

## **Altitudinal Diversity of Braconid Wasps (Hymenoptera: Braconidae) at Fraser's Hill, Pahang, Malaysia**

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### **ABSTRACT**

A diversity study of the subfamilies of Braconidae was conducted at Fraser's Hill, Pahang, Malaysia. Sampling took place at three different altitudes using Malaise traps: lower altitude (<500m), intermediate altitude (501-1000m) and higher altitude (>1000m). A total of 572 individuals of braconids were collected from the three altitudes and comprised 15 subfamilies: Agathidinae, Alysiinae, Blacinae, Braconinae, Cheloninae, Doryctinae, Euphorinae, Helconinae, Lysiterminae, Microgastrinae, Miracinae, Opiinae, Orgilinae, Pambolinae and Rogadinae. There were 435 individuals and 55 species, 84 individuals and 30 species, and 53 individuals and 26 species, with a diversity index of  $H' = 3.75, 2.91$  and  $3.01$ , representing the lower, intermediate and higher altitudes, respectively. The diversity index of the lower altitude ( $H' = 3.75$ ) was significantly different from the intermediate ( $p = 0.00, < 0.05$ ) and higher altitudes ( $p = 0.00, < 0.05$ ). There was no significant difference between the intermediate and higher altitudes ( $p = 0.86, > 0.05$ ). This is probably due to the variability of host and food availability in lowlands compared with highlands.

*Keywords:* Altitude, braconid, diversity, abundance

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### **INTRODUCTION**

Braconid wasps (Hymenoptera: Braconidae) is a large group of insects with more than 15,000 species distributed throughout the world (Hanson & Gauld, 2006; Quicke, 2014) and occupying different ecological habitats (LaSalle & Gauld, 1993; Shaw & Huddleston, 1991). Braconids are insects that have been proven to be of economic

and ecological interest (Billah et al., 2008; Whitfield, 2003). Most braconid species have the potential to be used as biological control agents due to their nature of parasitizing other insects (Gillott, 2005; Kimani-Njogu et al., 2001). They tend to have a specific range of hosts specialized by biological and behavioural adaptation (Janzen et al., 2003). The most common hosts for braconids are the larvae of Lepidoptera, Coleoptera and Diptera (Tripplehorn & Johnson, 2005). Braconids are also very sensitive to environmental disturbances which makes them good indicators of diversity and environmental stability (Shaw & Hochberg, 2001). However, in Malaysia, studies on this subfamily is remains undeveloped (Yaakop & Aman, 2013; Yaakop et al., 2009).

Insects usually occupy different niches in a certain habitat and play a functional role in maintaining the dynamics of the ecosystem (Goldstein, 1999). According to Rohner et al. (2015), insect abundance and diversity are affected by both altitude and latitude. For parasitoids, the geographic distribution depends on the plant communities and on their faunal history (Wiens & Donoghue, 2004). This is because of the difference in environmental conditions in lowlands and highlands (Lomolino, 2001).

Abiotic (e.g., temperature, humidity, etc.) and biotic (e.g., competition, migration, etc.) factors can affect the distribution of any species (Eitam et al., 2004). Temperature is a crucial factor that limits distribution

especially when altitude becomes a physical factor (Logan, 2001; Parmesan, 2006). Furthermore, wasp diversity is also closely influenced by altitude and latitude (Tan et al., 1990; van der Ent & Shaw, 1998), such that if any change in these factors may affect their diversity. Vegetation covers are strongly associated with insect accumulation, thus attracting various host insects to the wasps (Campan & Benrey, 2004; Sperber et al., 2004). The abilities of parasitoids differ in their surviving period based on the availability of hosts (Eitam et al., 2004). Fraser's Hill is one of the areas known for ecotourism, but there are some activities that can affect the biodiversity of insects. The objective of this study is to compare and measure the differences in braconid diversity and abundance between different altitudes at Fraser's Hill.

## METHODS

### Study Site

Sampling was conducted at Fraser's Hill in the state of Pahang, Peninsular Malaysia. A few areas have been developed into tourism destinations, but most of the hill forests are still protected and conserved. Three altitudes were selected for this study: lower (<500m), intermediate (501-1000m) and higher (>1000m). The lower altitude is composed of lowland dipterocarp forest, intermediate altitude of hill dipterocarp forest, and higher altitude of montane forest (WWF, 2001).

### Braconid Collection

Sampling was carried out by using Malaise traps, which are known for the best ways to capture flying insects passively (Mazon & Bordera, 2008). During the sampling, a total of nine Malaise traps were set up and placed at three different altitudes faced towards the sunlight. For each selected altitude three traps were set up for 30 days before the samples were collected. Sampled insects were then kept in 70% ethanol for further identification in the laboratory.

