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## AMPHIBIAN DIVERSITY AND ABUNDANCE FROM TWO STREAMS IN XUAN SON NATIONAL PARK

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**Abstract.** Vietnam is one of the most well-known countries in the world in terms of amphibians diversity. However, we have little information on whether or not amphibian populations are declining in this country. We use visual encounter surveys to obtain data on amphibian abundance and species diversity from two streams in Xuan Son National Park (stream 1 located in limestone mountain and stream 2 located in soil mountain). We recorded 14 species belonging to 11 genera and six families of anuran from survey streams. In two survey streams, the stream 1 exhibited higher amphibian abundance than the stream 2 while the stream 2 exhibited higher amphibian diversity than the stream 1. In 362 individuals recorded, males are more abundant than females in most of species at surveyed streams, the male: female sex ratio was nearly 3:2; especially in *Hylarana maosoneis*, the male: female sex ratio was nearly 6:1.

*Keywords:* Diversity, abundance, limestone mountain, soil mountain, Xuan Son National Park.

## 1. Introduction

Vietnam is one of the most well-known countries in the world in terms of amphibians diversity with a total of about 279 recognized species of amphibians (Frost, 2019 [1]). Currently, nearly one-fifth of Vietnamese amphibians are listed as threatened and one-third are listed as data deficient (IUCN, 2019 [2]). However, we have little information on whether or not amphibian populations are declining in Vietnam. In order to detect amphibian population declines in Vietnam, baseline information on amphibian population abundance and how it changes over time is urgently required (Rowley *et al.,* 2010 [3]), the establishment of amphibian population monitoring programs is a priority for Vietnam and Southeast Asia as a whole (Rowley *et al.,* 2010 [3]). Xuan Son National Park is located in the Phu Tho Province, northern of Vietnam. The total area of this nature reserve is approximately 34,000 ha, consisting of 15,000 ha of evergreen forest in the core zone and 19,000 ha of disturbed secondary forest. In terms of the

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amphibian species diversity, this national park is one richly studied areas in Vietnam: Tran et al. (2008) reported a total of 27 species [4], Nguyen & Nguyen (2009) added two new record species [5]; Nguyen *et al.* (2013) documented 40 species with 11 new records species [6]; Le et al. (2016) recorded the species *Microhyla marmorata* for the first time [7]; and Vassiliev (2015) listed 21 species of amphibians from primary forest near the villages Du and Lap [8]. However, there are no studies to assess the abundance and structure of amphibian populations in this national park.

In this study, amphibian population monitoring sites were established in Xuan Son National Park, one of the large protected areas in Vietnam. The park was selected due to its diverse and endemic amphibian fauna and increasing threat of habitat loss and modification. Results provided baseline information on amphibian population abundance and community structure within the park, and provide evidence of how Vietnamese amphibian communities in forested areas respond to environmental condition and habitat. The results will also allow the detection of ongoing or future amphibian population declines in the region. In addition, information gathered in this project will inform management decisions within the protected area, and in other Vietnamese protected areas.

## 2. Content

### 2.1. Material and Methods

*Field surveys.* Six field surveys (Figure 1) were conducted in two seasons: hot season from May 2017 to July 2017 and cold season from September 2017 to November 2017 in two streams in Xuan Son National Park, Phu Tho Province by Do Thi Yen, Le Trung Dung, Nguyen Thi Loc, Dao Ngoc Anh and Doan Manh Tung (Do *et al.*). Study streams, situated in evergreen forest above 400 m elevation, distance between study streams was 500 m. Site 1 (S1, from 21°09.175'N 104°55.864'E to 21°09.864'N 104°57.820'E, average 400 m elevation, the stream with habitat on both sides are evergreen forest with many shrubs and small trees on the limestone mountain). Site 2 (S2, from 21°09.443'N 104°57.038'E to 21°08.455'N 104°57.204'E, average 400 m elevation, small stream with habitat on both sides are evergreen forest with many shrubs and big trees on the soil mountain, near the residential area, many trees were cut down across the stream). At each stream, we conducted nocturnal visual encounter surveys along 300 m of the stream. For all individuals encountered, we recorded species, sex, snout-vent length (SVL), and body mass.

*Morphological characters*. Measurements were taken with a digital caliper to the nearest 0.1 mm. The following abbreviations were used: SVL: Snout-vent length; for webbing formula, we followed Glaw&Vences (2007) [9]. Sex was determined by gonadal inspection, nuptial pads on the forelimbs, vocal sac.

*Identification of amphibians*. The specimen, after analyzing the morphological data, which is classified according to the papers: Ohler & Duboi (2006) [10]; Taylor (1962) [11]; Smith (1943) [12]; Bain *et al.* (2006) [13], Ohler *et al.* (2011) [14]; and Hetch *et al.* (2013) [15]. The scientific name, common name of the species and the list for them which are arranged according to Nguyen *et al.* (2009) [16].

