# Malacofaunal diversity and distribution in the Masungi Georeserve in Luzon Island, Philippines

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any limestone ecosystems, particularly in tropical countries like the Philippines, still lack data regarding their malacofaunal diversity amidst extensive habitat destruction and modification, and uncontrolled tourism. To address this, the diversity and spatial distribution of land snails in the Masungi Georeserve in Rizal Province, Luzon Island were determined. A total of 120 5x5m quadrats were randomly set along six karst and six non-karst sites of the georeserve. Altitude, temperature, relative humidity, soil moisture, exchangeable soil calcium, number of trees, diameter of trees, canopy cover, and leaf litter depth were also determined per quadrat. A total of 1,283 land snails belonging to 45 species and 12 families were sampled. Three major land snail communities were distinguished by detrended correspondence analysis: the first group preferred cooler environments with high-calcium soil (karst), the second group dominated in warmer habitats with low-calcium soil (non-karst), and the third group was found in both habitat types. Species richness was highest (40 species/ H'=2.94) in the karst sites however non-karst sites had higher species evenness (J'=0.55). Species accumulation curve showed

\*Corresponding author Email Address: bkvaldez@up.edu.ph Date received: July 11, 2020 Date revised: January 18, 2021 Date accepted: January 31, 2021  $\alpha$ -dominated community and efficient sampling (completeness ratio=0.97). Analysis of similarity further confirmed significant differences in land snail community assemblages between its karst and non-karst sites. These results suggest the rich malacofauna of the Masungi Georeserve needs to be protected as a refugia of land snail diversity.

# KEYWORDS

Malacology, land snails, karst, diversity, distribution

# INRODUCTION

Land snails are among the fauna that occur in tropical rainforests and karst ecosystems. Karsts are regions that are primarily composed of limestone, an uplifted sedimentary rock made up of calcium carbonate (CaCO<sub>3</sub>), from carbonate bearing marine organisms as well as carbonate-rich parent material (UNESCO World Heritage Center 2001; Gilli 2011). Karst areas can accommodate high densities of land snails due to the presence of readily available CaCO<sub>3</sub>, which the land snails need for their growth and reproduction (Gardenfors 1992; Graveland et al. 1994; Schilthuizen and Vermeulen 2000). Karst ecosystems are also known to support many endemic land snail species due to their isolation which is ideal for speciation (Day and Urich 2000; Schilthuizen and Rutjes 2001; Griffiths and Herbert 2013; Uchida et al. 2013; Tanmuangpak et al. 2015; Foon et al. 2017). Hundreds of square kilometers of karsts in the Southeast Asia are considered as "arks of biodiversity" as well as centers of land snail speciation. In Thailand, all species of the genus *Sinoennea* were found in karst (Tanmuangpak et al. 2015). In Perak, Peninsular Malaysia, 34 species out of all the 122 species of land snails were unique to one out of the 12 surveyed limestone hills (Foon et al. 2017). Around 80% of all of Malaysia's land snails can be found on karst regions, which make up only 1% of the country's total land area (Schilthuizen 2000).

There are 35,000 km<sup>2</sup> of karst in the Philippines which is about 10% of the country's total land area (Restificar et al. 2006). Of these, only about 29% are protected by the Philippine government under laws such as the National Caves and Cave Resources Management and Protection Act in 2001 (Restificar et al. 2006). However, most of the government's actions to protect karst ecosystems are not primarily for the conservation of their biodiversity, which means that a karst area is protected for other reasons such as for ecotourism. Consequently, many local karst ecosystems are subjected to destruction and disturbance caused by anthropogenic activities. These include, limestone mining, harvesting of swiftlet nests, ecotourism, extraction of phosphate and guano, and hunting of bats for food (Hobbs 2004; Mickleburgh et al. 2009; Scheffers et al. 2012). Unfortunately, the impact of anthropogenic disturbance on the biodiversity of karst ecosystems in the Philippines is difficult to predict.

Among the karst ecosystems in Luzon Island is the Masungi Georeserve in Rizal Province which has a total area of about 1,500 ha. It has a history of quarrying, mining, land-grabbing and large-scale illegal logging activities until the 1990s. In 1993, the Department of Environment and Natural Resources proposed that the Masungi rock and its immediate area to be declared as a "strict nature reserve and wildlife sanctuary", however, it was not passed (DENR 1993). In 1996, a private group, the Masungi Georeserve Foundation, secured the spine of the rock formation and began the restoration and rehabilitation. In 2015, the area was opened to tourists, but only in limited numbers, and is now being developed as a geopark (Evangelista 2016). The georeserve has several cave systems, featuring various rock configurations, limestone towers, steep slopes, and montane rainforests. It has become a popular tourist destination, especially for hikers and mountaineers, due to its hanging bridges, rope courses, eco-trails, and viewing platforms, such as suspended metallic netting above the karst and those on top of the park's peaks (Evangelista 2016). It is home to various wildlife, such as insects, monitor lizards, snakes, birds, cloud rats, monkeys, and civets. There are also several plants endemic to the Philippines that can be found in the area, such as Jade vine (Strongylodon macrobotrys), narra (Pterocarpus indicus), palm trees (Arecaceae), and ipil (Intsia bijuga). The place also provides a source of livelihood to the local community by serving as park rangers (pers. obs.).

Land snails have been observed in Masungi Georeserve, but their diversity is not well documented. Thus, this research on the diversity and distribution of land snails is a pioneering undertaking. However, being an ecotourism site, the geopark is still being subjected to some disturbance. Activities such as mountain climbing, spelunking, and hiking can affect not just the area's geologic formations but also the flora and fauna thriving on them. This is especially true for land snails since anthropogenic disturbances can easily destroy their habitat and introduce invasive species into these habitats. With little knowledge of its biodiversity, it is essential to study its potential as an important karst ecosystem in the country. This study aimed to generate baseline information on the diversity and distribution patterns of macro (> 5 mm) and micro land snails (< 5 mm) in Masungi Georeserve. Data generated from this study will be vital in designing a comprehensive management and conservation plan for the malacofauna of the georeserve and could serve as a model for other karst environments in the country.

# MATERIALS AND METHODS

#### Sampling area

Masungi Georeserve (14°36'18" N and 121°20'19" E) is located in Rizal Province, Luzon Island, Philippines (Figure 1). It is 45 km east of Manila and consists of several cave systems, featuring various rock formations, limestone towers, steep slopes, and rainforests (Figure 1). Its total area is about 1,500 ha and, in its center, is the 10 km spine of limestone formations. The spine has two prominent peaks, the taller peak (600 masl) known as "Tatay" and the second, consisting of five interconnected peaks, is called "Nanay". The georeserve has a history of quarrying, mining, land-grabbing, and large-scale illegal logging activities. Most of these occurred during the early 1990s and have been stopped when the Masungi Georeserve Foundation bought the area in 1996. The Foundation rehabilitated the area while educating the tourists on the importance of nature conservation. They have protected it from illegal loggers, poachers, and land-grabbers, and initiated several tree-planting projects.

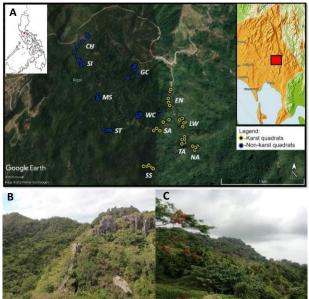


Figure 1: A) Distribution of the different sampling quadrats in the karst and non-karst areas of the Masungi Georeserve in Rizal Province, Luzon Island, Philippines (SS-600 steps; SA-Sapot; TA-Tatay; NA-Nanay; LW-Liwasan; EN-Masungi Entrance; GC-Garden Cottages; MS-Mahogany Stand; WC-Water Cascades; ST-Stream; SI-Silayan; CH-Chapel). B) Karst area of the Masungi Georeserve. C) Non-karst area of the Masungi Georeserve.

#### Sampling protocol

The sampling method for macro land snails used in this study was adapted from de Chavez and de Lara (2011) on their survey in Mount Makiling Forest Reserve, Philippines while protocol for micro land snails was adapted from the works of Clements et al. (2008), Schilthuizen and Rutjes (2001), and Uy et al. (2018). The center of the Masungi Georeserve, Masungi Rock, comprises the majority of the karst area of the georeserve while the forests surrounding Masungi Rock contains the non-karst areas. Six karst and non-karst areas in the Masungi Georeserve. The six karst sites are *600 steps (SS), Liwasan (LW), Tatay (TA), Nanay (NA), Sapot (SA)*, and the *Masungi Entrance (EN)*. On

the other hand, the six non-karst sites are *Garden Cottages (GC)*, *Mahogany Stand (MS)*, *Water Cascades (WC)*, *Stream (ST)*, *Silayan (SI)*, and the *Chapel (CH)*. In each site, five 5 x 5 m  $(25m^2)$  quadrats were set on accessible locations, at least 10 m away from the main trail (Figure 1). The quadrats were also at least 10 m away from each other to prevent pseudoreplication. Field sampling was conducted twice, once during the wet season (December 2017) and another during the dry season (June 2018). There were 60 quadrats established for each sampling period for a total of 120 quadrats (Supplementary Table 1).

