

Journal of Biodiversity and Environmental Sciences (JBES) ISSN: 2220-6663 (Print) 2222-3045 (Online) Vol. 25, No. 2, p. 20-30, 2024 http://www.innspub.net

RESEARCH PAPER

OPEN ACCESS

Diversity and microhabitat preferences of anuran species in the secondary forest and riverine areas in Tangub City, Misamis Occidental, Philippines

Ruth C. Maminta¹, Jesha Mea P. Sagario¹, Monaliza Joy Zaragoza-Magsayo²

¹College of Mathematics and Natural Sciences, Northwestern Mindanao State College of Science and Technology, Labuyo, Tangub City, Misamis Occidental, Philippines

²College of Agriculture and Environmental Studies, Northwestern Mindanao State College of Science and Technology, Labuyo, Tangub City, Misamis Occidental, Philippines

Article published on August 04, 2024

Key words: Anurans, Diversity, Microhabitat, Forest, Riverine

Abstract

The research study focused on the diversity and microhabitat preferences of anuran species in secondary forests and riverine areas in Tangub City, Misamis Occidental. It was particularly conducted in three different elevations (upper, mid, and lower) at Barangay Manga and Barangay Hoyohoy. Visual Encounter Survey and Capture-mark-release methods were used. Fifteen (15) species belonging to six (6) families were identified from 276 individuals captured, namely: Ansonia mcgregori, Ansonia muelleri, Ingerophrynus philippinicus, Rhinella marina, Fejevarya vittigera, Hoplobatrachus rugulosus, Limnonectes leytensis, Limnonectes magnus, Kalophrynus sinensis, Kaloula pulchra, Leptobrachium lumadorum, Megophrys stejnegeri, Leptomantis bimaculatos, Polypedates leucomystax, and Pulchrana grandocula, of which eleven species are endemics and four species are non-endemics. Both study areas attained moderate species diversity, however Barangay Hoyohoy showed a higher similarity index among all sampling sites. Most anuran species encountered preferred aquatic microhabitats, with Pulchrana grandocula as the most abundant species (69.99%) observed. Relative humidity as an environmental factor influenced its abundance in each sampling area. Primary threats such as human disturbances and habitat destruction were observed in Barangay Manga. Thus, researchers recommend conservation strategies and enforcement of policies to protect the study areas as well as anurans assemblage.

*Corresponding Author: Monaliza Joy Zaragoza-Magsayo 🖂 monalizajoy.zaragoza@nmsc.edu.ph

Introduction

Anurans are crucial in the food web, particularly for insect control, and serve as natural bioindicators of ecosystem health (Mohagan et al., 2018). Their sensitivity to environmental changes makes them valuable indicators of ecological integrity and watershed health. The Philippine archipelago is one of the most mega-diverse countries in the world, hosting a large number of endemic plant and animal species, particularly in its rainforests (Lavides et al., 2004). Mindanao, situated in the southern region of the Philippines, is considered home to various endemic species (Diesmos et al., 2015). According to Solania and Fernandez-Gamalinda (2018), there were 141 individuals of anurans recorded, of which 73% were Philippine endemics, and 36% were Mindanao faunal endemics. However, a large portion of Mindanao remains unexplored (Dela Torre and Nuñeza, 2021), and many new species are still being described (Siler et al., 2009).

Anurans utilize both terrestrial and aquatic microhabitats, exhibiting overlap in microhabitat preferences (Plaza and Sanguila, 2015). Anurans are most likely to seek cooler microhabitats with lower temperatures and higher relative humidity (Aureo and Bande, 2019). Their dependence on moist conditions and high humidity means that amphibian diversity is highest in regions with high precipitation and lower evaporative water loss (West, 2018). For decades, amphibian populations faced decline due to habitat destruction, overexploitation, quarrying, land conversion, and alteration of habitat that favors invasive species, which threaten native amphibian species (Mapi-ot et al., 2015). Currently, more than 150,300 species are recorded according to the IUCN Red List (2023), with over 42,100 species considered threatened with extinction, including 41% of amphibian species. Thus, the presence of potentially threatened anurans implies a need for conservation measures for species protection.

