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RESEARCH PAPER

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The leaf beetle (Coleoptera: Chrysomelidae) fauna of Mt. Manalmon in San Miguel, Bulacan, Philippines with reference to their host plants

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Abstract

Family Chrysomelidae is a highly speciose family that has been poorly studied in the Philippines. In this study, an inventory of chrysomelids was conducted in Mt. Manalmon within the protected area of Biak-na-Bato National Park located in San Miguel, Bulacan. Sampling was conducted seven (7) times in three (3) different elevations along vegetation within reach in a 200 m transect line using active collection method from August to November, 2017. A total of 250 specimens of Chrysomelidae were collected belonging to 4 subfamilies, 24 genera, and 30 species. Plants from which the leaf beetles were collected belong to 13 families, 19 genera, and 20 species. Species richness and abundance was highest in the lower montane and decreased with increasing altitude. Species diversity was highest in mid-montane (H' = 1.954), followed by the lower montane (H' = 1.935), and lowest in the upper elevation (H' = 0.659). The diversity of the family in the mountain was greatly attributed to the rich composition of host plants in the area. The species richness and diversity of Chrysomelidae fauna in Mt. Manalmon provides a baseline data for the conservation and the role of leaf beetles as a model group in tropical biodiversity.

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Introduction

Chrysomelidae (leaf beetles) is one of the largest family in the order Coleoptera with over 35,000+ species and more than 2,000 genera distributed worldwide except in the arctic regions (Jolivet *et al.*, 2009; Thormann *et al.*, 2016).

The sheer diversity of the family is accounted for their phytophagous habit, which means that their abundance is greater in the tropics because of the richness of plants compared to any parts of the world. This feeding characteristic makes chrysomelids an important component of tropical ecosystems (Lewinsohn and Roslin, 2008).

With this contribution of leaf beetles to a diverse world, the richness of Chrysomelidae communities is widely studied for biodiversity, environment quality, and as a model group for monitoring changes in natural areas. Leaf beetles generate a strong relationship with the environment, therefore it is possible to conduct research simultaneously in two highly diverse components-chrysomelids and their host plants, as well as the predominant ecological interaction of herbivory in the tropics (Sánchez-Reyes *et al.*, 2014; Gómez-Zurita *et al.*, 2016).

Recent climatic and environmental changes pose a great threat on the biodiversity of the family, an inventory to record and predict how organisms adapt to environmental changes is vital for baseline data and for future research (Sánchez-Reyes *et al.*, 2014).

The Philippines is a large archipelago (7,107 islands) in Southeast Asia with a land area of 300,000 km² and a high rate of endemism patterns both in flora and fauna (Zettel, 2014). Research on the diversity of Chrysomelidae and their interaction with host plants is poorly documented in the country.

They are generally known as pests of the agricultural industry that causes serious economic damage (Acevedo *et al.*, 2015), and most studies done on leaf beetles in the country are of relatively few taxonomic revisions (Bezděk, 2012, 2015).

The general lack of updated published studies and literatures of the species richness and diversity of Chrysomelidae in the Philippines makes it difficult task to compare the particular ecological characteristics and biogeographical distribution patterns of this family with other taxa in the country.

The diversity of leaf beetles is expected to be rich because of the tropical nature of the country; however, chrysomelid diversity is threatened because of anthropogenic disturbances such as habitat destruction and land conversion (Perry *et al.*, 2016). Therefore, an urgent need for biodiversity assessment on leaf beetles and their host plants is necessary for future conservation (Thormann *et al.*, 2016).

The study aims to document the poorly known Chrysomelid fauna and their host plants in the study area. Mt. Manalmon is part of the protected area of Biak-na-Bato National Park located in San Miguel, Bulacan in the island of Luzon which is considered a biodiversity hotspot and the best entomofaunistically explored island of the Philippines (Zettel, 2014).

The area provides suitable conditions for chrysomelids to thrive with dense vegetation covering majority of lands. Differences in richness and diversity of chrysomelids with increasing elevation were also described.

Materials and methods

Study Area

The study was conducted in Mt. Manalmon within the protected area of Biak-na-Bato National Park, San Miguel, Bulacan with elevation of 196+ meters (750+ ft.) above sea level (MASL) and coordinates of $15^{\circ}09'45.8''N 121^{\circ}05'27.9''E$ (Fig. 1).