*Species richness estimations.* The number of species at each site was estimated using the Jackknife formula (Krebs, 1999) [17].

$$\hat{S} \pm t_{\alpha} \times \sqrt{var(\hat{S})}$$

**Data analysis.** Data were analysed by Past statistic (PAST 3.14, Hammer *et al.*, 2001 [18]). The differences of overall amphibian abundance (number of individuals per survey) and diversity (number species per survey) at two surveyed streams and two rainfall categories were compared by using non-parameters analysis with Kruskal-Wallis rank sum test. The median difference is significant if p-value < 0.05. Boxplot graphs were used to examine the range of data with boxes encompassing 95% confident intervals for the data and middle lines are medians. Bars encompass values up to 1.5 interquartile range, open circules represent values > 1.5 interquartile ranges from the nearest quartile. Outliers appearing as dots outside boxes are values that deviate markedly from other members of the sample.



Figure 1. Position of the two surveyed streams in the centre of Xuan Son National Park

## 2.2. Results

## \* Amphibian species composition

During all surveys at two sites, 14 species belonging to 11 genera and six families of anuran were recorded, with 362 individuals counted. Of the anuran species recorded, 8 species were documented at the stream 1 while 9 species were recorded at the stream 2. Following the IUCN Red list 2019, four as Least Concern (LC) (Table 1).

Table 1. The composition of amphibian species at the surveyed sites

Stt	Species	Common name	IUCN (2019)	Stream 1	Stream 2
	Bufonidae				
1	Duttaphrynus melanostictus (Schneider, 1799)	Black-spined toad		X	
	Megophryidae				
2	Leptobrachella ventripunctata (Fei, Ye & Li, 1990)	Yunnan Asian Toad			Х
3	Megophrys major Boulenger, 1908	Major's Horned Toad	LC		Х
4	<i>M. microstoma</i> (Boulenger, 1903)	Asian Mountain Toad			Х
	Microhylidae				
5	Microhyla heymonsi Vogt, 1911	Heymon's Ricefrog	LC	Х	
	Dicroglossidae				
6	<i>Limnonectes bannaensis</i> Ye, Fei, ,Xie & Jiang, 2007	Banna Large- headed			Х
	Ranidae				
7	Amolops ricketti (Boulenger, 1899)	Chinese sucker frog			Х
8	<i>Odorrana</i> graminea (Boulenger, 1900)	Large Odorous Frog		X	X
9	Sylvirana maosonensis (Bourret, 1937)	Mao-Son Frog		X	
10	Rana johnsi (Smith, 1921)	John's Frog	LC	X	

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	Rhacophoridae				
11	Kurixalus bisacculus (Taylor, 1962)	Taylor's Tree Frog		Х	Х
12	Polypedates megacephalus Hallowell, 1861	Spot-legged Treefrog	LC	Х	Х
13	P. mutus (Smith, 1940)	Northern Treefrog			Х
14	Rhacophorus smaragdinus (Blyth, 1852)	White-lipped Treefrog		Х	

Some species appeared widespread, with Odorrana graminea, Kurixalus bisacculus, Polypedates megacephalus observed at all sites. In contrast, Duttaphrynus melanostictus, Microhyla heymonsi, Rana johnsi, Sylvirana maosonensis and Rhacophorus smaragdinus were only observed at stream 1, and Leptobrachella ventripunctata, Megophrys major, M. microstoma, Limnonectes bannaensis, Amolops ricketti and Polypedates mutus were only observed at the stream 2.

# \* Amphibian abundance

Trends in total amphibian abundance and species diversity

Amphibian abundance and diversity differed among streams. Stream 1 exhibited higher amphibian abundance and diversity than stream 2 (Table 2, Figure 2 & 3).





Figure 2. Boxplots showing the<br/>abundance of amphibian at each streamFigure 3. Boxplots showing the species<br/>diversity of amphibians at each stream

There was some unusually high abundance value recorded at streams 1 (outliers in Figure 2). These high values were associated with large breeding aggregations of *Polypedates megacephalus*.

Predicted total species diversity varied between sites (Table 3) approximately 50% of the species were predicted to have been found after 6 surveys (Table 3). Jackknife estimates also suggested that approximately 12 surveys were needed to detect 80% of the amphibian species present at a site (Table 3 and Figure 4).

	Abundance	Diversity
Chi-square	5.769	0.9231
Df	2	2
p-value	0.02	> 0.05

 Table 2. Kruskal-Wallis rank sum test for streams being significant different

 from each other in abundance and diversity

Table 3.	Jackknife	estimate	of sp	ecies	richness
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Parameter	Stream 1	Stream 2
Total number of species	8	9
Total number of surveys	6	6
Number of unique species	5	6
Jackknife estimate of species richness (for 95% confidence)	12.17±5.67	14±11.22

# \* The abundance of each species over time

Amphibian species were not detected at equal abundances throughout the year, and most species appeared to have distinct periods where they were most abundant at the surveyed streams (Figure 4).