#### Sampling site characterization

The sampling sites were characterized by determining the geographic coordinates for each of their quadrats and measuring the environmental variables prior to sampling of land snails during each fieldwork (Supplementary Tables 2 and 3). Geographic coordinates for each quadrat was determined using Garmin 12 Hand-held Global Positioning System (Garmin International, Inc., USA). The biotic variables were vegetation profile, canopy cover, number of trees, tree diameter at breast height, and leaf litter depth. Relative distribution of trees, grasses, vines, and herbs within the sampling sites was used to describe vegetation profile. Canopy cover was measured using a concave spherical densitometer (Forest Suppliers, Inc., USA). The number of trees within the quadrat was counted and their diameter at breast height (dbh) was measured. For leaf litter depth, five points (the corners and the center of the quadrat) were measured by inserting a wooden meter stick into the forest floor for each quadrat. The abiotic variables were altitude, air temperature, relative humidity, soil moisture, and soil calcium. Altitude was determined using Garmin 12 Hand-held Global Positioning System (Garmin International, Inc., USA). Air temperature and relative humidity were measured using a digital thermometer (Uni-T, Model UT333, China). Soil moisture was also measured by inserting a soil tester meter (Alotpower Soil Tester Meter) into the forest floor for each quadrat. Soil exchangeable calcium was determined by collecting approximately 2L topsoil at random points inside the quadrat. The samples were then submitted for analysis at the Service Laboratory of the Department of Soil Science, College of Agriculture, UPLB, using EDTA titration ammonium acetate extraction. Other significant information relevant to land snail habitat such as the presence of prominent karst outcrops and weather conditions were also documented onsite.

#### Collection and identification of samples

A thirty minute-sampling effort of 5 people per quadrat was allotted for searching for live snails and empty shells particularly on microhabitats such as bark and buttresses of trees, rotting logs, and undersides of plant leaves in each quadrat. To obtain the overall diversity and to increase the sampling size and statistical power of the data, live snails and empty shells were pooled together (de Winter and Gittenberger 1998; Cameron and Pokryszko 2004; Raheem et al. 2008). After photographing the samples, most of the live snails were returned to their original habitat while some live individuals including empty shells were collected and deposited at the UPLB Museum of Natural History (MNH) as vouchers (Supplementary Table 4). Empty shells and live snails were identified up to the species level, and their identification were confirmed using the published literatures of Faustino (1931), Bartsch (1932), and Springsteen and Leobrera (1986).

# Data analyses

Before performing parametric statistical analyses on all the quantitative data, they were tested for normal distribution using the one-sample Kolmogorov-Smirnov Test. Levene's Test was used to verify homogeneity of variance. The total number of species per quadrat was considered as species richness and individual counts of species per quadrat determined the species abundance. Abundance data was log-transformed prior to analysis to achieve normality and homogeneity of variance. Independent samples T-test was conducted to the species richness and abundance data gathered from the first and second sampling to determine whether there were significant differences between the dry and wet seasons. Since there were no significant differences between the sampling periods, the quantitative data from the two seasons were pooled together for more comprehensive analyses. All non-parametric and parametric statistical tests were performed using Statistical Package for Social Sciences (SPSS) for Windows (version 18, SPSS, Chicago, Illinois, USA).

Species diversity and evenness were determined using Shannon-Wiener Index of Diversity (Shannon and Weaver 1949), and species evenness (Simpson 1949), respectively. All indices were computed using Paleontological Statistics version 3.14 (Hammer et al. 2016). Species accumulation curve (SAC), a graph representing new species assembly relative to the sampling unit or efforts applied (Dove and Cribb 2006), was also generated using EstimateS ver. 8 (Colwell 2006). The measure of sampling effort used in this study was the number of individuals of each land snail species per quadrat. This was used to determine the richness estimation and diversity pattern ( $\alpha$ - or  $\beta$ - dominated) among the sampling sites. Completeness ratio, which is equal to the estimated number of species divided by the observed number of species, was calculated to determine sampling efficiency. This was different from sampling adequacy since sampling efficiency refers to the effectiveness of the sampling method while adequacy refers to the efficacy of the number of quadrats used. Incidence-based coverage estimator (SICE) was used to estimate the number of species based on the extrapolated scores obtained. The observed number of species (SOBS) was based on Mau Tao that assumes sampling without replacement should be done to include all samples in the event of randomization process. The shape of the SOBS can be used to describe the dispersal modes and richness partitioning of the species in the sample site.

The nine variables describing the environmental conditions (altitude, temperature, canopy cover, leaf litter depth, number of trees, tree diameter at breast height, relative humidity, soil moisture, and soil exchangeable calcium) were examined using principal component analysis (PCA). This enabled the extraction of the most important ecological gradients. PCA is very suitable for this purpose because it ensures that the maximum variability of the data points is visible (Chiba 2007). However, PCA is not ideal for analyzing community structure due to the unimodal species response curves along environmental gradients that are often exhibited by species. Thus, detrended correspondence analysis (DCA) was also used to analyze the land snail community structure. DCA assumes that species have unimodal species response curves. This was done by identifying clusters of ecologically similar species and similar locations in coordinate space. In addition, DCA allows the immediate relating of species to the samples where they can be found by simultaneously ordinating species and samples (Chiba 2007).

The average abundance of land snails from each sampling site was plotted against the scores of the top two principal components (environmental gradients), and further evaluated using quadratic regression analysis. These graphs enabled the examination of the relationship between abundance and the dominant environmental gradients. A matrix showing the locations of the land snail species was also calculated and used for DCA to compare the community structure between the karst and non-karst areas. Specifically, species richness and the diversity index of the sampling sites were plotted against the DCA scores, and were analyzed using exponential decay (single, 3 parameter) regression analysis. Both regression graphs were constructed using SigmaPlot v.10 (Systat Software, Inc.).

One-way analysis of similarity (ANOSIM) was done to check for significant differences in land snail community assemblages within habitat types and between the karst and non-karst areas. Similarity percentage analysis (SIMPER) was used to determine which land snail species can be used to indicate the presence of the habitat conditions of a karst and non-karst area. This analysis was also used to determine how similar the sampling sites within a habitat condition are to each other. ANOSIM and SIMPER were performed using Paleontological Statistics version 3.14 (Hammer et al. 2016).

# RESULTS

# Characteristics of the sampling sites

Six karst and six non-karst sites were sampled in Masungi Georeserve. The karsts were located on the Masungi rock itself while the non-karsts were in the surrounding forests. Nine environmental variables (altitude, temperature, canopy cover, leaf litter depth, number of trees, tree diameter at breast height, relative humidity, soil moisture, and soil exchangeable calcium) were used to characterize these areas. The karst sites are 600 steps (SS), Liwasan (LW), Tatay (TA), Nanay (NA), Sapot (SA), and Masungi Entrance (EN) (Supplementary Table 2). The 600 steps had the highest altitude (604.6 masl), highest tree diameter at breast height (59.04 cm), and highest leaf litter depth for the dry season (4.88 cm). It is slightly sloping, and man-made structures (benches and staircase) are present. Liwasan was one of the two sites with the highest relative humidity during the wet season (100%). There were evidences of former slash-and-burn practices in the form of old charcoal dumps. Tatay, one of the peaks in the georeserve, had the highest canopy cover among the karst sites. It has the highest relative humidity during the wet season (100%) and had the highest soil moisture for the wet season (8.12). Aside from being a part of the main trail of the georeserve, there are also some caves in this area. Nanay, had the highest soil calcium among the karst sites (29.95 cmolc/kg soil). The ground was slightly sloping, and the limestone formations had many clefts and crevices. This site is subjected to periodical cleaning wherein dead and fallen branches have been cleared. Sapot, one of the georeserve's tourist attractions. The main trail runs through this site and consists of concrete steps with bamboo. The last karst site, the Masungi Entrance, had the highest leaf litter depth (7.34 cm) and highest temperature (27.2 °C) for the wet season, as well as the highest relative humidity (78.23%) and highest soil moisture (3.92) for the dry season. There were also many limestone boulders and bamboo groves, and a few man-made pipes in this area.

The non-karst sites are *Garden Cottages* (*GC*), *Mahogany Stand* (*MS*), *Water Cascades* (*WC*), *Stream* (*ST*), *Silayan* (*SI*), and the *Chapel* (*CH*). The *Garden Cottages* had the highest tree diameter among non-karst sites (61.67%) (Supplementary Table 3). It also had highest relative humidity (95.74%) and soil moisture (6.92). Most of the quadrats had traces of slash-and-burn activities indicated by old charcoal dumps. *Mahogany Stand* had the highest altitude (530.2 masl), number of trees (6), leaf litter depth for both wet (6.52 cm) and dry (4.05 cm) seasons, and temperature for both wet (27.64 °C) and dry (29.54 °C) seasons. There were also evidences of past slash-and-burn practices. *Water Cascades*, with a series of small waterfalls, had the highest canopy cover for both wet (91.67%) and dry (88.93%) seasons. It also had the highest soil moisture for the

dry season (5.64) and the highest soil calcium (16.21 cmol<sub>c</sub>/kg soil) among the non-karst sites. The slope of the ground was very steep and there were many boulders scattered all over this site. The *Stream* had the highest relative humidity for the dry season (77.22%) among the non-karst sites. There were also evidences of previous anthropogenic activities such as old construction site for housing and a dump for construction materials. The *Silayan* site was slightly sloping and there were also evidences of former slash-and-burn activity. The *Chapel* site had an abundance of bamboo groves.

