2. Baited Traps at the SERC (source: [www.google.com/maps](http://www.google.com/maps)).

SWOT analysis is an early stage of the strategic planning process that helps planners define strategies and decisions on resource allocation for pursuing those strategies. In this case, determining internal and external factors is critical. This analysis also involves systematic thinking and comprehensive diagnosis of factors relating to a new product, technology, management, or planning. Hence, it provides knowledge regarding the situation and allows the design of procedures that may be deemed necessary for thinking in a strategic way (Shrestha et al., 2004; Lozano & Valles, 2007). The collected butterfly data was analysed using SWOT analysis after three weeks' observation at the SRL and SERC. The result of the SWOT analysis was tabulated in the Internal Factor

Estimate Matrix (IFEM) and External Factor Estimate Matrix (EFEM) tables. Each factor's weight was assigned a value of one.

## Results and Discussion

**Table 1.** List of species caught in baited traps at the SRL and SERC.

No.	Subfamilies	Species	SRL	SERC	Total number of individuals
1.	Charaxinae	<i>Prothoe franck</i>	4	0	4
2.	Danainae	<i>Euploea crameri</i>	1	0	1
3.		<i>Euploea phaenareta</i>	0	1	1
4.		<i>Ideopsis vulgaris</i>	0	1	1
5.	Heliconiinae	<i>Cirrochroa emalea</i>	1	0	1
6.	Limenitidinae	<i>Dophla evelina</i>	1	1	2
7.		<i>Lexias dirtea</i>	8	0	8
8.		<i>Lexias pardalis</i>	6	1	7
9.		<i>Parthenos sylvia</i>	1	0	1
10.		<i>Tanaecia aruna</i>	1	1	2
11.		<i>Tanaecia iapis</i>	1	0	1
12.	Morphinae	<i>Amathusia phidippus</i>	1	0	1
13.		<i>Zeuxidia amethystus</i>	0	1	1
14.	Satyrinae	<i>Elymnias panthera</i>	1	0	1
15.		<i>Melanitis leda</i>	0	1	1
16.		<i>Mycalesis fusca</i>	16	13	29
17.		<i>Mycalesis pitana</i>	20	8	28
18.		<i>Neorina lowii</i>	0	2	2
19.		<i>Orsotriaena medus</i>	1	0	1
20.		<i>Thaumantis noureddin</i>	1	2	3
Total			64	32	96

**Table 2.** Diversity, richness, and abundance of study sites at the SRL and SERC.

Sites	Diversity (Shannon-Weiner)	Richness	Abundance
Sukau Rainforest Lodge	0.8751	15	64
Sukau Ecotourism Research Centre	0.7892	11	32
Total		26	96

Based on Table 1, a total of 96 individuals from 20 species (6 subfamilies) were recorded during a sampling period of 5 days. The sampling area around the SERC was small, surrounded by houses and lodges at other villagers. However, *Euploea phaenareta*, *Ideopsis vulgaris*, *Melanitis leda*, *Neorina lowii*, and *Zeuxidia*

*amethystus* were found at that location. In contrast, *Amathusia phidippus*, *Cirrochroa emalea*, *Elymnias panthera*, *Euploea crameri*, *Lexias dirtea*, *Orsotriaena medus*, *Parthenos sylvia*, *Prothoe franck*, and *Tanaecia iapis* were found at the SRL. Additionally, *Dophla evelina*, *Lexias pardalis*, *Mycalesis fusca*, *Mycalesis pitana*, *Tanaecia aruna*, and *Thaumantis noureddin* were found at both sites. *Mycalesis fusca* and *Mycalesis pitana* had the most individuals caught in baited traps on this expedition, with a total of 29 and 28 individuals, respectively, followed by *Lexias dirtea* (8 individuals), *Lexias pardalis* (7 individuals), *Prothoe franck* (4 individuals), and *Thaumantis noureddin* (3 individuals). *Dophla evelina*, *Neorina lowii*, and *Tanaecia aruna* were all composed of 2 individual species. Meanwhile, all the remaining butterfly species were composed of 1 individual species.

Table 2 shows the determination of the diversity, richness, and abundance of the study sites at the SRL and SERC. Only 15 species with a total of 64 individual species were recorded at the SRL. Meanwhile, only 11 species along with a total of 32 individual species were recorded at the SERC. Apart from this, SRL showed a diversity index of 0.8751, whereas SERC showed a diversity index of 0.7892. Among both sites, SRL showed higher species in terms of diversity, richness, and abundance compared to SERC due to a large area of sampling where nine baited traps were set. Unfortunately, there were no Bornean endemic butterflies (Otsuka, 1988; Maruyama & Otsuka, 1991; Seki et al., 1991; Otsuka, 2001) found at the SRL and SERC as shown in Table 1. Notably, *Prothoe franck* found at the SRL is one of the 11 Nymphalidae species protected under the Wildlife Conservation Act 2010 (Act 716). This species is vulnerable to illegal trade due to its aesthetic appeal (UNEP-WCMC, 2012).

Accessing a canopy can be partially solved by using fruit-baited traps to lure canopy and fruit-feeding butterflies to the ground level as suggested by Tangah et al. (2004). However, fruit-baited traps only catch a specific butterfly guild that is attracted to rotting fruits. Consequently, most canopy butterflies cannot be studied in their entirety. There is also limited information about butterflies and any other species attracted to that traps, making it difficult to determine a trap sampling area. The advantage of using these traps is that it reduces field identification problems because all individuals can be spotted and easily identified. Besides that, this is an easy way to conduct a canopy study without the need to access the canopy (Barker & Sutton, 1997).



**Table 3.** IFEM for potential butterfly tourism products at the SRL and SERC.

No.	Internal Factors	Weight	Effectiveness Score	Final Score
<b>Strengths (S)</b>				
1.	Trails along the SRL and SERC are easily accessible and many open-field butterfly species can be seen.	0.30	4	1.20
2.	Unique scenery that features the real ecosystem of secondary forests.	0.20	4	0.80
3.	Available rare species of butterfly (i.e., <i>Prothoe franck</i> ).	0.20	3	0.60
<b>Weaknesses (W)</b>				
1.	No population study and impact assessment on the butterflies. Carrying capacity is unknown.	0.25	4	1.00
2.	No signage about butterfly information that includes potential sighting.	0.05	3	0.15
<b>Total</b>		<b>1</b>	<b>18</b>	<b>3.75</b>

Across the world, there are plenty of butterfly tours that have been mainstreamed. However, Sabah lacks butterfly tours. Few attempts have been made to use butterflies for environmental education (Takizawa et al., 2012; Chung, 2019). Total score values for IFEM were 3.75 as shown in Table 3. Based on the result, each factor's weight ranged from 0.05 to 0.30, and the effectiveness score ranged from 3 to 4 only. Three factors had been identified as strengths for butterfly tourism products based on three weeks' observation at both sites. The highest weight given to trails along the SRL and SERC as these were easily accessible and many open-field butterflies can be seen with a total of 1.20 as the final score, followed by unique scenery that features the secondary forest and available rare butterfly species with a total of 0.80 and 0.60, respectively. Two factors were highlighted as weaknesses. Signage about butterfly information was lower (0.15) compared with population study and impact assessment on the butterflies (1.00).

Along the boardwalk and trails at the SRL and SERC, many open-field and common butterfly species can be spotted like *Lexias pardalis*, *Lexias dirtea*, *Thaumantis noureddin*, *Mycalesis fusca*, and *Mycalesis pitana*. Rare species like *Prothoe franck* were seen during the expedition. Furthermore, because SRL and SERC were built around the secondary forest (Tropical Rainforest) that houses flora and fauna, all guests who stay at the SRL and SERC are able to enjoy this scenery. These benefits can provide an ecosystem balance in terms of the prevalence of butterflies' species and their dependability on the state of the environment (Ivinskis & Rimšaitė, 2004).

No population studies and impact assessments have been carried out at the SRL and SERC for butterfly studies recently. As a result, the carrying capacity of butterflies is still unknown and must be thoroughly investigated through scientific research. These could potentially have a direct impact on butterfly misinformation at the SRL and SERC. As an outcome, the butterfly information provided by the signage will be rejected.

**Table 4.** EFEM for potential butterfly tourism products at the SRL and SERC.

No.	External Factors	Weight	Effectiveness Score	Final Score
<b><i>Opportunities (O)</i></b>				
1.	An opportunity for the scientific study of butterflies to determine their population and carrying capacity.	0.20	4	0.80
2.	Increasing flowering plants along the boardwalk and trails for butterfly attraction.	0.10	3	0.30
3.	Butterfly specimens that have been preserved will be displayed in the SERC building.	0.30	4	1.20
4.	Providing pamphlets and brochures about butterfly information for tourists.	0.25	4	1.00
<b><i>Threats (T)</i></b>				
1.	Threat to butterfly species through forest fragmentation due to oil palm plantations near the SRL and SERC.	0.10	2	0.20
2.	Mass tourism activities that could cause habitat disturbance and noise pollution.	0.05	2	0.10
<b>Total</b>		<b>1</b>	<b>19</b>	<b>3.60</b>

Total score values for EFEM were 3.60 as shown in Table 4. Hence, each factor's weight ranged from 0.05 to 0.30, whereas the effectiveness score ranged from 2 to 4. There were four factors related to opportunities. Butterfly specimens that have been preserved received the highest score (1.20), followed by providing pamphlets and brochures for tourists (1.00), butterfly scientific study (0.80), and increasing flowering plants along the boardwalk and trails to attract butterflies (0.30). There were two factors related to threats. Threat to butterfly species due to oil palm plantations received a score of 0.20, while mass tourism activities received a score of 0.10 as the final score.

It is necessary to pay for the existence of butterflies in terms of maintaining and improving the village environment. Scientific studies of butterflies should be utilised to determine their current population and carrying capacity (Curtis et al., 2015), therefore, ensuring that butterfly diversity can be safeguarded. Increasing flowering plants along the boardwalk and trails can attract some

butterflies, adding to the scenery, a plus for tourists. When choosing flowers to attract butterflies, low lantana and hibiscus could be a few choices as recommended by Takezawa et al. (2012). It is suggested that orchards should be planted at open areas around the SRL's boardwalk and trail, as well as at the surrounding of the SERC area's trail, for butterfly observation and to keep bare wet spots for butterflies to absorb moisture. Bringing tourists to SERC to see an actual butterfly specimen could be proposed as part of environmental education, thus addressing butterfly conservation awareness. Such small tours are beneficial and appealing to SRL guests within the context of nature tourism products. Above all, the provision of signage, appropriate pamphlets and training guides are vital to attracting tourists. Such activities should serve as a key task for the Information Centre.

Butterflies on Borneo are diverse with many rare, endemic and restricted-range species that are dependent on the canopy forest and do not occur in oil palm or other crops (Otsuka, 1988; Maruyama & Otsuka, 1991; Seki et al., 1991; Otsuka, 2001; Benedick et al., 2006). However, the fragmentation of tropical rainforests could threaten butterflies at the SRL and SERC because both sites have been surrounded by oil palm plantations (Brunke et al., 2019). In support of this notion, there was some evidence of a reduction in genetic diversity of *Mycalesis orseis* following forest fragmentation in Sabah as reported by Benedick et al. (2007b). Environmental education about butterfly conservation awareness such as exhibitions and talk sessions are suggested to prevent such tragic deforestation. Moreover, it is possible for tourists to take small-scale butterfly tours at the SRL and SERC to reduce habitat disturbance and noise pollution.

## Conclusion

The present study has identified about 20 butterfly species of Nymphalidae from 6 subfamilies with 96 individual species at the SRL and SERC. SRL had a higher diversity index compared to SERC based on Shannon-Wiener Diversity. There is an opportunity for butterfly tourism products to be carried out at both sites based on a SWOT analysis. Butterfly diversity and potential for butterfly tourism products at both sites acted as the baseline data as no research had been conducted previously. Continuous studies on butterfly species are needed as some butterflies have not been caught and listed due to time and resource limitations at both sites.

The main issues at the SRL and SERC affecting the diversity of butterflies as potential nature tourism products have been addressed. Such weaknesses and

threats such as lack of research, no butterfly signage, forest fragmentation, and mass tourism activities have been given proper guidelines and action plans in the SWOT analysis for strengths and opportunities. These findings could help decision-makers and stakeholders develop butterfly tourism products at the SRL and SERC.

Both findings of butterfly diversity and SWOT analysis are compulsory for development of butterfly tourism products. Such valuable fauna, together with appropriate decision-making strategies, can be correlated with nature tourism products. As a long-term prospect, this will ensure a preserved heritage.

