

### **Research Article**

## Herpetofaunal Assemblages in the Lowland Regions of Sumatera Barat

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#### ABSTRACT

The habitat destruction and land-use changes caused the decline of animal composition in many tropical regions. Here, we study the diversity of herpetofauna in the lowland areas in Sumatera Barat, a midwestern province in Sumatera island, using a visual encounter survey method. The surveyed habitat included rubber plantations, streams, paddy fields, and peat swamps. We observed 338 individuals representing 44 species from 14 families of herpetofauna with almost 90% individuals were amphibians. Overall, the rubber plantations contained a higher number of species than other types of habitat. For amphibians, Ranidae and Dicroglossidae represented the first and the second highest both in the species and individual number. For reptiles, Agamidae and Colubridae or Gekkonidae accounted for the first and the second highest in the individual number while Colubridae and Scincidae consisted of the highest species number. Our data showed that the diversity index was mostly in moderate level except in paddy field. The species composition in rubber plantations were more similar to those of streams rather than paddy field or peat swamp Sago habitat.

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#### **INTRODUCTION**

Most of the decline of fauna diversity occured due to loss of habitat, habitat destruction, and habitat change (Stuart et al. 2004; Todd & Rothermel 2006). Those damages are mainly caused by the conversion of forests to areas for plantations. Indonesia is known to be the second largest rubber exporting country globally (Bruinsma & Food and Agriculture Organization of the United Nations 2003). In exchange, Indonesia lost many more primary forest land than Brazil (Margono et al. 2014). Among the five biggest islands in Indonesia, Sumatera is one of those experiencing the highest loss of primary forests (Gunarso et al. 2013). The conversion of forest land to plantation has raised many animal diversity problems. Changes in the vegetation structure and human activities lead to the loss of many intolerant disturbance species

(Drescher et al. 2016). The conversions also negatively impacted the species richness, abundance, and community structure (Fitzherbert et al. 2008).

Amphibians are the most endangered group of vertebrates by the habitat loss and overutilization (Stuart et al. 2004). On the other hand, reptiles, in the conservation perspective, also receive a little attention on how they response to agricultural activities (Tews et al. 2004). Herpetofauna communities are affected by changes in vegetation for example, from primary forest to plantation (Paoletti et al. 2018). Some anthropogenic activities like oil-palm replanting (Kurz et al. 2016), converting forest into plantation (Konopik et al. 2015) have been known decreasing the number of species, the richness, and abundance of frog communities. Specifically, leaf litter thickness and canopy cover have strongly determined the species richness and abundance (Whitfield & Pierce 2005; Wanger et al. 2009).

Several works partly or fully involving Sumatera Barat as the study site were reported (e.g. Inger & Iskandar 2005; Kurniati 2008; Teynie et al. 2010; Wostl et al. 2017). However, those studies mainly focused on biodiversity discoveries and conservation areas that are administratively regulated by national or local governments. Nonetheless, non-protected areas could not be neglected in terms of amphibians and reptiles conservation because they might contain more diverse and more abundant herpetofauna (Whitfield & Pierce 2005; Luja et al. 2017). Even though the herpetofauna inventory and community assessment outside the protected areas in Sumatera Barat are scarce, such studies seem to have already started to grow (Sumarmin et al. 2019; Nugraha et al. 2021).

This study aimed to analyze the diversity of amphibians and reptiles communities in the lowland regions of Sumatera Barat province. We chose lowland region because many areas are vulnerable to anthropogenic activities (Gunarso et al. 2013), likely impacting the structure of herpetofauna communities.

#### MATERIALS AND METHODS

#### The study sites

We chose four regions in this study that were located under 400 meters above sea level (masl) in Sumatera Barat Province (Figure 1). City of Padang (CP, 0°51'28.60"S; 100°19'57.07"E, elevation ca.4 masl) is a peat swamp area that is overgrown by Sago plants and is surrounded by the settlement. Sungai Barameh (SB, 1°1'54.71"S; 100°24'43.69"E, elevation ca. 83 masl) and Bukik Kasang (BK, 0°47'7.20"S; 100°21'8.59"E, elevation ca. 351 masl) comprise of streams and rubber plantations habitat. Lubuk Bonta (LB, 0° 31'10.16"S; 100°17'43.16"E, elevation ca. 222 masl) consists of stream, rubber plantation, and paddy field (Figure 2).

