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THESIS

AMPHIBIAN DIVERSITY AND STREAM ENVIRONMENTAL
VARIATION IN PRISTINE TROPICAL FOREST OF
KHAO YAI NATIONAL PARK



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Environmental variation influences directly amphibian diversity. A stream in tropical forest is habitat which supports forest amphibian diversity. Destruction and environmental change of stream strongly affect stream amphibian population. Thus this research aimed to study diversity of stream amphibian and relation with environmental variation. I compared species richness, abundance and environmental factors which may affect to distribution and assemblage of amphibian in stream of Khao Yai National Park. I selected three stream characteristics; cascade, rocky and sediment substrate and three landscape characteristics; higher slope area, lower slope area and hill near grassland. I encountered 6, 10 and 10 species in cascade, rocky and sediment substrate respectively and 7, 8 and 11 species in higher slope area, hill near grassland and lower slope area respectively. *Microhyla berdmorei* and *Hylarana nigrovittata* were the most number of individuals in dry season and rainy season respectively and shown tendency to be the dominant species since the distribution of these species influenced diversity. The Simpson's diversity and similarity indices revealed that the amphibian diversity and similarity was higher in the dry season, but the rainy season abundance was higher. NMDS revealed that some stream variables were the important factors to the assemblage of amphibian but did not tend to bear similar assemblage between both seasons. Species accumulation curves and diversity index indicated that the assemblage of many species was supported by some stream characteristic and habitat pattern, these were the rocky substrate and the lower slope area and these habitats indicated support to be beta diversity.

Student's signature

Thesis Advisor's signature

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AMPHIBIAN DIVERSITY AND STREAM ENVIRONMENTAL VARIATION IN PRISTINE TROPICAL FOREST OF KHAO YAI NATIONAL PARK

INTRODUCTION

Amphibians can be found in terrestrial habitats from tropical rainforests to the driest deserts on earth (Bentley, 1966), except Antarctica and high Arctic latitudes, from below sea level to altitudes above 4,500 m. Amphibians cannot live in marine environments, but they are found in nearly all types of aquatic habitat, from brackish estuaries and salty pans to rivers, streams, ponds, lakes, marshes, swamps and ditches. Since we still know little about amphibian ecology including pattern of spatial and temporal variability in the diversity (Pechmann *et al.*, 1991) and amphibian population has rapidly declined in many areas of the world due to environmental change and habitat destruction (Beebee and Griffiths, 2005), thus this is one of the interesting aspects to study and imply for conservation planning.

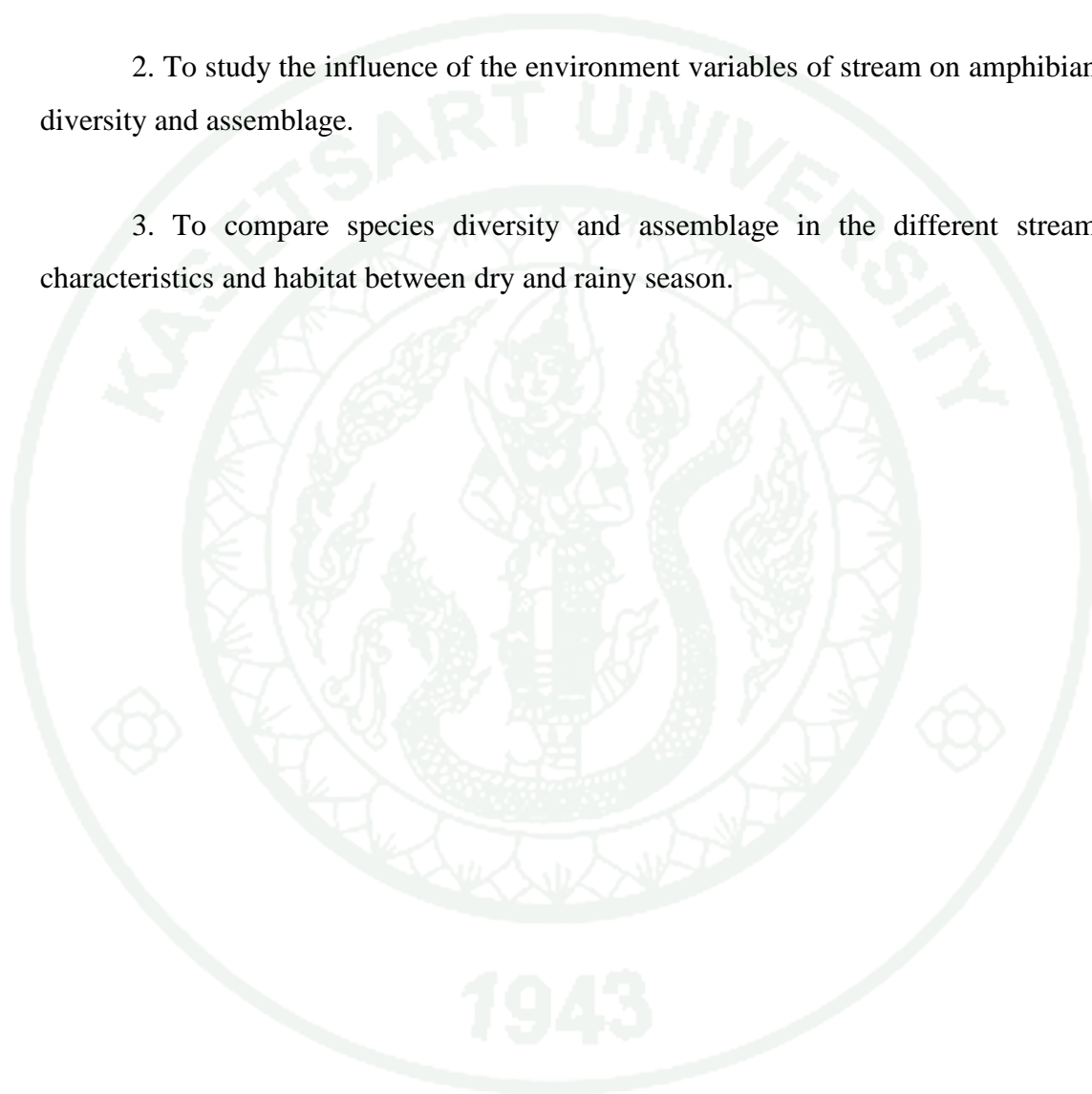
The life cycle of amphibians strongly depends on their surrounding environment. Land cover patterns, fragmentation patterns, connectivity of landscape elements, and spatial position of roads, water bodies and other landscape elements in the region are able to indicate the influence to an amphibian community structure and health at regional scales. Furthermore, the biotic interactions with other species, water quality, parasite burden, and contaminant levels in the environment exert a strong influence as well (Beasley, 2002).

Since development of most juveniles occur in water, their life cycle of amphibian strongly relate to aquatic habitats. Stream is an important lotic breeding habitat of many amphibians (Chan-ard, 2003). In Thailand and around the world, tropical stream components have been altered by many factors (human disturbance, agriculture, deforestation and climate change) and these factors affect the diversity of stream amphibian.

Previous studies showed that environmental and spatial factors were the important variables directly associated to amphibian population (Parris and McCarthy, 1999; Wilkins and Peterson, 2000; Parris, 2004; Gardner and Fitzherbert, 2007) and other fauna, such as mussel (Baldigo *et.al.*, 2007), Black fly (Pramual and Kuvangkadilok, 2009; Hamada and McCreadie, 1999; Hamada *et.al.*, 2002) and fish (Kadye *et.al.*, 2008). Thus these factors determined the distribution and diversity of aquatic organism and the variation of factors could be predicted in the different habitat. Although there are many studies of amphibian diversity and assemblage, most studies emphasize correlation between composition of amphibian population and spatial factors, but it is scarce in correlation of amphibian diversity and assemblage with the environmental variation of habitat especially stream in tropical forest. Thus study of environmental variables (physical and chemical) of habitat, particularly stream habitat, is interesting because these variables possibly influence amphibian diversity and assemblage. In this study, I investigated the environmental factors (physical and chemical factors) of stream and riparian area, and establish the following questions: (1) Do physiochemical condition of stream in the different characteristic and habitat differ? (2) Do environment variables of the different stream and habitat characteristics affect amphibian diversity?

OBJECTIVES

1. To study species diversity of amphibians that inhabited the difference of stream characteristics and habitat pattern.
2. To study the influence of the environment variables of stream on amphibian diversity and assemblage.
3. To compare species diversity and assemblage in the different stream characteristics and habitat between dry and rainy season.



LITERATURE REVIEW

Amphibian Diversity

The amphibians are members of class Amphibia that are divided into three orders: Gymnophiona (caecilians), Anura (frogs and toads), and Urodela (salamanders). Frogs and toads are the most broadly distributed, occurring throughout the globe, except in Antarctica, Greenland, Arctic regions of North America and Eurasia, and some oceanic islands. The greatest diversity of frogs and toads is in the neotropical region (Central America, South America, and the West Indies), where more than 2000 species occur. Salamanders inhabit nearly all northern Temperate Zones of the world. The greatest diversity of salamanders is in North America. Caecilians are found in most of the tropical regions of Southeast Asia, Africa, and Central and South America, except the dry areas and high mountains. Caecilians are primarily terrestrial, relying on moist soils for their water requirements (Lowe, 2009).

Amphibians are one of an example of organisms living at the interface between terrestrial and aquatic habitats. Most amphibians are strictly dependent from water for their larval development, and water for this group of animals is freshwater (although a few amphibians are able to tolerate high-salinity level) (Vences and Köhler, 2008). Since the life cycle of amphibians highly depends on water, thus aquatic habitat degradation affected the diversity of amphibians.

In total, 5,828 amphibians species have widely distributed throughout the world, 4177 are aquatic in that they live in the water during at least one life-history stage, and a further 177 species are water dependent and the end of 2005, there were a total of 168 species of caecilians, 514 salamanders and 5,146 frogs (Vences and Köhler, 2008).

The amphibian decline

Amphibians have strongly declined in many areas of the world and this tendency continually increased in the present (Beebee and Griffiths, 2005; Whiles *et al.*, 2006). Although habitat loss has impacted amphibians for decades, the other causes demonstrated influence on amphibian population decline. In present, the decline has various causes, but there has been widespread controversy in their cause significance (Pechmann *et al.*, 1991; Pechmann and Wilbur, 1994; Alford and Richards, 1999; Blaustein and Keisecker, 2002). Beebee and Griffiths (2005) suggested that the environmental contaminants, UV-B irradiation, emerging diseases, the introduced species, direct exploitation and climate change are possible causes of amphibian declines and strongly correlate with anthropogenic activities. Challenge in the study of environmental and spatial variation is being increasingly emphasized.

Since amphibians contribute considerably to biomass (Burton and Likens, 1975), therefore the reduction of amphibians must have important consequence for ecosystem because they are important prey and predators (Vallan, 2002). At the recent report from the IUCN's Global Amphibian Assessment indicates that a third of amphibian species (now estimated at over 5700) undergo severe declines or extinction, especially stream-associated amphibian (Stuart *et al.*, 2004). Since stream is an important habitat of many aquatic organisms, it therefore has high supports species diversity and several amphibian species depend on stream as breeding and laying area. Recently, many streams are greatly disturbed form anthropogenic activities such as alteration of stream channel morphology from filling the stream for cultivation, roads establishment and across stream and deforestation in riparian zone. These strongly affect life cycles of aquatic organisms, particularly aquatic insects, fishes and amphibians.

Correlation between amphibians and lotic system

Lotic system is a system of flowing water such as stream and river. Streams and riparian areas were the important habitat for many terrestrial animals, plants, and other organisms during some stage of their life cycles (Richardson and Moore, 2009). Many of the amphibians associated with lotic systems, both aquatic and terrestrial habitat, throughout their lives (Lowe, 2009). Since most amphibians have a complex life history with dependence upon two or more habitats to complete their life cycles: an aquatic breeding and larval habitat; and a terrestrial habitat, where juveniles and adults spend most of their time (Richardson *et al.*, 2002), thus the alteration of environmental factors of these habitat directly influence amphibian populations.

The many studies indicate spatial and environmental factors of stream and riparian zone influencing to amphibian assemblage, distribution and diversity. For example these studies, Parris and McCarthy (1999) reported that the composition of frog assemblages significantly correlated with stream size, understory vegetation in the riparian zone, the presence of palms and elevation and they found that broad forest type and the assemblage did not correlation. These results indicated that ground layer vegetation is more important for frogs than the nature of the forest overstorey. Keller *et al.*, (2009) studied the influence of spatial and environmental factors of stream habitats, including density of vegetation, stream and riparian characteristics and water volume, in rainforest and found that both factors determined of assemblage of amphibian and mostly affected to community structure and according with the study of Parris (2004) in forest streams in sub-tropical eastern Australia. These results could define amphibian guilds based on environmental characters (stream size and turbidity) with the non-metric multidimensional scaling (NMDS) technique as three discrete guilds, including waterfall stream species, calm stream species and large stream species. The variation of habitat characteristics influenced the amphibian occurrence. Wilkins and Peterson (2000) studied factors of headwater streams, including landform characteristics, channel characteristics, channel substrate and riparian habitat, to relate with salamander occurrence and abundance in second-growth Douglas-fir forest of southwestern Washington. The result suggested that

variance of habitat characteristics strongly influenced occurrence and abundance of headwater amphibians by determined occupancy of salamander at the difference level.

On the other hand, the variation of habitat did not influence the diversity and assemblage of some amphibian species because of ability of adaptation and tolerance. Maltchik *et al.*, (2008) studied species richness, abundance and composition of amphibian assemblage in a flooded riparian forest fragment of Sinos River of southern Brazil. They showed that the period of highest temperature associated with the highest species richness and abundance of terrestrial amphibians. In this study, amphibian richness and abundance did not change after flood events, showing amphibian could strongly resist to disturbance by floods of different durations. Leiuperidae and Leptodactylidae were higher abundance than the other, since ability of their adaptation such as they lay egg in floating foam in the riparian vegetation and fast larval development. Thus dominance of species related with ability to live and adaptation in habitat which was high variance.

The difference of elevation influenced amphibian assemblage and diversity. Phochayavanich *et al.*, (2010) studied stream frog assemblages at three elevations (800, 950, and 1250 m) in an evergreen forest, north central Thailand. This study was found that the average annual stream width, depth, and rate of water flow significantly differed among the three elevations and highest at the lowest elevation and according with the study of Parris and McCarthy (1999). The stream width and gradient at 800 and 950 m in elevation were similar and differed from at 1250 m. The analysis of the species diversity by the Shannon-Wiener index and the similarities of species by Morishita's similarity index revealed that frog diversity and similarity tended to be highest at the lowest elevation (800 and 950 m).