#### Land snail diversity, abundance, and distribution

A total of 45 species of land snails consisting of 1,283 individuals were sampled in the Masungi Georeserve (Tables 1 and 2; Figures 2-7). Thirty-nine species belonging to 12 families consisting of 1,159 land snails were sampled in the karst areas while 16 species belonging to 6 families consisting of 124 individuals were sampled in the non-karst region. Of the 1,283 land snails collected, 56 (4%) were live snails while 1,227 (96%) were empty shells. Also, of the 45 species sampled, 7 (15%) were invasive while 39 (85%) were species endemic to the Philippines. However, due to insufficient data on the identified species, further classification if they were endemic only to the georeserve or native species was not possible. The species classified as invasive were land snails that were not endemic to the Philippines and were either reported as pests or already established as invasive. Meanwhile, 37 (92%) of the land snail species in the karst area and 11 (69%) of the land snail species in the non-karst area were species endemic to the Philippines. Hypselostoma latispira masungiensis, has been proven to be a new subspecies in the Masungi Georeserve. It has a significantly larger shell and an additional apertural tooth compared to Hypselostoma latispira latispira of Baguio (Lipae et al. 2020). The species that were identified were either not listed in the IUCN red list or were listed as data deficient and least concern. The Shannon-Wiener diversity index (Table 5) of the karst area (H' = 2.94) was more diverse than in the non-karst (H' = 2.18). However, species evenness (Table 3) in the non-karst (J' = 0.55) was higher than in the karst (J' = 0.49). In the karst area, the site with the highest diversity index was in *Tatay* (H' = 2.90), while the lowest was along the Masungi Entrance (H' = 2.26). Nanay (J' = 0.72) had the highest species evenness among the karst sites while the lowest was in *Sapot* (J' = 0.54). In the non-karst area, the highest diversity index was in *Stream* (H' = 2.13) while the lowest was in Silayan (H' = 1.09). However, Silayan (J' =0.99) had the highest species evenness among the non-karst sites while *Chapel* (J' = 0.71) had the lowest.

In all sampling sites, Hemiglypta semiglobosa had the highest individual count (198) followed by Helicostyla metaformis (176) and Ryssota nigrescens (119). On the other hand, the species with the lowest individual counts were Chloraea dryope, Leptopoma perlucidum, Laevicaulis alte, and Sarasinula plebeia (1) followed by Allopeas sp.1, 2, and 3 (2) and Microcystina sp.2, Chloraea fibula, Obba listeri, Obba morongensis, Endodonta sp., Geotrochus mp. 2, and Moulinsia sp. (3). Among karsts, the highest individual counts were for Hemiglypta semiglobosa (170) and Helicostyla metaformis (147) while lowest for Chloraea dryope (1), Leptopoma perlucidum (1), and Laevicaulis alte (1). In the non-karst area, the highest individual counts were also for Helicostyla metaformis (29) and Hemiglypta semiglobosa (28) while the lowest were for Calocochlia sp. 1, Hemitrichiella setigera, and Sarasinula plebeia, all with only a single individual identified.

Table 1: Land snails sampled across the karst sites of the Masungi Georeserve (SS-600 steps; SA-Sapot; TA-Tatay; NA-Nanay; LW-Liwasan; EN-Masungi Entrance).

LAND SNAIL FAMILY & SPECIES	Status*		Karst					
A shokini Jaa		SS	SA	ТА	NA	LW	EN	Total
Achatinidae				_	-			
Allopeas sp.1	is	0	1	0	0	0	1	2
Lissachatina fulica (Ferussac, 1821)	is	0	0	0	0	0	6	6
Ariophantidae								
Macrochlamys indica (Benson, 1832)	npl	4	1	1	3	0	1	10
Macrochlamys sp.	npl	3	4	1	2	6	0	16
Microcystina sp.1	npl	2	1	4	5	7	0	19
Assimineidae								
Acmella mp.1	npr	0	6	0	0	3	0	9
Acmella mp.2	npr	0	0	4	5	17	0	26
Camaenidae								
Calocochlia sp. 1	npl	0	1	0	0	3	0	4
Calocochlia sp. 2	npl	3	0	10	0	0	0	13
Chloraea dryope (Broderip, 1841)	npl	0	0	1	0	0	0	1
Helicostyla carinata (Lea, 1840)	npl	1	1	3	0	3	0	8
Helicostyla metaformis (Ferussac, 1821)	npl	13	29	24	20	51	10	147
Helicostyla rufogaster (Lesson, 1831)	npl	15	7	5	7	5	4	43
Helicostyla woodiana (Lea, 1840)	npl	42	12	10	9	9	2	84
Obba lasallii (Eydoux, 1838)	npl	1	1	0	1	0	1	4
Obba listeri (Gray, 1825)	npl	3	0	0	0	0	0	3
Obba morongensis (von Moellendorff, 1890)	npl	0	0	3	0	0	0	3
Obba marmorata (von Moellendorff, 1890)	npl	0	0	0	3	0	2	5
Chronidae								
Kaliella sp.	npl	1	0	4	0	1	0	6
Hemiglypta cuveriana (Lea, 1852)	npl	14	14	6	0	10	1	45
Hemiglypta semiglobosa (Pfieffer, 1854)	npl	33	50	20	16	32	19	170
Hemitrichiella setigera (Sowerby, 1898)	npl	8	4	2	0	3	4	21
Ryssota nigrescens (von Moellendorff, 1888)	npl	29	20	13	18	12	5	97
Cyclophoridae								
Cyclophorus appendiculatus (Pfieffer, 1854)	npr	24	23	15	7	9	4	82
Cyclophorus reevei (Hidalgo, 1890)	npr	13	8	11	7	13	0	52
Leptopoma helicoides (Grateloup, 1840)								

Leptopoma perlucidum (Grateloup, 1840)	npr	0	0	0	0	1	0	1
Leptopoma sp.	npr	15	1	9	4	7	0	36
Cyclophorus sp.	npr	0	0	1	0	4	0	5
Endodontidae								
Endodonta sp.	npl	0	1	0	0	2	0	3
Helicarionidae								
Helicarion sp.	npl	0	2	0	1	1	0	4
Nanina sp.	npl	1	2	1	0	1	0	5
Hydrocenidae								
Georissa mp.	npr	29	0	0	10	0	0	39
Hypselostomatidae								
<i>Hypselostoma latispira masungiensis</i> (Lipae & de Chavez, 2020)	npl	11	0	6	3	0	0	20
Pupinidae								
<i>Moulinsia</i> sp.	npl	0	0	2	0	1	0	3
Trochomorphidae								
Videna metcalfei (Pfeiffer, 1845)	npl	8	9	5	4	5	1	32
Geotrochus mp.1	npl	12	12	10	29	33	2	98
Geotrochus mp.2	npl	3	0	0	0	0	0	3
Veronicellidae								
Laevicaulis alte (Ferussac, 1822)	is	0	0	1	0	0	0	1
TOTAL		293	214	182	165	242	63	1159

\*is - invasive species; npl - native pulmonate; npr - native prosobranch; mp - morphospecies

Table 2: Land snails sampled across the non-karst sites of the Masungi Georeserve (GC-Garden Cottages; MS-Mahogany Stand; WC-Water Cascades; ST-Stream; SI-Silayan; CH-Chapel).

Status*		Non-karst					
Status	GC	MS	WC	ST	SI	СН	Total
is	0	0	0	2	0	0	2
is	0	0	0	2	0	0	2
is	2	0	6	1	0	4	13
npl	0	0	0	3	0	0	3
is	4	0	0	0	0	0	4
npl	0	1	0	0	0	0	1
npl	0	0	3	0	0	0	3
npl	4	3	12	0	3	7	29
	is is npl is npl npl	GC is 0 is 2 npl 0 is 4 npl 0 npl 0 npl 0	GC         MS           is         0         0           is         0         0           is         2         0           npl         0         0           is         4         0           npl         0         1           npl         0         0	GC         MS         WC           is         0         0         0           is         0         0         0         0           is         2         0         6         0           npl         0         0         0         0           is         4         0         0         0           npl         0         1         0         0         3	Status*         GC         MS         WC         ST           is         0         0         0         2           is         0         0         0         2           is         2         0         6         1           npl         0         0         0         3           is         4         0         0         0           npl         0         1         0         0           npl         0         3         0         0	Status*         GC         MS         WC         ST         SI           is         0         0         0         2         0           is         0         0         0         2         0           is         0         0         0         2         0           is         2         0         6         1         0           npl         0         0         0         3         0           is         4         0         0         0         0           npl         0         1         0         0         0           npl         0         3         0         0         0	Status*         GC         MS         WC         ST         SI         CH           is         0         0         0         2         0         0           is         0         0         0         2         0         0           is         0         0         0         2         0         0           is         2         0         6         1         0         4           npl         0         0         0         3         0         0           is         4         0         0         0         0         0           npl         0         1         0         0         0         0           npl         0         3         0         0         0         0

Helicostyla rufogaster (Lesson, 1831)	npl	0	1	0	2	0	1	4
Obba marmorata (von Moellendorff, 1890)	npl	1	1	0	1	0	0	3
Chronidae								
Hemiglypta cuveriana (Lea, 1852)	npl	0	0	1	2	0	1	3
Hemiglypta semiglobosa (Pfieffer, 1854)	npl	0	3	8	1	3	13	28
Hemitrichiella setigera (Sowerby, 1898)	npl	0	1	0	0	0	0	1
Ryssota nigrescens (von Moellendorff, 1888)	npl	1	4	9	2	4	2	22
Cyclophoridae								
Leptopoma helicoides (Grateloup, 1840)	npr	0	0	0	0	0	4	4
Veronicellidae								
Sarasinula plebeian (P. Fischer, 1868)	is	0	0	1	0	0	0	1
TOTAL		12	14	40	16	10	32	124

\*is - invasive species; npl - native pulmonate; npr - native prosobranch; mp - morphospecies

Table 3: Land snail species diversity and evenness across the 12 sampling sites in the Masungi Georeserve (SS-600 steps; SA-Sapot; TA-Tatay; NA-Nanay; LW-Liwasan; EN-Masungi Entrance; GC-Garden Cottages; MS-Mahogany Stand; WC-Water Cascades; ST-Stream; SI-Silayan; CH-Chapel).