Streams in SB and BK were conformable in which they consistof many rocks with medium to large size. The width of the streams is about 8 to 9 meter. While stream in LB is smaller in width (3-5 meter), rocks are obtainable but the size is much smaller and most of them submerged in the



**Figure 1.** Locations of the study indicated by black-filled circle. Lubuk Bonta (LB), Bukik Kasang (BK), City of Padang (CP) and Sungai Barameh (SB).



**Figure 2**. Typical habitat in the study site. A - C: streams in BK, LB and SB, respectively. D - F: rubber plantations in BK, LB and SB, respectively. G: peat swamp overgrown by Sago plants in CP. H: paddy field in LB.

streambed. Rubber plantation in BK was not well-treated by the owner thus allowing the shrubs grew among the rubber trees. There was a small-sized stream in the middle of plantation with width of about 0.7 to 1 m and depth of about 0.3 to 0.5 m. In contrast, the rubber plantations in SB and LB were more well-maintained in which vast majority of space among rubber trees were dominated by leaf litter. In addition to rubber plantation in SB, there was a tiny flow of water with width of only about 0.2 m that sometimes no water found within it.

Paddy fields comprised of paddy plants in various stages: early planting, middle age, and post-harvested. Peat swamp contained Sago as the majority of plants surrounded by dense shrubs. Some areas had been destructed due to human activities like harvesting Sago.

#### Field survey and data collection

Two times survey was conducted in Sungai Barameh on 13<sup>th</sup> and 14<sup>th</sup> April, 2019. Third and fourth sampling were carried out in Bukik Kasang and Lubuk Bonta in April 21<sup>st</sup> and November 16<sup>th</sup>, 2019, respectively. Peat swamp habitat in the city of Padang was surveyed twice in November 23<sup>th</sup> and 30<sup>th</sup>, 2019.

We used the visual encounter survey technique for a known period of time (Dodd 2009) to explore areas in the study sites, where four to six persons searched systematically in the study area. The search was carried out from 8pm - 11pm by following the stream path for approximately 600-700 m during those three hours. Up to 5 m beside each stream was also surveyed with randomized walk. In each rubber plantation, the movement of surveyors was also randomized covering the area of about 400 to 500 m<sup>2</sup> after three hours searching. The fragmented Sago populations were represented approximately 400 to 500 m<sup>2</sup>, while paddy fields being surveyed were about 200 – 300 m<sup>2</sup> in size.

The searches were made in all possible areas including: inside the shrubs, under the rocks, logs, and leaf litter (Dodd 2009), tree stems, tree branches, and among low vegetations. The observed specimens were captured for documentation in the next morning. All the specimens were released back to the site where they were captured. Species identification was performed under the guideline books and articles related to Sumateran herpetofauna (e.g. Das 2015; Frost 2021; Inger & Iskandar 2005; Inger & Stuebing 1997; Iskandar 1998; Kurniati 2008; Teynie et al. 2010).

#### Data analysis

The number of species and individuals in each location were subjected for analysis of herpetofauna diversity indices including: Shannon-Wiener's heterogeneity index (H') (Krebs 1998), Margalef's species richness index (Dmg) (Magurran 2003), and Simpson's dominance index (D) (Magurran 1988); we used Jaccards's coefficient to compare species composition similarity among the study sites (Sokal & Sneath 1963) implemented in PAST v3.11 (Hammer et al. 2001).

The number of species and individual was divided into four habitat types: stream, rubber plantation, paddy field, and peat swamp Sago. Shannon -Wiener index is classified into three categories: low (< 1), moderate (1 < H' < 3), and high (> 3) (Odum 1994); dominance Simpson index is classified into three categories: low (0.00 < D < 0.30), moderate (0.30 < D < 0.60), and high (0.60 < D < 1.00) (Krebs 1999).

#### RESULTS

#### Species composition and sampling effort

Overall, we recorded 338 individuals, of which 306 were amphibians representing 26 species from 6 families, and 32 were reptiles representing 18 species from 8 families (Table 1 and Figures 6-7). Regardless to the locations, the overall species number in each type of habitat showed a variation. Totally, rubber plantation contained the highest number of species (n= 27) that differed slightly from stream (n= 24), while paddy field was the lowest (n= 2). Similarly, if splitted into amphibian and reptile groups, both were more abundant in rubber plantation with 16 and 11, respectively, than in any other types of habitat. The second richest habitat was stream that contained 15 species of amphibians and 9 reptiles. Meanwhile, no reptile was encountered in paddy field and only two species of amphibians were observed (Figure 3).