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**Appendix.** (A) *Amathusia phidippus*, (B) *Cirrochroa emalea*, (C) *Dophla evelina*, (D) *Parthenos sylvia*, (E) *Prothoe franck*, and the protected species under Wildlife Conservation Act 2010 [716]; (F) Set of butterfly specimen collection for environmental education purpose.







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**Research Article**

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## Ant Community Structure in Secondary Logged Forest of Malua Forest Reserve, Sabah, Borneo

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### Abstract

Ants are ecologically dominant and important in the functioning of an ecosystem. Thus, understanding their community structure has become fundamental in ecological studies. This study aims to examine the ant richness, abundance, and composition in the secondary logged forests of Sabah, Malaysia. Ground-based fogging was employed to collect canopy ants (n=38) and Winkler extraction method for leaf litter ants (n=63). A total of 12,810 ant individuals were collected, representing 389 morphospecies, 65 genera, and 11 subfamilies. The most species-rich subfamily for canopy and leaf litter ants were Formicinae (112 morphospecies, 49.34%) and Myrmicinae (116 morphospecies, 58.00%) respectively. *Polyrhachis* (56 morphospecies, 24.67%) was the most diverse genera in the canopy, while *Pheidole* (23 morphospecies, 11.50%) was the most speciose genera on the leaf litter. The most abundant species for canopy and leaf litter ants were *Dolichoderus* 1 (876 individuals) and *Carebara* 2 (1,215 individuals) respectively. The randomized species accumulation curves and species richness estimators reveal that additional sampling is required. We suggest that incorporating a variety of ant sampling methods and high sampling efforts are important to thoroughly sample the ant assemblage in an area.

**Keywords:** Formicidae, Borneo, secondary logged forest, species estimators, accumulation curve

### Introduction

Logged forests are often assumed to be degraded and fragmented lands that support limited taxonomic groups (Bihn et al., 2010). Their conservation value is neglected and subsequently make them extremely vulnerable to non-forest land-use conversion (Edwards et al., 2011; Edwards et al., 2014b). For instance, constant pressure from conservationists to restrict the conversion of primary forests into agricultural lands, and changes in policy to increase agricultural profitability, have caused governments as well as plantation agencies to shift their focus to logged degraded forests (Wilcove et al., 2010).

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However, growing literature suggests that ecologists have overestimated the damage of logging to biodiversity (Ramage et al., 2012) and logged forests can have astonishing value. Logged forests are able to retain a majority of their ecological functions such as hydrological processes, carbon sequestration, thermal buffering, and climate-regulation services (Putz et al., 2012; Edwards et al., 2014a; Senior et al., 2018), provided that the logging intensity is low (i.e., less than  $10\text{m}^3\text{ha}^{-1}$ , Burivalova et al., 2014); while supporting certain taxonomic groups which are vulnerable to plantation or barren land (Putz et al., 2012; Edwards et al., 2014a, b; Granados et al., 2016). For example, Edward et al. (2011) have reported that at least 75% of the dung beetle and bird species that persisted in the primary forest were also found within the twice logged tropical forests of Sabah, Borneo.

Ants are one of the ecologically diverse and ubiquitous groups among arthropods (Hölldobler & Wilson, 1990). They mediate not only a wide range of ecosystem functions (e.g., nutrient cycling, nutrient redistribution, soil turnover, and seed dispersal; Folgarait, 1998; Fayle et al., 2011), but also take part in many ecological interactions, including mutualism, predation and competition (Rico-Gray & Oliveira, 2007). Furthermore, they are known to be very responsive to their surrounding environmental changes, even at very small spatial scales (Tieda et al., 2017). Collectively, these make ants an ideal surrogate for arthropod taxa to study community structures in both degraded and natural habitats.

The ant fauna in Borneo is highly diverse (Pfeiffer et al., 2011). It is estimated at least 1,100 to 1,500 ant species exist in Borneo, with 1,000 species having been described, represented by 100 genera and 12 subfamilies (Pfeiffer et al., 2011; Fayle et al., 2014). Due to their ecological significance and contribution to community dynamics, a better understanding of ant community structure in logged forests of Borneo would greatly enhance our knowledge of the organization and dynamics of tropical ant communities in a disturbed area.

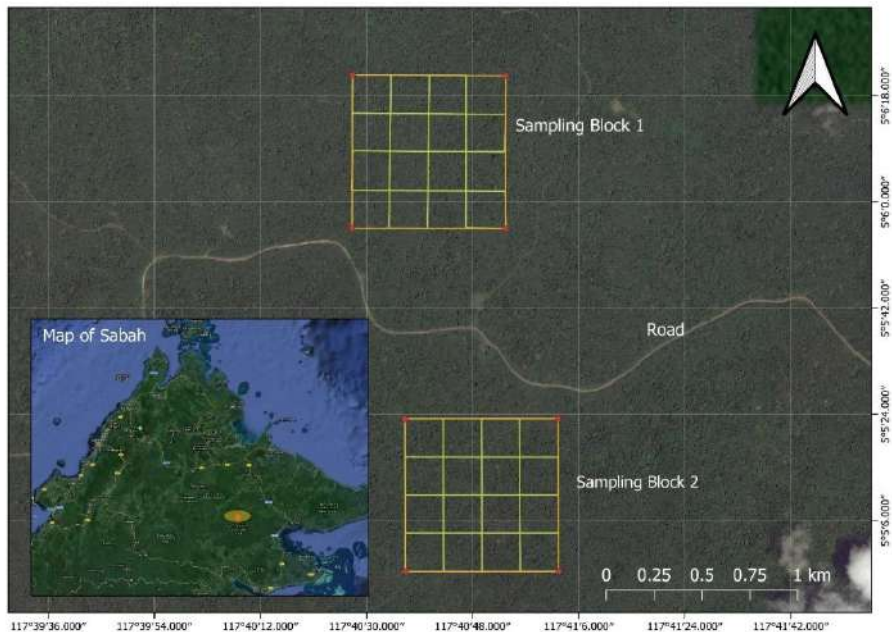
Here, we aim to investigate and reveal the ant community structure, specifically on species richness, ant abundance, and composition of canopy and leaf litter ants in Malua Forest Reserve (MFR), a little-known, remote secondary logged forest in Sabah, Borneo. Several past studies have been conducted to examine the ant communities in MFR. For example, in the study of the impact of climber-cutting silvicultural on arthropod groups, Dzulkifli (2014) reported that ants constituted the largest biomass in his samples. Using the Winkler method alone, Woodcock et al. (2011) recorded 196 species of leaf litter ants and found no

difference in terms of species richness between unlogged and logged forests in the Ulu-Segama Malua region. We believe this study will benefit future studies by providing additional information on ant communities in MFR to the existing literature, and subsequently adding conservation value to the forest as well as contributing to future conservation planning and management.

## Materials and Methods

### *Study site*

The study was conducted between June and September 2019 at Malua Forest Reserve (MFR, between E 116° 28' - E 118° 14' and between N 4° 14' - N 5° 18'), located at the South-eastern part of Sabah, Borneo (Figure 1). MFR was first gazetted in 1961 and was re-gazetted in 1984 as a Commercial Class II Forest Reserve. However, in 2006, the Sabah State Government grouped several neighbouring forests, including MFR as part of the Ulu-Segama Sustainable Forest Management Area. It is now managed by Yayasan Sabah and Sabah Forestry Department (Ulu-Segama Malua SFMP, 2020).



**Figure 1.** Map of the location of study site at Malua Forest Reserve, Sabah. The two square grids (in yellow) indicate the sampling blocks, each comprise 16 small grids or sampling plots. (Map made in QGIS 3.16)

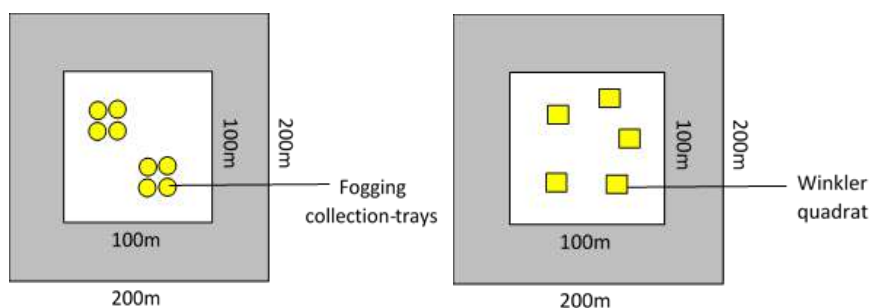
MFR has a flat topography with elevation ranging from below 100m a.s.l. to around 800m a.s.l. and many of its vegetation types is lowland mixed dipterocarp forest. The forest is dominated by valuable timber species mainly from the family Dipterocarpaceae (Ulu-Segama Malua SFMP, 2020). MFR is a selectively logged forest that has undergone logging rotation twice (i.e., between 1987 and 1991, between 2001 and 2007).

#### *Sampling design and sampling methods*

A total of two sampling blocks were created, each sized 800m x 800m within MFR. Each of the sampling blocks was divided into 16 small plots at 200m x 200m each (Figure 1). Of the 32 plots, 13 were selected to sample leaf litter ants, while 19 were chosen to collect canopy ants. The ground-based insecticide fogging method was employed to sample canopy ants while the Winkler extraction method (Bestelmeyer et al, 2000) for leaf litter ants.

#### *Canopy ant sampling*

Each small plot comprised of two replicates ( $n=38$ , 19 plots x 2 replicates) and each replicate contained four sets of funnel-shaped collection trays (Figure 2), each with a radius of  $1.2\text{m}^2$ , that was hoisted using ropes tied on the tree trunks. A bottle that contains 70% ethanol was placed at the centre of each collection tray to collect fallen arthropods. We followed Adis et al. (1999) protocol where fogging was performed before 06:00 for four minutes, using Thermal Fogging Machine (IGEBA TF-35) with fog mixture of cypermethrin-based insecticide and diesel (ratio 15:1). After the fogging, the collection trays were left for two hours. The fallen insects in the collection trays were brushed into the bottles using a flat decorator's brush (Floren et al., 2014).



**Figure 2.** Diagram illustrates an example of sampling plots and the allocation of replicates for (left) canopy ant sampling and (right) leaf litter ant sampling.

*Leaf litter ant sampling*

A total of five 1m x 1m quadrats were placed in each plot located in the sampling blocks (n=63, 13 plots x 5 replicates; Figure 2). Leaf litter ants were sampled before ground-based fogging, using the Winkler Extraction method (Bestelmeyer et al., 2000). This was to prevent the insecticide fog from killing the litter ants, which would potentially affect the results. The leaf litter and twigs within the quadrat were collected manually and put into the Winkler sifter. The sifter was shaken thoroughly for about four minutes. Next, the contents of the sifter were transferred to a mesh inlet sack and suspended in the Winkler sack. A 50ml container that was half-filled with 70% ethanol was affixed to the metal ring located at the bottom part of the Winkler sack. Winkler sacks were then hung out to dry for three days.

*Ant sorting, identification, and preservation*

Sorted ant samples were counted and identified to morphospecies following Fayle et al. (2014). The major workers, queens, and male ants were excluded from this study to minimize the error caused by vagrants; Andersen, 1995), while the worker ants (which are normally found; Andersen, 1995) were then identified to morphospecies or species, depending on the availability and certainty of the ant species classification information. Labelled specimens were stored in 1.5ml filled with 70% ethanol.

*Data analyses*

Basic analyses using ant abundance and richness were carried out in Microsoft Excel to describe the patterns of ant samples in MFR. The relative species richness for each genera and subfamily of canopy and leaf litter ants were calculated using the formula listed below:

*Relative species richness of genus A (%)*

$$= \frac{\text{Number of morphospecies present in genus A}}{\text{Total number of morphospecies from all genera}} \times 100\%$$

*Relative species richness of subfamily A (%)*

$$= \frac{\text{Number of morphospecies present in subfamily A}}{\text{Total number of morphospecies collected from all subfamilies}} \times 100\%$$

The sampling effort was assessed using the rarefaction and extrapolation curve (Hill's number of  $q=0$ ), with 1,000 randomization of species accumulation curves and 95% unconditional confidence intervals, based on the incidence data matrix (Colwell & Coddington, 1994). The reference sample sizes were doubled when extrapolating the curves (Chao et al., 2014).