Temporal pattern can influence abundance and diversity. Gardner *et al.*, (2007) studied spatial and temporal pattern of amphibian relative abundance and diversity in western Tanzania. The study areas were in woodland, seasonally flooded grassland, cultivated habitats and riverine (define as the area within 10 m adjacent permanent and intermittent river) and were studied across three seasons (dry, early wet and late

wet seasons). The sampling period has a greater influence richness and abundance of amphibians than spatial location of study site.

An environmental change from anthropogenic activities often affects population of organism. Since human occupancy associates with a river, especially a large one, which provides water, food and suitable conditions for transportation and recreation, thus these affect aquatic organisms at the margins of large river (Viana *et al.*, 1997). Souza and Eterovick (2010) studied relationship of environmental factors of rivers under varied impact levels to anuran assemblage, richness and distribution in southeastern of Brazil. The impact composed of human use and occupation at riparian habitat, invasive herbs and shrubs with isolated trees and intensive erosion visible through sediment deposits throughout the river channel. Their results Canonical Correspondence Analysis (CCA) indicated that soil types and riparian vegetation were main factors influencing amphibian distribution among bodies of water and species richness positively related to microhabitat and availability of preferred microhabitats.

Relationship among amphibians, water quality variables and substrate types

1. Water quality variables

Since life cycle of amphibians intimately relate to an aquatic environment, thus the quality of the water in which they live can affect their growth, development, and survival (Boyer and Grue, 1995). Because pollutants, waterborne pathogens, and global environmental changes can all affect water quality, these factors can in turn affect amphibians. Conversely, amphibians are important indicators of water quality, and are considered a sentinel species, meaning that what affects amphibians presently may affect other animal species in the future (Boyer and Grue, 1995; Blaustein and Wake, 1998).

The quality of water was important factors supporting an amphibian's survival. Amphibians are perhaps even more sensitive to water quality than many fishes because they are aware of water quality issues, often make the best amphibian

caretakers (Pramuk and Gagliardo, 2012). There were several variables determine water quality, for example pH, Dissolved Oxygen (DO), conductivity, transparency, water temperature, alkalinity, phosphorus, nitrogen, sulfate, etc.

The example of the some water quality variables influenced amphibian population:

1. pH

The pH of water is basically the proportion of hydrogen (H^+) and hydroxide (OH^-) ions in solution or water. The pH scale is logarithmic with each pH unit representing a 10-fold change in the number of hydrogen ions (Pramuk and Gagliardo, 2012). Natural pristine water sources generally have a pH between 6.5 and 8.5 (Ultsch *et al.*, 1999).

Many amphibians prefer a pH that is slightly basic. However, as pH requirements vary by species, a pH of 7 is recommended as a good starting point if the optimal pH is unknown. Some amphibians, such as aquatic caecilians and Pine Barrens treefrog (*Hyla andersonii*), preferred slightly acidic water but salamanders that inhabited in limestone aquifers, such as the Barton Springs salamander (*Eurycea sosorum*), required a slightly alkaline (basic) environment (Pramuk and Gagliardo, 2012).

Previous studies revealed that low pH effect on amphibians biology, including increased tadpole growth (Relyea, 2006), delayed hatching (Horne and Dunson, 1994), larval deformation (Beattie and Tyler-Jones, 1992), immunological alterations (Brodkin *et al.*, 2003), altered oviposition/reproductive behavior (Rowe and Dunson, 1993; Ortiz-Santaliestra *et al.*, 2007), and decreased survival (Dunson and Connell 1982; Rowe *et al.*, 1992; D'Amen *et al.*, 2007). These results demonstrated that amphibian populations were sensitive on the decrease of pH.

2. Dissolved Oxygen (DO)

DO is the amount of oxygen dissolved in water. The amount of oxygen required by an aquatic amphibian depends to a great part on its natural history. For example, lentic (pond-dwelling) species require less oxygen than lotic (stream dwelling) species (Pramuk and Gagliardo, 2012).

Previous studies revealed that the low DO affected to decreasing of body weight in adult and high DO supported growth rate in tadpole but the relative sensitivities of two stage were not significant when compared (Shrum, 2004). These indicated that the effect of alteration of DO differ to life stage.

3. Water temperature

Water temperature has extremely important ecological consequences and is also an important influence on water chemistry (Pramuk and Gagliardo, 2012). Temperature exerts a major influence on aquatic organisms with respect to selection/occurrence and level of activity of the organisms (Huk and Kühne, 1999; Pramuk and Gagliardo, 2012).

Since many amphibians lived in aquatic habitat, such as stream and pond, thus alteration of water temperature highly influenced adult phase and their larval development. These reasons were supported with the report of Beebee (1995) that revealed water temperature influence the timing of reproduction (breeding/egg laying), metamorphosis, dispersal, and migration

4. Conductivity

Conductivity is a measure of the ability of water to pass an electrical current. Conductivity in water is affected by the presence of inorganic dissolved solids and conductivity in streams and rivers is primarily affected by the geology of the area through which the water flows (USEPA, 2012).

The alteration of conductivity can negatively impact many freshwater inhabitants, including bacteria, plants, invertebrates, and vertebrates. Several studies demonstrated that the alteration of conductivity negatively impacted growth and development (Gomez-Mestre and Tejedo, 2003; Snodgrass *et al.*, 2008), level of hormone (corticosterone) and prey consumption (Chambers, 2009) behavior (Haramura, 2007; Karraker *et al.*, 2008; Collins and Russell, 2009), and survival (Collins and Russell, 2009) of amphibian.

2. Stream substrate

Stream substrate is the type of substance on the bottom of the stream. This widely varies from fine sediment, through various sizes of gravels and stones, and up to large rocks and boulders. The type of substrate in a stream is dependent on the geology of the area, but is subject to change as a result of water velocity and surrounding land use (USEPA, 2012). In slower flowing areas, such as pools, the substrate is often finer, whereas faster flowing areas usually have coarser substrates. In faster flowing areas, such as stream and river, these areas are able to support a wider range of aquatic invertebrate (Erman and Erman, 1984; Warren and Kraft, 2006), stream-breeding amphibians (Bisson *et al.*, 2002) and fish species (Valentine, 1995) as the coarse substrate provides a range of stable micro-habitats attractive to animals with differing habitat preferences and increases the level of oxygen in the water because coarse substrate was cause of water turbulence.

Because the type of stream substrate influences what kinds of animals live in the waterway, any change to the substrate has an effect on the vertebrate and invertebrate population. The difference of stream substrate was factor to determine diversity of stream-dwelling amphibians and some species were sensitive to substrate alteration (Welsh and Ollivier, 1998). The alteration of substrate negatively affected to population of stream-breeding amphibian, for example when fine sediments are introduced into the aquatic environment- their growth rates capacity to hold on to the rocky substrate decreased (Welsh and Ollivier, 1998; Wood and Richardson, 2009). Furthermore, the study of Bury *et al.*, (1991) suggested that large numbers of tailed

frogs (*Ascaphus truei*) had the highest percentages of cobble substrate and was negatively affected by stream sedimentation and according with study of Gradwell (1971). These indicated that type and alteration substrate could influence amphibian species.

Amphibians in Khao Yai National Park

The study of amphibian diversity in Thailand was started the first time by Günther (1860) and has highly flourished from study of Taylor in 1957 – 1962. In present, the diversity is continuously studied in many areas especially National Park.

Chan-ard (2003) reported that 141 species was found in Thailand and could be divided into 3 Orders (8 Families): Order Urodela (Family: Salamandridae), Order Gymnophiona (Family: Ichthyophiidae) and Order Anura (Families: Megophryidae, Bufonidae, Hylidae, Microhylidae, Ranidae and Rhacophoridae)

Khao Yai National Park is a UNESCO world heritage site and the first National Park in Thailand. The area covers 2,168 km² and more than 80% is forest (Srikosamatara and Hansel, 2004). The Park has diversity of plant communities, comprising five main vegetation types: tropical moist evergreen forest, dry evergreen forest, dry deciduous forest, hill evergreen forest and grassland (Srikosamatara and Hansel, 2004). Since the Park is situated in Dong Phrayayen Mountains, the Park is the source of five main rivers in the North-East of Thailand which these rivers originate from streams that remain completely in the Park thus Khao Yai National Park is an important watershed in Thailand.

Khao Yai National Park was one of areas that had the high diversity of amphibian. In 1982, Nabhitabhata and Noopakdee reported finding of 5 families, 7 orders and 11 species of amphibian in forest of KhaoYai National Park such as *Xenophrys parva*, *Bufo melanostictus*, *Occidozyga martensii*, *Limnonectes gyldestolpei*, *Hylarana nigrovittata*, *H. macrodactyla*, *Philautus* sp., *Polypedates leucomystax*, *Microhyla butleri* and *M. pulchra*. These families were Megophryidae,

Bufonidae, Ranidae, Rhacophoridae and Microhylidae. Sangtean (1999) reported that 5 families 23 species of amphibians in the Park were encountered and species that were encountered more from 1982 were *Fejervarya limnocharis*, *H. erytheraea*, *H. indeprensa*, *H. taipehensis*, *Rhacophorus verucosus*, *Chirixalus doriae*, *C. nongkhorensis*, *C. vittatus*, *Caiiuella guttutata*, *Kaloula pulchra*, *M. berdmorei*, *M. fissipes* and *Micryletta inoranta*.

Kongjaroen (2007) studied amphibian diversity to relate altitudinal distribution along Lam Ta Klong watershed area which was one of main stream of KhaoYai National Park. In total, 19 species of 11 genus 5 families was encountered and species that were encountered more from 1982 and 1999 were *X. lekaguli*, *L. kuhlii*, *H. cubitalis*, *M. heymonsi* and *Philautus parvulus*. This study was found that 700 MSL site had the highest individuals, the 400 MSL site had the highest species richness and the 1,000 MSL site had the highest diversity index.

MATERIALS AND METHODS

Materials

Equipments for determine study sites and field survey were Topographic Maps (1:50,000), GPS and Compass. The stream variables, including pH, DO, conductivity, water temperature, were measured by Electrochemistry C933 (CONSORT, Turnhout, Belgium) and air temperature and humidity were measured by digital thermometer. The stream velocity was measured by Flow Probe (Global Water, California, USA).

Methods

Study area

The study areas located in a pristine tropical forest of Khao Yai National Park (latitude 14° 05' to 14°15', longitude 101°05' to 101°50'). The Park has two seasons, i.e. dry and rainy seasons. An annual mean temperature and annual mean precipitation in the last 30 years (1975 - 2005) is 20.9 °C and 1161.5 mm respectively. I investigated fifty-four sampling sites (selected by topographic map, 1 : 50,000) which covered first or second order streams and riparian zone, elevate at 600 – 800 m above sea level (Figure 1). I conducted in two seasons; dry (January 2011-March 2011) and rainy season (August 2011 – October 2011).

Khao Yai National Park

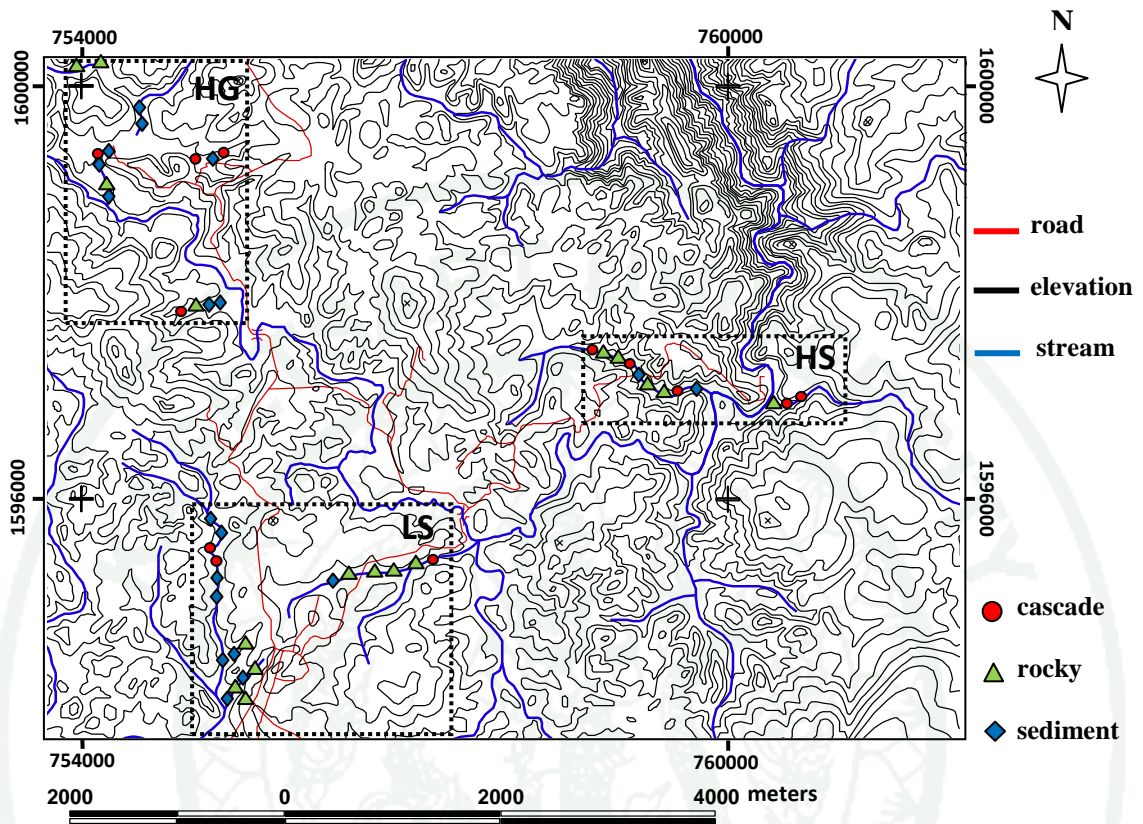


Figure 1 Map of study area in forest of Khao Yai National Park; HS is the higher slope area, HG is the hill near grassland and LS is the lower slope area.

I selected sampling sites based on stream heterogeneity. I divided streams into three characteristics: stream with cascade, stream with rocky bed and stream with sediment bed. Each characteristic of streams were subdivided into three type width: 0-1 m, 1-3 m and over 3 m. Number of each site is shown in Table 1.

Table 1 Number of sampling sites based on the difference of stream width and characteristic.

Stream width	Number of sampling sites		
	stream with cascade	stream with rocky bed	stream with sediment bed
>0-1	5	5	7
>1-3	3	7	7
>3	7	7	6
Total sites	15	19	20

Stream characteristics were determined three types:

1. Stream with cascade: The stream bed consists in cascades, rock and leaf litter. The stream velocity is higher than the other stream types. The speed of the water flow can also vary within a system and the water flow has high turbulence. In dry season, the turbulence and speed of water flow decrease.