### Species Identification

Identification of braconids was carried out using morphological characters based on taxonomic keys by Achterberg up to subfamily level and to genus and species where possible (Achterberg, 1993; Li et al., 2012; Li et al., 2013). The identification process utilized a stereo microscope, and a photograph of each specimen was taken with a Canon EOS 1000D attached to the stereo microscope (Carl Zeiss Stemi DV4).

### Data Analysis

Samples from different altitudes were analysed using PAST software to compare the diversity indices. Diversity Indices viz. Shannon diversity index ( $H'$ ), Evenness ( $E$ ) and Richness ( $R'$ ) were analysed to measure

the braconids' diversity and species richness at each altitude. The diversity of braconids at the three altitudes was then compared using t-test with  $p=0.05$ . Two-way clustering was conducted using computer software PCORD 6 to observe the similarities and differences between the species and altitudes.

### RESULTS

A total of 572 individuals were successfully collected throughout the sampling period. They represented 15 subfamilies from all altitudes: Agathidinae, Alysiinae, Blacinae, Braconinae, Cheloninae, Doryctinae, Euphorinae, Helconinae, Lysiterminae, Microgastrinae, Miracinae, Opiinae, Orgilinae, Pambolinae and Rogadinae (Table 1). The lower altitude (<500m) showed the highest number with 435 individuals and 55 species, followed by the intermediate altitude (501-1000m) with 84 individuals and 30 species. The higher altitude (>1000m) showed the lowest number with 53 individuals and 26 species (Table 1). The 15 subfamilies that were found at Fraser's Hill represent 32% of the 46 subfamilies recorded all over the world. Seven of the subfamilies were found in all three altitudes: Alysiinae, Braconinae, Cheloninae, Doryctinae, Microgastrinae, Opiinae and Orgilinae.

Table 1  
 Subfamilies and genera collected in three different altitudes in Fraser's Hill, Pahang, Malaysia

Subfamily	Genus	Altitude		
		<500m	501-1000m	>1000m
Agathidinae	<i>Aneurobracon</i> sp.	0	5	0
	<i>Therophilus</i> sp.	0	0	7
Alysiinae	<i>Dinotrema</i> sp.	0	5	1
	<i>Dacnusa</i> sp.	5	0	0
	<i>Heratemis</i> sp.1	4	2	0
	<i>Heratemis malayensis</i>	0	0	1
Blacinae	<i>Blacus</i> sp.1	1	1	0
	<i>Blacus</i> sp.2	3	3	0
	<i>Blacus</i> sp.3	6	2	0
Braconinae	<i>Bracon</i> sp.1	3	0	1
	<i>Bracon</i> sp.2	5	1	0
	<i>Bracon</i> sp.3	23	0	0
	<i>Braconinae</i> sp.	5	0	1
	<i>Gammabracon</i> sp.	0	1	0
	<i>Habrobracon</i> sp.	10	0	1
Cheloninae	<i>Vipio</i> sp.	6	0	1
	<i>Ascogaster</i> sp.	12	1	1
	<i>Chelonus</i> sp.	10	1	0
Doryctinae	<i>Phaneretoma</i> sp.	7	1	0
	<i>Rhoptrocentrus</i> sp.	6	0	1
	<i>Spathius</i> sp.1	7	0	1
	<i>Spathius</i> sp.2	10	0	0
	<i>Spathius</i> sp.3	8	0	0
Euphorinae	<i>Spathius</i> sp.4	3	1	0
	<i>Spathius</i> sp.5	9	0	0
	<i>Centistes</i> sp.	1	1	0
Helconinae	<i>Peristenus</i> sp.	8	0	0
	<i>Triaspis</i> sp.	5	2	0
Lysitermiinae	<i>Acanthormius</i> sp.	6	0	0
Microgastrinae	<i>Alloplitis</i> sp.	23	4	0
	<i>Apanteles</i> sp.1	2	21	5
	<i>Apanteles</i> sp.2	16	4	2
	<i>Choeras</i> sp.	4	4	4
	<i>Cotesia</i> sp.1	18	1	2
	<i>Cotesia</i> sp.2	4	0	1
	<i>Diolcogaster</i> sp.1	9	1	0
	<i>Diolcogaster</i> sp.2	21	0	0
	<i>Fornicia</i> sp.	7	0	2

Table 1 (continue)