Research beyond Mt. Malindang to lowland areas, where anuran populations are more vulnerable to disturbances, is needed (Nuneza *et al.*, 2010). Tangub

City, sheltered by Mt. Malindang and Panguil Bay, faces threats to its anuran habitats due to human population growth and deforestation (Nuneza et al., 2010). Barangay Hoyohoy shows potential as a tourist destination, while Barangay Manga is crucial for irrigation and water quality maintenance. Understanding anurans in these areas is essential for ecosystem health and conservation. To bridge this gap, researchers undertook a comprehensive study aimed to identify the diversity and microhabitat preferences of anuran species in secondary forest and riverine areas of Tangub City, Misamis Occidental. The findings serve as baseline data for policymakers improve conservation and to management efforts and highlight local awareness as to their role in maintaining anuran diversity.

Materials and methods

Study area

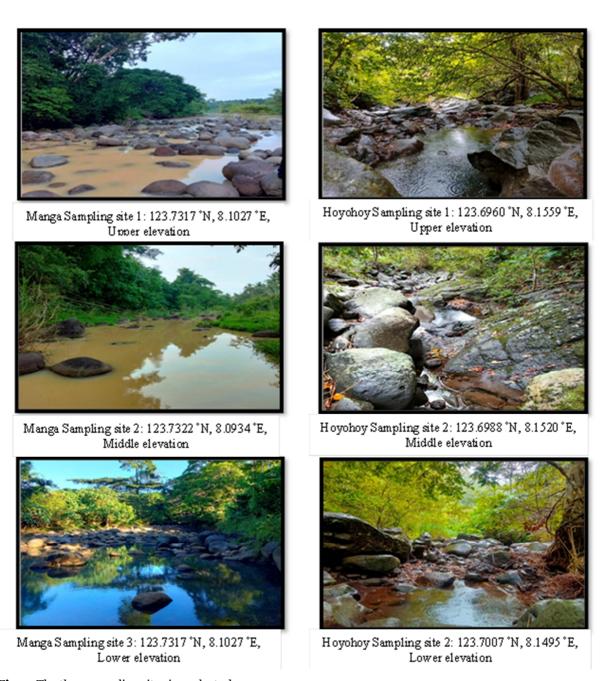
Tangub is a City in Misamis Occidental, Philippines that is situated approximately within 8° 4' North latitude and 123° 45' East longitude. The Panguil Bay Bridge, the longest bridge in Mindanao, faces Tangub City in the South, protected by the majestic Mt. Malindang in the North, bordered by Ozamis City in the East, and the municipality of Bonifacio in the West. Two Barangays were selected as sampling areas, Barangay Manga and Barangay Hoyohoy which lies at 8° 5' North Latitude, 123° 44' East Longitude with an estimated terrain elevation of 98 masl, and lies at 8° 9' North Latitude, 123° 42' East Longitude with an elevation of 735 masl, respectively. The secondary forest and riverine areas of Barangay Manga were surveyed examining three different elevations (Fig. 1).

Anuran sampling

The sampling period was conducted from November 2023 – January 2024 examining the high, middle, and low elevations of Barangay Manga and Hoyohoy (Fig. 2). A total of 360-man hours was allotted for each sampling site. Visual encounter survey (VES) and capture-mark-release methods (Cortico *et al.*, 2018) were employed. VES was used to capture anurans with the use of hands from 18:00 to 21:00.