Figure 2 Characteristic of stream with cascade

2. Stream with rocky bed: The stream bed consists in gravels, rocks and leaf litter. The velocity is nearby with cascade stream in rainy season but this is less turbulence.



Figure 3 Characteristic of stream with rocky

3. Stream with sediment bed: The stream bed consists in sediment and leaf litter. The velocity and turbulence were slower and lower than the cascade and rocky stream.



Figure 4 Characteristic of stream with sediment

The areas that covered the study sites were determined as the habitat of amphibian species. I divided the different habitat as three patterns: higher slope area, hill near grassland and lower slope area. Number of sampling sites is shown in Table 2.

Table 2 Number of study sites, sampling based on the difference of habitat pattern.

Habitat pattern	Number of sampling sites
higher slope area	12
hill near grassland	22
lower slope area	20

Amphibian sampling method

Each site was stream body 20 m lengths and riparian zone 10 x 20 m (Figure 5). I sampled specifically in sampling sites which cover environmental heterogeneity. Thus these plots depicted the dissimilarity of environmental habitat characteristics. I selected stream variable as major factors for this study. I measured chemical and physical variables of stream, including pH, conductivity, DO, stream velocity, water temperature and stream depth, and measured riparian variable, including percentage cover of understorey vegetation, since these variables were environmental and spatial variables that determine water quality and environment of amphibian habitat. The water quality and physical variables were collected at the center of the stream three ranks in each 5 m interval (Figure 5) and available values from three ranks were calculated as average of these variables.

The survey period in each site was 30 minutes in diurnal (07.00-17.00 hr) and nocturnal (19.00-24.00 hr). All captured amphibians at stream body and riparian zone and were identified and released at the same site. I recorded number of species and individuals encountered in each site. All collected amphibians were identified based on Taylor (1962) and Chan-ard (2003).

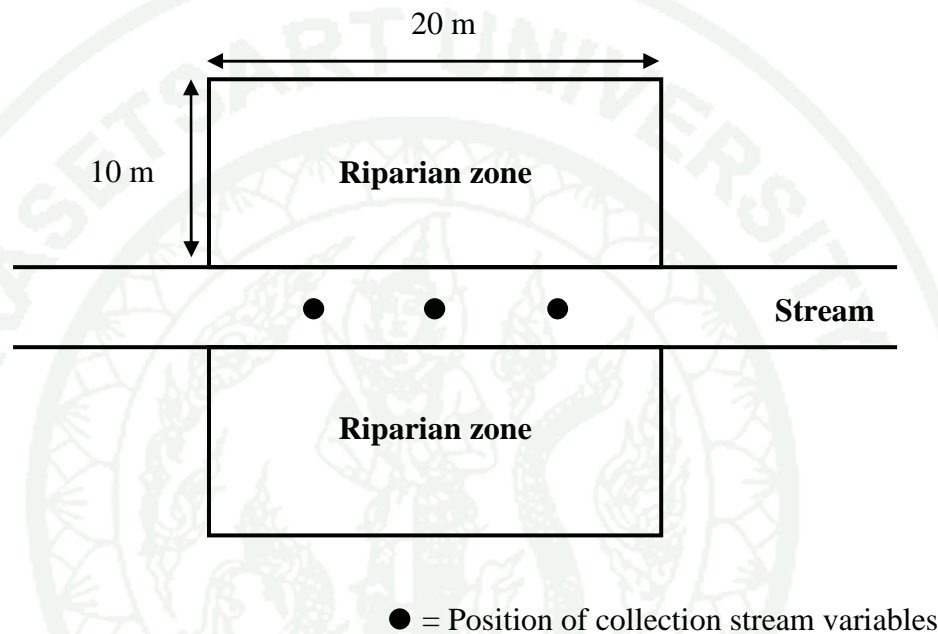


Figure 5 Detail of sampling plot in surveyed stream.

Environmental variables of stream and riparian area

Environmental variables of streams including water temperature, conductivity, DO, pH, velocity and stream depth are measured. I measured environmental variables within riparian areas, including percentage cover of understorey vegetation, humidity and air temperature. These variables were compared between stream characteristics, between habitat pattern and between seasons. I selected these variables because they were variables that determine water quality and environmental characteristic of organisms which inhabit in/near stream.

I measured the percentage cover of understorey vegetation from riparian vegetation under 1 m height in sampling plots with quadrat 1 x 1 m. The quadrat was

divided into four tables (50 x 50 cm). They were placed on the ground away from bank of the stream 2 and 6 m respectively (Figure 6). The percentage cover of each quadrat was calculated from average of percentage of each table using visual estimation.

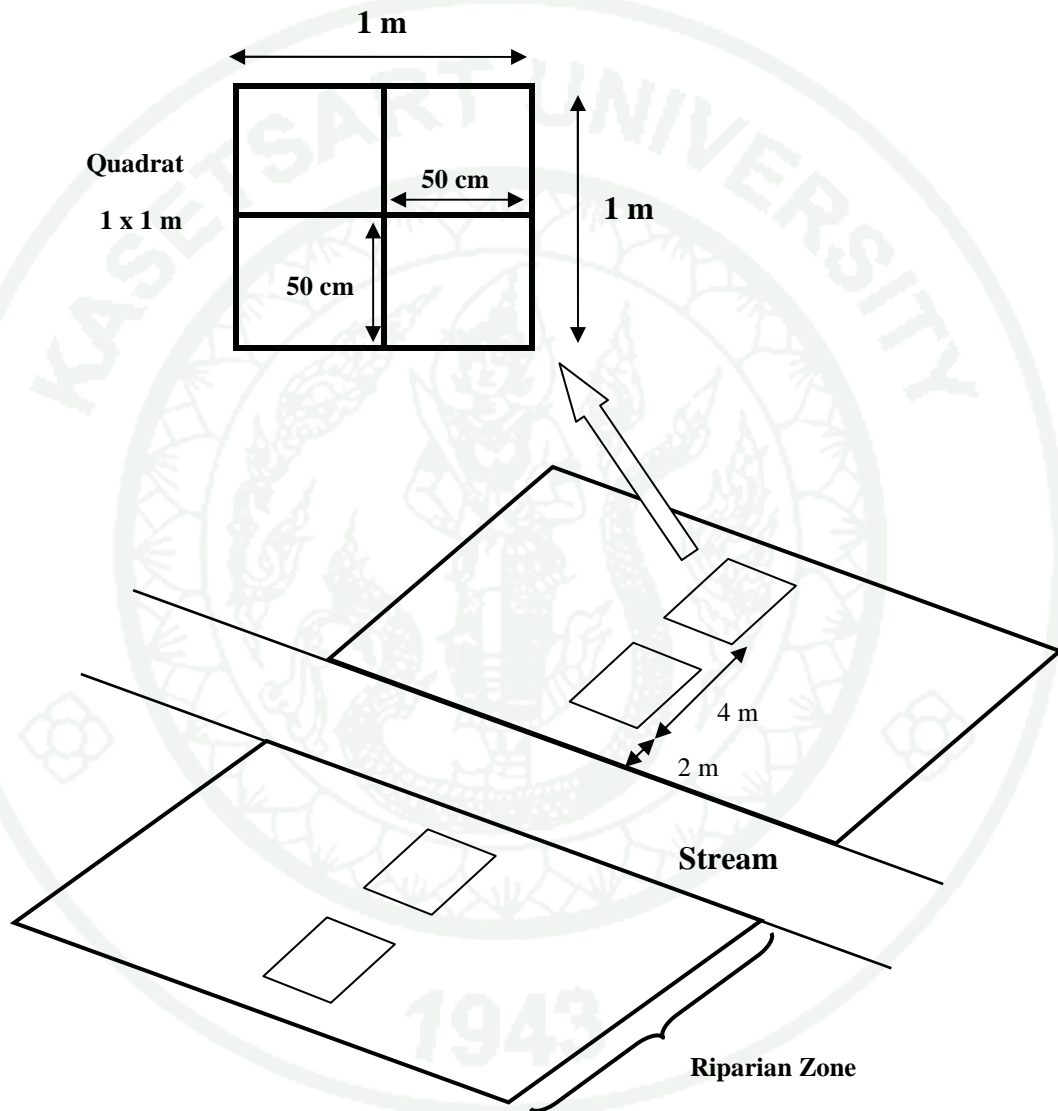


Figure 6 The position of quadrat 1 x 1 m sampling plots. They were center of riparian zone away from bank of the stream 2 and 6 m respectively.

Data analysis

1. Diversity indices

A diversity index is a mathematical measure of species diversity in a community. I used the Simpson's diversity index (D) (Equation 1). This is a dominance index because it gives more weight to common or dominant species and responds most strongly to changes in the proportional abundance of the most common species (Whittaker, 1965; Nagendra, 2002). In this case, a few rare species with only a few representatives do not affect the diversity. As species richness and evenness increase, so diversity increases.

$$D = 1 - \left(\sum \frac{n(n-1)}{N(N-1)} \right) \quad \text{----- (1)}$$

Where n = the total number of organisms of a particular species

N = the total number of organisms of all species

The value of D ranges between 0 and 1. With this index, 1 represents infinite diversity and 0, no diversity.

Simpson's Evenness Index (E_D) is a measure of the relative abundance of the different species making up the richness of an area and expresses how evenly the individuals are distributed among the different species, ranges from 0 (not even) to 1 (completely even) (Smith and Wilson, 1996) (Equation 2).

$$E_D = D/S \quad \text{----- (2)}$$

Where D = Simpson's diversity index

S = the number of species or species richness

2. Analysis of similarity of amphibian species

Comparing species composition and diversity of two or more species assemblage, similarity or dissimilarity indices provided quantitative bases of assessment (Magurran, 2004). I calculated the similarity between species using Sorensen's similarity index. This index measures similarity in species composition for two sites, A and B (Equation 3).

$$C_s = \frac{2ab}{a + b} \text{----- (3)}$$

where a = the number of species found in site A

b = the number of species in site B

ab = the number of species shared by the two sites.

Ranges from 0 (no species overlap) to 1 (complete overlap)

3. Analysis of relationship between species richness and sampling sites

Relationship between species richness and number of sampling sites was analyzed by species accumulation curve. The species accumulation curve (or sample-based rarefaction curve) showed the accumulation of species when increases the numbers of sites and shows the rate at which new species are found within a community and can be extrapolated to provide an estimate of species richness (Oksanen, 2011). Species accumulation curve was conducted by using R package *BiodiversityR* (Kindt, 2013; R Development Core Team, 2011). This package computes expected species accumulation curves and create the graph of the number of observed species as a function of some measure of the sampling effort required to observe them by using the analytic method developed by Kindt *et al.*, (2006) (Equation 4), with 95% confidence intervals

$$E(S_n) = \sum_{i=1}^S (1 - P_i) \quad \text{----- (4)}$$

where $P_i = \binom{N - f_i}{n} / \binom{N}{n}$

where $E(S_n)$ = The expected number of species $E(S_n)$ in n samples

N = the number of a total of sites

S = the total number of species encountered in the survey

f_i = the total number of sites where species i occurs

We can construct 95% confidence intervals with approximate variance estimator (s^2) that was developed by Jari Oksanen (Equation 5)

$$s^2 = p_i(1 - p_i) + 2 \sum_{i=1}^s \sum_{j>i} (r_{ij} \sqrt{p_i(1 - p_i)} \sqrt{p_j(1 - p_j)}) \quad \text{----- (5)}$$

where r_{ij} is the correlation coefficient between species i and j .

4. Analysis of difference of stream environmental variables

The difference of environmental variables of stream characteristics and habitat pattern (landscape characteristic) were tested through analysis of variance (ANOVA) and Kruskal-Wallis test. ANOVA is a statistical procedure for testing the hypothesis that the mean among two or more variables are equal, under the assumption that the sampled variables are normally distributed and have similar variances. Kruskal-Wallis test is a non-parametric ANOVA for testing the mean among variables that are not normally distributed and/or have difference of variances. The levels of significance of testing are .05

5. Analysis of species assemblage and correlation with environmental variable

Non-metric multidimensional scaling (NMDS) was used to analyze correlation between the amphibian assemblage and environmental variables of stream and between the amphibian assemblage and three stream characteristics in two seasons by the R package named “vegan” (Oksanen *et al.*, 2013). The NMDS was used to analyze this research because NMDS could describe species response to environmental factors associated with sites used by individual species (McCune and Grace, 2002), and provided a better fit to ecological data than may be obtained using other ordination techniques (Kenkel and Orloci, 1986).

5.1 Community dissimilarity

NMDS is an ordination method well suited to data that are non-normal or are on arbitrary, discontinuous and assumptions of data are non-linear relationships. NMDS can use ecologically meaningful ways of measuring community dissimilarity and a good dissimilarity measure has a good rank order relation to distance along environmental gradients (Oksanen, 2011). NMDS is a numerical technique that differs in several ways from nearly all other ordination methods which are analytical. In NMDS, a small number of axes are explicitly chosen prior to the analysis and the data are fitted to those dimensions; there are no hidden axes of variation and NMDS iteratively seek a solution and stops computation when an acceptable solution has been found, or it stops after some pre-specified number of attempts (Holland, 2008; Oksanen, 2011).

NMDS start with a matrix of data consisted in n rows of samples and p columns of variables, such as taxa for ecological data. All pairwise distances matrix among samples is calculated with dissimilarity measure. The NMDS ordination is performed on this distance matrix in species space (Holland, 2008; Oksanen, 2011). For this study, the distance was calculated by Bray-Curtis dissimilarity (D_{jk}) (equation 6).

$$D_{jk} = \frac{\sum_i |x_{ji} - x_{ki}|}{\sum_i (x_{ji} + x_{ki})} \quad \text{----- (6)}$$

where D_{jk} = Bray-Curtis dissimilarity

X_{ji} = the number of individuals of species i in sample j

X_{ki} = the number of individuals of species i in sample k

NMDS sought an ordination in which the possible distances between all pairs of samples were in rank-order agreement with their dissimilarities in species composition. The number of m dimensions was chosen for the ordination. NMDS began by constructing an initial configuration of the samples in the m dimensions. This initial configuration could consist of an entirely random arrangement of the samples. The final ordination was partly dependent on this initial configuration, thus approach was to perform several ordinations, each starting from a different random arrangement of points, and to select the ordination with the best fit (Holland, 2008).