The incidence-based estimator (Chao2) and first-order Jackknife estimator (Jackknife1) were used to estimate the sampling completeness of this study (Colwell et al., 2004; Gotelli & Colwell, 2011). Both Chao2 and Jackknife1 species estimators have been widely used to correct or reduce the bias found in the number of observed species in a sample (e.g., rare, and undiscovered species; Gwinn et al., 2016). Chao2 operates by deriving a lower bound of undiscovered species richness with reference to the number of singletons and doubletons (Chao, 1984), while Jackknife1 uses the presence and absence of unique species in a sample to predict the undetected species (Smith & van Belle, 1984). Software EstimateS version 9.1.0 (Colwell, 2013) was employed to compute the rarefaction and extrapolation curve as well as to calculate Chao2 and Jackknife1.

## Results

We identified a total of 12,810 ant individuals in MFR using the Ground-based Fogging and Winkler Extraction Method. They are represented by 389 morphospecies, 65 genera, and 11 subfamilies (Table 1). Canopy ants recorded were 5,104 individuals (227 morphospecies and 39 genera, Table 2) while leaf litter ants were documented to be 7,706 individuals (200 species, 49 genera, Table 3).

**Table 1:** The total number of individuals, morphospecies, genera, and subfamily of canopy and leaf litter ants collected in MFR.

	Canopy Ants	Leaf Litter Ants	Total
Individuals	5104	7706	12,810
Morphospecies	227	200	389
Genera	39	49	65
Subfamily	5	10	11

**Table 2:** Summary of total morphospecies and abundance of canopy ants obtained for each genus in MFR.

Subfamily	Genus	Total Morphospecies	Total Abundance
Dolichoderinae	<i>Dolichoderus</i>	10	1324
	<i>Tapinoma</i>	2	6
	<i>Technomyrmex</i>	8	212
Formicidae	<i>Anoplolepis</i>	1	1
	<i>Camponotus</i>	35	769
	<i>Cladomyrma</i>	1	2
	<i>Dinomyrmex</i>	1	4

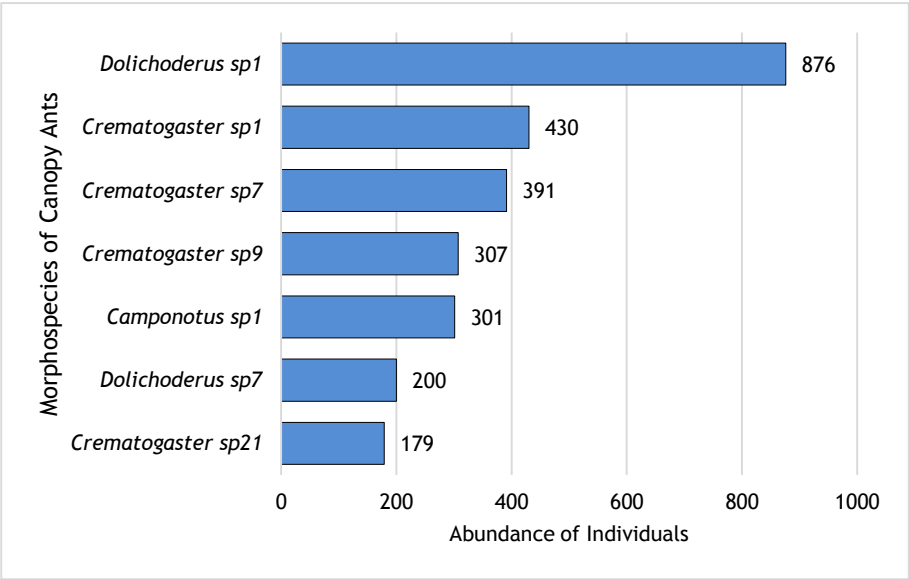
	<i>Echinopla</i>	4	9
	<i>Myrmoteras</i>	1	1
	<i>Nylanderia</i>	5	47
	<i>Paratrechina</i>	1	1
	<i>Parapatrechina</i>	5	34
	<i>Polyrhachis</i>	56	394
	<i>Prenolepis</i>	1	12
	<i>Pseudolasius</i>	1	6
Myrmicinae	<i>Acanthomyrmex</i>	1	1
	<i>Cardiocondyla</i>	4	14
	<i>Carebara</i>	6	150
	<i>Cataulacus</i>	1	5
	<i>Crematogaster</i>	23	1544
	<i>Dilobocondyla</i>	1	5
	<i>Lophomyrmex</i>	1	1
	<i>Meranoplus</i>	2	19
	<i>Monomorium</i>	4	33
	<i>Myrmecaria</i>	3	64
	<i>Paratopula</i>	1	2
	<i>Pheidole</i>	8	245
	<i>Rhopalomastix</i>	2	3
	<i>Solenopsis</i>	1	3
	<i>Strumigenys</i>	5	11
	<i>Tetramorium</i>	11	55
	<i>Vollenhovia</i>	5	30
	<i>Vombisidris</i>	6	25
Ponerinae	<i>Diacamma</i>	2	43
	<i>Hypoponera</i>	2	6
	<i>Leptogenys</i>	1	1
	<i>Odontomachus</i>	1	1
	<i>Platythyrea</i>	1	2
Pseudomyrmicinae	<i>Tetraponera</i>	3	19
<b>TOTAL</b>		<b>227</b>	<b>5104</b>

**Table 3.** Summary of total morphospecies and abundance of leaf litter ants obtained for each genus in MFR.

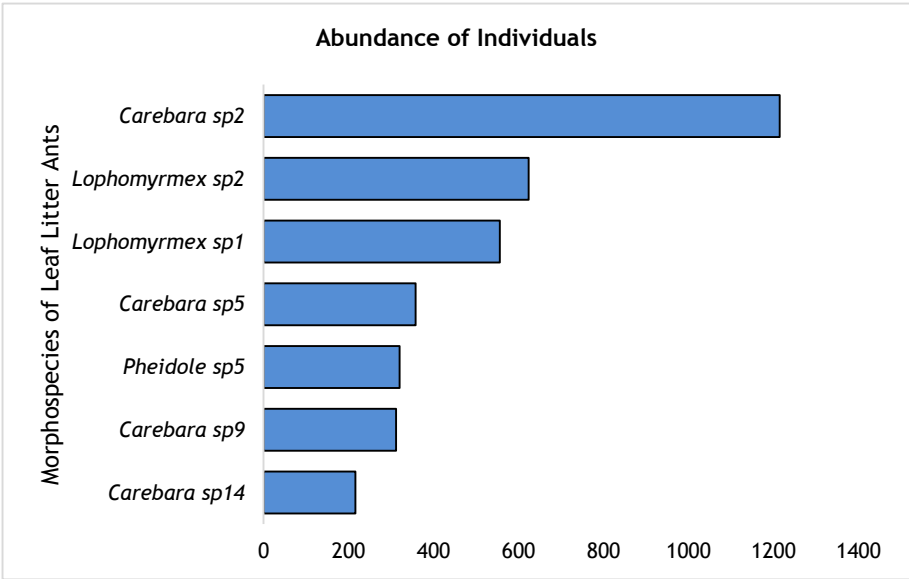
Subfamily	Genus	Total morphospecies	Total abundance
Aenictinae	<i>Aenictus</i>	3	27
Amblyoponinae	<i>Prionopelta</i>	1	51
Cerapachinae	<i>Cerapachys</i>	4	22
Dolichoderinae	<i>Loweriella</i>	1	3
	<i>Tapinoma</i>	2	5
	<i>Technomyrmex</i>	3	17
Dorylinae	<i>Dorylus</i>	1	1
Ectatomminae	<i>Gnamptogenys</i>	2	2
Formicinae	<i>Camponotus</i>	1	1

	<i>Echinopla</i>	1	1
	<i>Euprenolepis</i>	1	1
	<i>Nylanderia</i>	6	398
	<i>Paratrechina</i>	1	2
	<i>Polyrhachis</i>	5	18
	<i>Pseudolasius</i>	4	189
Myrmicinae	<i>Acanthomyrmex</i>	1	1
	<i>Aphaenogaster</i>	2	2
	<i>Calyptomyrmex</i>	3	13
	<i>Cardiocondyla</i>	1	3
	<i>Carebara</i>	12	2281
	<i>Crematogaster</i>	6	50
	<i>Dacatinops</i>	1	2
	<i>Eurhopalothrix</i>	3	43
	<i>Lophomyrmex</i>	4	1306
	<i>Mayriella</i>	1	2
	<i>Monomorium</i>	5	41
	<i>Myrmecina</i>	1	1
	<i>Myrmoteras</i>	1	27
	<i>Pheidole</i>	23	1381
	<i>Pristomyrmex</i>	1	2
	<i>Proatta</i>	1	26
	<i>Pyramica</i>	1	1
	<i>Recurvidris</i>	5	71
	<i>Solenopsis</i>	3	181
	<i>Strumigenys</i>	17	543
	<i>Tetramorium</i>	21	328
	<i>Vollenhovia</i>	2	4
	<i>Vombisidris</i>	1	2
Ponerinae	<i>Anochetus</i>	6	36
	<i>Centromyrmex</i>	1	2
	<i>Cryptopone</i>	1	4
	<i>Emeryopone</i>	1	1
	<i>Hypoponera</i>	18	368
	<i>Leptogenys</i>	3	6
	<i>Odontomachus</i>	1	2
	<i>Odontoponera</i>	1	2
	<i>Pachycondyla</i>	5	113
	<i>Ponera</i>	8	113
Proceratiinae	<i>Discothyrea</i>	3	10
<b>TOTAL</b>		<b>200</b>	<b>7706</b>





**Figure 3** Top seven most abundant morphospecies of canopy ants arranged in descending order.



**Figure 4.** Top seven most abundant morphospecies of leaf litter ants arranged in descending order.

Genus *Polyrhachis* (56 morphospecies, 24.67%) was the most speciose genus for canopy sampling (Table 2), while *Pheidole* (23 morphospecies, 11.50%) was the most diverse in leaf litter sampling (Table 3). *Dolichoderus* sp1 (876 individuals, Figure 5) was the most abundant ant species in the canopy; *Carebara* sp2 (1,215 individuals) was most abundant in leaf litter (Figure 6). Lastly, the most species-rich subfamily for canopy and leaf litter ants were Formicinae (112 morphospecies, 49.34%) and Myrmicinae (116 morphospecies, 58.00%) respectively (Table 4).

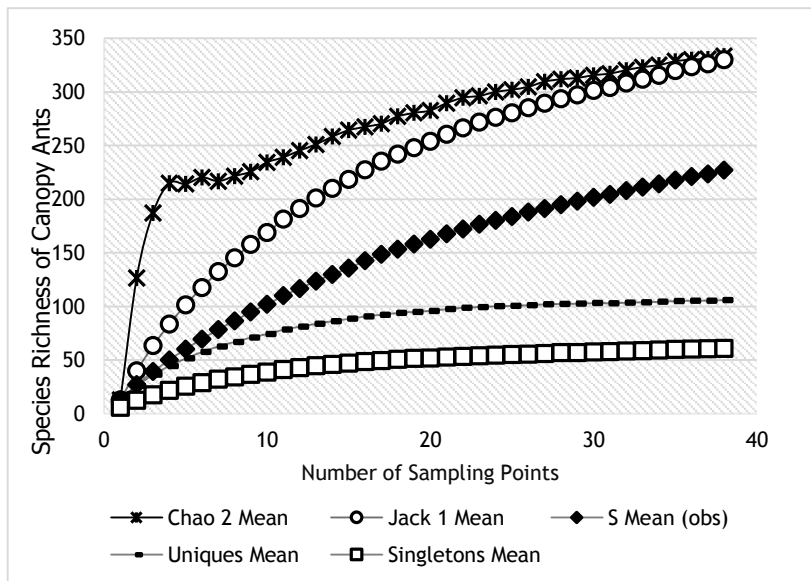
**Table 4.** Number of species and relative species-abundance found in each subfamily for canopy and leaf litter ants.