Microhabitat preferences of anurans were also noted. The microhabitats were classified into three types: Type I – Arboreal microhabitat refers to anurans observed from the ground (5-10 m) which includes branches and stems of trees/plants, leaves and leaf axils; Type II – Ground microhabitat if anurans seen directly on the ground (0-5 m) or on rotting logs and tree buttresses; Type III – Aquatic microhabitat if anurans were found on streams, rivers, creeks, and standing bodies of water (Ates and Delima, 2008;

Toledo-Bruno *et al.*, 2017; Vidal *et al.*, 2018). Morphometric measurements were performed using a vernier caliper followed by Dela Torre and Nuñeza (2021) measurements. A Wildlife Gratuitous Permit was secured with permit number R10-2024-35. The identification and verification of species was referred to the works of Diesmos *et al.* (2015) and with the help of an expert. A list of endemic and threatened species was generated based on published data and IUCN (2023) criteria.



 ${\bf Fig.}\ {\bf 1.}$ The three sampling sites in each study area



Fig. 2. Anuran speices found in the sampling areas. Legend: A- Ansonia mcgregori, B- Ansonia muelleri, C-Ingerophrynus pilippinicus, D- Rhinella marina, E- Fejevarya vittigera, F- Hoplobatrachus rugulosus, G-Limnonectes leytensis, H-Limnonectes magnus, I-Kalophrynus sinensis, J-Kaloula pulchra, K-Leptobrachium lumadorum, L- Megophrys stejnegeri, M- Leptomantis bimaculatus, N- Polypedates leucomystax, O- Pulchrana gradocula

Determination of environmental parameters

The environmental parameters were determined in each sampling area across elevation gradients. In describing the general conditions of each sampling site, water, air and soil temperature, relative humidity and soil pH were frequently recorded. Instrument used were field thermometer, sling psychrometer and a 5-in-1 water quality tester, respectively.

Data analysis

Data were analyzed the gathered using Paleontological Statistics (PAST) Software version 4.03. The study used the seriation analysis to determine the anuran species composition between sampling areas and among sampling sites while, diversity indices such as Shannon-Weiner, evenness and dominance were measured to determine species richness, variation in the abundance per species and uniformity of species within the community. Bray-Curtis index was further used to calculate the similarities in species among sampling sites in the two study areas. And Canonical Correspondence Analysis (CCA) was performed to examine the significantly influential environmental parameters to anuran species abundance.

Results and discussion

Species composition

A total of 15 anuran species were found in the two study areas (Table 1). Of these, 9 anuran species were found in Barangay Manga, however, these species were not collected across different elevations as shown in Fig. 3. For instance, in the upper elevation, 7 species were documented including *Ingerophrynus* philippinicus, Rhinella marina, Hoplobatrachus rugulosus, Limnonectes magnus, Kaloula pulchra, **Polypedates** leucomystax, and Pulchrana grandocula. While, in the middle elevation, 5 species were identified, 4 of which are similar to the species found at the upper elevation (R. marina, H. rugulosus, K. pulchra, and P. leucomystax) along with Limnonectes leytensis. Moreover, 5 anuran species were also collected in the lower elevation with 4 similar species from the upper and middle elevation (R. marina, H. rugulosus, K. pulchra, and P.

leucomystax) and an additional 1 different species, Fejevarya vittigera.