The ordination is sensitive to the number of dimensions and the appropriate number of dimensions is the interpretability of the ordination (Holland, 2008). We need to find out the lowest number of NMDS axes that adequately represent the data and choosing few dimensions force multiple axes of variation to be expressed on a single ordination dimension (Holland, 2008). Thus we can use stress to assess the departure of the NMDS configuration from the actual data (Lau, 2008). The stress is a statistic of goodness of fit. It is a function of non-linear monotone transformation of observed dissimilarities and ordination distances (Oksanen, 2011). Stress can be calculated in several ways, with one of the most common being Kruskal's Stress (S) (equation 7)

$$S = \sqrt{\frac{\sum_{h,i} (d_{hi} - \hat{d}_{hi})^2}{\sum_{h,i} \hat{d}_{hi}^2}} \quad \text{----- (7)}$$

where d_{hi} is the observed dissimilarities between samples h and i , and \hat{d}_{hi} is ordination distances from the regression.

The value of stress is used for considering the goodness of fit: a small stress value indicates a good fitting solution, whereas a high value indicates a bad fit (Kruskal, 1964; Oksanen, 2011). Kruskal (1964) provided guidelines for the interpretation of the stress value with respect to the goodness of fit (Table 3)

Table 3 Stress and goodness of fit for NMDS

Stress	Goodness of fit
> .20	poor
.10	fair
.05	good
.025	excellent
.00	perfect

We found an appropriate number of dimensions by scree plot which could be used as a guide in deciding on the number of dimensions required and. The scree plot is a plot of minimum stress against number of dimensions and the stress rapidly decreases as we increased the number of dimensions (Rabinowitz, 1975). Furthermore, scree plot can identify the point beyond which additional dimensions do not substantially lower the stress value (Holland, 2008). The stress and scree plot were analyzed by R package *vegan* (using function *stressplot*) and package *ecodist* (using function *nmds*) respectively (Oksanen *et al.*, 2013; Lau, 2008).

5.2 Environmental interpretation

The ordination technique often uses to explain ecological characteristics of a species, or study site. The most used method of interpretation is to fit environmental vectors on to ordination, which allows quantification of the strength of relationships through the correlation coefficient (r^2) (Dargie, 1984).

The fitted vectors are arrows with the interpretation: (1) The arrow points to the direction of most rapid change in the environmental variable. This is called the direction of the gradient. (2) The length of arrow is proportional to the correlation between ordination and environmental variable. This is called the strength of the gradient. The fitting environmental vectors were analyzed by R package *vegan* (using function *envfit*). The function *envfit* can also add vectors into an ordination plot, and it tests the 'significance' of fitted vectors with permutation tests (Oksanen, 2004).

The printed output of function *envfit* gives the direction cosine of the vectors (NMDS1 and NMDS2) which are the coordinates of unit length arrows and r^2 gives the squared correlation coefficient. The axes should be scaled by the square root of r^2 . The significances ($Pr > r$), or *P*-values are based on random permutations of the data (Oksanen, 2011). For this study, I used 1000 random permutations.

RESULTS AND DISCUSSION

Results

Amphibians diversity in different stream substrate and habitat

I encountered amphibians 772 individuals in 11 species of one order, three families and six genuses, respectively as shown in Table 4.

Table 4 Amphibians species detected during survey in the dry and rainy seasons.

Order	Family	Genus	Species	
Anura	Ranidae	<i>Occidozyga</i>	<i>Occidozyga martensii</i>	
		<i>Fejervarya</i>	<i>Fejervarya limnocharis</i>	
		<i>Limnonectes</i>	<i>Limnonectes kuhlii</i>	
			<i>Limnonectes gyldenstolpei</i>	
		<i>Hylarana</i>	<i>Hylarana nigrovittata</i>	
		<i>Hylarana erythraea</i>		
	Mycrohylidae	<i>Microhyla</i>		<i>Microhyla berdmorei</i>
				<i>Microhyla fissipes</i>
				<i>Microhyla heymonsi</i>
				<i>Microhyla butleri</i>
Rhacophoridae	<i>Polypedates</i>	<i>Polypedates leucomystax</i>		

Description and distribution of amphibians

1. *Occidozyga martensii* Peters, 1867



Figure 7 *Occidozyga martensii*

Source: <http://commons.wikimedia.org/wiki/>

It is a short, thick frog with a broad head. Body is olive-grey to brown, sometimes with reddish or orange line down the back. Small bumps are widely separated across the skin. Head is short and rounded. Tongue is round, though that is difficult to discern in the field. Underside is paler than rest of body. Tadpoles are small and usually bury in the mud (Taylor, 1962; IUCN, 2012; AmphibiaWeb, 2013).

It primarily occurs in streamside puddles, seepages, lowland plains and other damp patches along streams and rivers, as well as rain puddles and small pools, in areas of forest on slight to moderate slopes, in areas of seasonal rainfall. Tadpoles apparently live largely buried in soft puddle substrate. It is not known from level lowland plains (where *O. lima* occurs). In China it has also been recorded from paddy fields (IUCN, 2012).

This study, *O. martensii* was found at rocky and sediment substrates in both seasons and the hill near grassland in the dry season and the hill near grassland and the lower slope area in the rainy season.

2. *Fejervarya limnocharis* Boie, 1835



Figure 8 *Fejervarya limnocharis*

Source: <http://commons.wikimedia.org/wiki/>

It is a medium-sized frog with a long narrow snout and raised bump on its back. A yellow vertebral stripe is seen in lines across the entire top of the body. Body is brown or gray with a yellow, tan, or green stripe down the middle that may be wide, narrow, or completely absent. Often has dark brown or black markings as well, especially on the lips. Underside is white. Skin warty above, the warts often confluent into irregular, longitudinal folds, a supratympanic fold from eye to axilla. Male with loose gular region, with brown or blackish W-shaped mark, fore limbs stronger, with pad like subdigital tubercles under first finger. Tadpoles are oval-shaped with a tail twice as long as the body. Body is brown or gray on top and silver on the bottom. (Taylor, 1962; AmphibiaWeb, 2013).

It inhabits most open wet habitat types, including river floodplains, wet agriculture areas such as rice fields, ditches, marshes, parks, gardens and other habitats and in closed-canopy forest (although this is rare in some regions). Its breeding and larval development take place in various wetland habitats (IUCN, 2012)

This study, *F. limnocharis* was found at all stream substrates and habitat patterns in both seasons.

3. *Limnonectes kuhlii* (Tschudi, 1838)



Figure 9 *Limnonectes kuhlii*

Source: <http://commons.wikimedia.org/wiki/>

It is a medium sized stocky frog. Adult males have a grotesquely broad head and two fangs in mouth. Both males and females with short arms and beary legs. The tips of toes and fingers are slightly swollen. The skin of the back and sides has many small bumps. The eardrum is not visible. The upper surface are brown or dark brown. The underside is usually white, purple is red. The unique of this species is adult male bigger than the females (Taylor, 1962; AmphibiaWeb, 2013).

This species is a lives along the edges of small, moderately fast streams with rocky bottom. It is found in small, clear streams in evergreen forest in hilly areas. It breeds in small tributary streams and the larvae develop in quiet pools along these streams. It does not wander from the vicinity of streams. In China it has also been found in still-water ponds and streams among fields (IUCN, 2012)

This study, *L. kuhlii* was found at all stream substrates and habitat patterns in both seasons.

4. *Limnonectes gyldenstolpei* (Boulenger, 1916)



Figure 10 *Limnonectes gyldenstolpei*

Source: <http://www.siamensis.org>

It has short and squat body, large and broad head. Head is distinctly wider than body. The males have a distinctive bump towards the back of the head. Eye is smaller than tympanum which is separated from eye by distance equal to length of eye. Foot has three-fourths webbed, tips of toes dilated into small but distinct terminal disc (Taylor, 1962).

It inhabits small streams in evergreen or dense deciduous forest. It breeds in streams by larval development (IUCN, 2012).

This study, *L. gyldenstolpei* was found at the sediment substrates and the lower slope area in the dry season and rocky substrates and the higher slope area in the rainy season.

5. *Hylarana nigrovittata* Blyth, 1856



Figure 11 *Hylarana nigrovittata*

This species exhibits the typical shape of the Ranidae family. It has a slender body, pointed snout, moderately large eyes, long hind legs and prominent external ear-drums. It is easily identified by the broad dark stripe which runs along each flank from the tip of the snout, through the eye to the base of the hind leg. The dorsal surface is medium brown, sometimes mottled, and the lower flank and belly are creamy yellow to white. The hind legs are patterned with dark, irregular bars or are mottled. The tympanum is distinct and a dorsolateral fold is present. Rictal and humeral glands are present as well (Taylor, 1962; Manthey and Grossmann, 1997).

It inhabits gentle streams in evergreen forest (including evergreen galleries in deciduous forest areas). Tadpoles live in quiet stream sections. It is generally not a commensal of humans, though it can be found close to settlements if the streams and surrounding forest are intact. It has occasionally been seen in drains in towns (IUCN, 2012).

This study, *H. nigrovittata* was found at all stream substrates and habitat patterns in both seasons.

6. *Hylarana erythraea* Schlegel, 1837



Figure 12 *Hylarana erythraea*

Source: <http://amphibia.my>

It is a medium-sized frog with smooth skin. Body is green, brown, or both, with a narrow cream-to-yellow stripe extending from behind each eye to the rear legs and a white stripe lower down along the sides. Upper stripe can sometimes have black edges. Head is long and narrow. Legs are especially long, with long half-webbed toes. Underside is white. Tadpoles have oval bodies and deep tails that taper to a tip. Their bodies and tails are green or brown with dark speckling, and the tails sometimes have a cream stripe. (Taylor, 1962; IUCN, 2012; AmphibiaWeb, 2013).

It is characteristic of vegetated floodplain ponds and is abundant in agricultural areas, ditches along roads, irrigation ponds, etc. It breeds only in stagnant waterbodies (IUCN, 2012).

This study, *H. erythraea* was found at all stream substrates in the dry season and the rocky and sediment substrates in the rainy season and found at the higher and lower slope area in the dry season and the lower slope area in the rainy season.

7. *Microhyla berdmorei* Blyth, 1856



Figure 13 *Microhyla berdmorei*

This species is a small frog but it has long legs and can make impressive jumps. The coloration is made up by shades of brown, that is brownish grey or pinkish above; a darker, light-edged, houghlass shaped mark on head and anterior part of the back, beginning between the eyes, sometimes continued on the hind part of the back; black spots on the sides of the limbs and occasionally on the flanks, the black spots on the limbs forming more or less indistinct cross-bars; a chevron-shaped black marking on the anal region. Note the conspicuous markings across the shoulder and between the eyes. Fingers bear small adhesive disks, more expanded ones are on the toe tips. Tympanum presents (Taylor, 1962; AmphibiaWeb, 2013).

It inhabits various types of moist evergreen forest, including monsoon and perennial rainforest types, being generally associated with hilly regions, and often found near streams. It also occurs in secondary growth. Breeding mainly takes place in still pools (IUCN, 2012).

This study, *M. berdmorei* was found at all stream substrates and habitat patterns in both seasons.

8. *Microhyla fissipes* Boulenger, 1884



Figure 14 *Microhyla fissipes*

It is a small, squat frog with a pointed nose that gives the entire body a triangular shape. Light brown, reddish-brown, or gray on top with an “ornate” dark marking on the back. The relative color and shape of the marking can be highly variable. Some individuals have a series of thin lines outside the markings that mirror it. Often has a light stripe on the side from the eye to the leg. Underside is creamy white. Tadpoles are nearly transparent with a “guitar” shape and a protruding mouth (Taylor, 1962; IUCN, 2012; AmphibiaWeb, 2013).

It occupies a number of habitat types including lowland scrub forest, grassland, agricultural land, pastureland and urban areas. Sub-fossorial in habit, it is also found in forest floor leaf-litter. It is mostly a nocturnal species that is only active diurnally during the rainy season. It breeds in temporary rain pools and other bodies of still water. It can occur in modified areas, such as non-intensively farmed agricultural land (IUCN, 2012).

This study, *M. fissipes* was found at all stream substrates in both seasons and found at the hill near grassland and the lower slope area in the dry season and the lower slope area in the rainy season.

9. *Microhyla heymonsi* Vogt, 1911



Figure 15 *Microhyla heymonsi*

Source: <https://aark.portal.isis.org>

It is a tiny squat frog with a pointed nose that gives the entire body a triangular shape. Light brown on top, sometimes with a thin light line going down the back and have other faint markings on the body and legs. Sides are characteristically dark, though sometimes the dark coloration only appears on the very top of the sides. Underside is creamy white. Tadpoles are approximately 1.5 cm long, with a “guitar” shape and a protruding mouth. They are dark in the middle and transparent elsewhere. (Taylor, 1962; IUCN, 2012; AmphibiaWeb, 2013).

It inhabits disturbed areas such as riverbanks, gardens, fire-maintained grassy areas, paddy fields and savannah forest as well as secondary vegetation. It breeds in temporary rain puddles, paddy fields, ditches, marshes and slow-flowing streams (IUCN, 2012).

This study, *M. heymonsi* was found at the rocky substrate in the dry season and the rocky and sediment substrate in the rainy season and found at the higher slope area in the dry season and all habitats in the rainy season.

10. *Microhyla butleri* Boulenger, 1900



Figure 16 *Microhyla butleri*

Source: <http://commons.wikimedia.org>

This species has a small head and long and slender hind limbs. Its back and sides are dark brown with a wavy yellowish stripe along each side of its back. Skin is smooth or with small smooth warts above. There is an oblique yellowish streak behind each eye and dark brown bands on the hind limbs. Tympanum is hidden (Taylor, 1962; Baker and Lim, 2008).

It is species of the forest edge, occasionally encountered on the forest floor of primary forest, but most often heard in massive choruses at forest edge puddles and pools. It is also known occasionally from plantations, tall shrublands and cultivated fields. It breeds in relatively permanent still waters, such as grassy pools, marshes, ponds and paddy fields in hilly areas (IUCN, 2012).

This study, *M. butleri* was found at the sediment substrate in both seasons and the hill near grassland and the lower slope area in both seasons.

11. *Polypedates leucomystax* Gravenhorst, 1829



Figure 17 *Polypedates leucomystax*

Source: <http://frogsforborneo.org>

It has the characteristic large head, flattish body, long legs and toes, and adhesive toepads of a treefrog. Body coloration is variable, but is usually brown or gray with a dark W-shaped mark on top of the head and a black stripe on the front of the side. Its eggs are laid next to ponds in a foamy mass glued to overhanging vegetation. Once hatched, the young tadpoles fall into the water below to start a new life. Tadpoles are up to 5 cm long with a tall tail-fin that narrows to a point. They are grey-green to brown on the top, silver on the bottom, and have a dark stripe on the side of the tail. The eyes are far to the sides and mouth is on the bottom (Taylor, 1962; IUCN, 2012; AmphibiaWeb, 2013).