Subfamily	Number of Species Represented by Canopy Ants (%)	Number of Species Represented by Leaf Litter Ants (%)
Formicinae	112 (49.34)	19 (9.50)
Myrmicinae	85 (37.44)	116 (58.00)
Ponerinae	7 (3.08)	45 (22.50)
Dolichoderinae	20 (8.81)	6 (3.00)
Pseudomyrmicinae	3 (1.32)	0 (0.00)
Cerapachinae	0 (0.00)	4 (2.00)
Proceratiinae	0 (0.00)	3 (1.50)
Aenictinae	0 (0.00)	3 (1.50)
Ectatomminae	0 (0.00)	2 (1.00)
Dorylinae	0 (0.00)	1 (0.50)
Amblyoponinae	0 (0.00)	1 (0.50)
<b>TOTAL</b>	<b>227 (100)</b>	<b>200 (100)</b>

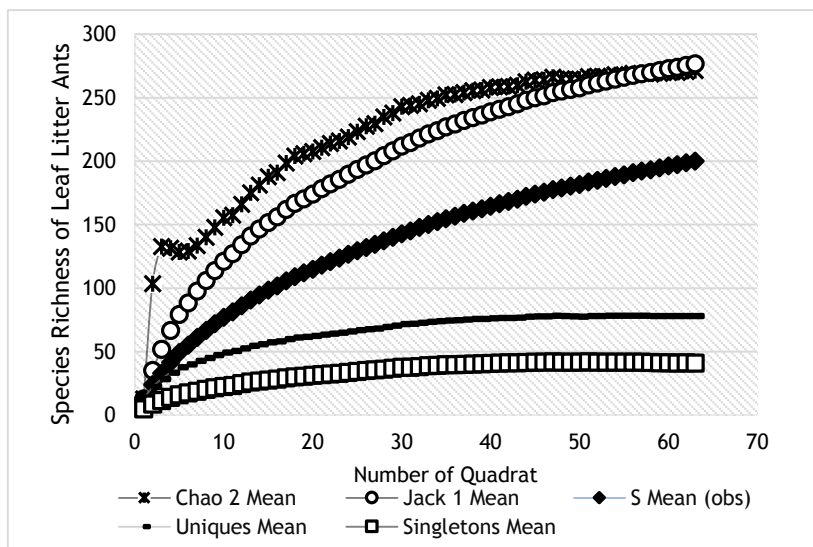
Species richness estimators of Chao2 and Jackknife1 predicted that 68.17% to 68.79% of canopy ant species in MFR were collected, while 72.20% to 73.80% for leaf litter ant species (Table 5). The randomized accumulation curves and the species estimators reveal that additional sampling is required to attain a higher percentage value in the sampling completeness of ant richness in MFR. Of the 227 species of canopy ants recorded, 61 species (26.87%) were singletons, and 106 species (46.70%) were unique. On the other hand, leaf litter ants (i.e., a total of 200 species) recorded 41 (20.50%) singletons and 78 (39.00%) uniques.

**Table 5.** Estimated species richness calculated by Chao2 and Jackknife1 estimators. The observed species richness ( $S_{obs}$ ) for both canopy and leaf litter ants were divided with each richness estimator to obtain the percentage of sampling completeness.

	$S_{obs}$	Chao2 $\pm$ SD	%	Jackknife1 $\pm$ SD	%
Canopy ants	227	333 $\pm$ 27	68.17	330 $\pm$ 14	68.79
Leaf litter ants	200	271 $\pm$ 21	73.80	277 $\pm$ 21	72.20



**Figure 5:** Randomized accumulation curves for canopy ant species richness collected ( $n=38$ ) in Malua Forest Reserve. Curves are given for randomized observed species richness (S Mean), species richness estimators (Chao2 and Jackknife1), and separately for singletons and uniques.



**Figure 6.** Randomized accumulation curves for leaf litter ant species richness collected ( $n=63$ ) in Malua Forest Reserve. Curves are given for randomized observed species richness (S Mean), species richness estimators (Chao2 and Jackknife1), and separately for singletons and uniques.

## Discussion

Borneo ant fauna comprises 12 subfamilies with a minimum of 97 genera (Pfeiffer et al., 2011). In this study, a total of 11 ant subfamilies were recorded, except the family Leptanillinae that was represented by 65 genera. Two thirds of the species collected were in subfamily Myrmicinae and Formicinae for both canopy and leaf litter ants. This is consistent with previous studies done in Borneo, where approximately 70% of the ant species collected belonged to these subfamilies (Sukimin et al., 2010; Yusah et al., 2012; Pfeiffer et al. 2011; Klimes et al., 2015). With about 6,500 described species in the world, Myrmicinae is the most diverse and largest subfamily of ants; followed by Formicinae, the second largest, comprising nearly 3,030 described species (Boudinot, 2015). Not surprisingly, two groups of subfamilies were dominant in this study.

Moreover, we found that *Polyrhachis* was the most genera-rich genus for canopy ants in this study and this is supported by Yusah et al. (2012) and Klimes et al. (2015), where they also found similar results in the tropical forests of Borneo. On the contrary, *Pheidole* was the most speciose genera for leaf litter ants. Not only ecologically dominant, Economo et al. (2014) also noted that the genus is one of the most hyper-diverse species of all the ants in the world and can be found in most of the tropical biomes.

Randomized accumulation curves and species estimators indicate that the sampling completeness of both canopy and leaf litter ants are far from complete. Houadria and Menzel (2021) also detected similar pattern in the subterranean ants of MFR. Although sampling completeness of ant communities can be improved by extensive sampling efforts and incorporating multiple sampling methods (Agosti et al., 2000; King & Porter, 2005), Pfeiffer et al. (2011) argued that in the tropics, many ant species are rare with cryptic behaviour. Thus, it is usual that the accumulation curves of tropical ants do not reach 85% (e.g., Floren & Linsenmair, 2000; Pfeiffer & Mezger, 2012).

By using Winkler extraction and ground-based fogging, our study offers an overview of the pattern of assemblage structure of canopy and leaf litter ants in MFR. We believe this study will form a foundation for future research on ant diversity and abundance, particularly in the secondary forest of Sabah, Borneo. Finally, to thoroughly sample ant assemblage in an area, higher sampling efforts combined with multiple sampling methods are recommended for future ant research.

## Acknowledgements

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## Research Article

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# A Bird Survey of Sungai Kangkawat Research Station, Imbak Canyon Conservation Area, Sabah.

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## Abstract

A bird survey was conducted at the Sungai Kangkawat Research Station (117° 3'34.593"E, 5° 4'29.187"N), Imbak Canyon Conservation Area (ICCA), Sabah from 5<sup>th</sup> until 8<sup>th</sup> October, 2018. The objective of this survey was to produce a preliminary checklist of bird species in the study area. The primary method used was mist-netting with occasional observation from vocal and visual identification. A total of 30 mist-nets were deployed for 11 hours (0630-1730 hr.) for four consecutive days with a final accumulation of 1840 net/hours. Overall, 59 species comprising 23 families were recorded throughout the survey. Of these, 114 individuals were netted representing 41 species from 13 families. The most abundant species caught was the Little Spiderhunter *Arachnothera longirostra* with 26 individuals. Additionally, 18 species from 10 different families were identified via opportunistic sighting and vocalisation. This includes one Critically Endangered (Helmeted Hornbill, *Buceros vigil*), three Vulnerable (i.e., Rhinoceros Hornbill *Buceros rhinoceros*, Large-billed Blue Flycatcher *Cyornis caeruleus* and Blue-headed Pitta *Pitta baidii* and 20 other Near-threatened species. Four endemic species were recorded namely the Bornean Blue Flycatcher *Cyornis superbus*, White-crowned Shama *Copsychus stricklandi*, Black-headed Pitta *Pitta ussheri* and Blue-headed Pitta *Pitta baidii*. We expect higher diversity of birds in ICCA because the species accumulation curve did not reach an asymptote until the last day of sampling. This suggests additional trapping effort with point counts along predetermined transects should be considered for future surveys.

**Keywords:** Birds survey, biodiversity, Imbak Canyon, mist-netting, endemic

## Introduction

The term biodiversity hotspots specifically refer to; currently 35 identified biological rich areas around the world that have lost at least 70 percent of their original habitat (Mittermeier et al., 2011). A biodiversity hotspot is a biogeographic region, playing a significant role to hold a large amount of biodiversity, as a reservoir, yet being knocked down with massive threats through anthropogenic activities, resulting in catastrophic population lost (Brooks et al., 2002; Mittermeier et al., 2011). Borneo Island is one of the eight hottest global biodiversity hotspots in Southeast Asia, along with Madagascar, Philippines, Brazil's Atlantic Forest, Caribbean, Indo-Burma, Western Ghats and Sri Lanka, and lastly, the Eastern Arc and Coastal Forest of Tanzania or also known as Kenya (Dowksza, 2002). Being the third largest island in the world after Greenland and Guinea, with a size of more than half a million square kilometers, covering an area slightly larger area than Texas, Borneo is home to some of the world's most diverse rainforest and Southeast Asia's last intact forest with extraordinary biodiversity; habitat to marvelous creatures like the orangutans, clouded leopard and sun bears. Despite this richness, the forest of Southeast Asia still encounters dramatic changes due to human anthropogenic activities for over the past 40 years (Curran et al., 2004; Sodhi et al., 2004)

Sabah, is a Malaysian state that occupies part of the northern Borneo Island. It is famed with its 4,095 m tall Mount Kinabalu, Malaysia's highest peak, crowned with distinctive granite spires. Sabah is also known for its beaches, rainforest, coral reefs, and abundant wildlife, mostly within protected areas including parks and reserves. In total, there are 93 named protected areas in the State. The number seems many, with approximately 75% of the land area under forest cover but much of these experience high selective logging problems in various regeneration stages (Reynolds et al., 2011). Imbak Canyon Conservation Area (ICCA) was in 2009 gazetted as a protected area in Sabah, a Class I Forest Reserve that functions to safeguard water supplies, soil fertility and environmental quality. It is located 300km from Kota Kinabalu, at central Sabah and has an area of approximately 30,000 hectares of undisturbed forest, with a 25km long valley. ICCA provides numerous biodiversity and serves as an excellent field of study to many researchers around the world, linking two other protected areas, the Maliau Basin and Danum Valley conservation areas. ICCA is also the biggest pure lowland dipterocarp forest remaining in Sabah and serves as the water catchment area for the longest river in Sabah, the Kinabatangan River and its river system. Although these three areas are connected with each other, ICCA is different as it consists of undisturbed pristine lowland rainforest.

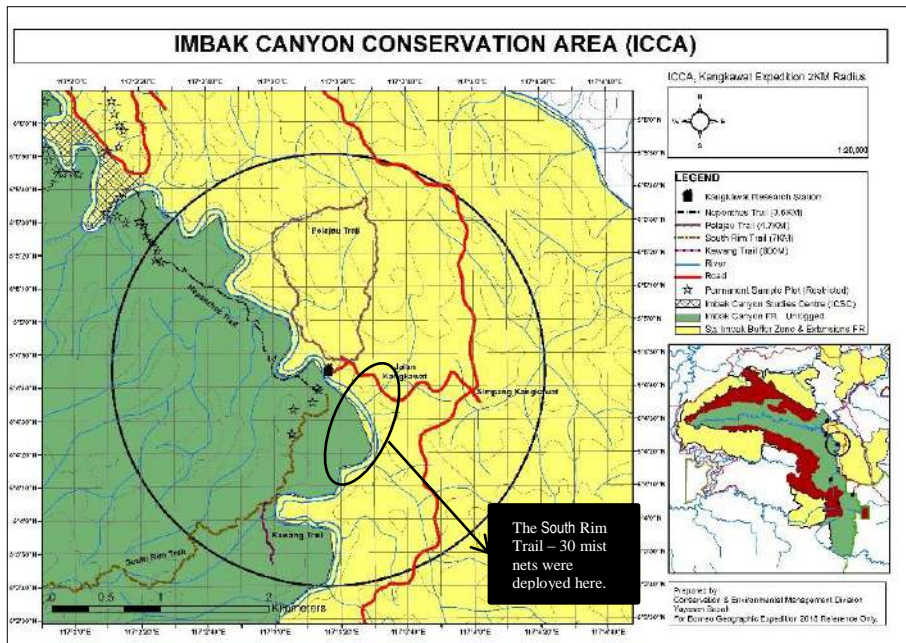
Birds are one of the biodiversity components and play an important role in maintaining a balanced ecosystem, providing numerous ecological services. Not only that, birds have been widely used by many researchers as they can act as effective biological indicators since they are able to respond quickly towards any changes that happen in their environment (Sodhi et al., 2005; Yap et al., 2007; Gilbert et al., 2018). According to Phillipps and Phillipps (2014) up until now, the recorded avifauna in Borneo is made up of at least 669 bird species of which 51 are endemic. Back in 2010, an unpublished record of an expedition is ICCA reported a total of 71 bird species including the Scarlet-rumped Trogon *Harpactes duvaucelii* and Oriental Bay Owl *Phodilus badius*. However, the data was considered as incomplete and needed more input, according to one of the participants (Emmor, 2011). This clearly shows that there are still limited bird studies to provide significant information in Sabah particularly in ICCA. Thus, the aim of this survey is to produce a preliminary checklist of the bird species that exist in the ICCA, Sabah.