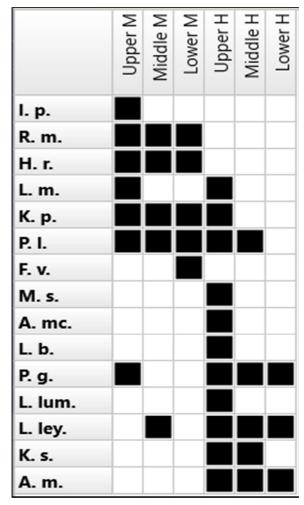


Fig. 3. The Serration Analysis shows the species composition of anuran species in each sampling area and among the sampling site. Legend: I. p.= Ingerophrynus philippinicus, R. m.= Rhinella marina, H. r.= Hoplobatrachus rugulosus, L. m.= Limnonectes magnus, K. p.= Kaloula pulchra, P. l.= Polypedates leucomystax, F. v.= Fejevarya vittigera, M. s.= Megophrys stejnegeri, A.mc.= Ansonia mcgregori, L. b.= Leptomantis bimaculatos, P. g.= Pulchrana grandocula, L. lum= Leptobrachium lomadurom, L. ley= Limnonectes leytensis, K. S.= Kalophrynus sinensis, A.m.= Ansonia muelleri. Upper Manga (Upper M), Middle Manga (Middle M), Lower Manga (Low M), Upper Hoyohoy (Upper H), Middle Hoyohoy (Middle H), and Lower Hoyohoy (Low H). Shaded boxes (a) indicate the presence of anuran species while empty boxes (a) denote the absence of anurans.

Table 1. List of Anuran Species, their Ecological status, Conservation status, and Distribution range of Barangay Manga and Barangay Hoyohoy

Species	Family	Common name	Local name	Ecological Status*	Conservatio status**	nDistribution range**
Ansonia mcgregori		Mueller's Toad	Baki sa bato	ME	LC	Mindanao
Ansonia muelleri Ingerophrynus philippinicus Rhinella marina Fejevarya vittigera	Bufonidae	McGregor's toad Philippine toad Cane toad Luzon wart frog, Philippine grass frog	Baki sa bato	ME	LC	Dinagat; Mindanao
			Camprag	PE	LC	Balabac; Busuanga; Culion; Dumasan; and Palawan
			Camprag	NE	LC	Throughout the Philippines
			Bak-bak	PE	LC	Throughout the Philippines
Hoplobatrachus rugulosus Limnonectes		Chinese edible frog Small disked	Bak-bak	NE	LC	Caluya; Luzon; Mindanao; and Panay
leytensis	Dicroglossidae		Bak-bak	PE	LC	Throughout the Philippines
Limnonectes magnus		Giant Philippine Frog Mindanao fanged frog	Bak-bak	PE	NT	Basilan; Biliran; Bohol; Camiguin Sur; Dinagat; Leyte; Mindanao: and Samar
Kalophrynus sinensis	Microhylidae	Philippine sticky frog	Baki/ Bolprag	ME	LC	Basilan, Bohol, Camiguin Sur, Culion, Dinagat, Leyte, Mindanao, and Samar
Kaloula pulchra		Asian painted frog Mindanao litter frog Mindanao horned frog	Bullfrog	NE	LC	Cebu; Luzon; Mindanao; and Palawan
Leptobrachium lumadorum Megophrys stejnegeri	Megophryidae		Baki	ME		Dinagat; Mindanao; and Basilan
			Baki	ME	LC	Dinagat; Mindanao; and Basilan
Leptomantis bimaculatos Polypedates leucomystax	Rhacophoridae	Asiatic tree frog	Baki	PE	LC	Southern Luzon, Bohol, and Mindanao
		frog	Bigwak	NE	LC	Throughout the Philippines
Pulchrana grandocula	Ranidae	Big-eyed frog	Baki	ME	LC	Mindanao

*Ecological Status: Alcala and Brown (1998); Diesmos *et al.* (2015); Gamolo (2023) **Conservation Status: IUCN, 2023 LC= Least Concern, NT=Near Threatened, ME= Mindanao Endemic, PE= Philippine Endemic, NE=Non-endemic

Whereas, Barangay Hoyohoy with an average elevation of 735 masl, there were 11 species recorded. These species were all found at the upper elevation (Limnonectes magnus, Kaloula pulchra, Polypedates leucomystax, Megophrys stejnegeri, Ansonia mcgregori, Leptomantis bimaculatos, Pulchrana grandocula, Leptobrachium lumadorum, Limnonectes leytensis, Kalophrynus sinensis, and Ansonia muelleri). Meanwhile, only 5 species were found in the middle elevation comprising P. leucomystax, P. grandocula, L. leytensis, K. sinensis, and A. muelleri which are all

observed at the upper elevation. And 3 species were collected in the lower elevation which is identical to the species obtained in the upper and middle elevations, *P. grandocula*, *L. leytensis*, and *A. muelleri*. Out of the 15 total recorded species, 5 were identified as common to both secondary forest and riverine areas in the two study locations including *L. magnus*, *K. pulchra*, *P. leucomystax*, *P. grandocula*, and *L. leytensis*.