Looking more detail at species or individual number in each type of habitat in all locations, the richest species number in rubber plantation was in BK (n= 25), differed significantly from rubber plantations in SB and LB at 5 and 7 species, respectively. However, in the stream habitat, BK had the lowest number of species (n= 7) differed markedly from stream in LB with 17 species and stream in SB with 12 species. Although peat swamp Sago habitat comprised of 12 species, the number of individual was the highest among other habitat with 103 individuals. The lowest individual number was found in the stream of BK with only 9 individuals (Figure 3).

Among amphibians, the ranid group was the most abundant with a slight below 50% of the total (n= 156) followed by Dicroglossidae (n= 104), Bufonidae (n= 37), Microhylidae (n= 4), Megophrydae (n= 3), and Rhacophoridae (n= 2). Similar figure to the individual number, Ranidae and Dicroglossidae were represented the highest species number with 8 and 7, respectively (Figure 4).

While among reptiles, Agamidae was the most abundant (n= 11 individuals) ), followed by Colubridae (n= 6), Gekkonidae (n= 6), Scincidae (n= 5), Geomydidae (n= 1), Lacertidae (n= 1), Varanidae (n= 1), and Viperidae (n= 1) (Figure 4). Although Agamidae was the most abundant among individuals, Colubridae and Scincidae accounted for the richest species number (4 species). None of the encountered species was in the Threatened or Data Deficient status under IUCN red list, yet *Limnonectes blythii* and *Cyclemys dentata* were listed as near threatened (NT) species. In addition, two amphibian species were known to be endemic to Sumatera Island (*Wijayarana sumatrana* and *Chalcorana rufipes*).

In the first attempt of the survey, we found 5 species in the rubber plantation in SB. The second survey was in the stream habitat of SB and we observed 9 species. The number of species reached the highest point in rubber plantation of BK at 15 species, then gradually decreased until the final survey. However, overall, the species accumulation curve showed an upward **Table 1.** List of the species observed in the study including the number of individual. SB: Sungai Barameh; BK: BukikKasang; LB: Lubuk Bonta; CP: City of Padang; N: number of specimens. Typed in bold: endemic to Sumatera.

Family	Species	Rubber Plantation			Stream			Peat Swamp Sago	Paddy Field
		SB	BK	LB	SB	BK	LB	СР	LB
Bufonidae	Duttaphrynus melanostictus	3	5	1	0	0	0	0	0
	Ingerophrynus divergens	0	1	0	0	0	0	0	0
	Leptophryne borbonica	2	4	0	3	0	0	0	0
	Phrynoidis aspera	0	6	2	2	3	5	0	0
Dicroglossidae	Fejervarya cancrivora	0	6	0	5	0	5	3	9
	Fejervarya limnocharis	0	4	0	0	0	0	0	18
	Limnonectes blythii	0	4	0	4	2	3	12	0
	Limnonectes kuhlii	0	4	0	3	5	0	0	0
	Limnonectes macrodon	0	0	0	0	0	2	0	0
	Occidozyga lima	0	0	0	0	0	0	6	0
	Occidozyga sumatrana	0	0	0	0	0	1	8	0
Megophrydae	Megophrys nasuta	0	2	0	0	0	0	0	0
	Leptobrachium cf. hasseltii.	0	1	0	0	0	0	0	0
Microhylidae	Kaloula baleata	0	0	1	0	0	0	0	0
	Microhyla sp1.	0	0	0	0	0	0	2	0
	Microhyla sp2.	0	1	0	0	0	0	0	0
Ranidae	Amnirana nicobariensis	0	0	0	0	0	0	17	0
	Chalcorana parvaccola	2	5	0	4	0	4	37	0
	Chalcorana rufipes	0	7	1	4	0	4	0	0
	Wijayarana sumatrana	0	0	0	0	4	3	0	0
	Hylarana erythrea	3	6	2	0	3	4	3	0
	Odorrana hossi	0	0	0	2	6	5	0	0
	Pulchrana glandulosa	0	0	0	0	0	0	11	0
	Pulchrana sundabarat	0	8	1	10	0	0	0	0
Rhacophoridae	Polypedates leucomystax	0	0	0	0	0	1	0	0
I.	Polypedates macrotis	0	0	0	0	0	1	0	0
Agamidae	Aphaniotis fusca	1	1	0	2	0	0	0	0
0	Bronchocela cristatella	0	3	0	0	0	1	0	0
	Gonochepalus grandis	0	0	1	0	0	2	0	0
Colubridae	Coelognathus radiatus	0	1	0	0	0	0	0	0
	Dendrelaphis haasi	0	0	0	0	0	1	0	0
	Dendrelaphis pictus	0	0	0	0	0	2	ů 0	0 0
	Xenochrophis trianguligerus	0 0	1	0	1	0	0	ů 0	ů 0
Gekkonidae	Cyrtodactylus sp.	0 0	0	0	3	0	Ő	ů 0	0 0
	Hemidactylus frenatus	0	0	0	0	0	1	1	0
	Hemiphyllodactylus typus	0	1	0	0	0	0	0	0 0
Geoemvdidae	Cvclemvs dentata	0 0	1	0	0	0	0	ů 0	ů 0
Lacertidae	Takvdromus sexlineatus	0	1	0	0	0	0	0	0
Scincidae	Eutropis multifasciata	0	0	0	0	0	0	2	0
	Eutropis rugifera	0	1	0	0	0	0	0	0
	L voosoma howringii	0	0	0	0	0	0	1	0
	Spenomorphus sp.	0	1	0	0	0	0	0	0
Varanidae	Varanus salvator	0	1	0	0	0	0	0	0
Viperidae	Trotidolaemus waoleri	0	0	0	0	1	0	0	0
Total m	mber of individuals	11	76	0	13	1 24	15	103	27
Total number of individuals		5	25	7	тэ 12	2 <del>-</del> ∓ 7	+3 17	105	2