## Material and Methods

### *Study site*

The study area is located at the eastern part of the canyon near Sungai Kangkawat and is named Sungai Kangkawat Research Station (117° 3'34.593"E, 5° 4'29.187"N). Sampling was conducted over four consecutive days from 5<sup>th</sup> until 8<sup>th</sup> October, 2018. Located to the north of Maliau Basin Conservation Area in central Sabah, the canyon encompasses a range of rainforest habitats within a 10 km long valley (250 m asl) where the Sungai Kengkawat Research Station is in the eastern part of the canyon near Kangkawat River (Figure 1).

Being one of the Yayasan Sabah Forest Management Areas in Central Sabah, Malaysia Borneo with an area of 30,000 ha, ICCA is one of the most important protected primary forests in Southeast Asia aside from Maliau Basin and Danum Valley conservation areas with an area of 588 km<sup>2</sup> and 438 km<sup>2</sup> respectively (Reynolds et al., 2011). The vegetation in and around the Sungai Kangkawat Research Station is predominantly characterised by a pristine lowland mixed dipterocarp forest. Other forest types located within walking distance of 2-10 km radius from the Kangkawat River comprise of upland mixed dipterocarp forest, riverine forest, lower montane kerangas forest and limestone forest. A logged over forest is located nearby about 0.5-1 km from the base camp site.



**Figure 1** Map of Sungai Kangkawat Research Station. Mist nets were deployed at the South Rim Trail.

### *Data collection*

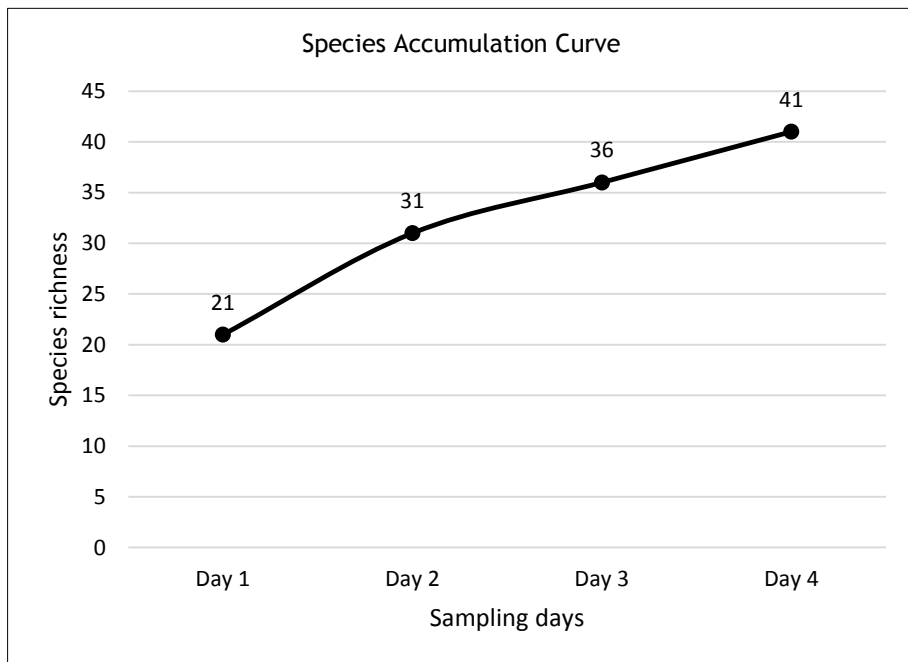
Throughout the sampling period, a total of 30 understorey mist-nets (15m x 2.5m x 18mm mesh) were set up along the South Rim Trail at 100-150 m intervals and were checked at two hours interval. Twenty nets were left open for 11 hours (0630 - 1730 hr.) and another 10 nets were left open for 24 hours to trap fruit bats. All mist nets were set at least 0.5 m above the ground to reduce the risk of captured birds at the bottom shelf being exposed to ground-dwelling predators like monitor lizards (Arif & Mohd-Azlan, 2014). The total net effort for the mist netting was 1840 net hours.

Birds caught from the nets were immediately transferred into cloth bags prior to the banding and measuring process. Captured birds were measured on site and released immediately to avoid any casualties. Ten morphological characters were recorded, including tarsus (TR), bill length (BL), bill depth (BD), bill width (BW), head bill (HB), wing length (WL), wing span (WS), tail-anus (TA), total length (TL) and weight (W). All bill/feather-based characters were measured using a 30-cm stiff metal ruler and a Mitutoyo® digital caliper (Japan) while weight was measured using PESOLA® light spring-loaded scales model (Switzerland); 50 g, 100 g and 300 g. In addition, molting (MO) and brood patch

(BP) stages were also noted (if any). The birds were ringed accordingly based on their tarsus size with custom aluminum rings. The reason of putting the ring on is so that any recaptured individuals would not be counted twice. All bill-related measurements were standardized to the nearest 0.01 mm. Birds were identified following Phillipps and Phillipps (2014) and Myers and Allen (2009). Additionally, opportunistic bird survey was also conducted via visual and aural observation.

## Results and discussion

A combined total of 59 species from 24 different families were identified over four consecutive sampling days from 5<sup>th</sup> until 8<sup>th</sup> of October, 2018 (Table 1). Comparatively, 41 species were netted while and additional number of 18 species were recorded either by visual observation or vocalization. In detail, 113 individuals were netted and ringed to avoid any recaptured individuals being count twice (Arif & Mohd-Azlan, 2014).



**Figure 2.** Species accumulation curve with number of species recorded during the sampling period.

Based on the species accumulation curve, a total of 21 species were recorded on the first day of sampling (Figure 2). On the second day, the list rose by 10 species, whilst another ten species were recorded on the final two days of

sampling. Although the species accumulation did not reach the asymptote, the curve nonetheless showed species addition the very final day of the sample period. This suggests that longer sampling period may provide a better insight of the bird diversity in ICCA.

The Little Spiderhunter *Arachnothera longirostra* was recorded as the most abundant species, with 26 individuals caught. This was followed by Grey-chested Jungle Flycatcher *Rhinomyias umbratilis*, Yellow-bellied Bulbul *Criniger phaeocephalus*, Scaly-crowned Babbler *Malacopteron cinereum*, and Rufous-winged Philentoma *Philentoma pyrrhopterum*. The largest family was represented by Timaliidae which consists of 11 species such as the Black-capped Babbler *Pellorneum capistratum*, Browned Fulvetta *Alcippe brunneicauda*, Rufous-crowned Babbler *Malacopteron magnum*, Scaly-crowned Babbler *Malacopteron cinereum*, Short-tailed Babbler *Trichastoma malaccense*, Sooty-capped Babbler *Malacopteron affine*, Striped-wren Babbler *Kenopia striata*, and White-chested Babbler *Trichastoma rostratum*. The second largest family was dominated by the Muscicapidae family with a total of 5 species recorded including the endemic Bornean Blue Flycatcher *Cyornis superbus*.

Taken collectively, the total number of species recorded during this survey was considerably modest given that the sampling period was only limited to four days. Our mist netting effort yielded a high abundance of understorey birds like the Bulbuls (Family: Timaliidae). This group of birds are shy ground-dwellers that occupy various forest types with favourable density of trees (Forshaw, 1991; Lindsell, 2001). A Siberian Blue Robin (Turdidae) known as a migratory insectivorous species breeding in Eastern Asia across to Japan, was netted. This species winters in South-east Asia including Peninsular Malaysia, Borneo and Thailand, in order to have a continuous diet of insects (Forshaw, 1991).

Some notable observations include the frequent calls of the Great Argus Pheasant *Argusianus argus* and Helmeted Hornbill *Buceros vigil* which were heard almost every day throughout our observation. Also, the Rhinoceros Hornbill *Buceros rhinoceros* was also sighted regularly throughout the sampling period. This indicates that ICCA serves as a high-quality habitat as these birds are known to be very sensitive towards habitat change. These interesting species were categorized as Critically Endangered (Helmeted Hornbill) and Vulnerable (Rhinoceros Hornbill) and (Great Argus Pheasant) on the IUCN (2021). This could probably be caused by difficulties in searching for suitable habitat as they are being threatened with extensive habitat loss and ongoing hunting pressure.

Table 1. The list of bird species recorded in Sungai Kangkawat Imbak Canyon Conservation Area (ICCA)

Family	Local Name	Scientific Name	WCE 1997	IUCN	Mn	Obs	Dist
Accipitridae	Crested Serpent Eagle	<i>Spilornis cheela</i>	P	LC	-	/	CR
Alcedinidae	Oriental Dwarf Kingfisher	<i>Ceyx erithaca</i>	NP	LC	1	-	CR
	Rufous-backed Kingfisher	<i>Ceyx rufidorsa</i>	NP	LC	4	-	CR
Bucerotidae	Helmeted Hornbill	<i>Buceros vigil</i>	P	CR	-	/	SR
	Rhinoceros Hornbill	<i>Buceros rhinoceros</i>	P	VU	-	/	LCR
Cisticolidae	Ashy Tailorbird	<i>Orthotomus ruficeps</i>	NP	LC	-	/	CR
Corvidae	Crested Jay	<i>Platylophus galericulatus</i>	NP	LC	-	/	CR
Dicaeidae	Yellow-breasted Flowerpecker	<i>Prionochilus maculatus</i>	NP	LC	2	-	R
Dicruridae	Greater Racquet-tailed Drongo	<i>Dicrurus paradiseus</i>	NP	LC	-	/	CR
Eurylaimidae	Black-and-red Broadbill	<i>Cymbirhynchus macrorhynchus</i>	NP	LC	-	/	CR
	Green Broadbill	<i>Calyptomena viridis</i>	NP	NT	2	-	CR
Irenidae	Asian Fairy-bluebird	<i>Irena puella</i>	NP	LC	-	/	CR
Megalaimidae	Blue-eared Barbet	<i>Megalaima australis</i>	NP	LC	-	/	CR
	Red-crowned Barbet	<i>Megalaima rafflesii</i>	NP	NT	-	/	CR
	Yellow-crowned Barbet	<i>Megalaima henrici</i>	NP	NT	-	/	SMR
Meropidae	Red-bearded Bee-eater	<i>Nyctornis amictus</i>	NP	LC	-	/	CR
Monarchidae	Asian Paradise Flycatcher	<i>Terpsiphone paradisi</i>	P	LC	1	/	CR
	Pied Fantail	<i>Rhipidura javanica</i>	NP	LC	-	/	LR
	Spotted Fantail	<i>Rhipidura perlata</i>	NP	LC	2	-	LR
Muscicapidae	Bornean Blue Flycatcher	<i>Cyornis superbus</i>	NP	LC	2	-	E
	Grey-chested Jungle Flycatcher	<i>Rhinomyias umbratilis</i>	NP	NT	7	-	R
	Hill Blue Flycatcher	<i>Cyornis banyumas</i>	NP	LC	2	-	SR
	Large-billed Blue Flycatcher	<i>Cyornis caerulatus</i>	NP	VU	2	-	SR
	Rufous-tailed Jungle Flycatcher	<i>Rhinomyias ruficauda</i>	NP	LC	1	-	R
Nectariniidae	Little Spiderhunter	<i>Arachnothera longirostra</i>	NP	LC	26	-	CR
	Long-billed Spiderhunter	<i>Arachnothera robusta</i>	NP	LC	-	/	SR
	Purple-naped Sunbird	<i>Hypogramma hypogrammicum</i>	NP	LC	2	-	CR
Phasianidae	Crested Partridge	<i>Rollulus rouloul</i>	P	NT	-	/	CR
	Great Argus Pheasant	<i>Argusianus argus</i>	P	VU	-	/	LRF