Three sampling sites with different vegetation types and elevation were established which consist of the lower montane (200-400 ft.), mid-montane (400-600 ft.), and upper montane (600-750 ft.). All shrubs, grasses, other herbaceous vegetation, and trees within reach were sampled in each site.

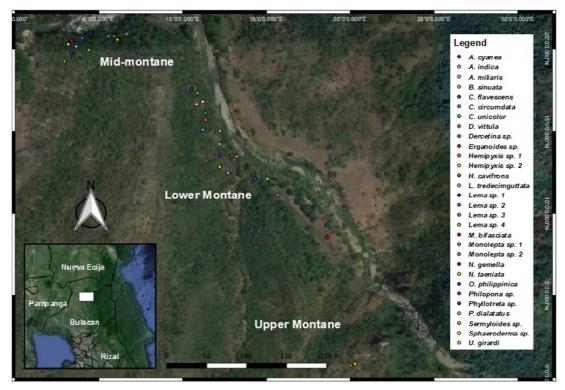


Fig. 1. Map of the study area showing the three sites with different elevation levels and the locations of the specimens collected. The inset shows the location of Mt. Manalmon (white shade) within the province of Bulacan (Created using Google Earth data on QGIS).

Sampling Methods

Sampling was conducted along a 200 m transect line using standardized entomology methods (active collection), visual encounters, and photo documentation (Nikon d5300) from 6:00 to 12:00 hrs. Adult beetles were collected on sight from plants. Handpicking of specimens was often used. Sweep netting was used to catch species that can quickly escape by flying or jumping (some Alticinae). Beat sheeting was used to collect numerous individuals in one plant. Aspirators were used to collect small specimens. Leaves, stems, and flowers (if available) of plants were collected from which the leaf beetles are found. Chrysomelids were killed using 70% ethyl acetate and was card-mounted for longer storage.

Plants were pressed on the field. Both beetle and plant specimens were stored in the biological collections room of Bulacan State University, City of Malolos, Bulacan, Philippines. Information on host plants were derived from previous literatures and personal observations.

Taxonomic Determination

Identification was done by observing the specimens under a Motic stereo microscope and comparison with available taxonomic keys and monographs on Chrysomelidae (Kimoto, 1989; Kimoto, 2000; Medvedev and Moseyko, 2002; Warchałowski, 2011; Staines, 2015; Borrowiec and Świętojańska, 2017). Plant specimens were sent to the Research Center for Applied and Natural Sciences Herbarium at the University of Sto. Tomas, España Blvd., Sampaloc, Manila, Philippines for identification.

Statistical Analysis

Abundance was calculated using the number of individuals per each site. Species abundance was divided into five categories 1.) very common (more than 10 individuals); 2.) common (six to 10 individuals); 3.) rare (three to five specimens); 4.) doubletons (two specimens); 5.) singletons (one specimen only) (Sánchez-Reyes *et al.*, 2014). Shannon-Weiner Index for comparing diversity in each elevation was determined using Bio Pro software version 2.0.

Results and discussions

Species Richness and Host Plants of Chrysomelidae in Mt. Manalmon

A total of 250 specimens of Chrysomelidae were collected belonging to four (4) subfamilies, 24 genera, and 30 species. Galerucinae was the most species rich subfamily with 16 registered species, representing 53.33% of total richness followed by subfamily Cassidinae with nine (9) species (30%). Lower richness was recorded for subfamilies Criocerinae with four (4) species (13.33%) and Eumolpinae with only one (1) species (3.33%). All adult leaf beetles

were found on angiosperm plants, mostly dicotyledons. Chrysomelids were collected from 20 species of plants belonging to 19 genera and 13 families. Damage part of the plants were taken as indicators that the beetles really fed on the plant. The greatest number of Chrysomelid species were collected from families Convolvulaceae, Fabaceae, Lamiaceae, and Poaceae. A comprehensive review on host plants of the family is found in Jolivet and Hawkeswood (1995). Most species of plants identified in this study are those that have already been confirmed as hosts from previous authors (Table 1).