As observed in the study, the number of species decreases as the elevation decreases. This finding

suggests a consistent decrease in the species count across all sampling sites of both selected study areas. This observation was supported by different studies, for instance, species such as A. mcgregori only thrive at higher elevations (Cortico et al., 2018), and M. stejnegeri species, which had a particular microhabitat preference that was only observed in streams and ground at high elevation (Solania and Fernandez-Gamalinda, 2018). On the contrary, the species *F. vittigera* was noted by Delima-Baron *et al.* (2022) as one of the species that can thrive well in low-elevation, agricultural areas, and highly disturbed habitats. Results from all the sampling sites can be associated with the findings of Dela Torre and Nuñeza (2021), who observed higher anuran species richness at high elevations.

Table 2. Species diversity in the two sampling areas, Barangay Manga and Barangay Hoyohoy Tangub City, Misamis Occidental

	Barangay Manga	Barangay Hoyohoy	
No. of species	9	11	
Number of individuals	131	145	
Shannon-Weiner diversity	1.804	1.164	
Evenness	0.675	0.2912	
Dominance	0.194	0.5066	

Diversity indices of anuran species

The diversity of anuran species between the two study areas slightly varied as shown in Table 2, Barangay Manga showed moderate species diversity (H'=1.804), indicating an average diverse species composition. Similarly, Barangay Hoyohoy exhibited moderate species diversity with a calculated value of H'=1.164. This area is located in an upland area, and 11 out of 15 species were found. However, having a high number of captured species does not guarantee having high species diversity. This is primarily due to the dominance of Pulchrana grandocula, with a total of 101 individuals found. This highlights the high abundance of P. grandocula across all elevation sites ranging from 592 to 735 masl. Notably, P. grandocula exhibits a wide distribution, spanning elevations from 60 to 570 masl (Decena et al., 2023), and has been observed in a flowing stream within a secondarygrowth forest at approximately 700 masl (Venturina

et al., 2020). P. grandocula was observed along the streams, perching on rocks and logs, predominantly favouring an aquatic microhabitat. As reported by Aureo and Bande (2019), P. grandocula is a species dependent on forests or streams. This infers that its abundance, particularly in the middle elevation site, was primarily what they mostly inhabit. The evenness values differ as well, primarily due to the variation in the abundance of individuals per species within the community of anurans accounted in each study area. Barangay Manga is categorized as having unstable community of anurans (E = 0.675), indicating there is an uneven distribution in the number of individuals between species. The presence and abundance of invasive species such as H. rugulosus, R. marina, and K. pulchra contributed to this community of anurans to become unstable. Conversely, in Barangay Hoyohoy, the evenness index (E = 0.2912) falls into the category of a depressed community, suggesting a less balanced distribution of individuals between species. During the sampling, it was observed that the area was less disturbed by humans due to its dangerous terrain. Nevertheless, the evenness of species was higher in Barangay Manga than in Barangay Hoyohoy. These significant differences between the two study areas were probably due to the apparent dominance of P. grandocula (82.5%). Furthermore, Barangay Manga indicates a lower level of dominance (D = 0.194) of certain species against others. Generally, dominance arises when one or several species assert control over the environment and its conditions, thereby impacting the associated species. On the other hand, Barangay Hoyohoy falls into the category of moderate dominance (D = 0.5066), suggesting a moderate level of dominance among species. However, this result contradicts the observation made by Nuñeza et al. (2017), as their study reported a dominance index value of 0.5102, signifying the highest dominance index and suggesting the presence of a dominant anuran species. Thus, this explains that a more or less distribution of anuran species is likely influenced by certain parameters such as elevation gradients, microhabitat preferences, and availability of water and food (e.g. insects) (Ngilangil et al., 2014).