Habitat type	Sampling site	Number of quadrats*	Total number of species	Н'	J'
	SS	10	25	2.79	0.65
	SA	10	24	2.55	0.54
	ТА	10	27	2.90	0.67
Karst	NA	10	20	2.66	0.72
	LW	10	26	2.66	0.55
	EN	10	15	2.26	0.64
	Total	60	39	2.94	0.49
	GC	10	5	1.45	0.85
	MS	10	7	1.77	0.84
	WC	10	7	1.68	0.77
Non-karst	ST	10	9	2.13	0.94
	SI	10	3	1.09	0.99
	СН	10	7	1.61	0.71
	Total	60	16	2.18	0.55
	Grand total	120	45	2.99	0.43

\*combined quadrats of 1<sup>st</sup> and 2<sup>nd</sup> sampling

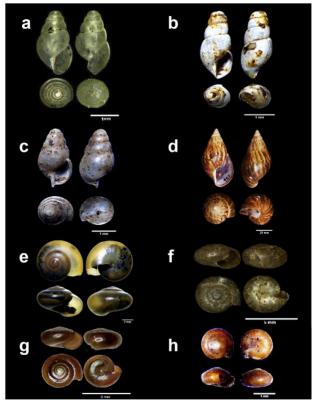


Figure 2: Land snails of Masungi Georeserve: a) *Allopeas* sp.1, b) *Allopeas* sp.2, c) *Allopeas* sp.3, d) *Lissachatina fulica*, e) *Macrochlamys indica*, f) *Macrochlamys* sp., g) *Microcystina* sp.1, h) *Microcystina* sp.2.

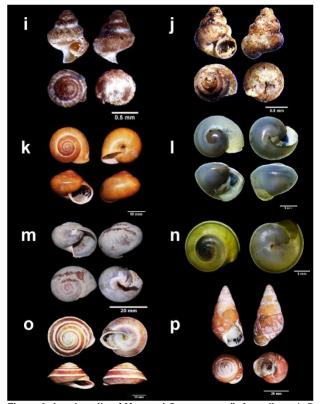


Figure 3: Land snails of Masungi Georeserve: i) Acmella sp.1, j) Acmella sp. 2, k) Bradybaena fodiens, I) Calocochlia sp. 1, m) Calocochlia sp. 2, n) Chloraea dryope, o) Chloraea fibula, p) Helicostyla carinata.

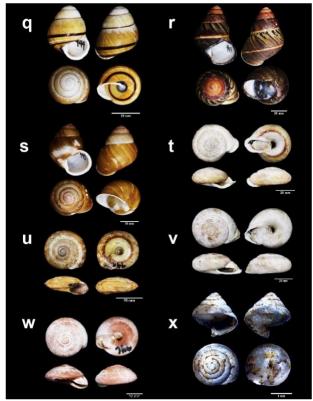


Figure 4: Land snails of Masungi Georeserve: q) Helicostyla metaformis, r) Helicostyla rufogaster., s) Helicostyla woodiana, t) Obba lasallii, u) Obba listeri, v) Obba marmorata, w) Obba morongensis, x) Kaliella sp..

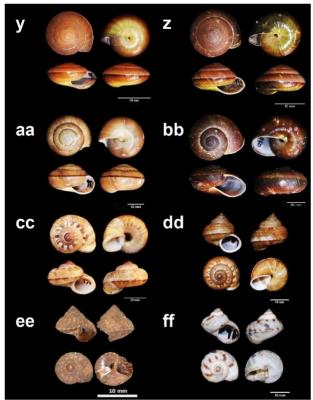


Figure 5: Land snails of Masungi Georeserve: y) Hemiglypta cuveriana, z) Hemiglypta semiglobosa, aa) Hemitrichiella setigera, bb) Ryssota nigrescens, cc) Cyclophorus appendiculatus, dd) Cyclophorus reevei, ee) Cyclophorus sp., ff) Leptopoma helicoides.

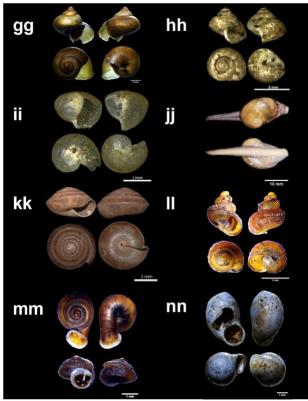


Figure 6: Land snails of Masungi Georeserve: gg) Leptopoma perlucidum, hh) Leptopoma sp., ii) Endodonta sp., jj) Helicarion sp., kk) Nanina sp., II) Georissa mp., mm) Hypselostoma latispira masungiensis, nn) Moulinsia sp..

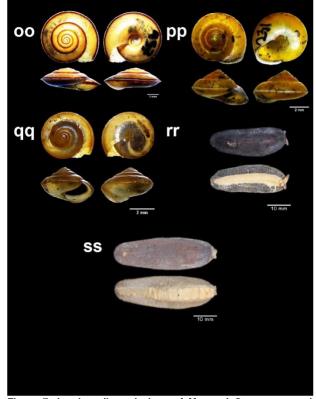


Figure 7: Land snails and slugs of Masungi Georeserve: oo) Videna metcalfei., pp) Geotrochus mp. 1, qq) Geotrochus mp. 2, rr) Laevicaulis alte, ss) Sarasinula plebeia.

#### Species accumulation curve

Sample- and individual-based species accumulation curves (SACs) showed that land snail assemblages in the karst area and the whole Masungi Georeserve exhibited an α-dominated community characterized by rapid asymptotes (Figure 8). An adominated community means that fewer quadrats and individuals were needed to capture the entire species diversity. This indicates that many species can be found in a relatively small area. This also suggests that the sampling sites were saturated, niche assembled and locally rich. On the other hand, the sample-based and individual-based SACs for the non-karst area revealed that the land snail assemblage exhibited a βdominated community characterized by late asymptotes. This means that more quadrats and individuals were needed in order to capture species diversity. This indicates that only a few species can be found in a relatively small area. This also suggests that the sampling sites were unsaturated, dispersal assembled and regionally enriched (Dove and Cribb 2006). The samplebased and individual-based SACs showed very high completeness ratios for the karst area (0.93) and the whole Masungi Georeserve (0.97). Meanwhile, the non-karst area's SACs showed the lowest completeness ratio (0.77).

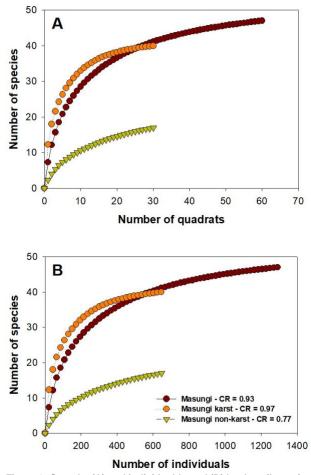


Figure 8: Sample- (A) and individual-based (B) land snail species accumulation curve in the two habitat conditions in Masungi Georeserve. High completeness ratio (CR) indicates effective sampling.

# Comparison of land snail community assemblages in karst and non-karst areas

The Principal component analysis (PCA), incorporating all karst and non-karst areas of the Masungi Georeserve, showed that the first and second PC axes accounted for 61% of the total variation explained by the measured environmental variables (Table 4).

Component loadings	PC 1	PC 2	
Environmental Variables			
Altitude	0.413	0.348	
Temperature	-0.418	0.333	
Canopy cover	0.047	-0.505	
Leaf litter depth	0.258	0.539	
Number of trees	-0.192	0.207	
Tree diameter at breast height	-0.248	0.282	
Relative humidity	0.397	-0.263	
Soil moisture	0.393	0.161	
Soil calcium	0.419	0.074	
Variance explained by components (Eigenvalue)	3.626	1.890	
Percentage of total variance explained	40.287	20.997	

Only components with an eigenvalue of >1.0 were used since these would explain more variance than the original individual variables. Components of the PCA are axes that lie in the directions of variation in the data. The component with the highest eigenvalue lies along the axis of maximum variation in the data thus this component will account for most of the variation that was observed (Wallisch et al. 2014). The lower the eigenvalue, the less is the variation that a component can account for. A component with an eigenvalue of < 1.0 indicates that it accounts for less variation compared to the original variables. The highest loading scores in the first principal component (PC1) corresponded to habitats with a higher soil calcium but lower temperature. PC1 is then interpreted as temperature and soil calcium gradient. Meanwhile, the highest loading scores in the second principal component (PC2) corresponded to a habitat with a higher leaf litter depth but lower canopy cover. The PC2 is interpreted as the canopy cover and leaf litter depth representing the productivity gradient.