It is a very adaptable opportunist and commensal, occurring from beach vegetation through all manner of human habitats (such as agricultural areas, ditches, artificial ponds and lakes, gardens, even in houses) and natural edge habitats to closed primary forest. It appears to be dependent on human activities to create suitable habitats. There might be separation among habitat axes among the component species in the *leucomystax* complex (IUCN, 2012).

This study, *P. leucomystax* was found at only the rocky substrate in the lower slope area in the rainy season.

Abundance of amphibians

I collected 179 individuals of 10 species in dry season (Appendix Table A2). Species encountered were *M. berdmorei*, *M. fissipes*, *H. nigrovittata*, *F. limnocharis*, *H. erythraea*, *M. heymonsi*, *M. butleri*, *L. kuhlii*, *L. gyldenstolpei* and *O. martensii*. Species which were observed all stream characteristics were *M. berdmorei*, *M. fissipes*, *H. nigrovittata*, *F. limnocharis*, *H. erythraea* and *L. kuhlii*. Species which were observed all habitat patterns were *M. berdmorei*, *H. nigrovittata*, *F. limnocharis*, and *L. kuhlii*. *M. berdmorei* was the most number of individuals ($n = 72$, 40.22%). The number of individual per sampling sites of all characteristics and habitats was shown in Figure 18 and 19 respectively.

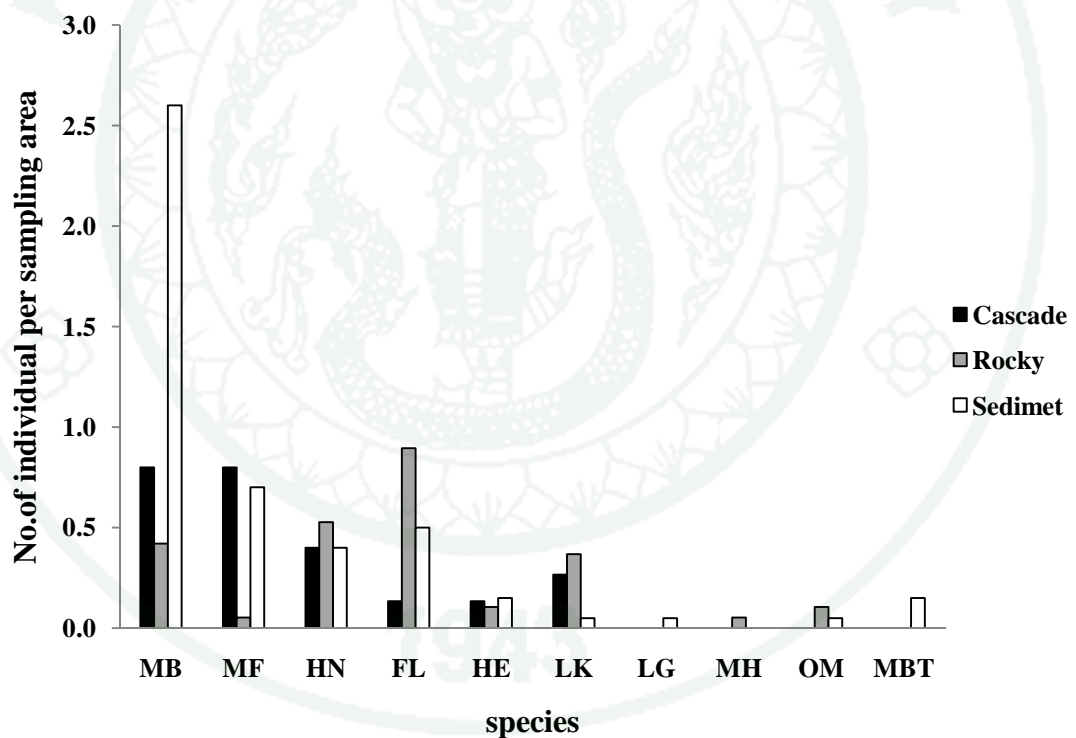


Figure 18 Number of individual per sampling sites in different stream substrate in dry season; cascade is the stream with cascade, rocky is the stream with rocky bed and sediment is the stream with sediment bed. Abbreviations of species were *M. berdmorei* (MB), *M. fissipes* (MF), *H. nigrovittata* (HN), *F. limnocharis* (FL), *H. erythraea* (HE), *L. kuhlii* (LK), *L. gyldenstolpei* (LG), *M. heymonsi* (MH), *O. martensii* (OM) and *M. butleri* (MBT).

According to species abundance, I classified all species into two groups. First, the group that was observed all stream characteristics was *M. berdmorei*, *M. fissipes*, *H. nigrovittata*, *F. limnocharis*, *H. erythraea* and *L. kuhlii*. Second, the group that was observed some characteristic was *H. erythraea*, *L. gyldenstolpei*, *M. heymonsi*, *O. martensii* and *M. butleri*.

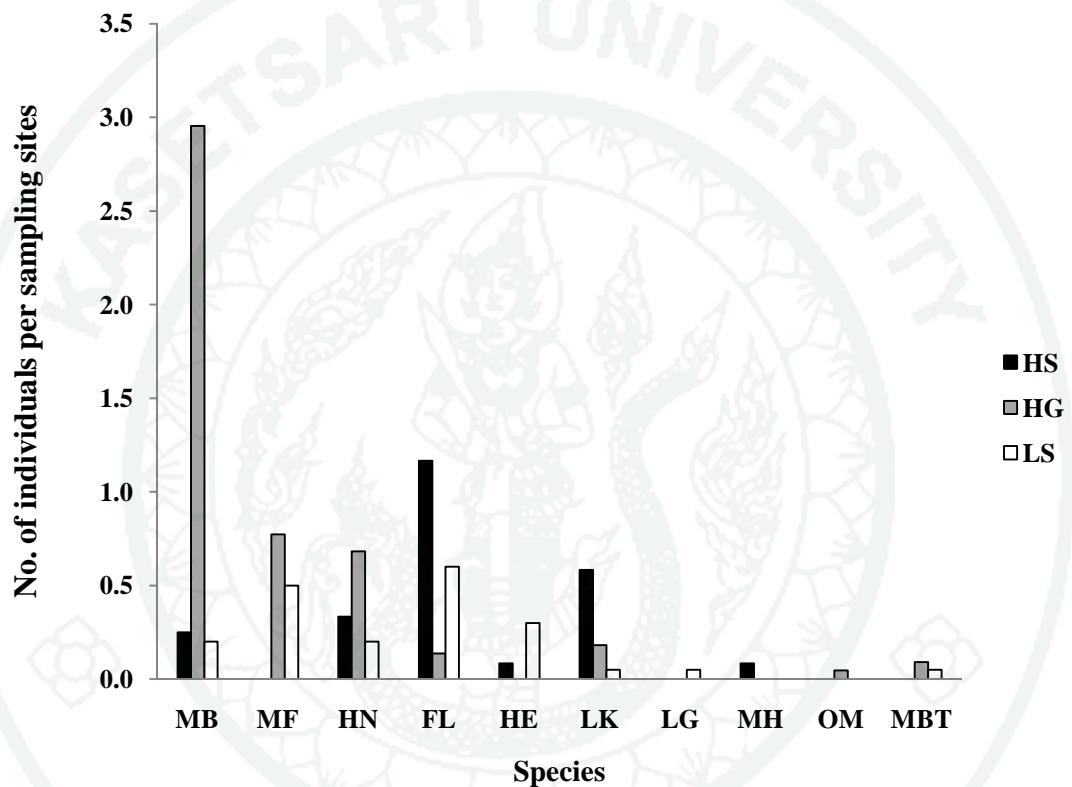


Figure 19 Number of individual per sampling site in different habitat patterns in dry season; HS is the higher slope area, HG is the hill near grassland and LS is the lower slope area. Abbreviations of species were *M. berdmorei* (MB), *M. fissipes* (MF), *H. nigrovittata* (HN), *F. limnocharis* (FL), *H. erythraea* (HE), *L. kuhlii* (LK), *L. gyldenstolpei* (LG), *M. heymonsi* (MH), *O. martensii* (OM) and *M. butleri* (MBT).

I classified all species into two groups. First, the group that was observed all habitats was *M. berdmorei*, *H. nigrovittata*, *F. limnocharis* and *L. kuhlii*. Second, the group that was observed some habitat was *M. fissipes*, *H. erythraea*, *L. gyldenstolpei*, *M. heymonsi*, *O. martensii* and *M. butleri*.

I collected 593 individuals of 11 species in rainy season (Appendix Table A5). I found *M. berdmorei*, *M. fissipes*, *H. nigrovittata*, *F. limnocharis*, *H. erythraea*, *M. heymonsi*, *M. butleri*, *L. kuhlii*, *L. gyldenstolpei*, *P. leucomystax* and *O. martensii*. Species which were observed all stream characteristics were *M. berdmorei*, *M. fissipes*, *H. nigrovittata*, *F. limnocharis* and *L. kuhlii*. Species which were observed in all habitat patterns were *M. berdmorei*, *H. nigrovittata*, *F. limnocharis* and *L. kuhlii*. *H. nigrovittata* was the most number of individual (n = 294, 49.58%). Figure 20 and 21 showed the number of individuals per sampling sites of all characteristics and habitats respectively.

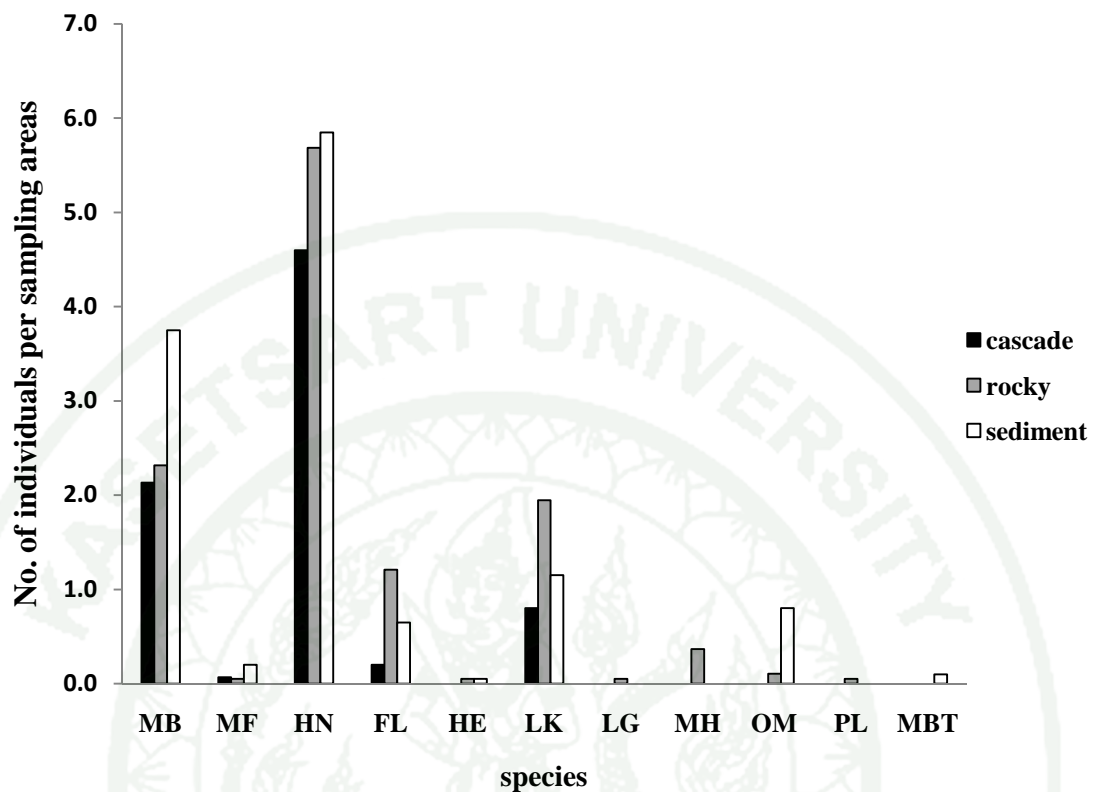


Figure 20 Number of individual per sampling sites in different stream substrate in rainy season; cascade is the stream with cascade, rocky is the stream with rocky bed and sediment is the stream with sediment bed. Abbreviations of species were *M. berdmorei* (MB), *M. fissipes* (MF), *H. nigrovittata* (HN), *F. limnocharis* (FL), *H. erythraea* (HE), *L. kuhlii* (LK), *L. gyldenstolpei* (LG), *M. heymonsi* (MH), *O. martensii* (OM) and *M. butleri* (MBT).

Resemble with dry season, I classified all species into two groups. First, the group that was observed all stream characteristics was *M. berdmorei*, *M. fissipes*, *H. nigrovittata*, *F. limnocharis* and *L. kuhlii*. Second, the group that was observed some habitat was *H. erythraea*, *L. gyldenstolpei*, *M. heymonsi*, *O. martensii*, *P. leucomystax* and *M. butleri*.

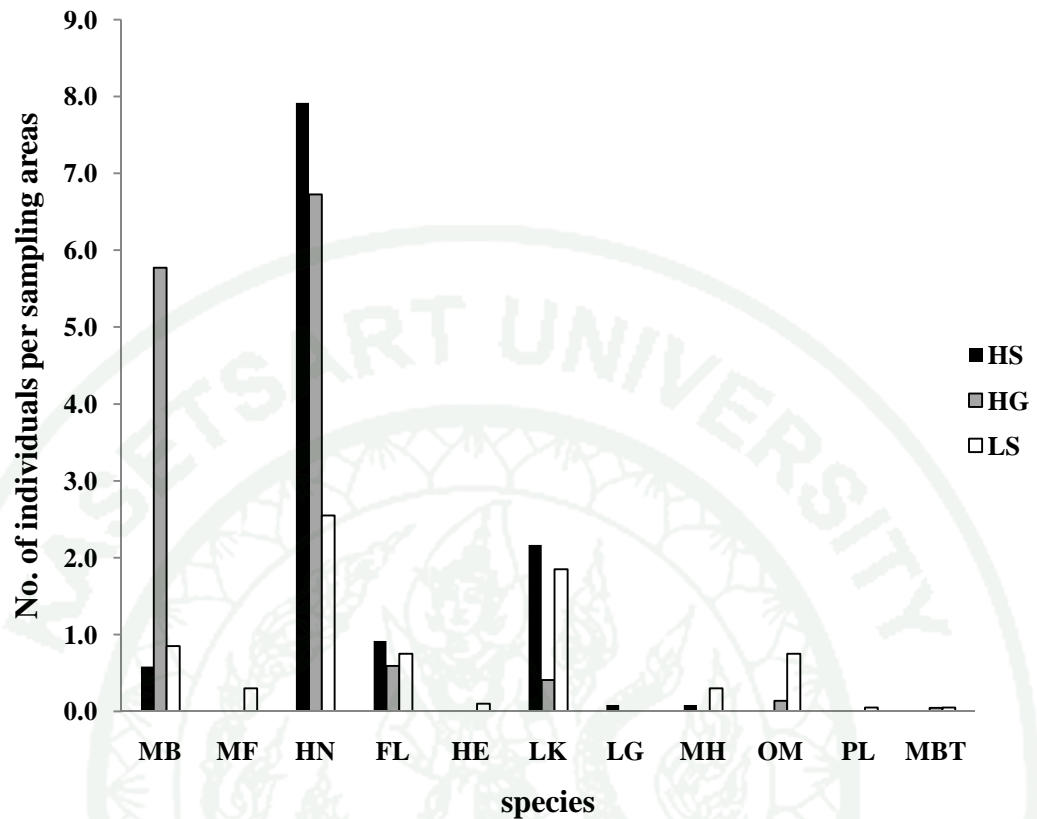


Figure 21 Number of individual per sampling site in different habitat patterns in rainy season; HS is the higher slope area, HG is the hill near grassland and LS is the lower slope area. Abbreviations of species were *M. berdmorei* (MB), *M. fissipes* (MF), *H. nigrovittata* (HN), *F. limnocharis* (FL), *H. erythraea* (HE), *L. kuhlii* (LK), *L. gyldenstolpei* (LG), *M. heymonsi* (MH), *O. martensii* (OM) and *M. butleri* (MBT).