# Figure 1. Month of year when each species was most abundant at all surveyed sites

Amphibian species composition was temporally partitioned. During two seasons, some species were most active at the same time as other species (*Megophrys microstoma, Kurixalus* 

*bisacculus and Polypedates megacephalus* were most abundant in rain season) while *Sylvirana maosonensis* was most active in both of two seasons (Figure 4).

For each species, times of peak abundance were overall get highest in May and Jun. According to previous studies, the majority of amphibian species reproduce strongly in the months from March to May, a few species of tree frogs were active in September and October. This study began in May and recorded the most amphibian species (nine species). However, in *Sylvirana maosonensis* and *Megophrys microstoma*, patterns of abundance differed from other species. From July to November, there were more individuals of *Sylvirana maosonensis* found at stream 1.

### \* Amphibian sexual and age structure

We collected data and build chart whether the population of each species more than 20 individuals. Males are more abundant than females in most of species at surveyed streams, the male: female sex ratio was nearly 3:2. This was particularly evident in *Sylvirana maosonensis*, where the male: female sex ratio was nearly 6:1.



Figure 5. The sex and age structure of each species over time, at all surveyed streams

## 2.3. Discussion

### \* Amphibian species composition

This study revealed no evidence of population declines in a montane tropic forest in Southeast Asia. Although amphibian species diversity and abundance fluctuated throughout the year, as is common in amphibian communities globally (Pough *et al.*, 1998 [19]), no overall population trends were observed over the 6-month survey period. Although amphibian declines have occurred globally in tropical montane habitats (Alford & Richard, 1999 [20]), the lack of population monitoring in Southeast Asia has made it impossible to determine whether declines are occurring in Southeast Asia as they are in other parts of the world (Rowley *et al.*, 2010 [3]). In Vietnam, there is only one study about population monitoring in the South and this study is the first such amphibian population monitoring in the North.

While there was no evidence of amphibian population declines at the disturbed sites, habitat modification is likely to have an impact on population abundance, particularly of certain species. Maybe overall abundance will remain but species which most sensitive to habitat change may be lost (Alford & Richard, 1999 [20]). For example, changing vegetation structure due to a shift in land use practices caused the decline of *Bufo calamita* population in Britain and provided conditions under which the common toad *Bufo bufo* became a successful competitor (Beebee, 1977 [21]). Similarly, in west-central Florida, USA, while four species *Bufo quercicus, Scaphiopus h. holbrookii, Hyla femoralis* and *H. gratiosa* decreased because of residential development, three species of ranids, *Rana utricularia, R. grylio,* and *R. catesbeiana* were found in higher abundances (Delis *et al.*, 1996 [22]). This study, which was over a relatively short time and only at two streams, may not provide the overall status of amphibian communities. Therefore, continued population monitoring and expanding monitoring to other sites are recommended to determine with more certainty if amphibian populations are declining in the area or not.

## \* Amphibian abundance and species diversity

Each species uses particular habitats depending on its ecological and biological characteristic (Duellman & Trueb, 1994 [23]). Species may be distributed according to their thermal and hydric tolerances (Paula & Ivan, 2000 [24]; Skelly *et al.*, 1999 [25]; Well, 2007 [26]). For example, *Microhyla heymonsi* was only recorded in the stream 1 in microhabitats like mosses and grasses while *Megophrys microstoma* was found in the stream 2 in habitat of flowing stream and shrubs.

Our findings also emphasize the important of repeated surveys of a single site in order to get more adequate assessment of total amphibian diversity at a site. For example, after 6 surveys, we were still finding species of amphibian that were not previously recorded for that stream. Repeated surveys at single sites are rare in Vietnam, with most studies relying on surveys over a very short time (Nguyen *et al.*, 2009 [16]).

# \* Amphibian sexual and age structure

Most species detected during surveys aggregated at streams at particular times of the year with the number of males mostly higher than females. The higher number of males may reflect that males was actually more abundant than females or simply that males can be found easier than females by their calls, or are otherwise more detectable during surveys. Environmental conditions and habitat change may influence the distribution of sexes over time (Emlen & Oring, 1977 [27]). For example, *Dendrobates pumilio* was observed to have different sex ratios when secondary forest replaced primary forest (Prohl, 2001 [28]). In this survey, it remains unclear which factors are

responsible for difference in sex ratios among species and sites and over time.

## 3. Conclusions

A total of 14 amphibian species are recorded in the study area, belonging to 11 genera and six families. Of the anuran species recorded, eight species were documented at the stream 1 while nine species were recorded at the stream 2.

The abundance and diversity of amphibians were significant different among streams. Stream 1 exhibited higher amphibian abundance than stream 2 while stream 2 exhibited higher amphibian diversity than stream 1 (Jackknife estimate of species richness in stream 1 as  $12.17\pm5.67$  and in stream 2 as  $14\pm11.22$ ).

Males are more abundant than females in most of species at surveyed streams, the male: female sex ratio was nearly 3:2; especially in *Hylarana maosoneis*, the male: female sex ratio was nearly 6:1.

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