There was no significant linear correlation between the number of individuals/25m<sup>2</sup> and the PC1 scores of the sites ( $R^2 = 0.37$ , P > 0.05). The relationship was represented by a concave regression curve  $f = 86.46 + 42.54x + 6.16x^2$  (Figure 9). This pattern nevertheless showed an increase in the number of individuals as PC1 score (temperature and soil calcium) increases. Karst sites were clustered and have higher PC1 scores and greater numbers of individuals while non-karst sites were spread out and mostly have lower PC1 scores and fewer individuals. Meanwhile, for PC2, there was also no significant linear correlation between the number of individuals/25m<sup>2</sup> and the scores of the sites ( $R^2 = 0.35$ , P > 0.05). The relationship was represented by a concave regression curve f = 59.62 - 15.92x +27.30x<sup>2</sup>. No clear pattern can be observed for the PC2 scores and the number of individuals/25m<sup>2</sup>. Both the karst and non-karst sites have relatively similar PC2 scores, but the karst sites have

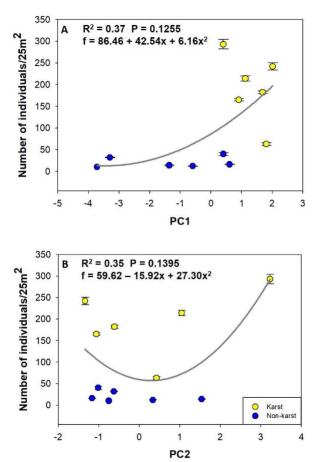


Figure 9: Scatter plots between the average number of individuals of land snails at each site in Masungi Georeserve (± SE) and (A) PCA 1 scores (soil calcium and temperature gradient) and (B) PCA 2 scores (productivity gradient). Gray line signifies best fit curve. n=12

higher number of individuals/25m<sup>2</sup> than the non-karst sites. These results suggested that the environmental variable gradients across the sites probably did not cause very significant difference in land snail relative abundance in the Masungi Georeserve.

Detrended correspondence analysis (DCA), performed for all sites in the karst and non-karst sites of the Masungi Georeserve, extracted three axes. After correlation analysis, it was observed that only the DCA axis 1 (DCA1) was highly correlated with PC1 scores of the sites. The DCA1 scores of the sites were negatively correlated with the PC1 (R = -0.92, P < 0.05). This means that DCA1 represented species that were strongly influenced by soil calcium and temperature gradient across the sampling sites. The matrix created for the DCA (Figure 10) enabled the discrimination of distinct ecological groups of land snails where they have the greatest abundance. The first group, the karst-associated land snails (blue convex hull and dots), was characterized by having lower DCA1 scores while the second group, the non-karst- associated land snails (red convex hull and dots), was characterized by having higher DCA1 scores. However, a third group, the intermediates (species that can thrive in both karst and non-karst habitat), was also observed (orange convex hull and dots) but was not clearly discriminated since it overlapped with the first.

Species richness and Shannon diversity index (*H'*) were then plotted against the DCA1 scores (Figure 11). Initial linear regression analysis showed no significant correlation between species richness and DCA1 scores ( $R^2 = 0.11$ , P > 0.05) as well as between Shannon diversity index (*H'*) and DCA1 scores ( $R^2$ 

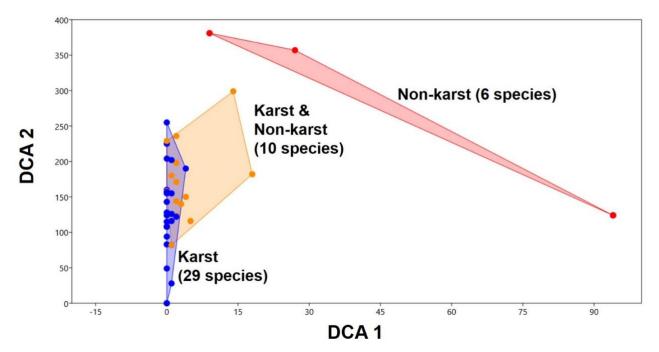


Figure 10: Detrended correspondence analysis (DCA) of the land snail species at different sites in Masungi Georeserve. Land snail communities were organized into three groups marked by convex hulls. DCA1 – soil calcium and temperature gradient. n = 45

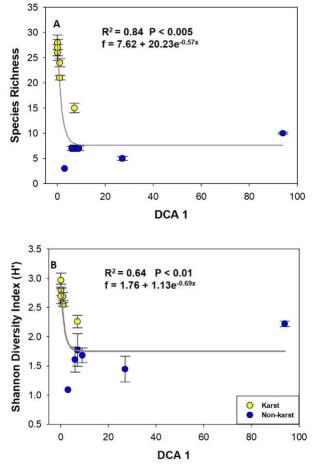


Figure 11: Scatter plots between DCA 1 scores (land snail community assemblage) of the sites and average ( $\pm$  SE) (A) species richness and (B) Shannon diversity index. Gray line signifies best fit curve. n=12

= 0.13, *P* > 0.05). However, when exponential decay regression analysis was performed, significant correlation was generated between species richness and DCA1 scores ( $R^2 = 0.84$ , *P* < 0.05) as well as between species diversity and DCA1 scores ( $R^2 = 0.64$ , *P* < 0.01). The relationship between species richness and DCA scores was represented by the regression curve  $f = 7.62 + 20.23e^{-0.57x}$ , while the relationship between species diversity and DCA1 scores was represented by the regression curve  $f = 1.76 + 1.13e^{-0.69x}$ . Thus, these showed that the land snail species richness and diversity decreased exponentially as the DCA1 scores increased.

One-way analysis of similarity (ANOSIM) was conducted to validate the results of the DCA. In ANOSIM, a high R value (> 0.30) would mean that the sampling sites within an area are different from each other and a low R value (< 0.30) would mean that the sites are similar. The p value will then confirm if the R value is significant or not. The R and p values for all the sites showed that the karst have significantly different habitat conditions compared to the non-karst (Table 3). The R values also indicated that the quadrats in the karst area exhibited similar habitat conditions. The same can also be interpreted from the R value of the non-karst area (Table 5).

Table 5: Analysis of similarity (ANOSIM) test for significant differences between karst and non-karst areas in Masungi Georeserve.

Areas	R	p-value	Total number of quadrats (n)
Karst	0.27	0.0001	60
Non-karst	0.17	0.0052	60
Overall	0.36	0.0001	120

La	and snail species	Average dissimilarity	Contribution (%)	Cumulative (%)	Mean abundance in karst	Mean abundance in non-karst
1.	Hemiglypta semiglobosa	12.89	14.6	14.6	6	1
2.	Helicostyla metaformis	11.71	13.27	27.87	5	1
3.	Ryssota nigrescens	7.48	8.47	36.34	3	1
4.	Cyclophorus appendiculatus	6.14	6.95	43.29	3	0
5.	Helicostyla woodiana	5.70	6.55	49.84	3	0
6.	Helicostyla rufogaster	3.88	4.40	54.24	1	0
7.	Chloraea geotrochus	3.71	4.21	58.44	2	0
8.	Cyclophorus reevei	3.61	4.09	62.53	2	0
9.	Leptopoma sp.	3.044	3.45	65.98	1	0
10.	Hemiglypta cuveriana	2.833	3.21	69.19	2	0

Similarity percentage analysis (SIMPER) was conducted for both the karst and the non-karst sites. The overall average dissimilarity of the areas was 88.28% signifying that the habitat condition in the karsts is indeed different from that of the nonkarsts. The land snail with the highest percent contribution (14.6%) to the dissimilarity between the two habitat types was *Hemiglypta semiglobosa* followed by *Helicostyla metaformis* (13.27%) and *Ryssota nigrescens* (8.47%) (Table 6). The 10 species identified by SIMPER were the land snail species that contributed most to the dissimilarity out of the 45 species. These species account for 69.19% of the observed average dissimilarity between the habitat conditions. All the species discriminated in SIMPER were more abundant in the karst area than in the non-karst.

# DISCUSSION

The Masungi Georeserve has the higher diversity (45 species and 12 families) compared to other protected sites in the Luzon Island including Mt. Makiling (24 species and 9 families), Mt. Banahaw (34 species and 11 families), and Pamitinan Protected Landscape in Rodriguez, Rizal (29 species and 13 families) (de Chavez and de Lara 2011; RRB Agudo unpublished observations; PPNL Mendoza unpublished observations; KMG Perez et al. unpublished observations; Uy et al. 2018). In addition, Faustino (1930) only listed 30 species in Rizal, Philippines while 39 species belonging to 12 families were documented from 13 localities in Sabah, Malaysia inclusive of three limestone outcrops (Uchida et al. 2013). However, it should be noted that some differences in species diversity may have been caused by differences in the sampling protocol. Even though the georeserve experienced habitat destruction in the past, it was observed that the number of invasive species was relatively low (15%) which is similar to a disturbed karst area near Istanbul, Turkey where 12.5% of the sampled land snails were invasive species (Örstan et al. 2005).

Majority of the land snails sampled in the georeserve were empty undamaged shells indicating natural death. It is most likely that these shells were amassed within 3 to 5 years since bioerosion is relatively fast in tropical regions. However, although they were only empty shells, they can still reflect the diversity of land snails in the area due to the low vagility and restricted distribution of land snails. This means that the presence of empty shells would also signify the presence of live representatives of these land snails.