I classified all species into two groups. First, the group that was observed all habitats was *M. berdmorei*, *H. nigrovittata*, *F. limnocharis* and *L. kuhlii*. Second, the group that was observed some habitat was *M. fissipes*, *H. erythraea*, *L. gyldenstolpei*, *M. heymonsi*, *O. martensii*, *P. leucomystax* and *M. butleri*.

Species richness of amphibians

Between two seasons, species richness in three stream characteristics is shown in Figure 22.

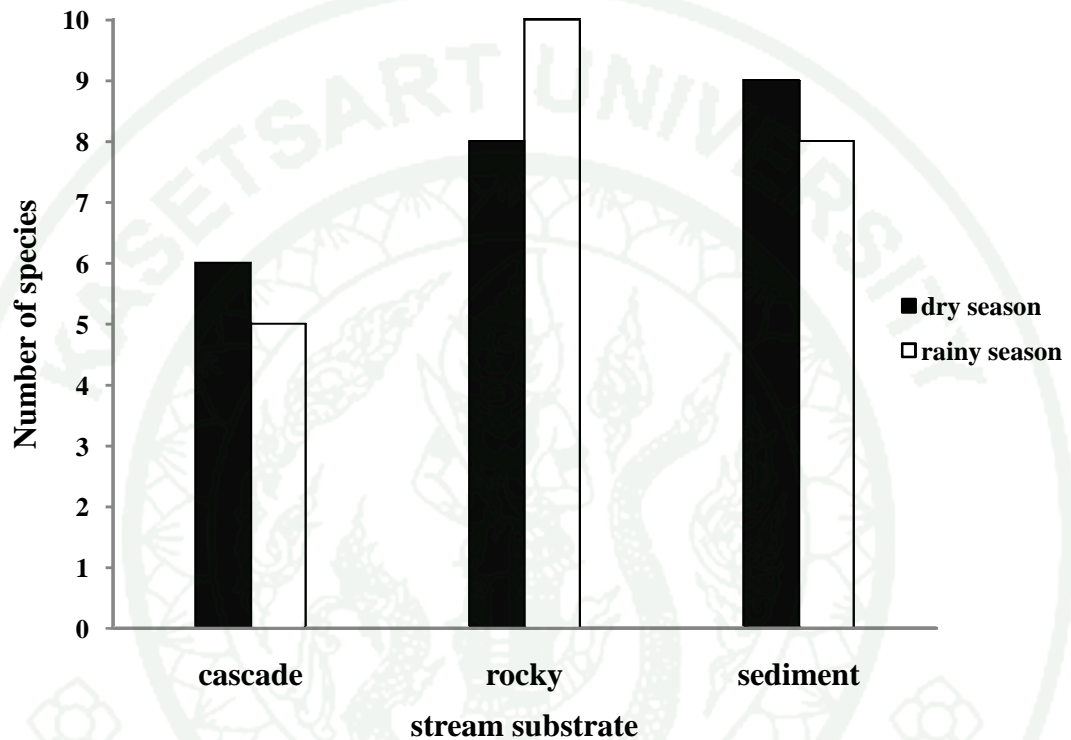


Figure 22 Species richness of amphibians in different substrate between two seasons; cascade is the stream with cascade, rocky is the stream with rocky bed and sediment is the stream with sediment bed.

In total, 10 and 11 species were collected to three stream substrates in rainy season and dry season respectively. No species richness between two seasons was equal. Species richness in stream with cascade and sediment was higher in the dry season than that in the rainy season, however, species richness in the stream with rocky was higher in the rainy season than that in the dry season.

Between two seasons, species richness in three habitat characteristics is shown in Figure 23.

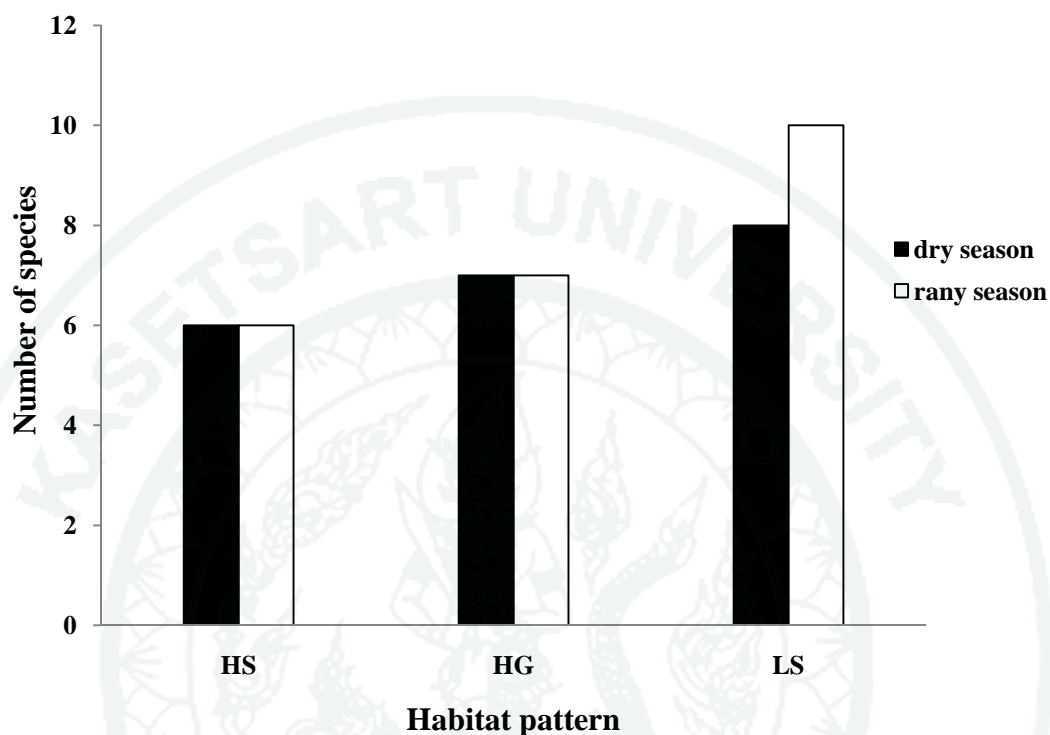


Figure 23 Species richness of amphibians in different habitat between two seasons; HS is the higher slope area, HG is the hill near grassland and LS is the lower slope area.

In total, 10 and 11 species were collected to three stream habitats in rainy season and dry season respectively. The majority of species richness between two seasons was equal, except lower slope area. Species richness at lower slope area was higher in the rainy season than that in the dry season.

The consideration in two seasons, The number of amphibian species which encountered in cascade, rocky and sediment substrate were 6, 10 and 10 species respectively and in the higher slope area, the hill near grassland and the lower slope area were 7, 8, 11 species respectively. The most number of species were encountered at lower slope area in two seasons, thus this habitat might support assemblage of stream-dwelling amphibians.

Comparison of amphibian abundance and species richness in the different stream substrate and habitat.

Total richness and abundance of amphibian in the different substrates and habitat pattern in the dry season is shown in Table 5 and 6 respectively.

Table 5 The number of species in different stream substrates and habitat patterns in the dry season.

		Number of species		
		Substrates		
Habitats		Cascade	Rocky	Sediment
	Higher slope area	4	6	1
	Hill near grassland	5	4	5
	lower slope area	1	4	8

The comparison of among substrates, the numbers of species in rocky substrate were approximate in the three habitats. In contrast, numbers of species in other substrates were higher in some habitats. This result indicated that species richness in rocky substrate did not depend on the different of habitats.

The comparison of among habitats, the numbers of species in hill near grassland were approximate in the three substrates. In contrast, numbers of species in other habitats were higher in some substrates. This result indicated that species richness in hill near grassland did not depend on the different of substrates.

Table 6 The number of individuals in different stream substrates and habitat patterns in the dry season. Values in parentheses are number of individuals per substrates.

		Number of individuals		
		Substrates		
		Cascade	Rocky	Sediment
Habitats	Higher slope area	6 (1.2)	23 (4.6)	1 (0.5)
	Hill near grassland	30 (4.3)	17 (2.8)	60 (6.7)
	lower slope area	2 (0.7)	5 (0.6)	32 (3.6)

The comparison of among substrates and habitats, the numbers of individuals in all substrates and habitats were higher in some habitats and some substrates respectively. These results indicated that abundance in all substrates and habitats depended on the difference of habitats and substrates.

Total richness and abundance of amphibian in the different substrates and habitat pattern in the rainy season is shown in Table 7 and 8 respectively.

Table 7 The number of species in different stream substrates and habitat patterns in the rainy season.

		Number of species		
		Substrates		
		Cascade	Rocky	Sediment
Habitats	Higher slope area	4	6	1
	Hill near grassland	4	4	6
	lower slope area	2	8	6

The comparison of among substrates, the numbers of species in cascade substrate were approximate in the three substrates. In contrast, numbers of species in other substrates were higher in some habitats. This result indicated that species richness in cascade substrate did not depend on the different of habitat, although species richness was less other.

The comparison of among habitats, the numbers of species in hill near grassland were approximate in the three substrates. In contrast, numbers of species in other habitats were higher in some substrates. This result indicated that species richness in hill near grassland did not depend on the different of substrates.

Table 8 The number of individuals in different stream substrates and habitat patterns in the rainy season. Values in parentheses are number of individuals per substrates.

		Number of individuals		
		Substrates		
		Cascade	Rocky	Sediment
Habitats	Higher slope area	48 (9.6)	90 (18.0)	3 (1.5)
	Hill near grassland	67 (9.6)	71 (11.8)	163 (18.1)
	lower slope area	2 (0.7)	64 (8.0)	85 (9.4)

The comparison of among substrates and habitats, the numbers of individuals in all substrates and habitats were higher in some habitats and some substrates respectively. These results indicated that abundance in all substrates and habitats depended on the difference of habitats and substrates.

Diversity index of amphibians

The diversity of stream amphibians was measured by Simpson's index. The diversity index of all characteristics and habitats are shown in Figure 24 and 25 respectively.

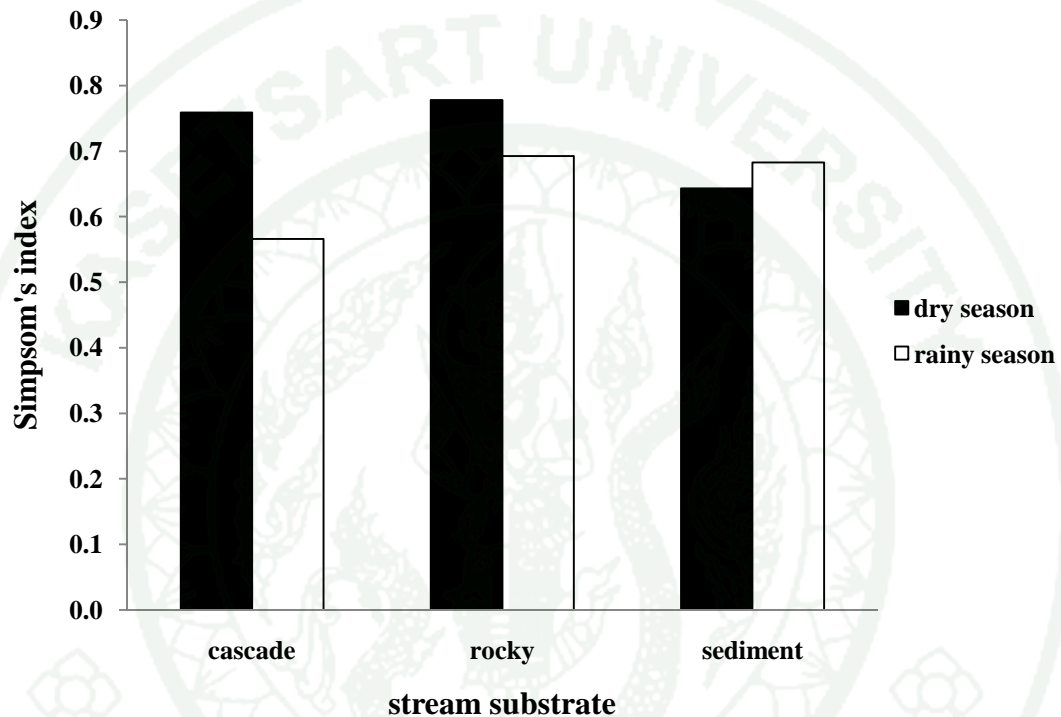


Figure 24 Simpson's index of amphibians in different substrates between two seasons; cascade is the stream with cascade, rocky is the stream with rocky bed and sediment is the stream with sediment bed.

Simpson's index of amphibians in cascade, rocky and sediment were 0.759, 0.778 and 0.643 respectively in the dry season and 0.566, 0.693 and 0.683 respectively in the rainy season (Figure 24). Simpson's index of amphibians in stream with cascade and with rocky bed in the dry season was higher than in the rainy season, except stream with sediment bed. Comparison of both seasons, the index was highest in rocky substrate since the species richness of this substrate was high and the numbers of individual of each species were approximate more than sediment when

compared. The result indicated that rocky substrate tended supporting diversity of stream amphibian.