Subfamily	Genus	Altitude		
		<500m	501-1000m	>1000m
	<i>Glyptapanteles</i> sp.	7	1	1
	<i>Microgaster</i> sp.	9	0	2
	<i>Parapanteles</i> sp.	39	4	5
	<i>Pholetesor</i> sp.	7	1	1
	<i>Wilkinsonellus</i> sp.	15	7	3
Miracinae	<i>Centistidae</i> sp.	5	0	1
Opiinae	<i>Apodesmia</i> sp1	8	0	0
	<i>Biosteres</i> sp.	0	1	2
	<i>Bitomoides</i> sp.	7	2	0
	<i>Opiinae</i> sp.1	7	0	0
	<i>Opiinae</i> sp.2	6	0	0
	<i>Opiinae</i> sp.3	9	0	0
	<i>Opius</i> sp.1	5	0	0
	<i>Opius</i> sp.2	9	0	0
	<i>Opius</i> sp.3	4	0	0
	<i>Orientopius</i> sp.	4	2	4
	<i>Psytalia</i> sp.	7	1	0
Orgilinae	<i>Orgilus</i> sp.	5	1	0
	<i>Stantonia</i> sp.	3	0	1
Pambolinae	<i>Pambolus</i> sp.	0	0	1
Rogadinae	<i>Aleiodes</i> sp.	1	0	0
	<i>Canalirogas</i> sp.	6	2	0
	<i>Rogas</i> sp.1	1	0	0
	<i>Rogas</i> sp.2	4	0	0
	Total	435	84	53

Table 2

Number of individuals, species and diversity indexes sampled from Fraser's Hill, Pahang, Malaysia. Small letter a and b indicates significant difference at  $p = 0.05$

	<500m	501-1000m	>1000m
No. of Individuals	435	84	53
No. of genus	55	30	26
Shannon (H')	3.75 <sup>a</sup>	2.91 <sup>b</sup>	3.01 <sup>b</sup>
Richness (R')	8.89	6.55	6.30
Evenness (E)	0.77	0.61	0.78

This study shows that Microgastrinae has the highest abundance in both species (14) and individuals (257), and can be found in all altitudes. The most dominant genus is also from the same subfamily, *Apanteles*, with 50 individuals. In terms of the Shannon diversity index, the lower altitude has the highest  $H'$  of 3.75, compared to the intermediate ( $p=0.00, < 0.05$ ) and higher ( $p=0.00, <0.05$ ) altitudes, while there is no significant difference between the intermediate and higher altitudes ( $p=0.86, >0.05$ ) (Table 2).

Based on the two-way cluster analysis, there are three groups that represent more than 75% similarities (Group 1, 2, 3) (Figure

1). Group 1 has the highest number of species (50) which are mostly found in the lower altitude even though some of them can also be accommodated in intermediate and higher altitudes. Group 2 has three species, *Aneurobracon* sp.1, *Gammabracon* sp.1 and *Dinotrema* sp.1 which appear in the intermediate altitude. *Dinotrema* sp.1 also occurs in the higher altitude but at weak probability. Meanwhile, Group 3 has four species, *Biosteres* sp.1, *Heratemis malayensis*, *Pambolus* sp.1 and *Therophilus* sp.1, concentrated in the higher altitude, but there is one, *Biosteres* sp.1, that was also found in the intermediate altitude.

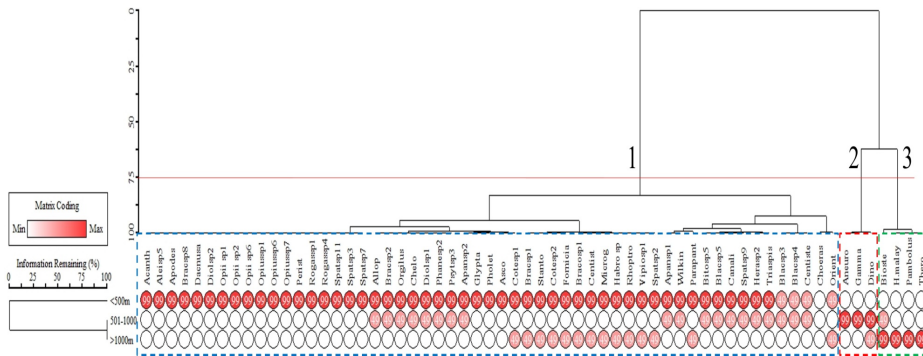


Figure 1. Two-way cluster analysis (dendrogram) of the braconid species collected from different altitudes in Fraser’s Hill

**DISCUSSION**

In this study, 15 braconid subfamilies were found in Fraser’s Hill, Malaysia based on Yu et al. (2012). Previous studies from Sivinski et al. (2000) in Mexico, Fernandez-Triana et al. (2013) in New Zealand and Fernandez-Triana et al. (2016) in Costa Rica also discovered some subfamilies

in highland areas, such as Alysinae, Doryctinae, Microgastrinae, and Opiinae, that were similar to this study. A few species from subfamily Opiinae, such as *Fopius arisanus*, are thought to be limited in low temperatures (Snowball & Lukins, 1964), while subfamily *Doryctinae* *Doryctobracon crawfordi* is sensitive to higher temperatures

than the host, *Anastrepha ludens* (Sivinski et al., 2000). However, there is no solid proof to support that subfamilies Alysiinae, Doryctinae, Microgastrinae, and Opiinae found in this study have been recorded in highlands, because most of the previous studies found them only in lowlands (Abu El-Ghiet et al., 2014; Gadelha et al., 2012; Jimenez-Peydro & Peris Felipo, 2011). This may be a new record for diversity of braconid wasps in highlands in Malaysia, especially for Fraser's Hill.