The observed malacofaunal diversity can be attributed to the Masungi Georeserve's relatively vast karst area that has a complex terrain of fissures, clefts, and outcrops. This yields a multitude of ecological niches that can support a high species diversity (Clements et al. 2006). Previous studies have demonstrated that there were higher densities and endemicity of mollusks in karst due to the readily available calcium source needed for egg production and shell formation (Gardenfors 1992; Graveland et al. 1994; Schilthuizen and Vermeulen 2000). Another factor that may have facilitated the possible malacofaunal recovery is the rehabilitation and conservation efforts of the Masungi Georeserve Foundation which acquired the area in 1996. The foundation closed the land to the public for 19 years which has possibly allowed its biodiversity to reestablish. Even today as some parts of the reserve have been opened to the public, the number of visitors allowed each day are tightly controlled. Hiking trails were also constructed to cause the least modifications in the natural environment.

The species richness and abundance of land snails were greater in the karst area than in non-karst. However, species evenness was higher in the non-karst which could be due to the disparity between the individual snail counts of various species in the karst and non-karst habitats. The species with the highest individual counts (Hemiglypta semiglobosa and Helicostyla metaformis) were the same for both karst and non-karst. However, in the karst sites, 170 individuals were observed for Hemiglypta semiglobosa and only 28 in the non-karst sites. Meanwhile, 147 individuals of Helicostyla metaformis were collected in the karst sites and only 29 in the non-karst sites. The dominance of these two species may be attributed to their habitat preference and shell coloration. Hemiglypta semiglobosa dwells on the soil surface and blends its coloration to probably evade predation. Meanwhile, Helicostyla metaformis is an arboreal snail and has a shell with a bright coloration. These may help the snail to avoid predation by keeping out of the reach of many terrestrial predators and deterring them through its brightly colored shell.

The SACs further revealed that the karst and non-karst areas differed in the type of diversity pattern. It is important to note that sites with  $\alpha$ -dominated community recover faster from population depletion than those with a  $\beta$ -dominated community (Shackell et al. 2012). This is because an  $\alpha$ -dominated diversity indicates species that are interactive thus a locally depleted population can be replenished by neighboring populations. Since karsts exhibited  $\alpha$ -diversity, it is possible that the forests were repopulated by snails originating from adjacent limestone outcrops. On the other hand, land snail populations in non-karst, which has  $\beta$ -diversity, will take longer time to recover from the depopulation caused by habitat destruction. The effects of this difference were evident in the disparity in species richness and

abundance between the two areas. The high completeness ratios obtained indicate that the sampling method employed during the two fieldworks can be considered enough to capture the entire malacofaunal diversity. However, it was observed that the sampling efficiency in the karst sites was higher than that of the non-karst sites. This difference may be due to the identical sampling plot size utilized for the karst and non-karst. Identical sampling methods may yield unrepresentative data sets and, in the sampling of different habitat types, result in one habitat having higher sampling efficiency than the other. The higher efficiency in karst may be due to the smaller sampling plots needed to capture substantial proportions of the malacofaunal diversity since karsts exhibit high land snail species density. On the other hand, the lower efficiency in non-karst indicates that more sampling plots should be used due to the low malacofaunal diversity exhibited by this habitat condition (Liew et al. 2008).

The land snails were discriminated into three ecologically different groups. The first group preferred cooler habitats with rich soil calcium (karst), while the second group thrived best in warmer patches with poor soil calcium (non-karst). A third intermediate group was identified comprising of species found in both the karst and non-karst. Land snail species that were exclusive in the karst sites constituted 64.44% (29 out of the 45) of the species found in the Masungi Georeserve. On the other hand, 13.33% (6 out of the 45 total) species were found only in non-karst sites. The remaining 22.22% (10) species were found in both the karst and non-karst sites. Since desiccation is one of the most effective limiting factors for land snails, there were more species in the sites with low temperature where the likelihood of desiccation is low (Chiba 2007). The high amounts of soil calcium in these areas allow more species to thrive and maintain their populations. Land snails that were found only in non-karst sites were probably those that can survive warmer temperatures. The remaining members were invasive species (Bradybaena fodiens and Lissachatina fulica) which are adapted to wide range of environmental conditions, or species with a small shell (Chloraea fibula), and in the case of Sarasinula plebeia., is shell less.

The DCA against diversity graphs showed that as the distance from the karst area increases, the species richness of land snails tends to decline drastically. Also, the presence of an intermediate group of land snail species, which overlapped with the karst-associated snails, could indicate possible evidence of their dispersal from the karst towards the surrounding non-karst region. These results highlighted the differences in habitat conditions between the karst and non-karst areas. The habitat condition in the karst area is ideal wherein land snails can successfully thrive. It has been observed in previous studies that karst areas can host high densities of land snails primarily due to the high amounts of readily available calcium in these locations (Gardenfors 1992; Graveland et al. 1994; Schilthuizen and Vermeulen 2000). Remarkably, since the georeserve experienced widespread habitat destruction in the past, the high species richness and diversity concentrated in the karst area demonstrated that karst ecosystem can indeed function as a refuge for land snails.

The results from ANOSIM further supported the above findings that there was a difference in land snail assemblages between karst and non-karst areas. The ANOSIM, based on the R and p values, showed that the habitat condition of the karst sites is significantly different from that of the non-karst sites. Although the karst and non-karst areas were adjacent to each other, karst area is delineated by limestone outcrops and cliffs that act as major dispersal barriers. Landscapes with these features have more dissimilar community assemblage than in open or topographically similar regions (Garcillán and Ezcurra 2003).

Another factor that may have contributed to the variation in community assemblage is the low vagility of land snails. There is a faster decay in similarity as distance increases for organisms with low vagility since there is lower dispersal across the sites, resulting in heterogeneous communities (Soininen et al. 2007). Based on SIMPER, the land snail species that contributed greatly to the community dissimilarity was *Hemiglypta semiglobosa*. Aside from this, nine other species that had significant effects on the community dissimilarity were identified. It was also seen that these 10 species that affected the dissimilarity greatly were all observed in greater abundance in the karst sites. These then imply that such species can indicate the habitat conditions of a karst area. Thus, these land snails can be used as indicator species that can help identify if a certain locality has conditions similar to that of a karst ecosystem.

This study suggests that the land snails of the Masungi Georeserve, particularly those in the karst area, exhibits possible resilience against habitat destruction. This is only a possibility since there is no quantitative data on the diversity in the area before habitat destruction, and also no data on the degree and frequency of habitat destruction. The results showed that although the Masungi Georeserve was subjected to various anthropogenic disturbances in the past, it harbored very high land snail diversity. The resiliency of the land snails could also suggest that they have fast recovery allowing them to replenish their populations in disturbed habitats. These observations were supported by the  $\alpha$ -dominated community exhibited in the karsts. The endemic land snail species of the georeserve appear to be unaffected by the presence of invasive snail species. This may have been due to their resistance to perturbations caused by invasive snail species or that they occupy different niches. Human activities introduce many invasive species that either compete with the native snails for resources or eat the native snails (Uchida et al. 2013; Nurinsiyah et al. 2016). However, in the case of the Masungi Georeserve, only 15% of the present land snail species and 2% of the total individuals sampled were invasive. This could be due to the lack of environmental niches that invasive species could occupy. Another possibility is that the native land snail species of the georeserve have become adapted to disturbances in the environment due to the highly dynamic ecological conditions of the area. The georeserve is located in Luzon Island, an ancient oceanic island that has experienced episodes of major environmental perturbations and regenerations for thousands of years. This may have caused more sensitive species to become extinct, leaving more resilient species to thrive (Rickart et al. 2011).

# CONCLUSION AND RECOMMENDATIONS

Overall, this study demonstrated the very rich malacofauna of the Masungi Georeserve. The land snail diversity was higher in the karst area, where there is an abundance of available soil calcium and cooler temperature enough to prevent rapid desiccation of land snails, compared to the non-karst area. Karsts are ideal habitats for land snails that are important in maintaining land snail biodiversity thus further emphasizing the need to protect and conserve these areas. Considering the habitat destruction that previously occurred, these results also proved that Philippine karsts are also arks of land snail biodiversity. This further emphasizes the need to protect the Masungi Georeserve and other karst areas of the Philippines.

The inclusion of the cave systems of the Masungi Georeserve is recommended to further malacofaunal diversity studies of the area. Since the place is now a popular ecotourism destination, it is also necessary that the effects of anthropogenic disturbance on the species richness and abundance of land snails be investigated. Further studies of its malacofauna should also incorporate molecular techniques to examine the genetic integrity and evolutionary history. It is also recommended to conduct related studies on other karst areas in the Philippines in order to validate the findings of this study.

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# CONFLICT OF INTEREST

The authors have no conflicts of interest.

# CONTRIBUTION OF INDIVIDUAL AUTHORS

BK Valdez is the principal author and conducted the sampling and the statistical analyses. JA Parcon helped in the sampling and processing of the snail samples. ERC de Chavez is the research advisor who helped in the design, sampling, data analysis and writing of the manuscript.