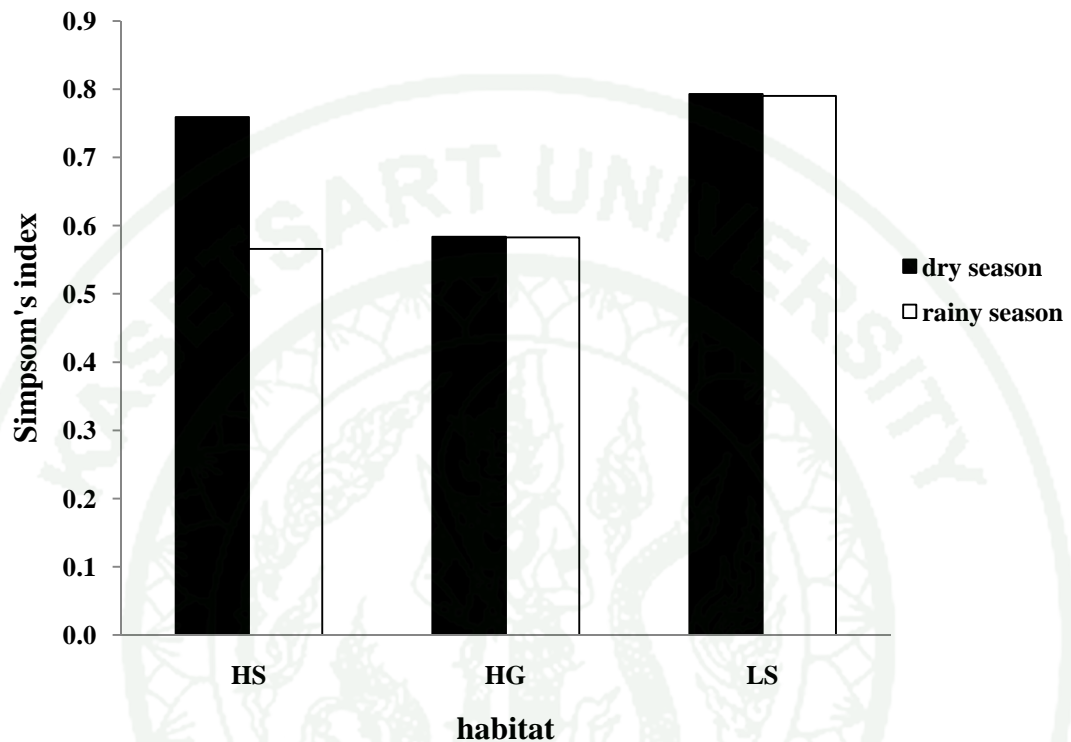


Figure 25 Simpson's index of amphibians in different habitats between two seasons; HS is the higher slope area, HG is the hill near grassland and LS is the lower slope area.

Simpson's index of amphibians in the higher slope area, the hill near grassland and the low slope area were 0.759, 0.584 and 0.793 respectively in the dry season and 0.566, 0.583 and 0.790 respectively in the rainy season (Figure 25). Simpson's index of amphibians in higher slope area was higher in the dry season than in the rainy season but other habitats were similar. Comparison of both seasons, the index was highest in the lower slope area since the species richness of this habitat was highest and the numbers of individual of each species were approximate more than other. The result indicated that the lower slope area tended supporting diversity of stream amphibian.

Evenness index of amphibians

The evenness index of stream amphibians was measured by Simpson's evenness index. The evenness index of all characteristics and habitats are showed in Figure 26 and 27 respectively.

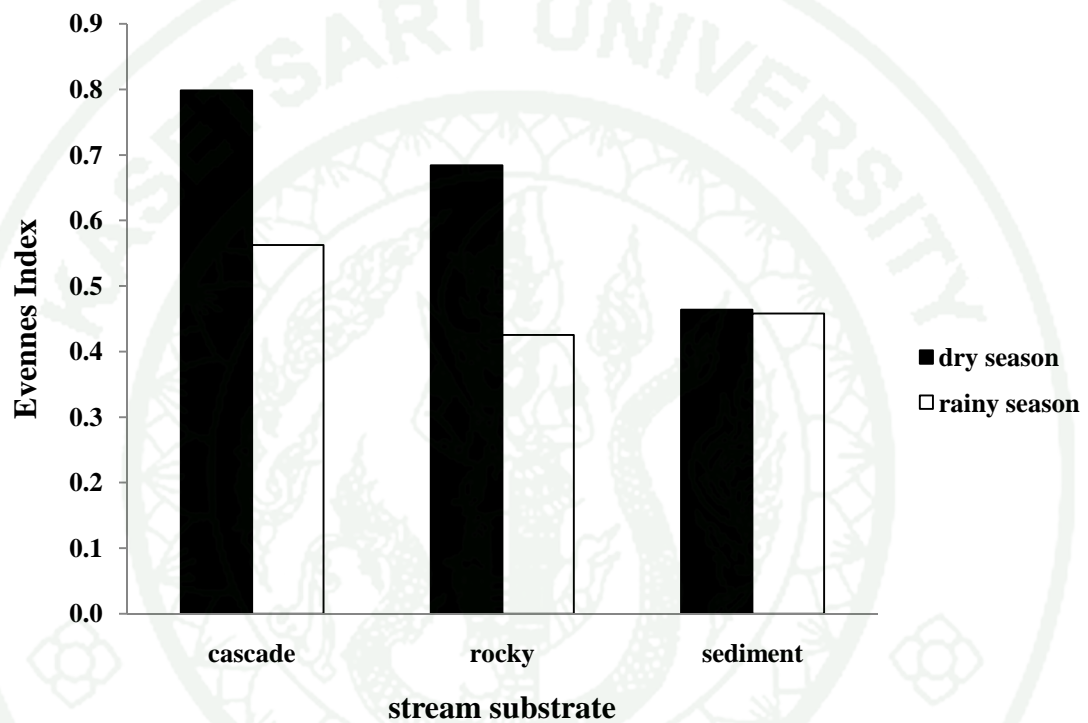


Figure 26 Simpson's evenness index of amphibians in different substrates between two seasons; cascade is the stream with cascade, rocky is the stream with rocky bed and sediment is the stream with sediment bed.

Simpson's evenness index of amphibians in cascade, rocky and sediment were 0.798, 0.684 and 0.464 respectively in the dry season and 0.563, 0.425 and 0.458 respectively in the rainy season (Figure 26). Evenness index of amphibians in cascade and rocky was higher in the dry season than in the rainy season, except sediment. Evenness index of cascade was higher than other both seasons. This result indicated that individuals are distributed evenly in stream with cascade.

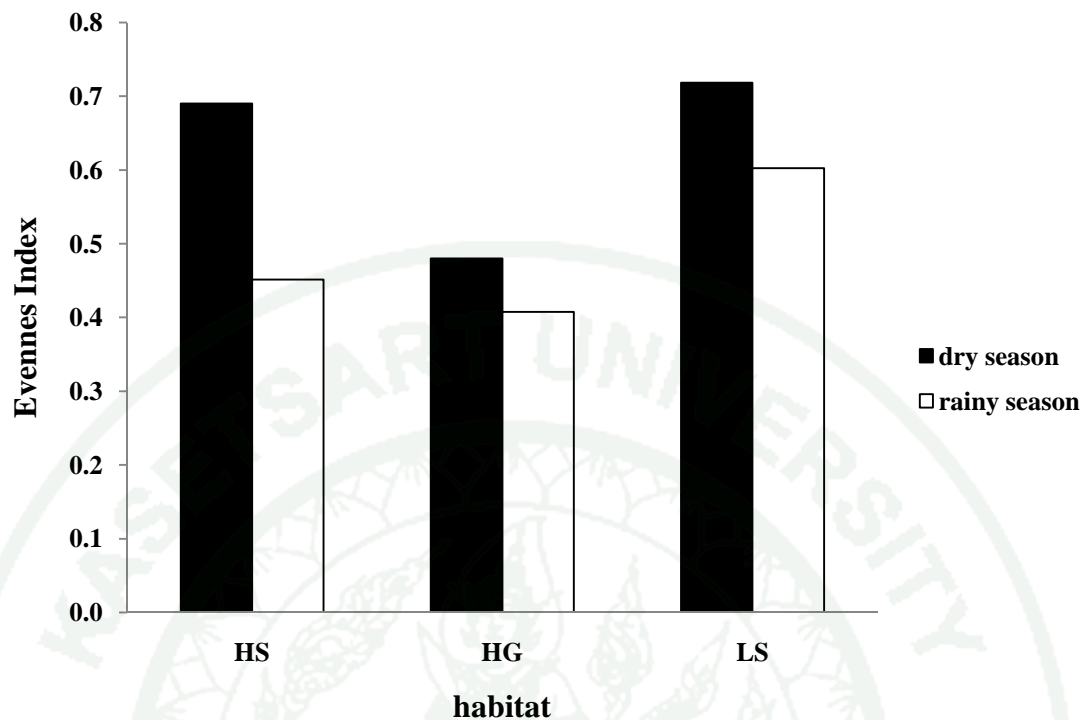


Figure 27 Simpson's evenness index of amphibians in different habitats between two seasons; HS is the higher slope area, HG is the hill near grassland and LS is the lower slope area.

Simpson's evenness index of amphibians in the higher slope area, the hill near grassland and the lower slope area were 0.690, 0.480 and 0.718 respectively in the dry season and 0.451, 0.408 and 0.603 respectively in the rainy season (Figure 27). Evenness index of amphibians in all habitats was higher in the dry season than in the rainy season. Evenness index of the lower slope area was higher than other both seasons. This result indicated that individuals are distributed evenly in the lower slope area.

Similarity of amphibian species

The comparison of similarity between stream characteristics and habitats was analyzed by Sørensen similarity index. The similarity index of pair of characteristics and habitats is shown in Table 9 and 10 respectively.

Table 9 Sørensen similarity index of amphibian between substrates; C is the stream with cascade, R is the stream with rocky bed and S is the stream with sediment bed.

Pair of substrates	Sørensen similarity index	
	Dry season	Rainy season
C-R	0.86	0.67
C-S	0.80	0.77
R-S	0.82	0.78

Sørensen similarity index revealed that the similarity of amphibian species between cascade and rocky substrates in the dry season and between rocky and sediment substrates in the rainy season were highest and the similarity of species was higher in the dry season than in the rainy season. This result indicated that species assemblage between substrates in the dry season was similar more than the rainy season.

Table 10 Sørensen similarity index of amphibian between substrates; HS is the higher slope area, HG is the hill near grassland and LS is the lower slope area.

Pair of habitats	Sorensen similarity index	
	Dry season	Rainy season
HS-HG	0.62	0.67
HS-LS	0.71	0.63
HG-LS	0.80	0.75

Sørensen similarity index revealed that the similarity of amphibian species between the hill near grassland and the lower slope area were highest in both seasons and the similarity of species was higher in the dry season than in the rainy season,

except between the higher slope area and the hill near grassland. This result indicated that species assemblage between habitats in the dry season was similar more than the rainy season.

Pattern of relationship between species richness and sampling sites

Relationship between number of species encountered and sampling efforts (number of sampling plots) in different substrates and habitats were examined by species accumulation curves and 95% confidence interval (CI) based on standard deviation (s) (calculated form square root of variance; s^2) in the dry season (Figure 28 and 29).

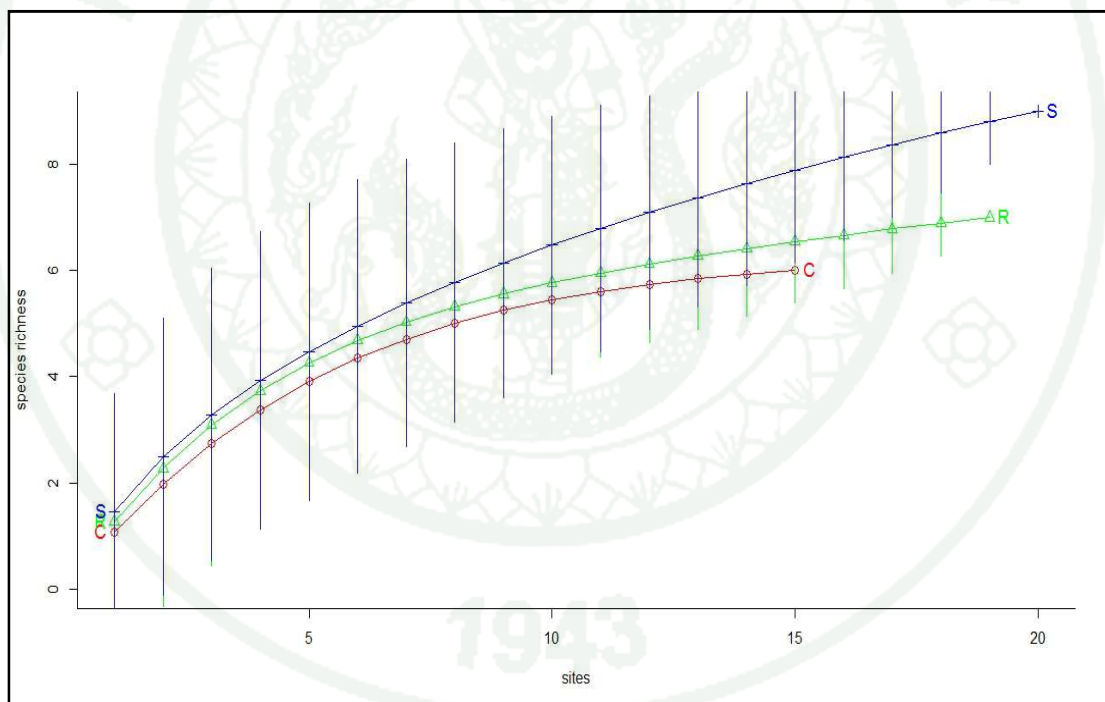


Figure 28 Species accumulation curves of amphibians in different substrates in the dry season; C is the stream with cascade, R is the stream with rocky bed and S is the stream with sediment bed. The bars indicate the 95% confidence interval (CI) based on standard deviation.

In the dry season, species richness in the cascade substrate was lower than in the rocky and sediment substrates. The species richness in the sediment and rocky increased to more number of sampling plot, but species richness tended to be stable in the cascade habitat. This result indicated that if sampling efforts increased, the probability of amphibian species encountered in the rocky and sediment substrates would increase in the survey.