For overall diversity, the lower altitude (<500m) showed the highest number of braconid individuals compared to both intermediate and higher altitudes (>501m). The lower altitude is said to have denser and more varied vegetation (Adam et al., 2011; Barbieri Junior & Dias, 2012) and generally more abundant insect species groups (Lessard et al., 2011; Nufio et al., 2010; Sanders & Rahbek, 2012). In addition, the altitudinal study of Janzen et al. (1976) shows that species richness of parasitic Hymenoptera was high at 200m is congruent with our study in lower altitudes below <500m is because in most habitats, plant communities determine the physical structure of the environment and hence the distribution and species interactions (Saaksjarvi et al., 2004). The number of small plants such as shrubs and herbs declines as altitude increases, which is another reason lower altitudes has greater diversity of host plants and wasps (Khairiyah et al., 2013).

Higher altitude tends to have more adaptive and specific plants compared

to the lower altitude. According to this study, *Apanteles* sp. Were found in all three altitudes. This may be due to its high adaptive character. Besides limited plant variation at higher altitudes, and the construction of tourist conveniences contribute to less braconid abundance and diversity compared to lower ground. Less adaptive braconid species will not survive at higher altitudes (Shaw & Hochberg, 2001), leaving the area less sensitive and adaptive groups. Environmental changes will diminish the host population negatively affecting the braconid population as the habitat area becomes undersized and more disturbed (Bobo et al., 2006; Malmivaara et al., 2002; Roy et al., 2001).

An urbanized forest area that experiences destruction will lead to habitat and species loss (Blair & Launer, 1997; Brown et al., 1996). Poor adaptive species are fragile and cannot survive such disturbances, which will be forced to either leave for other favourable areas, or die (Lien, 2013). The environment at higher altitudes are also influenced by weather conditions and can play an important role in soil characteristics, the composition of plants, population structure and invertebrate composition (Willig et al., 2011). In addition, high altitude environments have low productivity rates (Mani, 2013; Stevens, 1989). Environmental changes can shrink the host population and negatively affect the wasps' population (Idris & Hasmawati, 2002).

Microgastrinae are known for its vast diversity and distribution besides being the most conspicuous group attacking

caterpillars. As exclusive caterpillar parasitoids, the Microgastrinae are one of the most economically important natural enemies of Lepidoptera. This is proved by the utilization of more than 100 species of Microgastrinae in the biological control of lepidopteran pests in industries (Whitfield, 1997). According to Kahuthia-Gathu (2013), the genera *Apanteles* are found to be the common parasitoid for the larvae of *Plutella xylostella*, diamondback moth, the most destructive insect of cruciferous plants throughout the world (Talekar & Shelton, 1993). In Malaysia, these caterpillars are non-native pests of cruciferous crops that were frequently found to be parasitized by a braconid wasp in the genus *Apanteles* (Furlong et al., 2013). Cruciferous plants are economically important being the most common diet in various cultures (Shelton, 2001). The genera *Apanteles* is one of the dominant species with the highest parasitism recorded in reducing the *P. xylostella* population. Besides that, *Heratemis malayensis* has been discovered in highland altitudes corresponding to the taxonomic study by Yaakop et al., (2009) who found the species in the highlands of Bukit Larut, Perak, Malaysia.

Based on the two-way cluster analysis, Group 1 is mostly concentrated at lower altitudes. This is due to the availability of food, temperature and humidity conditions, found in the lowlands (Sanders & Rahbek, 2012). Their numbers could be higher during the rainy season as young new leaves grow, and provide more food for

insects (Barbieri Junior & Dias, 2012; Young, 2012). Compared with the lowlands, Groups 2 and 3 are more likely to have a similar habitat which increases in altitude. Species of insects are less rich as the altitude increases because higher altitudes have lower habitat niches, limitation in host plants, and deficiency of food sources (Lomolino, 2001; McCain & Grytnes, 2010). In addition, the good flying ability of braconids makes them capable of finding a suitable habitat for their development (Araujo et al., 2004). From our study, although we can still find a few species of braconid in the higher altitudes, their diversity and abundance tell us that the lowlands are a preferable habitat for the braconid.

## CONCLUSION

The braconid wasp's diversity and population were higher in lower altitude mainly because of the availability of food sources. The presence of hosts directly invite the wasps to parasitize and continue their life cycle. Microgastrinae is the most abundant subfamily as the species is widely distributed. As the altitude increases wasp diversity decreases due to less productive vegetation.

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