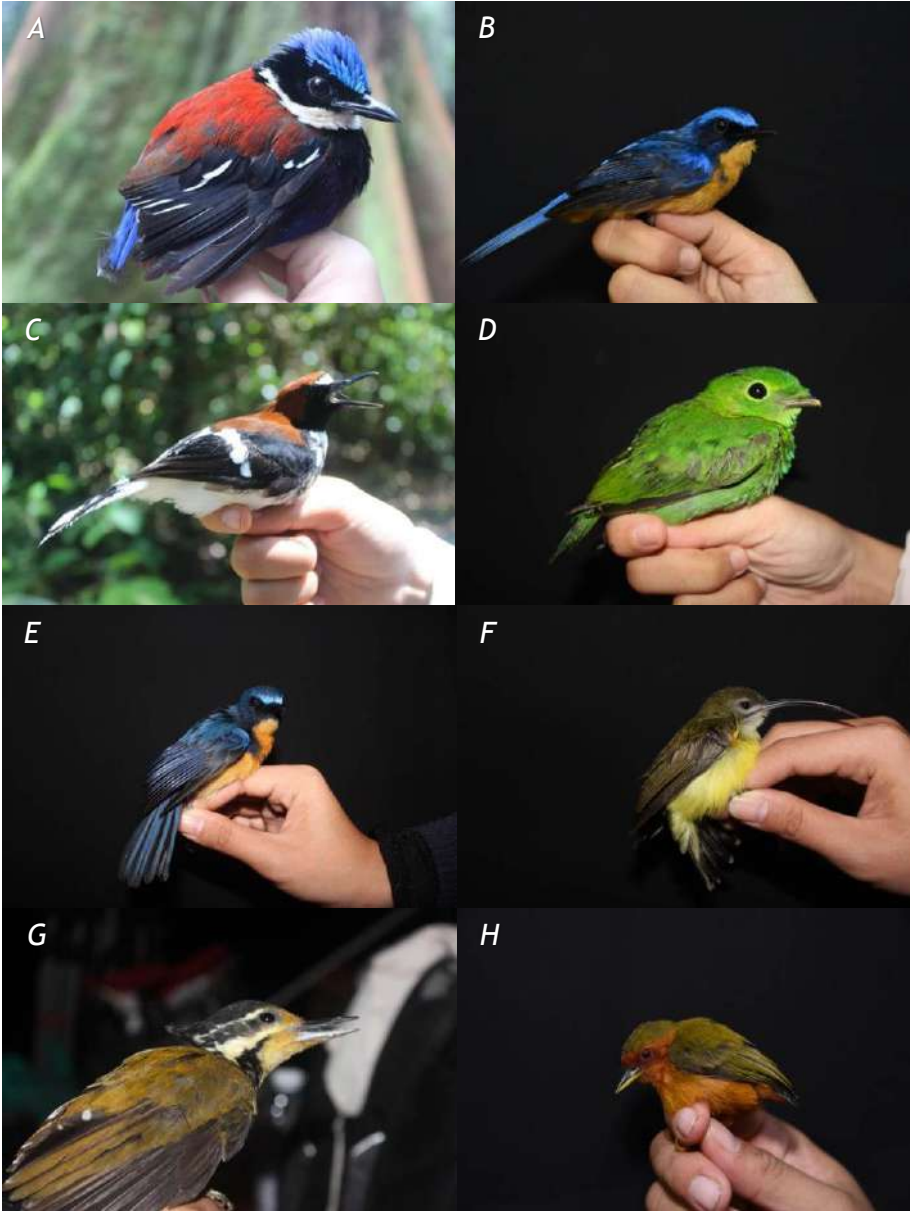
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Table 1. (Continued)

Picidae	Buff-necked Woodpecker	<i>Meiglyptes tukki</i>	NP	NT	1	-	CR
	Maroon Woodpecker	<i>Blythipicus rubiginosus</i>	NP	LC	1	/	CR
	Olive-backed Woodpecker	<i>Dinopium rafflesii</i>	NP	NT	1	-	SR
	Rufous Piculet	<i>Sasia abnormis</i>	NP	LC	2	-	CR
Pittidae	Black-crowned Pitta	<i>Pitta ussheri</i>	NP	NT	-	/	LE
	Blue-headed Pitta	<i>Pitta baudii</i>	P	VU	1	/	LE
	Garnet Pitta	<i>Erythropitta granatina</i>	NP	NT	1	-	LR
Pycnonotidae	Grey-cheeked Bulbul	<i>Alphoixus tephrogenys</i>	NP	VU	2	-	CR
	Hairy-backed Bulbul	<i>Tricholestes criniger</i>	NP	LC	2	-	CR
	Yellow-vented bulbul	<i>Pycnonotus goiavier</i>	NP	LC	1	-	CR
	Yellow-bellied Bulbul	<i>Griniger phaeocephalus</i>	NP	LC	6	-	LR
Sturnidae	Hill Myna	<i>Gracula religiosa</i>	P	LC	-	/	CR
Timaliidae	Black-capped Babbler	<i>Pellorneum capistratum</i>	NP	LC	1	-	CR
	Brown Fulvetta	<i>Alcippe brunneicauda</i>	NP	NT	2	-	CR
	Rufous-crowned Babbler	<i>Malacopteron magnum</i>	NP	NT	2	-	CR
	Scaly-crowned Babbler	<i>Malacopteron cinereum</i>	NP	LC	6	-	CR
	Short-tailed Babbler	<i>Trichastoma malaccense</i>	NP	NT	1	-	CR
	Sooty-capped Babbler	<i>Malacopteron affine</i>	NP	NT	4	-	CR
	Striped-wren Babbler	<i>Kenopia striata</i>	NP	NT	1	-	SR
	White-chested Babbler	<i>Trichastoma rostratum</i>	NP	NT	1	-	cr
	Grey-headed Babbler	<i>Stachyris poliocephala</i>	NP	LC	1	-	LR
	Chestnut-rumped Babbler	<i>Stachyris maculata</i>	NP	NT	3	-	CR
	Chestnut-winged Babbler	<i>Stachyris erythroptera</i>	NP	LC	3	/	CR
Trogonidae	Scarlet-rumped Trogon	<i>Harpactes duvaucelii</i>	NP	NT	3	-	LLR
	Chestnut-naped Forktail	<i>Enicurus ruficapillus</i>	NP	NT	1	-	LR
	Siberian Blue Robin	<i>Luscinia cyane</i>	NP	LC	2	-	SWV
	White-crowned Forktail	<i>Enicurus leschenaulti</i>	NP	LC	1	-	LR
	White-crowned Shama	<i>Copsychus stricklandi</i>	NP	LC	3	/	CE
Tytonidae	Oriental Bay Owl	<i>Phodilus badius</i>	P	LC	1	-	SR
Vangidae	Rufous-winged Philentoma	<i>Philentoma pyropteron</i>	NP	LC	5	-	LR

Note that WCE 1997 = Wildlife Conservation Enactment, P = Protected, NP = Not Protected, IUCN = IUCN Red Data list (2021-1); NE = Not Evaluated, DD = Data Deficient, LC = Least Concern, NT = Near Threatened, V = Vulnerable, EN = Endangered, CR = Critically Endangered, EW = Extinct in the Wild and EX = Extinct. Mn. = Number of Individuals mist-netted, Obs = Species recorded by observation, Dist = Distribution, CR = Common Resident, SR = Scarce Resident, LCR = Locally Common Resident, R = Resident, SMR = Sub montane Resident, \*E = Endemic, LR = Local Resident, SWV = Scarce Winter Visitor, CE = Common Endemic, LRF = Local Resident Forest, LLR = Lowland Resident







**Figure 3.** A) Blue-headed Pitta, B) Bornean Blue Flycatcher, C) Chestnut-naped Forktail D) Green Broadbill E) Hill Blue Flycatcher F) Little Spiderhunter G) Olive-backed Woodpecker H) Rufous Piculet I) Rufous-backed Kingfisher J) Scarlet-rumped Trogon K) Siberian Blue Robin L) White-crowned Forktail M) White-crowned Shama N) Rufous-winged Philentoma

## Conclusion and Recommendation

The Imbak Canyon Conservation Area is a critical area for bird conservation because the protected areas remain intact as a high-quality habitat. This ensures that ICCA as a whole continues to support a large number of significant endemics and rare species like the Helmeted Hornbill, Garnet Pitta, Blue-headed Pitta, Great Argus and White-rumped Shama. Higher bird richness was expected within ICCA if a longer sampling period is considered. This was supported by the species accumulation curve that depicted an increasing curve until the last day of sampling. Future sampling efforts should include comprehensive bird surveys

with ecological perspectives including temporal and spatial considerations of the bird community in the upper-storey and forest canopy. Furthermore, multiple sampling strategies that incorporate canopy mist-netting method with visual sightings and aural survey recordings should be considered to increase capture rate and sampling efficiency.

## Acknowledgment

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## Research Article

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# Balance environmental obligations and socio-economic compulsion; the case of Kadamaian's indigenous community proficiency needs

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## Abstract

The community of Kadamaian is expected to have heightened sense of environmental and biodiversity awareness and an obligation towards protection and conservation since the community occupies an area adjacent to Sabah's premier World Heritage Site i.e. Kinabalu Park. Of course, with support from the Sabah government, Kadamaian's community is compelled to exploit their natural surroundings as primary resources to better their socio-economic position to be at par with other communities elsewhere in Sabah. Will Kadamaian's community be able to balance the obligation and the compulsion to exploit? An empirical study in a scientific expedition that was carried out in late 2019 at Kadamaian-Kota Belud answers this question. The unit of analysis in this study is the social relationship between Kadamaian's community and its social artifacts. The documentation technique, direct observation technique and the interview technique were used to collect data, while data analysis was carried out according to the structuration theory using the triangulation method. The findings of this study showed the existence of three (3) interdependent social relationships that influenced Kadamaian's community strive to balance environmental and socio-economic issues. While these relationships have somewhat contributed to the establishment of a non-governmental organization i.e. Kadamaian Biocultural Protocol Committee that focused on the environmental obligation of Kadamaian's community, other small enterprising network focused on socio-economic compulsion. The implication of these relationships is discussed in the article.

**Keywords:** Structuration, Behavioural sink

## Background

Sabah is world renowned for its environmental contributions. From the many types of environmental issues, Sabah has been quite successful in addressing two (2) environmental issues namely deforestation and biological diversity conservation. With many key areas protected using the concept of fortress conservation, the government is now transitioning towards rights-based conservation in the hope of creating buffer zones for the existing protected

areas as well as increasing contributions of indigenous communities in conservation through the concept of Other Effective Area-based Conservation Measures. Indigenous community support is needed. But this transition is only possible if the respective community finds it worthwhile (Schulze & Suratman, 1999). The root factor for such a change is the community's collective assessment of the potential benefits to be gained by having a relationship with the government and complying with the government's plan versus the types of risks involved. If the cost of a relationship is higher than its reward, the relationship will face challenges (Emerson, 1976, p. 345), which would eventually lead to a situation where the government has no support for its plan. Hence, for a government that wants to transition from fortress conservation to rights-based conservation, it is important to determine whether a community in Sabah can transition and whether the cost that a community would need to bear during such transition is acceptable.

## Introduction

Kadamaian is an area covering some 445 km<sup>2</sup> located to just the west of the renowned Kinabalu Park - Malaysia's first site listed under UNESCO's World Heritage Convention. It is in a rural region of Sabah. Development in and around Kadamaian has been ongoing from as early as the 1940s with the establishment of the Kadamaian routes, which were upgraded from footpaths of the indigenous peoples inhabiting the area. Socio-politically, Kadamaian falls within the district of Kota Belud - a district with a population of about 95,000 and a third of that population stays in the Kadamaian area. The largest town in Kadamaian is Taginambur, which is about 10 km from Kota Belud town and about 20 km from Tuaran town. There are 76 villages in Kadamaian, and its inhabitants comprise the Kadazandusun, Bajau and Irranun. While majority in Kota Belud are Muslim, the communities of Kadamaian are mainly Christians who still appreciate some of their ancestral pagan beliefs. Socio-economically, many of the inhabitants of Kadamaian are either farmers, civil servants or tourism entrepreneurs.

Geographically, Kadamaian is an undulating area with many rivers. It is an important water catchment area to the Northwest region of Sabah. The Kadamaian and Wariu rivers flow to this area. Although Kadamaian is an important water catchment area, more than 60% of Kadamaian's inhabitants do not enjoy treated water supply and have to rely on gravity water supply. Along with other matters, Kadamaian's communities are considered far behind urban communities in terms of its capitalistic strengths. Nevertheless, since it is just outside of the Kinabalu World Heritage Site and its inhabitants may possess

certain unique indigenous knowledge, Kadamaian has an important role to play and its community is obligated to support Kinabalu Park's environmental and biodiversity conservation efforts. There are a few conservation acts that are community driven. Among these are the implementation of the tagal system. Among natives, this is inbuilt, but now most require consolidation since there is a need for that consolidation to network with government and non-governmental organizations. The question is, does Kadamaian's community have the relevant knowledge to fulfill their obligations and still pursue socio-economic interests? A community's education approach to the act as an environmental custodian is unique. How do we study education needs of a community? While scientific research may be preferred, it is seldom carried out simply because it is technically just too laborious. The general approach in assessing a community's educational needs is the establishment of a technical committee that would eventually produce a technical report, but this approach is not free from political criticism. Understanding matters such as social network and material flows may prove useful because it ties together environmental practices (Flikke, 2014). Hence, to explore the question about a community's ability to balance demands and obligations, we need to know local culture and traditions, and if the concept of conservation is a component to these cultures and traditions.