Table 3. Microhabitat preferences of anuran species in the secondary forest and riverine areas of Barangay Manga and Barangay Hoyohoy in all sampling sites

Species	Family	Barangay Manga			Barangay Hoyohoy		
		Upper	Mid	Low	Upper	Mid	Low
Ansonia mcgregori					Aq		
Ansonia muelleri	Bufonidae				Aq	Aq,	Aq
Ingerophrynus philippinicus	Bulomdae	Aq, G					
Rhinella marina		Aq, G	Aq, G	G			
Fejevarya vittigera				Aq			
Hoplobatrachus rugulosus	Dicroglossidae	Aq, G	Aq	G			
Limnonectes leytensis	Dicrogiossidae	_	Aq		Aq	Aq	Aq
Limnonectes magnus		Aq	_		Aq	_	-
Kalophrynus sinensis	Mianahylidaa				G		
Kaloula pulchra	Microhylidae	G	G	G	G		
Leptobrachium lumadorum	Megophryidae				G		
Megophrys stejnegeri					G		
Leptomantis bimaculatos	D1 1 11				A		
Polypedates leucomystax	Rhacophoridae	A, G	A	A, Aq	A	A	
Pulchrana grandocula	Ranidae	Áq		, 1	Aq	Aq	Aq
Total no. of species		7	5	5	11	4	3

A= Type-I Arboreal Microhabitat, G= Type II- Ground Microhabitat, Aq= Type-III Aquatic Microhabitat

Microhabitat preferences of anuran species

All microhabitat preferences of anurans were represented across all sampling sites. Most encountered species were recorded in Type III aquatic microhabitats, referring to areas near or within bodies of water such as slow-moving streams, rivers, wallows, swamps, and ponds. Of 15 captured species, 3 species (*I. philippinicus, R. marina*, and *H. rugulosus*) inhabit both type II and III microhabitats. Additionally, only one species, *P. leucomystax*, was observed to inhabit all microhabitats but this species mostly preferred arboreal microhabitats (Table 3).

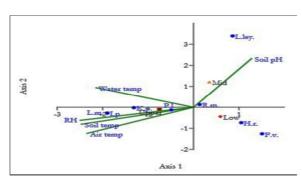


Fig. 4. Canonical Correspondence Analysis shows the association of anuran species in relation to environmental parameters in the three sampling sites of Barangay Manga. Legends for anuran species: L.m. (*Limnonectes magnus*), I.p. (*Ingerophrynus philippinicus*), K.p. (*Kaloula pulchra*), P.l. (*Polypedates leucomystax*), R.m. (*Rhinella marina*),

L.ley. (*Limnonectes leytensis*), H.r. (*Hoplobatrachus rugulosus*), F.v. (*Fejevarya vittigera*). Legends for 5 environmental parameters: A.temp (Air temperature), S.temp (Soil temperature), RH (Relative Humidity), SpH (Soil pH), and W.temp (Water temperature)

Environmental parameters and anuran species association

Among the 9 identified species, distinct preferences for environmental conditions were observed in Barangay Manga (Fig. 4). In the upper elevation site (located on the left lower side of the central axis), species such as L. magnus, I. philippinicus, K. pulchra, and P. leucomystax were observed highly associated with increasing values of air, soil temperature, and relative humidity. This means that these species thrive well in these specific environmental parameters. These findings concur with the results of Aureo and Decena (2022), particularly for P. leucomystax, identified as a disturbance-tolerant species, implying that this species was found in areas with increasing soil temperature. Similarly, P. leucomystax was found to have a significantly strong positive association with relative humidity (Simon et al., 2022). In the middle elevation, reflected on the right upper side of the central axis, a high value of soil pH (7.46) was identified as presumably a favourable environmental