Table 1. Overview of Chrysomelidae in Mt. Manalmon and the list of plants from which they were collected. (* = plant association not recorded in other studies).

Chrysomelid Species	Plant Species	Plant Family	Previous Literature	
Subfamily Galerucinae Latreille, 1802		_		
Altica cyanea Weber, 1801	<i>Ludwigia octovalvis</i> (Jacq.) Raven	Onagraceae	Biondi and D'Alessandro, 2010	
Aulacophora indica (Gmelin, 1790)	Calopogonium muconoides Desv.	Fabaceae	Lee and Beenen, 2015	
Coeligetes unicolor Jacoby, 1895	Lantana camara L.*	Verbenaceae	-	
Dercetina sp.	<i>Prueraria montana</i> (Lour.) Merr.*	Fabaceae	-	
Erganoides sp.	Centrosema pubescens Benth.	pubescens Benth. Fabaceae Beenen and		
Hemipyxis sp. 1	<i>Chromolaena odorata</i> L. R.M. King & H. Rob.		Biondi and D'Alessandro, 2010	
<i>Hemipyxis</i> sp. 2	Viburnum sp. L	Adoxaceae	Biondi and D'Alessandro, 2010	
Hoplosaenidea cavifrons (Duvivier, 1885)	Streblus asper Lour.	Moraceae	Nadein and Bezděk, 2014	
	Urena lobata L.	Malvaceae	Nadein and Bezděk, 2014	
Monolepta bifasciata (Hornstedt, 1888)	Litsea glutinosa Lam.	Lamiaceae	Nadein and Bezděk, 2014	
Monolepta sp. 1	Litsea sp. Lam	Lamiaceae	Nadein and Bezděk, 2014	
Monolepta sp. 2 Nisotra gemella (Erichson, 1834)	Litsea sp. Lam Urena lobata L.	Lamiaceae Malvaceae	Nadein and Bezděk, 2014 Biondi and D'Alessandro, 2010	
Philopona sp.	Streblus asper Lour.*	Moraceae	Bioliul allu D'Alessaliulo, 2010	
Phullotreta sp.	Elaeagnus pungens C.P.	Elaegenaceae	-	
Phylotreta sp.	Thunb.*	Elaegellaceae	-	
Sermiloydes sp.	Aporosa sp. Blume*	Phyllanthaceae	_	
Sphaeroderma sp.	Elephantopus sp. L	Asteraceae	Biondi and D'Alessandro, 2010	
Subfamily Cassidinae Gyllenhal, 1813	Порнанориз эр. П	Tisteraceae	Dional and Diffestulato, 2010	
Aspidimorpha miliaris (Fabricius, 1775)	<i>Ipomoea</i> sp. L	Convolvulaceae	Borrowiec and Świętojańska, 2017	
Basiprionota sinuata (Olivier, 1790)	Premna odorata Blanco.	Lamiaceae	Borrowiec and Świętojańska, 2017	
Callispa flavescens Weise 1891	Bambusa blumeana Schult	Poaceae	Staines, 2015	
Cassida circumdata Herbst, 1799	<i>Ipomoea</i> sp. L	Convolvulaceae	Borrowiec and Świętojańska, 2017	
Dactylispa vittula (Chapuis 1876)	Paspalum conjugatum P.J.	Poaceae	Chaboo, 2007	
	Bergius			
<i>Laccoptera tredecimguttata</i> Wagener, 1877	<i>Ipomoaea</i> sp. L	Convolvulaceae	Borrowiec and Świętojańska, 2017	
Notosochanta taeniata (Fabricius, 1801)	Millettia pachycarpa Benth.	Fabaceae	Borowiec and Świętojańska, 2017	
Oncocephala philippinica Uhmann 1937d	<i>Ipomoea</i> sp. L	Covolvulaceae	Chaboo, 2007	
<i>Uroplata girardi</i> Pic, 1934 Subfamily Criocerinae Latreille, 1807	Lantana camara L.	Verbenaceae	Staines, 2015	
Lema sp. 1	<i>Paspalum conjugatum</i> P.J. Bergius	Poaceae	Vencl & Leschen, 2014	
Lema sp. 2	Paspalum conjugatum P.J. Bergius	Poaceae	Vencl & Leschen, 2014	
Lema sp. 3	Unknown	Unknown	-	
Lema sp. 3	Unknown	Unknown	-	
Subfamily Eumolpinae Hope, 1840				
Phytorus dialatatus Jacoby, 1884	<i>Barringtonia</i> sp. J.R Forst & G. Forst*	Lecythidaceae	-	