### ***Communication Media as a Factor of Social Relationship***

One could form an understanding about how a community organizes and functions in a particular environment by studying the community's artifacts. This is possible when Marshall McLuhan's theoretical perspective about a medium is drawn upon. According to McLuhan (1964), mediums are instruments or tools that extend a man's interest; mediums are extensions of man. The message of this extension either is a message about enhancement, a message about obsolesces, a message about retrieval, and a message about reversal (McLuhan, 1964). The four (4) message possibilities are knowledge as the Laws of Media (McLuhan, 1964). Of course, a medium (pl. media) does not contain within itself any message; whatever is in a medium are merely forms of arts. The medium IS the message (McLuhan, 1964). Because of McLuhan's perspective, instruments and tools act as mediums to communicate a particular aspect of the said community. Thus, communication media studies are about the studies of media, its effect on society and its relationship with its surroundings. Moreover, while this field of study is commonly associated with commerce and the modern environment, it could also be associated to socialization and the traditional environment e.g. environmental education and nature-based tourism.

### ***Studying the educational needs of a community in Sabah***

Under the Malaysian education law, every child is required to finish primary education. Secondary level education or training is at discretion. Parents have a choice to send their children to public schools, private schools or training institutions and centres. Some of these education and training providers offer environmental education as extra-curriculum. It all depends on the providers since environmental education is not compulsory.

Education is important. There is huge literature on its importance. This literature shows that an absence of education and the low level of education can lower a family's household income since the absence of education and the low level of education are factors of job permanency (Zurina, Hukil & Norinsan, 2018). Hence, the question about the importance of education to a community is indeed a rhetorical question. It is an enabler, and people are willing to migrate in pursuit of education. It has been well documented that youth migrate to urban areas to pursue education (Haji Abbas & Binsin, 2008, p. 347). However, the implication of this migration is on the community. The question about why some people place more importance on education than others is not a rhetorical question. An answer to that question comes in many ways and one of them is through the Social Exchange Theory.

The Social Exchange Theory is also an example of an example of a human capital theory. This theory, which modeled the interpretation of a society based on a series of interactions between people, states that people's interactions e.g. teacher-student interaction and trainer-trainee interaction are based on estimates of rewards and punishments. According to this view, our interactions are determined by the rewards or punishments that we expect to receive from others excluding the educators and facilitators, which we evaluate using a cost-benefit analysis model (whether consciously or subconsciously). This theory was described by Richard M. Emerson in 1976, the same scholar that gave us the stakeholder theory. The importance of this theory is that it provides a justification for the need to migrate in pursue of education.

Forwarded by Anthony Giddens, the Structuration Theory is a way of explaining what constitutes a society. According to Giddens (1984), a society has three different properties or dimensions namely the authority dimension, the development dimension and the facilitation dimension. In the context of Malaysia, this theory was first used to explore the contribution of Imbak Canyon to educational opportunities. For education at Imbak Canyon, the Education Ministry forms the authority dimension, while Yayasan Sabah and PETRONAS



respectively form the development and facilitation dimensions. In Kadamaian's case, modality is the intermediary to the educational relationship between agencies operating at Kadamaian and the agents of Kadamaian. A structuration matrix is a matrix that illustrates a society's blueprint and how that blueprint affects the society. Each dimension consists of three social concepts namely the structure, the modalities or realities and the interactions. With Giddens' theory, determining an agency, an agent and/or a norm is made easy. More importantly, Giddens' theory helps clarify a society's internal network. Of course, Malaysia has not yet appreciated Structuration Theory as a grounding theory to understand social network and how material flows in that network, and thus knows if scientific research could explore a community's educational needs. In the context of Kadamaian's community involvement in conservation, one could easily determine the community's educational capabilities and needs by drawing from the Structuration Theory; and ultimately answer the question about balancing the role of environmental obligations.

## Methodology

The unit of analysis in this study is the social relationship between the Kadamaian's community and its social artifacts. The unit of analysis is the mediums of Kadamaian's community. The research location is a rural region and it is noted in the following map.

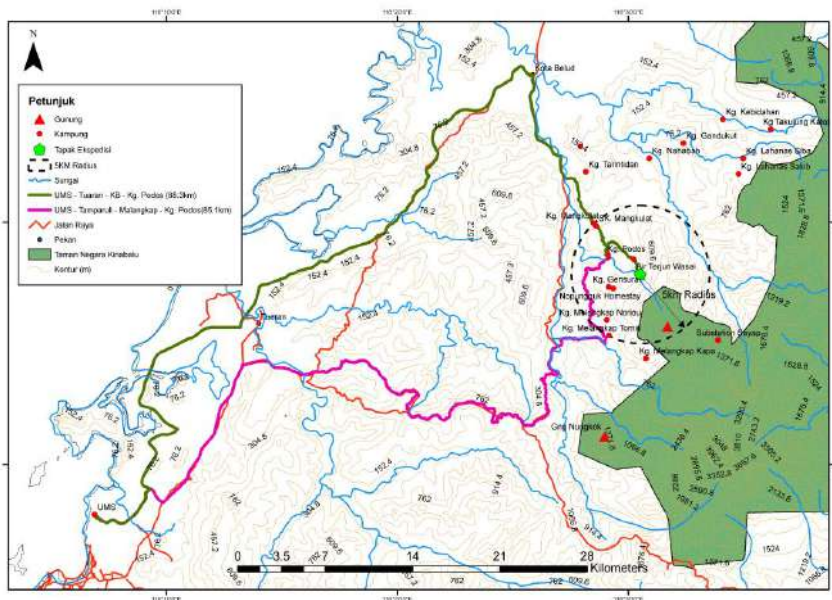


Figure 1: Research Location

The places where data collection was carried out include Malangkap village, Podos village, Bayayat village, and Taginambur town within the dotted circle adjacent to a protected landmass under Sabah Parks. An empirical study was done during a scientific expedition that was carried out from 14<sup>th</sup> to 25<sup>th</sup> October, 2019 known as the Borneo Geographic Expedition. This expedition was organized by the Institute for Tropical Biology and Conservation, supported by Sabah Parks.

As described earlier, mediums are extensions of man (McLuhan, 1964); and in this study, data about Kadamaian's mediums were collected using the documentation technique, direct observation technique and the interview technique. Documentation technique was applied on existing electronic media platforms that contain information about the research location such as *JTYWMA Pro Geospatial Mapping Application*, *Google Image*, *Facebook*, *Youtube®* and *Tripadvisor®*. Direct observation technique was carried out *ad hoc* at tourist spots. Since the research location is stated as a rural region, the focus of the collection was on extra-ordinary mediums that exist at the research location. The interview technique was carried out on two groups of people namely the residents of Kadamaian and the community developers and leaders associated to Kadamaian. Open-ended questions on casual lifestyle was given to the former, and socio-economic development of Kadamaian was given to the latter.

Data analysis was carried out according to the Structuration Theory using the triangulation method. Explanation-building technique according to McLuhan's Laws of Media was used to analyse the collected data.

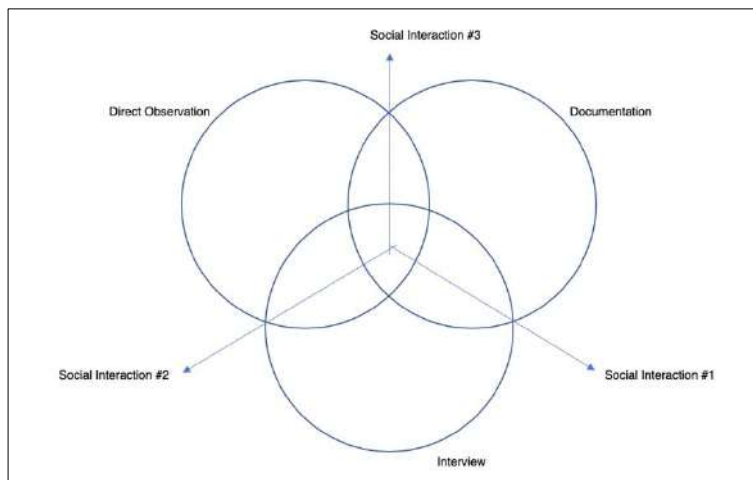


Figure 2: Conceptual framework

Figure 2 shows an analytical triangulation of three different data collection techniques. The explanation building technique used to analyze data will produce at least three (3) social interactions, which will be reported in the following section.

## Results and Discussion

In a recent socio-economic development study by the Institute for Development Studies Sabah (IDS) on Kadamaian, much of the output was focused on structural matters that regional and local authorities could monitor. Human capital development (Mat et al., 2015) e.g. entrepreneurship and job creation, which require an understanding about humanities, was confirmed in an interview by a member of the research team. But the interview also uncovered certain aspects of humanities like the level of environmental awareness in Kadamaian's community, sustainable traditional usage of Kadamaian's resources, the adoption of environmentally friendly technology, while the community members' readiness were prioritized the least. In addition, aspects relating to environmental pressures and conservation matters were also uncovered to be least prioritized. Existing strength in environment related enforcement may have led IDS away from environment related aspects. But without knowing the understanding about the environment among the Kadamaian community, there will always be a challenge to conservation.

Drawing from the Structuration Theory, the data was collected and analyzed. The result of the analysis is illustrated in the following matrix.

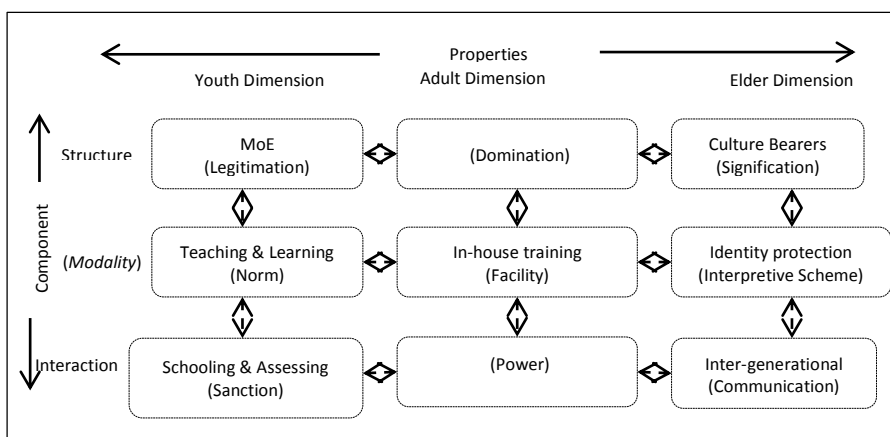


Figure 3. Structuration matrix of Kadamaian's education network (adapted from Giddens (1984))

The above Figure 3 shows Kadamaian's education network. It has three (3) social relationship interdependent patterns i.e. dimensions. The first dimension is about a relationship between community elder members with their surroundings. The relationship focuses on the elders' need to maintain their traditions and culture through any means possible. The second dimension is between Kadamaian's youths and their surroundings. That relationship focuses on the youths wanting to migrate since they technically have no authority or power and their acquired knowledge from schooling does not motivate them to remain. The last dimension is about a relationship between adults and their surroundings, and the focus is on the limited conservation activities that adults participate in. In Figure 3, education is categorized under the component of modality, and under the Youth Dimension it is carried out through a specific interaction due to the existence of a governing entity. A similar but rather loose matter exists under the Adult Dimension as in-house training, while within the Elder Dimension, education has evolved into a practice i.e. the protection of an identity. The structuration matrix is essential because it highlights three (3) aspects in relation to a community's conservation obligation and its ability to fulfill that obligation that is worth discussing. First, the older community members of Kadamaian have hierarchical needs. Second, Kadamaian's youths have no need for environmental education, and last is the inability of the adult group to balance socio-economic demands and conservation obligations.