parameter for the Philippine endemic species L. leutensis and the invasive species, R. marina. L. leytensis were commonly found in slow-moving streams or swamps as observed during sampling. The presence of these two species in mid-elevation denotes that they can survive in riverine areas such as slow-moving streams, stagnant water bodies such as ponds or swamps, and with moderate canopy cover. Moreover, the species H. rugulosus and F. vittigera were noted in the lower elevation site, as evident on the right lower side of the central axis, indicating that these two species flourish in disturbed habitats without any dominating environmental factors. And as depicted on the left upper side of the central axis, no anuran species were found to favor an increase in water temperature.

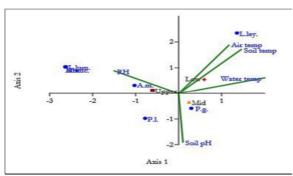


Fig. 5. Canonical Correspondence Analysis shows the association of anuran species in relation to environmental parameters in the three sampling sites of Barangay Hoyohoy. Legends for anuran species: L.m. (Limnonectes magnus), I.p. (Ingerophrynus philippinicus), K.p. (Kaloula pulchra), (Polypedates leucomystax), R.m. (Rhinella marina), L.ley. (Limnonectes leytensis), H.r. (Hoplobatrachus rugulosus), F.v. (Fejevarya vittigera). Legends for 5 environmental parameters: A.temp (Air temperature), S.temp (Soil temperature), (Relative Humidity), SpH (Soil pH), and W.temp (Water temperature)

On the other hand, in Hoyohoy, species such as *A. muelleri*, *A. mcgregori*, *L. lumadorum*, *L. bimaculatos*, and *K. sinensis* were found in upper elevation site located left upper side of the central axis (Fig. 5). These species were closely associated with high relative humidity environmental factor.

Additionally, these species likely inhabit lowtemperature and moist environments with close canopy cover, forest duffs, leaf litters, and shrubs surrounded by the presence of various plant and tree species. The riverine areas (streams) were described as having pristine water with huge rocks mostly covered with mosses and ferns. Meanwhile, only one species P. grandocula was observed favoring the soil pH in the middle elevation situated in the right lower side of the central axis. P. grandocula species were reported to be widely distributed in dipterocarp forests, favoring increasing relative humidity, elevation, canopy cover, litter depth, as well as soil pH (Aureo and Decena, 2022). Further, the presence of L. leytensis in the lower elevation located in the upper right of the central axis suggest a preference in an area with increasing air, water, and soil temperatures. Interestingly, the species P. leucomystax has no preferred environmental parameters, inferring that this anuran can thrive in any conditions with high tolerance of varying undisturbed and disturbed areas Supsup et al. (2016), such as in agricultural areas (Sanguila et al., 2016).

Conclusion

The trend of anuran species richness has been recorded in this study to decrease as elevation gradients decrease, and vice versa. The decreasing species richness primarily occurs in lowland areas which are greatly influenced by human disturbances. Both study areas exhibited moderate species diversity. For instance, Barangay Manga obtained a moderate diversity mainly because it was situated in a lowland area or elevation where it was classified as a residential area with prominent human disturbances such as hunting of frogs as a food source, altered habitat, and land conversion. Aside from human disturbances, Barangay Hoyohoy has a different case given that it was situated in an upland area or highly elevated area, hence described as undisturbed. The result of having moderate species diversity was primarily due to the dominance of Pulchrana grandocula among all species identified. Human disturbances such as frog hunting and habitat alteration greatly affect anuran species diversity.

Thus, minimizing further habitat destruction, conservation efforts in both study areas should be implemented.

Acknowledgments

The authors acknowledge the DENR Region 10 for the issuance of a Gratuitous Permit (GP), and Mr. Ralphinno G. Sanchez for the verification of the identification of the anuran species.

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