Galerucinae (Fig. 2) feed on a wide spectrum of dicotyledons. The genus Aulacophora Chevrolat, 1836 are known to feed on familes Cucurbitaceae and Fabaceae. A. indica is mostly associated with Cucurbitaceae (Lee and Beenen, 2015). However, it was observed feeding on C. muconoides (Fabaceae). Species from the genus Coeligetes Jacoby, 1884 and Dercetina Gressitt and Kimoto, 1963 were collected from families Fabaceae and Verbenaceae but was not previously recorded in other studies to be associated with these families. Species from Erganoides Jacoby, 1903 were mostly associated with Fabaceae (Lee and Beenen, 2010). H. cavifrons was collected from families Malvaceae and Moraceae and was the only chrysomelid observed to feed on two different plant species and family. Species from the genus Monolepta Chevrolat, 1837 were collected from Lamiaceae (Nadein and Bezděk, 2014).

One species from the genus *Sermyloides* Jacoby, 1884 was observed to feed on *Aporosa* Blume but is not recorded from other studies. Alticini from the genus *Altica* Geoffroy, 1762 and *Hemipyxis* Chevrolat, 1836 are polypagous and has been associated with herbaceous plants and shrubs belonging to many plant families (Biondi & D'Alessandro, 2010).

Species from these genera were mostly collected on families Adoxaceae, Asteraceae and Onagraceae. *N. gemella* was observed feeding on *U. lobata* (Malvaceae) in large numbers. *Philopona* sp. was observed feeding on *S. asper* (Moraceae) and *Phyllotreta* sp. on *E. pungens* (Elaeagnaceae), both species were not known to feed on these families. Species from the genus *Sphaeroderma* Stephens, 1831. are known to be associated with family Asteraceae, *Sphaeroderma* sp. was observed feeding on *Elepanthopus* sp.



Fig. 2. Subfamily Galerucinae. *A. cyanea* (A); *A. indica* (B); *C. unicolor* (C); *Dercetina* sp. (D); *Erganoides* sp. (E); *Hemipyxis* sp. 1 (F); *Hemipyxis* sp. 2 (G); *H. cavifrons* (H); *M. bifasciata* (I); *Monolepta* sp. 1 (J); *Monolepta* sp. 2 (K); *N. gemella* (L); *Philopona* sp. (M); *Phyllotreta* sp. (N); *Sphaeroderma* sp. (O).

418 | Hinolan et al.

Cassidinae (Fig. 3) were mostly collected from families Convolvulaceae, Fabaceae, Lamiaceae, Poaceae and Verbenaceae. Cassidines are more associated on families Convolvulaceae while Hispines prefer monocotyledons as their host plants (Chaboo, 2007; Staines, 2015, Borrowiec and Świętojańska, 2017). A. miliaris, C. circumdata, L. tredecimguttata and O. philippinica were mostly collected in Ipomoea sp. (Convolvulaceae). N. taeniata was collected from M. pachycarpa (Fabaceae), while B. sinuata was collected from P. odorata (Lamiaceae). C. flavescens was collected from B. blumeana and D. vitulla on P. conjugatum, both from family Poaceae. U. Girardi was collected from L. camara (Verbenaceae). All members of Cassidinae were observed feeding on the plants from which they were collected. Criocerinae (Fig. 3) are mostly associated with monocotyledonous families (Vencl and Leschen, 2014), Two (2) species (Lema sp. 1 and Lema sp. 2) were collected and associated in the nutrition with P. conjugatum (Poaceae). Lema sp. 3 and Lema sp. 4 were the only species of chrysomelids that were collected without any plant association in this study. Most species of Eumolpinae (Fig. 3) are attracted on latex plants (Asclepiadaceae and Apocynaceae) (Jolivet and Verma, 2008). P. dialatatus which represents the only eumolpine species in the study was collected from Barringtonia sp. without any feeding activity observed. Species found in this study with new plant association serves as a baseline data. This requires more detailed observations, critical food selection experiments, and food preference studies in the field and under laboratory conditions for a possibility of new host plant records.