**Figure 3**. Species number of amphibian and reptile collectively (left) and species number of in each habitat (right). Rubber= rubber plantation; SB, BK, LB= refer to the study sites mentioned in the previous section.



Figure 4. The number of species (left chart) and individual (right chart) in each family.

trajectory with a drastic uptick to a slight more than 45 species at the end of study (Figure 5).



**Figure 5**. Species accumulation curve from each VES-sampling effort. Two visits in SB (sampling effort 1 and 2), one visit in BK and in LB (3 and 4), and two visits in CP (5 and 6).

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Figure 6. List of some anuran species that were observed during the study. Images are not to scaled. A. A. nicobariensis, B. H. erythraea, C. K. baleata, D. O. Sumatrana, E. F. limnocharis, F. C. rufipes, G. P. glandulosa, H. I. divergens (juvenile), I. P. macrotis, J. O. lima, K. C. parvaccola, L. P. signata, M. Leptobrachium sp., N. P. leucomystax, O. L. kuhlii, P. W. Sumatrana, Q. O. hosii, R. P. aspera, S. M. nasuta, T. L. macrodon.



Figure 7. List of some reptile species that were observed during the study. Image are not to scaled. a. *T. wagleri*, b. *A. fusca*, c. *B. cristatella*, d. *H. typus*, e. *G. grandis*, f. *C. dentata*.

#### Notes on habitat use at the time of observation

Ranidae is generally found along the stream banks or in the middle of streams. However, *W. sumatrana* was sometimes found in a distance away from the stream (up to 5 meters). Some species were found in the amplexus position, such as *W. sumatrana*, *L. blythii*, and *Leptophryne borbonica*. Although many *P. sundabarat* were found on the rocks in the middle or in the edge of the streams, there was one individual found in the middle of the rubber plantation (15 meters away from the nearest stream). *Pulchrana glandulosa* distributed exclusively in the peat swamp Sago, often found perching on the

midribs of the Sago plants or on the its stem that has been cut. The dicroglossid group of the genus *Limnonectes* and *Occidozyga* (in all study sites except CP) was mainly found in the puddle of the stream edge where half portion of the body submerged in the water. Whereas, in the peat swamp Sago, *Limnonectes blythii* perch near the tree of Sago or perch on the cut stem of Sago, while *Occidozyga* spp. never used the same substrate instead of staying in the shallow water. The species of *F. cancrivora* and *F. limnocharis* were commonly found in paddy fields. Megophrydae members were found in more open grassy areas near the streams (5 meters in distance) with some temporary pools resulted from human and animal activities. Whereas, a species of *Kaloula baleata* (Microhylidae) was found perching on a rubber tree about 3 meters high above the ground.