# REFERENCES

- [UNESCO] United Nations Educational, Scientific and Cultural Organization World Heritage Center (2001) UNESCO geoparks programme: A new initiative to promote a global network of geoparks safeguarding and developing selected areas having significant geological features. 30 p.
- Bartsch P. The Philippine land mollusks Cochlyostyla Rufogaster and Obba Marmorata and their races. United States National Museum Bulletin 100, 1932: 329–342.
- Cameron R and Pokryszko BM. Land mollusk faunas of Bialowieza forest Poland, and the character and survival of forest faunas in the North European Plain. J Moll Stud 2004; 70: 149–164.
- Chiba S. Species richness patterns along environmental gradients in island land molluscan fauna. Ecology 2007; 887: 1738–1746.
- Clements R, Sodhi NS, Schilthuizen M, Ng PKL. Limestone karst of Southeast Asia: Imperiled arks of biodiversity. BioScience 2006; 569: 733–742.
- Clements R, Ng PK, Lu XX, Ambu S, Schilthuizen M, Bradshaw CJ. Using biogeographical patterns of endemic land snails to improve conservation planning for limestone karsts. Biological Conservation 2008; 141(11): 2751–2764. Colwell RK. Interpolating, extrapolating, and comparing incidencebased species accumulation curves. Ecology 2006; 85: 2717-2727.
- Day MJ and Urich PB. An assessment of protected karst landscapes in Southeast Asia. Cave and Karst Science 2000; 27: 61–70.

- de Chavez ERC and de Lara AV. Diversity and spatial distribution patterns of macro land snails on Mount Makiling Forest Reserve, Philippines. Asia Life Sciences 2011; 201: 185–201.
- de Winter A.J and Gittenberger E. The land snail fauna of a square kilometer patch of rainforest in southwestern Cameroon: high species richness, low abundance, and seasonal fluctuations. Malacologia 1998; 40: 231–250.
- Department of Environment and Natural Resources (1993) Administrative Order No. 33
- Dove ADM and Cribb TH. Species accumulation curves and their application in parasite ecology. Trends in Parasitology 2006; 22: 568–574.
- Evangelista C (2016) Exploring Rizal's Masungi Georeserve. http://lifestyle.inquirer.net/245409/exploring-rizals-masungigeoreserve (Accessed 30 November 2017).
- Faustino L. Summary of Philippine land shells. Philippine Journal of Science 1930; 48: 85-198.
- Foon JK, Clements GR, Liew T. Diversity and biogeography of land snails Mollusca, Gastropoda in the limestone hills of Perak, Peninsular Malaysia. ZooKeys 2017; 682: 1–94.
- Garcillan PP and Ezcurra E. Biogeographic regions and βdiversity of woody dryland legumes in the Baja California peninsula. J. Veg. Sci. 2003; 14: 859–868.
- Gardenfors U. Effects of artificial liming on land snail populations. J. Appl.Ecol 1992; 29: 50–54.
- Gilli E. Karstology: karsts, caves, and springs: Elements of fundamental and applied karstology. CRC Press, Florida 2011: 244 pp.
- Graveland J, van der Wal R, van Balen JH, van Noordwijk AJ. Poor reproduction in forest passerines from decline of snail abundance on acidified soils. Nature 1994; 368: 446–448.
- Griffiths OL and Herbert DG. New species of land snails Mollusca: Gastropoda from two isolated karst formations in central western Madagascar: TsingyBeanka and Antsingimavo, with additional notes on other regional endemics. African Invertebrates 2013; 54 (1): 1–48.
- Hammer Ø, Harper DAT, Ryan PD. PAST: Paleontological Statistics software package for education and data analysis. Paleontologica Electronica 2016; 4 (1): 1–9.
- Hobbs JJ. Problems in the harvest of edible birds' nests in Sarawak and Sabah, Malaysian Borneo. Biodiversity and Conservation 2004; 13: 2209–2226.
- Lipae HB, Estabillo AL, Fontanilla IKC, de Chavez ERC. A new subspecies of microsnail from Masungi Georeserve, Rizal Philippines. Philippine Journal of Systematic Biology 2020; 14 (3): 1-12.
- Liew T, Clements R, Schilthuizen M. Sampling micromolluscs in tropical forests: one size does not fit all. Zoosymposia 2008; 1: 271–280.
- Mickleburgh S, Waylen K, Racey P. Bats as bush meat: a global review. Oryx 2009; 43: 217–234.

- Nurinsiyah AS, Fauzia H, Hennig C, Hausdorf B. Native and introduced land snail species as ecological indicators in different land use types in Java. Ecological Indicators 2016; 70: 557–565.
- Örstan A, Pearce TA, Welter-Schultes F. Land snail diversity in a threatened limestone district near Istanbul, Turkey. Animal Biodiversity and Conservation 2005; 28 (2): 181–188.
- Raheem DC, Nag GSF, Peerce RC, Mapatuna Y, Kariyawasam L, Eggleton P. Structure and conservation of Sri Lankan landsnail assemblages in fragmented lowland rainforest and village home gardens. Journal of Applied Ecology 2008; 45: 1019–1028.
- Restificar SDF, Day MJ, Urich P. Protection of Karst in the Philippines. Acta Carsologica 2006; 35 (1): 121–130.
- Rickart EA, Balete DS, Rowe RJ, Heaney LR. Mammals of the northern Philippines: tolerance for habitat disturbance and resistance to invasive species in an endemic insular fauna. Diversity and Distributions 2011; 17: 530–541.
- Scheffers BR, Corlett RT, Diesmos A, Laurance WF. Local demand drives a bushmeat industry in a Philippine forest preserve. Tropical Conservation Science 2012; 5: 133–141.
- Schilthuizen M and Rutjes HA. Land snail diversity in a square kilometer of tropical rainforest in Sabah, Malaysian Borneo. Journal of Molluscan Studies 2001; 67 (4): 417–423.
- Schilthuizen M and Vermeulen JJ. The land snails of the Tabin limestone hill. In: Mohamed, M., Schilthuizen, M., Andau, M. (eds.), Tabin Limestone Scientiftc Expedition 2000. UniversitiMalaysia Sabah, 180 pp.
- Schilthuizen M. The evolution and conservation of limestone biotas in Malaysia, with special reference to land snails. Borneo 2000, Environment, Conservation, and Land 2000; 358–362.
- Schilthuizen M. Land snail conservation in Borneo: Limestone outcrops act as Arks. Journal of Conchology, Special Publication 2004; 3: 149–154.
- Schilthuizen M. Life on a block of limestone: Evolutionary, ecological, and geological dynamics of isolated malacofaunas on tropical karst. ScriptaGeologica 2011; 143: p. 27.
- Shackell NL, Fisher JAD, Frank KT, Lawton P. Spatial scale of similarity as an indicator of metacommunity stability in exploited marine systems. Ecological Applications 2012; 221: 336–348.
- Shannon CE and Weaver W. The Mathematical Theory of Communication. Urbana: University of Illinois Press 1949.
- Simpson EH. Measurement of Diversity. Nature 1949; 163, 688.
- Soininen J, Mcdonald R, Hillebrand H. The distance decay of similarity in ecological communities. Ecography 2007; 30: 3–12.
- Springsteen FJ and Leobrera FM. Shells of the Philippines. Carfell Seashell Museum, Phillipines 1986.
- Tanmuangpak K, Dumrongrojwattan P, Tumpeesuwan C, Tumpeesuwan S. Sinoennea loeiensis, a new species of

diapherid microsnail Pulmonata: Streptaxoidea: Diapheridae from PhuPha Lom Limestone Hill, Loei Province, Northeastern Thailand. Raffles Bulletin of Zoology 2015; 63: 293–300.

- Uchida R, Yahya BE, Ahmad AH, Sapaat A, Tomiyama K. Land snail fauna of Sabah, Borneo, Malaysia. Venus 2013; 71 (1-2): 49–59.
- Uy BBV, Cadiz NM, de Chavez ERC. Ecological Patterns of Micro Land Snails along the Altitudinal Gradients of Mount Makiling, Philippines. J. ISSAAS 2018; 24 (2): 30–42.
- Wallisch P, Lusignan ME, Benayoun MD, Baker TI, Dickey AS, Hatsopoulos NG. MATLAB for Neuroscientists. Second edition. Elsevier Inc. 2014, 305–315.

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Sampling Site	Quadrat Code	Geographic position
600 Steps (SS)	SSQ1	14°35'39.35"N 121°18'59.42"E
	SSQ2	14°35'39.52"N 121°19'0.86"E
	SSQ3	14°35'39.96"N 121°19'01.46"E
	SSQ4	14°35'39.18"N 121°19'02.69"E
	SSQ5	14°35'38.81"N 121°19'03.25"E
Sapot (SA)	SAQ1	14°35'52.98"N 121°19'03.43"E
	SAQ2	14°35'56.25"N 121°19'07.24"E
	SAQ3	14°35'52.94"N 121°19'5.91"E
	SAQ4	14°35'53.81"N 121°19'04.62"E
	SAQ5	14°35'57.13"N 121°19'9.59"E
Tatay (TA)	TAQ1	14°35'50.60"N 121°19'15.79"E
	TAQ2	14°35'49.68"N 121°19'15.78"E
	TAQ3	14°35'49.49"N 121°19'14.83"E
	TAQ4	14°35'48.37"N 121°19'15.75"E
	TAQ5	14°35'47.86"N 121°19'14.71"E
Nanay (NA)	NAQ1	14°35'46.92"N 121°19'21.03"E
	NAQ2	14°35'46.45"N 121°19'20.24"E
	NAQ3	14°35'46.22"N 121°19'20.55"E
	NAQ4	14°35'46.99"N 121°19'18.98"E
	NAQ5	14°35'45.48"N 121°19'19.84"E
Liwasan (LW)	LWQ1	14°35'57.03"N 121°19'13.94"E
	LWQ2	14°35'57.69"N 121°19'14.98"E

 Supplementary Table 1: Coordinates of the quadrats established for each of the sampling sites.