Fig. 3. Subfamily Cassidinae. *A. miliaris* (A); *B. sinuata* (B); *C. flavescens* (C); *C. circumdata* (D); *D. vitulla* (E); *L. tredecimguttata* (F); *N. taeniata* (G); *O. philippinica* (H); *U. girardi* (I). Subfamily Criocerinae. *Lema* sp. 1 (J); *Lema* sp. 2 (K). Subfamily Eumolpinae. *P. dialatatus* (L).

419 | Hinolan et al.

Abundance and Variation of Chrysomelidae Species in Different Elevations

The abundance of leaf beetles was significantly different among the three sites. The greatest richness and abundance was recorded on the lower montane and decreased with increasing altitude. In the lower montane, a total of 19 species with 167 individuals were collected accounting for 66.8% of total abundance. The upper montane has the lowest abundance and diversity with only two species with 27 individuals and the mid-montane with 11 species with 56 individuals representing 22.4%. Diversity and evenness was highest in the mid-montane with a value of 1.954 in the Shanon-Weiner index, followed by the lower montane with a value of 1.935. Low diversity was observed in the upper montane with a value of only 0.659 (Table 2). Abundance and richness was highest on the lowest elevation and decreased with increasing altitude. L. tredecimguttata and H. cavifrons were the only two

chrysomelids to be found on two different elevation levels. *N. gemella* (72 individuals) and *C. flavescens* (32 individuals) were the two most dominant species found on the lowest elevation, *Phyllotreta* sp. (18 individuals) was the most numerous number of individual collected on mid elevation, while *Hemipyxis* spp. (27 individuals) were the only chrysomelids collected on the highest elevation.

Five (5) species were categorized as "very common" each with greater than 10 specimens accounted for 62.8% of the total abundance. These very common species were *N. gemella* (Galerucinae), *C. flavescens* (Cassidinae), *Phyllotreta* sp. (Galerucinae), *Hemipyxis* sp. 2 (Galerucinae), and *Monolepta* sp.1. Six (6) species were considered common occupying 14.8% of the total abundance. Eight (8) species were considered rare. Four (4) species were doubletons occupying 3.2% of total abundance, and six (6) species were collected as singletons occupying 2.4%.

Table 2. Abundance and Altitudinal Variation of Chrysomelidae in Mt. Manalmon.

Chrysomelid Species	Lower Montane 200-400 ft	Mid- Montane 400-600 ft	Upper Montane 600- 750 ft	Abundance	Percentage (%)						
						Altica cyanea Weber, 1801	5	-	-	Rare	2
						Aspidimorpha miliaris (Fabricius,	4	-	-	Rare	1.6
1775)											
Aulacophora indica (Gmelin, 1790)	5	-	-	Rare	2						
Basiprionota sinuata (Olivier, 1790)	3	-	-	Rare	1.2						
Callispa flavescens Weise 1891	32	-	-	Very	12.8						
				Common							
Cassida circumdata Herbst, 1799	4	-	-	Rare	1.6						
Coeligetes unicolor Jacoby, 1895	6	-	-	Common	2.4						
Dercetina sp.	1	-	-	Singleton	0.4						
Dactylispa vittula (Chapuis 1876)	8	-	-	Common	3.2						
<i>Erganoides</i> sp.	1	-	-	Singleton	0.4						
Hemipyxis sp. 1	-	-	10	Common	4						
<i>Hemipyxis</i> sp. 2	-	-	17	Very	6.8						
				Common							
Hoplosaenidea cavifrons (Duvivier,	4	2	-	Common	2.4						
1885)											
Laccoptera tredecimguttata	3	6	-	Common	3.6						
Wagener, 1877											
Lema sp. 1	-	4	-	Rare	1.6						
<i>Lema</i> sp. 2	-	1	-	Singleton	0.4						
<i>Lema</i> sp. 3	-	1	-	Singleton	0.4						
<i>Lema</i> sp. 4	-	1	-	Singleton	0.4						
Monolepta bifasciata (Hornstedt,	3	-	-	Rare	1.2						
1888)											
Monolepta sp. 1	-	11	-	Common	4.4						
Monolepta sp. 2	1	-	-	Singleton	0.4						
Nisotra gemella (Erichson, 1834)	79	-	-	Very	31.6						
				Common							
Notosochanta taeniata (Fabricius,	2	-	-	Doubleton	0.8						
1801)											
Oncocephala philippinica Uhmann				Doubleton	0.8						