Among reptiles, Colubrid snakes were mainly found on tree branches adjacent to streams (3.5 meters above the ground and about 3 meters from the stream) except for *C. radiatus* which was found on the roof of a woody house. *Xenochropis trianguligerus* and *Gonocephalus grandis* were observed on tree branches above the stream (0.5 to 4 meters high above it). Another agamid (*B. cristatella*) was observed on a tree branch about 0.5 m high above the ground. Asian leaf turtle was found under water of the small stream (width of 0.7 m; depth of 0.3 m) in the middle of the rubber plantation in BK.

#### Diversity indices assessment and similarity

In general, most of habitat type in all locations had the moderate level of heterogeneity ranging from 1.55 to 2.36. The highest level of heterogeneity was in rubber plantation in BK (2.36) followed by stream in LB at 2.13, while the lowest level was in the paddy field in LB with only 0.64. The highest level of richness was in rubber plantation in BK (5.54), while stream in LB placed in the second higest with 4.20. Again, paddy field represented the lowest value of richness at 0.30. Dominance index values indicated that no species dominated in most of all types of habitat in all locations (range values of 0.12 to 0.22), yet the value in the paddy field was relatively higher than others (value of 0.56) (Table 2). Regarding the similarity of herpetofauna communities, all rubber plantations were more conformable for each other, separated from all streams habitat. The pattern of similarity in rubber plantation and stream groups was the same where BK was more similar to SB than LB. Community in paddy field and peat swamp Sago separated from those rubber plantations and streams (Figure 8).

#### DISCUSSION

The previous inventory carried out in Sumatera Barat region was completed by previous studies, e.g. Inger & Iskandar (2005), Kurniati (2008), Wostl et al. (2017), and Nugraha et al. (2020). Their inventories mainly focused on conservation forests that administratively managed and protected either by local or national government such as national park, nature reserve, or



Table	<b>2</b> . Diversit	y indices	of herpetofauna	communities	in each	type of habitat	in Sumatera	Barat. SB	: Sungai	Barameh,
BK: Bu	kik Kasang	g, LB: Lu	buk Bonta, and (	CP: City of Pa	dang.					

Habitat Type	Location	Heterogeneity	Richness	Dominance	
Rocky stream	SB	2,05	2,92	0,12	
Rocky stream	BK	1,70	1,89	0,17	
Rocky stream	LB	2,13	4,20	0,08	
Rubber plantation	SB	1,55	1,67	0,22	
Rubber plantation	BK	2,36	5,54	0,06	
Rubber plantation	LB	1,65	2,73	0,16	
Paddy field	LB	0,64	0,30	0,56	
Peat swamp Sago	СР	1,80	2,37	0,19	



**Figure 8**. Dendrogram of similarity of the herpetofauna species composition between each habitat. Rubber= rubber plantation; SB, BK, LB= refer to the study sites mentioned in the previous section, paddy = paddy field in LB and Sago = peat swamp habitat in CP.

protected forest. However, our knowledge of herpetofaunal communities assessment from non-protected areas in Sumatera Barat started to accrue and has been opened publicly, for example, Nugraha et al. (2021) provided a cheklist of amphibians and reptiles species in a tourism area, Harapan et al. (2020) analyzed the potential distribution of the secretive species of *Ichthyopis*, and Sumarmin et al. (2019) inventoried the anuran species in a paddy field. Our current study provided information on herpetofaunal communities that specifically analyze the diversity index for some regions of lowland habitat in Sumatera Barat. Diversity estimation is important for future protection and management (Snodgrass et al. 2000) and the effect of habitat changes assessment through time (Dodd 2009).

Regarding the sampling effort, the species accumulation curves did not show a plateau trend in the end of survey attempt, thus it can be deemed that more sampling effort would perhaps yield more number of species. Overall, regardless to the locations, rubber plantation contained more species number than other types of habitat. It was similar to what Paoletti et al. (2018) found in Jambi, eastern part of Sumatera that considered the rubber plantations might offer more niche for amphibian species diversity.