 Sampling Site
 Quadrat Code
 Geographic position

	LWQ3	14°35'54.4"N 121°19'15.15"E
	LWQ4	14°35'55.87"N 121°19'15.68"E
	LWQ5	14°35'56.17"N 121°19'16.37"E
Masungi Entrance (EN)	ENQ1	14°36'02.7"N 121°19'09.87"E
	ENQ2	14°36'03.91"N 121°19'09.45"E
	ENQ3	14°36'05.44"N 121°19'09.33"E
	ENQ4	14°36'08.33"N 121°19'10.15"E
	ENQ5	14°36'09.07"N 121°19'10.41"E
Garden Cottages (GC)	GCQ1	14°36'18.96"N 121°18'55.58"E
	GCQ2	14°36'18.41"N 121°18'55.08"E
	GCQ3	14°36'17.45"N 121°18'55.47"E
	GCQ4	14°36'14.77"N 121°18'56.43"E
	GCQ5	14°36'14.12"N 121°18'52.48"E
Mahogany Stand (MS)	MSQ1	14°36'07.48"N 121°18'39.91"E
	MSQ2	14°36'06.68"N 121°18'39.33"E
	MSQ3	14°36'06.17"N 121°18'39.81"E
	MSQ4	14°36'05.88"N 121°18'39.45"E
	MSQ5	14°36'05.34"N 121°18'39.73"E
Water Cascades (WC)	WCQ1	14°35'58.27"N 121°18'56.93"E
	WCQ2	14°35'59.13"N 121°18'56.64"E
	WCQ3	14°35'59.59"N 121°18'56.97"E
	WCQ4	14°35'59.53"N 121°18'57.96"E
	WCQ5	14°35'59.14"N 121°18'58.22"E

Stream (ST)	STQ1	14°35'52.93"N 121°18'44.85"E
	STQ2	14°35'52.73"N 121°18'45.38"E
	STQ3	14°35'52.85"N 121°18'44.09"E
	STQ4	14°35'52.67"N 121°18'43.62"E
	STQ5	14°35'53.19"N 121°18'42.68"E
Silayan (SI)	SIQ1	14°36'19.95"N 121°18'31.96"E
	SIQ2	14°36'20.32"N 121°18'32.48"E
	SIQ3	14°36'20.65"N 121°18'31.54"E
	SIQ4	14°36'21.61"N 121°18'31.20"E
	SIQ5	14°36'22.20"N 121°18'30.49"E
Masungi Chapel (MC)	MCQ1	14°36'25.41"N 121°18'28.84"E
	MCQ2	14°36'27.62"N 121°18'29.98"E
	MCQ3	14°36'29.83"N 121°18'32.33"E
	MCQ4	14°36'32.03"N 121°18'34.68"E
	MCQ5	14°36'32.80"N 121°18'34.26"E

Supplementary Table 2: Characteristics of each of the karst sampling sites (SS-600 steps; SA-Sapot; TA-Tatay; NA-Nanay; LW-Liwasan; EN-Masungi Entrance). Values were averaged from the quadrats of each sampling site.

Characteristics	Sampling Sites					
	SS	LW	ТА	NA	SA	EN
Altitude (masl)	604.6	508.2	532	546.4	562.6	540.4
Number of trees	5	4	5	5	6	3
Tree diameter at breast height (cm)	59.04	32.75	31.81	23.37	21.08	47.03
Canopy Cover (%)						
Wet season	77.5	88.33	89.17	85.83	84.16	83.33
Dry season	74.17	85.6	86.93	81.87	81.33	80.77
Leaf Litter Depth (cm)						
Wet season	6.9	5.76	5.15	3.9	5.68	7.34

Dry season	4.88	3.13	3.36	1.53	4.3	2.28
Temperature (°C)						
Wet season	24.68	24.42	24.86	22.88	24.02	27.2
Dry season	30.84	28.82	29.14	30.54	31.16	26.88
Relative humidity (%)						
Wet season	99.8	100	100	95.5	94.8	88.8
Dry season	56.58	74.1	72.68	71.12	62.62	78.34
Soil moisture						
Wet season	6.72	6.24	8.12	4.82	7.68	6.92
Dry season	3.12	3.4	3.04	3.24	3.72	3.92
Soil calcium (cmolc/kg soil)	28.3	26.35	25.7	29.95	27.94	27.80

Supplementary Table 3: Characteristics of each of the non-karst sampling sites (GC-Garden Cottages; MS-Mahogany Stand; WC-Water Cascades; ST-Stream; SI-Silayan; CH-Chapel). Values were averaged from the quadrats of each sampling site.

Characteristics	Sampling Sites						
	GC	MS	WC	ST	SI	СН	
Altitude (masl)	447.4	530.2	426.2	402.6	335.8	339	
Number of trees	5	6	4	4	5	5	
Tree diameter at breast height (cm)	61.67	40.55	44.54	29.90	53.65	45.74	
Canopy Cover (%)							
Wet season	81.67	83.33	91.67	81.67	82.5	86.67	
Dry season	78.6	80.03	88.93	78.6	86.47	82	
Leaf Litter Depth (cm)							
Wet season	4.16	6.52	5.43	4.1	5.34	4.76	
Dry season	2.97	4.05	3	2.63	2.86	2.37	
Temperature (°C)							
Wet season	26.7	27.64	26.24	26.12	27.17	26.18	
Dry season	27.86	29.54	28.54	27.14	28.28	28.28	
Relative humidity (%)							
Wet season	95.74	85.74	90.98	94.54	90.74	92.76	
Dry season	75.88	74.82	76.32	77.22	74.7	75.36	
Soil moisture							
Wet season	6.92	6.4	6.04	6.24	6.7	6.14	
Dry season	3.44	3.76	5.64	4.4	2.76	3.52	
Soil calcium (cmolc/kg soil)	5.15	4.63	16.21	13.89	7.87	6.24	

# Supplementary Table 4: Accession number for each of the land snail species found in the Masungi Georeserve.

Allopeas sp. 1	
mopeus sp. 1	UPLBMNHLS 062
Allopeas sp. 2	UPLBMNHLS 06
Allopeas sp. 3	UPLBMNHLS 06
Lissachatina fulica (Ferussac, 1821)	UPLBMNHLS 06
Macrochlamys indica (Benson, 1832)	UPLBMNHLS 06
Calocochlia sp. 2	UPLBMNHLS 06
Macrochlamys sp. 2	UPLBMNHLS 06
Microcystina sp. 1	UPLBMNHLS 06
Microcystina sp. 2	UPLBMNHLS 07
Bradybaena fodiens (Ferussac, 1822)	UPLBMNHLS 07
Calocochlia sp. 1	UPLBMNHLS 07
Chlorea dryope (Broderip, 1841)	UPLBMNHLS 07
Chlorea fibula (Reeve, 1842)	UPLBMNHLS 07
Helicostyla carinata (Lea, 1840)	UPLBMNHLS 07
Helicostyla metaformis (Ferussac, 1821)	UPLBMNHLS 07
Helicostyla rufogaster (Lesson, 1831)	UPLBMNHLS 07
Helicostyla woodiana (Lea, 1840)	UPLBMNHLS 07
Obba lasallii (Eydoux, 1838)	UPLBMNHLS 07
Obba listeri (Gray, 1825)	UPLBMNHLS 08
Obba morongensis (von Moellendorff, 1890)	UPLBMNHLS 08
Obba marmorata (von Moellendorff, 1890)	UPLBMNHLS 08
Kaliella sp. 1	UPLBMNHLS 08
Leptopoma sp.	UPLBMNHLS 08
Hemiglypta cuveriana (Lea, 1852)	UPLBMNHLS 08
Hemiglypta semiglobosa (Pfieffer, 1854)	UPLBMNHLS 08
Hemitrichiella setigera (Sowerby, 1898)	UPLBMNHLS 08
Ryssota nigrescens (von Moellendorff, 1888)	UPLBMNHLS 08
Cyclophorus appendiculatus (Pfieffer, 1854)	UPLBMNHLS 08
Cyclophorus reevei (Hidalgo, 1890)	UPLBMNHLS 09
Cyclophorus sp.	UPLBMNHLS 09
Leptopoma helicoides (Grateloup, 1840)	UPLBMNHLS 09
Leptopoma perlucidum (Grateloup, 1886)	UPLBMNHLS 09
Endodonta sp.	UPLBMNHLS 09

Helicarion sp.	UPLBMNHLS 095
Acmella mp. 1	UPLBMNHLS 096
Acmella mp. 2	UPLBMNHLS 097
Georissa mp.	UPLBMNHLS 098
Hypselostoma latispira masungiensis (Lipae & de Chavez, 2020)	UPLBMNHLS 099
Moulinsia sp.	UPLBMNHLS 100
Geotrochus mp. 1	UPLBMNHLS 101
Geotrochus mp. 2	UPLBMNHLS 102
Nana sp.	UPLBMNHLS 103
Videna metcalfei (Pfieffer, 1845)	UPLBMNHLS 104
Laevicaulis alte (Ferussac, 1822)	UPLBMNHLS 105
Sarasinula plebeian (P. Fischer, 1868)	UPLBMNHLS 106