J. Bio. & Env. Sci.	20	1
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8

Chrysomelid Species	Lower Montane 200-400 ft	Mid- Montane 400-600 ft	Upper Montane 600- 750 ft	Abundance	Percentage (%)
1937d					
Philopona sp.	-	1	-	Singleton	0.4
Phyllotreta sp.	-	18	-	Very	7.2
				Common	
Phytorus dialatatus Jacoby, 1884	2	-	-	Doubleton	0.8
Sermiloydes sp.	-	8	-	Common	3.2
Sphaeroderma sp.	-	3	-	Rare	1.2
Uroplata girardi Pic, 1934	2	-		Doubleton	0.8
Total No. of Individuals	167	56	27		100
Total No. of Species	19	11	2		
Shannon-Weiner Diversity (H')	1.935	1.954	0.659		

The diversity of the family varies differently with increasing elevation where greatest abundance and density was found on the lowest elevation and species richness was found decreasing in the middle and highest elevation (Sánchez-Reyes *et al.*, 2014; Thormann *et al.*, 2016). The results in this study showed that greatest species richness was found on the lowest elevation with some species were concentrated in large numbers (*N. gemella* and *C. flavescens*), thus reducing diversity. Diversity was highest in the mid-montane where most species were collected as doubletons and singletons, thus increasing the value of diversity.

The pattern of diversity in this study can be explained by the composition of host plants in each site which is also different in each elevation. This is clearly an important factor for Chrysomelidae since they are almost an exclusively phytophagous group (Gavrilović and Ćurčić, 2013). There is a strong evidence that the single most important factor contributing to high species diversity of tropical herbivores is the diversity of plants and that the single most important factor in the abundance and diversity of these beetles is clearly the presence of their host plants (Lewinsohn and Roslin, 2008; Novotny et al., 2012). This factor can also be used to predict total herbivore richness in tropical forests and montane ecosystems (Basset et al., 2012). Each site in the study area have different species compositions of host plants which reflected equally different communities and abundances of leaf beetles among the three sites. Most chrysomelids were also found feeding on the young leaves of their host plant (Whitefield et al., 2012).

Although some abiotic factors could also affect the diversity of leaf beetles such as altitude and temperature (Dahlhoff *et al.*, 2008; Schmitt and Rönn, 2011).

Shanon-Weiner diversity was highest in mid-montane (H'= 1.954) followed by the lower montane (H'= 1.935) while the upper montane had the lowest diversity (H'= 0.659). The high species diversity and abundance in the lowest elevation is attributed mainly to the abundance of host plants of chrysomelids in the area. Schowalter (2011) notes that Shannon-Weiner values generally fall in the range 1.5 and 3.5, rarely surpassing 4.5. Based on this scale, total diversity of Chrysomelidae in Mt. Manalmon is high (H'= 2.608). These results can be explained in part by the geographic location of the study area; Mt. Manalmon in Biak-na-Bato National Park is part of the Sierra Madre mountain range located in Luzon Island which is the largest island and the best entomofaunistically explored region in the Philippines (Zettel, 2014).

Conclusion

The present work is one of the first site specific area studies of Chrysomelidae and their host plants conducted in Mt. Manalmon. The information presented here provides baseline data that allow for comparisons of the diversity and species richness of Chrysomelidae on a regional and national scale. This could be used as an initial step to analyze the potential use of Chrysomelidae as an indicator group of biodiversity in the Philippines.

Recommendations

An increase effort of sampling towards a more complete inventory of the family is highly recommended. This would allow an easier comparison with the fauna of other nearby regions. The effects of anthropogenic disturbances on Chrysomelidae diversity could also be studied.

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