This matrix adds clarity to all social exchanges that take place when members of a community analyze the cost and benefit of a matter like education. This matrix enables us to determine the education needs in appreciating of the concept of conservation in daily lives. In this study, Kadamaian's land-use practises are quite rigid. The older generation still carry out their duties. This generation wants their land to be conserved, and the younger generation is to deal with conservation and the commercialisation of its results. The latter is where the problem lies. Not knowing how to commercialise and monetise those resources have forced the younger generation to seek opportunities elsewhere. The local government is assisting but their assistance is focused on commercial resource preparation only. It is believed that once the physical development of Kadamaian has taken off in accordance to Kadamaian's master plan, the scale between socio-economic demands and environmental obligations may be tipped. However, unless education is localized to the needs of the land and youths are recruited to be next in line in defending and implementing, it will be difficult to balance that scale.

***Community's Hierarchy Needs***

As introduced earlier, Kadamaian is west of Kinabalu Park, and has a very undulating landscape. A satellite image of the area (refer Appendix A) shows a distinct difference between privately owned lands outside of the park and Kinabalu Park. Forest cover outside the park is patchy whereas the forest cover within the park is full. This patchiness shows land-use of some sort, and more prominent where there is road access. Existing Sabah State laws denote a native having the right to be awarded at any one time a piece of state land up to 15 acres or about 6.1 hectares for agriculture-based use. From publicly accessible database i.e. JTUWMA Pro - land authority online database available to the public, most properties at Kadamaian are noted to be less than 15 acres. The land sizes range from one hectare to six hectares. The database also gives us the understating that land titles near road access have already been issued. From direct observation, land-use at Kadamaian is limited to agriculture and tourism activities. While there are other land-use e.g. rock quarrying, the use is negligible by comparison Kadamaian's overall land-use, and this was confirmed through interviews. A federal agency may only be brought into Kadamaian for assistance if the land available for development is more than 25 acres. To fulfill such requirement, it would make sense for native landowners to pool their resources and work together. Some of these lands have already been placed under a programme of *Lembaga Industri Getah Sabah* to support the development of the rubber industry. Of course, this does not imply that everyone in Kadamaian is a farmer. This suggests that Kadamaian's natural environment is relatively intact. As a result, environmental pressures and the need for conservation obligations are minimal at Kadamaian.

Kadamaian is one of three constituencies in the district of Kota Belud. As a constituency and based on the data about eligible voters of the Malaysian general election of 2013, there are 15,903 voters comprising 85.8% of all the eligible voters in Kadamaian. Based on statistical information available in the public domain, the population of Kota Belud is Kadazans 45% and Bajau 45%. With the population pyramid of Sabah resembling a triangle, we can assume that Kadamaian's population pyramid is also assumed to be somewhat resembling a triangle. Thus, Kadamaian's population can be divided into three functional groups and colloquially referred as a) the elderly generation, b) the adults and c) the younger generation. While this is a simplistic way of categorizing collected responses, this generational categorization is an important analytical step to the understanding of the concept of conservation at Kadamaian. With the conservation of biological diversity being linked to the ownership of the land, the educational relationship is best explored through generational

categorization. Data collection to determine the demography of Kadamaian was not carried out and not within the scope of this study. Nevertheless, Sabah's population pyramid being regarded triangle-shaped, it should be noted that in terms of generational size, it follows the equation below,

$$Y_t > Ad > El \text{ -----} \quad (Eq\ 1)$$

The following table lists the difference of these three (3) functional groups.

**Table 1:** Demography of Kadamaian's population based on functionality

Particular	Generation		
	Youth (Yt)	Adult (Ad)	Elder (El)
Age Range (Year Old)	≤ 34 YO	> 35 YO and ≤ 54 YO	> 55 YO
General Occupation	Students, Casual & contract based	White & Blue Collar	Farmer

The data collected during the study showed that the older generation namely those 60 years old or older are generally involved in land-use management. Since the older generations were assumed to be largely brought up as farmers, adults of Kadamaian may either be white or blue collar workers. Few would be farmers. As for the younger generation namely those below the age of 18 years old, public policies dictate their attention on formal education. With primary education being compulsory and secondary or higher education being optional and based on capabilities, only a handful of Kadamaian's youths are assumed to have contractual or casual employment. The rest are students.

Schulze & Suratman (1999) stated that people make a choice based on what they perceive as being worthwhile. Certain old people are described as living libraries. But this type of library is becoming obsolete in modern time. They are slowly fading away, and to ensure their relevancy, they become authoritative. Adults mainly think about getting by socio-economically. With regards to the elders, their interaction is intergenerational. It was important to note that while the Sabah Government had a ministry that focused on education between May 2018 to October 2020 known as the Ministry of Education and Innovation and another ministry focusing on the youths namely the Ministry of Culture, Youth and Sports, the fact that education and training is under the purview of the federal government, the development of human capital be it for environment education or otherwise is less focused in Sabah.

As stated earlier, generational categorization is an important analytical step. Besides land ownership, generational categorization gives an understanding

about a person's prior knowledge. In general, elders are farmers and obviously possess prior knowledge acquired through years of experience working on their land; their prior knowledge gives them an understanding about the concept of conservation. The current situation suggests occupations at Kadamaian do not have a high or urgent need for conservation-based knowledge. Kadamaian's media presence revolves around a few themes. One of the themes is about the need to maintain its existing status quo. In an interview with K1, it was apparent that the status quo that needs maintaining is traditions and culture. Hence, the elders are viewed as culture bearers, and they focus on the preservation of their community's identity. To do this preservation, they are willing to carry out an intergenerational relationship.

The concept of a sustainable culture as advocated by Japan is important. To be sustainable, a community establishes structures which in turn continue actions. Here, there is formation of a tradition. Whether this tradition really supports conservation, only time will tell since different generations have different needs and there is no harmony between them. But this has now been taken over by the Biocultural Protocol Committee.

#### ***Difficulty in Balancing socio-economy and environment***

As stated earlier, Kadamaian's media presence revolves around a few themes. The first theme was described previously, and two major conservation activities associated to the theme were uncovered. The formation of a biocultural administration body is one of them, while the other is the implementation of a popular traditional conservation activity i.e. the *Tagal*. While *Tagal* is taken as a symbol of conservation, the role and power of adults at Kadamaian is unclear other than as supporters of the elders.

As for the second theme, it is about Kadamaian's tourism appeal. The tourism appeal is driven by authentic spaces such as structure camping sites and photo opportunity spots. These spaces are different from normal spaces communities generally establish. Authentic spaces cost money, time, and effort to develop. This is higher for Kadamaian's adults without the appropriate knowledge or experience.



**Figure 3.** Kadamaian's extra-ordinary as found on the Internet

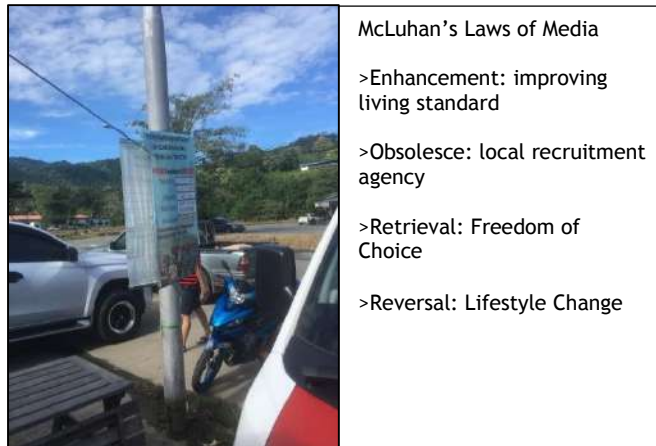
With all of this, it remains to be seen if Kadamaian's community has the capacity to sustain that balance between socio-economic demands and environmental obligations. Hence, what appears is more need for knowledge to commercialise and monetise the resources of Kadamaian.

***Kadamaian's youths have no need for a wholistic socio-economic and environmental understanding.***

In literature, Kadamaian's environmental issues are derived from its abundant water resource and its proximity to Sabah's World Heritage Site i.e. the Kinabalu Park. Its challenges however are not related to the mentioned attributes but instead related to its socio-economic structures particularly for the youths of Kadamaian. Environment related challenges have been overshadowed since Kadamaian's indigenous community is not dependent on the mentioned attributes; many members of the community have access to treated water and have land outside of Kinabalu Park. The socio-economic challenge that has overshadowed environmental challenges has also forced youths to migrate out of Kadamaian, and youths are motivated to leave for jobs in other parts of Malaysia. This is evident through posters and other advertisements for vacancies in Peninsular Malaysia.

McLuhans' Law of Medias state that a medium is capable of giving out one of four messages i.e. enhancement, obsolesce, retrieval or reversal, and this is not about the artform within the medium. In relation to employment bunting banners for factory vacancies in Peninsular Malaysia that was found in one of Kadamaian's town, the most concerning message is that people are expected to have a change of lifestyle, as this change will influence the way conservation is carried out.





**Figure 4.** Tetrad of Employment Bunting Banner Medium found in Taginambur town (adopted from McLuhan (1964))

In fact, from interviews, some youths had already migrated to other countries such as South Korea, to earn a living. Some of these youths did send money home. Because of this, it would not be a surprise to see others following suit. The trend of youths migrating to urban areas is an alarming trend worldwide. Youths migrate to these areas as there are job and education opportunities and an enticing wage handout (Haji Abbas and Binsin, 2008, p. 347). The lure of lucrative income is a great driver to migration. In socio-economic context, there are push and pull factors to the migration of youths to urban areas. According to Haji Abbas and Binsin (2008, p. 347), while there are many pull factors some of which have already been introduced, there are three (3) major push factors for youth to migrate out of their community. These factors are the prevalence of poverty, the lack of job opportunities and the limitation of basic structural necessities (Abbas and Binsin, 2008, p. 347). In urban areas, there are a lot of jobs as well as education opportunities for youths to fit into those jobs. Despite this, there is no guarantee of job opportunities; the chance of landing into a job is 50/50. Among the Kadamaian community, many aspects of Kadamaian's socio-economic structures have already been explored; and this trend is worrying so much so that the Sabah State government has taken steps to slow this trend down by focusing on developing facilities to aid Kadamaian's human capital development. To address this socio-economic challenge, the Kadamaian Development Master Plan (2020-2035) was unveiled in January 2020. In an interview with the planners, information showed the plan was to support the perceived entrepreneurial spirit of youths in Kadamaian. It is believed that developing the needed facilities would eventually spur human capital

investment and/or development. Regardless, human capital development is not the focus at Kadamaian. Hence, while the push-pull factors provide an explanation to youth movement including the highlighting of the importance of education, the push-pull factors cannot be used to explain educational needs particularly environmental education needs. The youths simply need to find money to have power or a voice in their community.

An example of a human capital theory is the Social Exchange Theory, and according to this theory, the action taken is based on a calculation of benefits outweighing costs. This theory not only explains why Kadamaian's youths migrate, it also implies environmental education needs of the community at Kadamaian. To be precise, the youths have no need for environmental education since they do not wish to remain in Kadamaian. Only those willing to remain are those who may need socio-economic knowhow while leveraging on environment understanding.

## Conclusion

Kadamaian is an indigenous populated area adjacent to a protected area, the Kinabalu Park World Heritage Site. Because of such proximity, Kadamaian's population is expected to be knowledgeable on matters concerning the conservation of biological diversity. Kadamaian's conservation of the environment and its biological diversity is expected to relate greatly with land-use management and product development. In fact, being an indigenous community living adjacent to a protected area, the community is expected to practice socio-economic development that is compatible with the concept of conservation, regardless of interest of community members. Since Kadamaian is regarded by other regional authorities as a buffer to Malaysia's first world heritage site, its community must have the ability to balance socio-economic demands and environmental obligations. However, they have not improved their socio-economic standing beyond regular approaches because they have no knowledge about the commercialisation of conservation and the monetisation of those natural resources. Hence, to support the government's move from fortress conservation to rights-based conservation, sustainability-based knowledge is needed in Kadamaian. The question about a community's obligation and ability to carry out conservation amid their socio-economy eventually led the community to establish a non-governmental organization (NGO). In the case of Kadamaian's indigenous community, the NGO is called the Melangkap Biocultural Protocol Committee (MBPC). But for MBPC to fulfill its obligation, Kadamaian's youths should be actively involved in conservation. This is where Kadamaian's

problem lies - the migration of youths out of the community. This study suggests that the migration is a behavioural sink in Kadamaian, and its adults need to play a role to resolve this behavioural sink from bring destructive to the community. Since this study was not able to detail the adults' social dimension of dominance and interactions prompting the need for more studies, one possible approach to resolve it is the introduction of a more encompassing concept of conservation apart from the currently popular *Tagal*. A criterion of such an approach is its support towards the sustainability of a community's traditions and culture.

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