Splitting to each type of habitat, the different number of species in each habitat could be explained through several reasons. For rubber plantation, the number of species in BK differed significantly from SB and LB. It most likely that BK rubber plantation was not well-treated by the owner compared to SB and LB, thus more understory vegetation existed there. Moreover, there is a small-sized stream in the middle of plantation that certainly plays an pivotal role in attracting a number of amphibians and reptiles. Conversely, SB and LB rubber plantations lack of such properties. Although there was a small water flow in rubber plantation of SB, it might be unsufficient for herpetofaunal needs because sometimes it contains no water. For stream habitat, however, the number of species in BK was lower than SB and BK. It might be due to the amount of water it had. Stream in BK in our visit time had much water as we hardly stepped on large rocks in it. The edge of stream is also too high to see during the survey, hence we likely surveyed that area less than in SB or LB. In peat swamp Sago area, the species number is quite comparable to SB and LB streams. Although the area was sometimes disturbed by harvesting activities, the disturbance is might be relatively much fewer because it is not a plantation. Hence, peat swamp Sago area provided better understory vegetation, more permanent puddles and most likely more humid than common type of plantations like rubber or oil palm. The abundance correlated with type of plantation. Oil palm plantations become the most inhabited plantation by large number of amphibians because the harvesting activities by using trucks often made basins filled by water for amphibian eggs. In contrast, rubber plantation contained more species numbers due to vegetation structure that are more stable and diverse (Paoletti et al. 2018).

Based on the heterogeneity index value, most of surveyed habitat was classified as moderate (1 < H' < 3). The lowest heterogeneity level was found in paddy field with the value of 0.64 and the highest was in rubber plantation in BK with the value of 2.36. As stated before that the more diverse of habitat in BK may allow herpetofaunal communities to survive and to develop well in the area. On the contrary, paddy field is considered to be the most disturbed habitat because the area is highly modified for plantation purposes. High human activities can reduce the diversity of habitat in an area (Hassan & Hassan 2019) which can directly affect the level of diversity and the abundance of herpetofauna (Carpio et al. 2015). In addition to habitat diversity, the quality of abiotic factors especially water also determines the survival of herpetofauna. It affects the survival of tadpoles, growth, maturation, and physical development (Dodd 2009). For comparison, the diversity level of herpetofauna in plantations and urban areas have been revealed by some authors. Samitra & Rozi (2020) revealed a moderate level of diversity of herpetofauna in rice field and river in Southern Sumatera; Maulidi et al. (2019) also showed that herpetofaunal diversity in Borneo in a rubber plantation was in moderate level.

Likewise for richness level, high level of heterogeneity corresponded to the high level of richness. Dominance index showed that there was no species dominated in all habitat, but paddy field might be getting risk by high modification on the land by the farmers. The richness index in our study site in paddy field was higher than that of in Samitra & Rozi (2020).

The similarity analysis clearly showed that same habitat grouped together, separated from the different type of habitat. It means that the herpetofauna composition in rubber plantations remained similar regardless to the locations. Herpetofauna composition in rubber plantations were more comformable with those in streams, leaving the other types of habitat outside the group. Either in streams or rubber plantations, SB and BK were more similar than those to LB. The elevation of LB was higher than SB and BK that might affect the species composition differentiation among those habitat. For example, we found rhacophorids (*P. leucomystax* and *P. macrotis*) and *Dendrelaphis* spp. only in LB. Another study showed that there was a variety of species composition along geographic elevational gradient. It might be also caused by the elevation-derived abiotic parameters such as temperature and humidity (Sasaki et al. 2005).

#### CONCLUSION

The diversity level of herpetofaunal communities in the lowland habitat was revealed. Most of habitat type supported moderate level of heterogeneity index except paddy field that was categorised as low. The highest point in heterogeneity index was in a rubber plantation as well as the richness index. The herpetofaunal communities in paddy field might be disturbed by reguler activities of the farmers as the richness index hit the lowest score and the dominance index peaked the highest. We found that rubber plantations contained the highest number of species followed by streams, peat swamp Sago, and paddy field. Regarding to the number of species per family, the families of Ranidae and Agamidae represented the most abundant group for amphibian and reptile, respectively.

#### **AUTHORS CONTRIBUTION**

F.A.D.N. designed, collected, analyzed the data and supervised all the process, F.K. designed the research and wrote the manuscript, R.S. collected the data, wrote and revised the manuscript, A.M.K analyzed the data and prepared the figures and tables, A.P.A revised the manuscript.

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#### **CONFLICT OF INTEREST**

There is no conflict of interest.

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