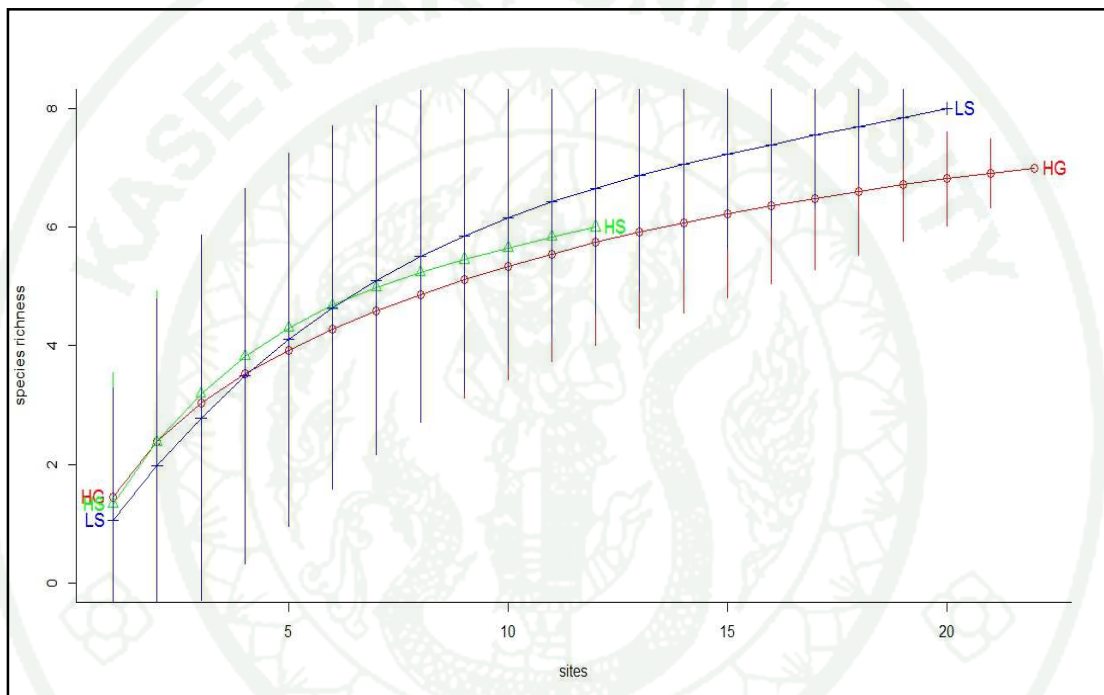


Figure 29 Species accumulation curves of amphibians in different substrates in the dry seasons; HS is the higher slope area, HG is the hill near grassland and LS is the lower slope area. The bars indicate the 95% confidence interval (CI) based on standard deviation.

In the dry season, species richness in the lower slope area was higher in the higher slope area and the hill near grassland. The species richness of all habitats increased to more number of sampling plot. This result indicated that if sampling efforts increased, the probability of amphibian species encountered in all substrates would increase in the survey.

Relationship between number of species encountered and sampling efforts (number of sampling plots) in different substrates and habitats were examined by species accumulation curves and 95% confidence interval (CI) based on standard deviation (s) (calculated from square root of variance; s^2) in the rainy season (Figure 30 and 31).

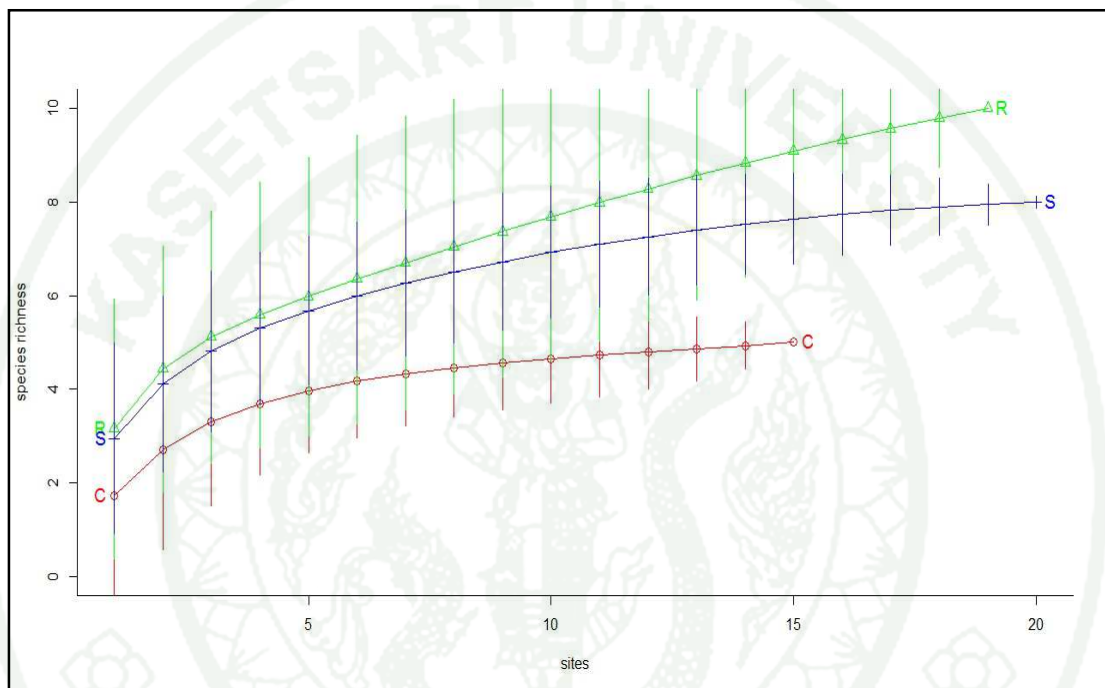


Figure 30 Species accumulation curves of amphibians in different substrates in the rainy season; C is the stream with cascade, R is the stream with rocky bed and S is the stream with sediment bed. The bars indicate the 95% confidence interval (CI) based on standard deviation.

In the rainy season, species richness in the rocky substrate was higher than in sediment and cascade substrates. The species richness in rocky increased to more number of sampling plot, but species richness tended to be stable in the sediment and cascade substrates. This result indicated that if sampling efforts increased, the probability of amphibian species encountered in the rocky substrates would increase in the survey.

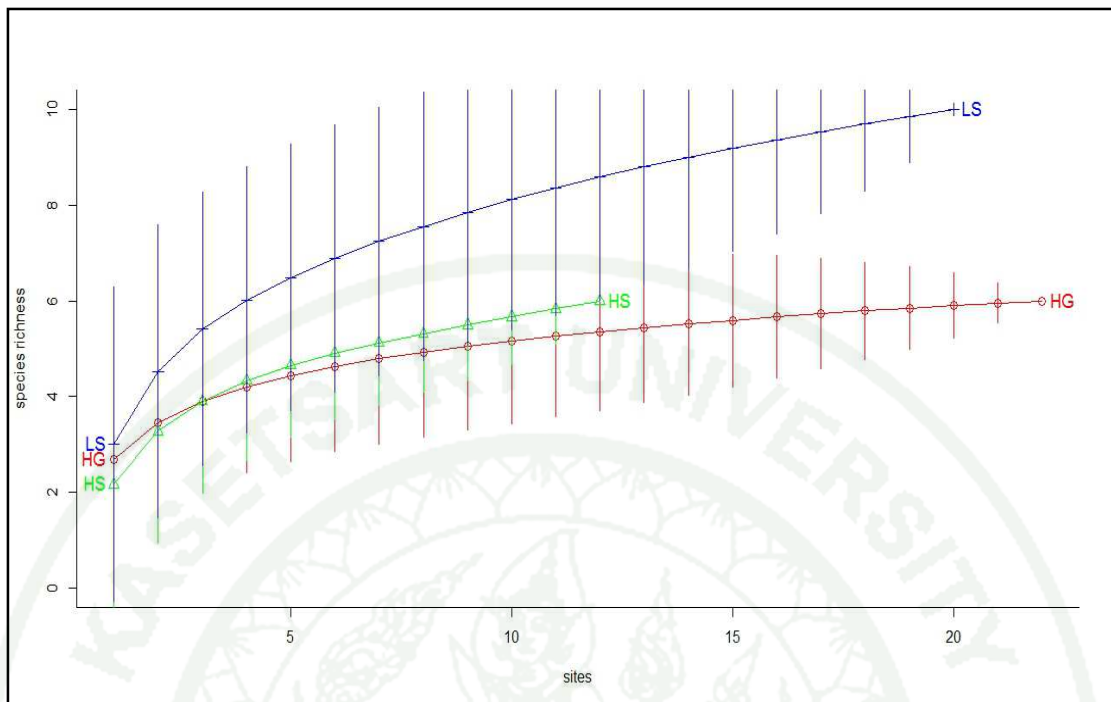


Figure 31 Species accumulation curves of amphibians in different substrates in the rainy seasons; HS is the higher slope area, HG is the hill near grassland and LS is the lower slope area. The bars indicate the 95% confidence interval (CI) based on standard deviation.

In the rainy season, species richness in the lower slope area was higher than in the higher slope area and the hill near grassland. The species richness in the lower and higher slope areas increased to more number of sampling plot, but species richness tended to be stable in the hill near grassland. This result indicated that if sampling efforts increased, the probability of amphibian species encountered in the lower and higher slope areas would increase in the survey.

Environmental variables of stream and riparian areas

In the dry season, the mean air temperature and humidity was 20.67 °C (SE = ± 0.41) and 49.36 % (SE = ± 0.74) respectively. The mean of stream variables, comprising of stream depth, pH, DO, conductivity, velocity and water temperature in all stream characteristics and habitat were shown in Table 11 and 12 respectively.

Table 11 Mean with standard error (\pm SE) of environmental variables between stream characteristics in the dry season; cascade is the stream with cascade, rocky is the stream with rocky bed and sediment is the stream with sediment bed.

Variables	Mean (\pm SE)			Test statistic	P-value
	Cascade	Rocky	Sediment		
% Cover	27.35 (± 3.03) ^a	24.18 (± 2.35) ^a	23.52 (± 2.29) ^a	0.891 [†]	0.350
pH	6.67 (± 0.10) ^a	6.50 (± 0.10) ^a	6.70 (± 0.09) ^a	0.072 [†]	0.790
DO (mg/l)	6.47 (± 0.15) ^a	5.96 (± 0.16) ^a	6.35 (± 0.16) ^a	0.121 [†]	0.729
Conductivity (μ S/cm)	69.13 (± 15.04) ^a	74.23 (± 10.97) ^a	67.02 (± 10.69) ^a	1.091 ^{††}	0.580
Velocity (Ft/s)	0.08 (± 0.03) ^a	0.02 (± 0.05) ^a	0.05 (± 0.05) ^a	5.050 ^{††}	0.080
Temperature (°C)	22.04 (± 0.20) ^a	22.53 (± 0.26) ^a	22.32 (± 0.25) ^a	0.438 [†]	0.511
Depth(cm)	18.49 (± 2.59) ^a	13.04 (± 1.09) ^{ab}	10.72 (± 1.06) ^b	10.550 [†]	0.002

* Values marked with ^a and ^b indicate the substrates that insignificantly differences between the variables were marked by the same signal.

[†]F values for ANOVA analysis

^{††}H value for Kruskal-Wallis test

The result of statistical analysis revealed that means of percentage cover of understory vegetation, pH, DO, conductivity, velocity and water temperature were not significantly different among characteristics at levels of significance are .05, except stream depth (Table 11). There are differences of mean of depths among characteristics by Turkey test. Depth between stream with cascade and stream with sediment bed differed significantly ($P = 0.005$) but depth between stream with cascade and stream with rocky bed and depth between stream with rocky bed and stream with sediment bed differed insignificantly ($P = 0.098$ and 0.453 respectively).

Table 12 Mean with standard error (\pm SE) of environmental variables between habitats in the dry season; HS is the higher slope area, HG is the hill near grassland and LS is the lower slope area.

Variables	Mean (\pm SE)			Test statistic	P-value
	HS	HG	LS		
% cover	29.89 (\pm 3.23) ^a	24.80 (\pm 2.74) ^a	21.79 (\pm 2.10) ^a	3.805 [†]	0.057
pH	7.14 (\pm 0.05) ^a	6.45 (\pm 0.08) ^b	6.49 (\pm 0.12) ^b	21.429 ^{††}	<0.001
DO (mg/l)	6.06 (\pm 0.14) ^a	6.48 (\pm 0.12) ^a	6.10 (\pm 0.17) ^a	0.030 [†]	0.864
Conductivity (μ S/cm)	114.81(\pm 17.78) ^a	41.02(\pm 1.67) ^b	48.78 (\pm 7.85) ^b	39.741 [†]	<0.001
Velocity (Ft/s)	0.02 (\pm 0.01) ^a	0.05 (\pm 0.03) ^a	0.05 (\pm 0.05) ^a	1.573 ^{††}	0.455
Temperature ($^{\circ}$ C)	22.71 (\pm 0.35) ^{ab}	21.85 (\pm 0.22) ^a	22.60 (\pm 0.19) ^b	0.038 [†]	0.846
Depth(cm)	14.05 (\pm 2.28) ^a	12.20 (\pm 1.21) ^a	15.12 (\pm 2.11) ^a	0.256 [†]	0.615

* Values marked with ^a and ^b indicate the habitats that insignificantly differences between the variables were marked by the same signal.

[†]F values for ANOVA analysis

^{††}H value for Kruskal-Wallis test

The means of percentage cover of understorey vegetation, DO, velocity and stream depth were not significantly different among habitats at levels of significance are .05 (Table 12). There are differences of means of pH, conductivity and water temperature among habitats by Turkey test. The pH and conductivity of study area between higher slope area and other area differed significantly, but between hill near grassland and lower slope area differed insignificantly (pH; $P= 0.743$ and conductivity; $P= 0.242$). P -value of pH and conductivity between higher slope area and hill near grassland are < 0.001 and between higher slope area and lower slope area are < 0.001 .

These result indicated that in the dry season, the different stream characteristics influenced stream depth but the different habitats influenced pH and conductivity. Since cascade often possessed pool habitat, thus cascade was deeper than other and pH and conductivity might be affected from the geology of the higher slope area.

In rainy season, the mean air temperature and humidity was 24.8°C ($\text{SE} = \pm 0.09$) and 68.85% ($\text{SE} = \pm 0.93$) respectively. The mean of stream variables, comprising of stream depth, pH, DO, conductivity, velocity and water temperature in all stream characteristics and habitat were shown in Table 13 and 14 respectively.

Table 13 Mean with standard error (\pm SE) of environmental variables between stream characteristics in the rainy season; cascade is the stream with cascade, rocky is the stream with rocky bed and sediment is the stream with sediment bed.

Variables	Mean (\pm SE)			Test statistic	P-value
	Cascade	Rocky	Sediment		
% cover	39.02 (\pm 3.72) ^a	36.35 (\pm 4.61) ^a	36.60 (\pm 3.89) ^a	0.131 [†]	0.719
pH	6.24 (\pm 0.13) ^a	6.15 (\pm 0.13) ^a	6.16 (\pm 0.08) ^a	1.220 ^{††}	0.543
DO (mg/l)	6.97 (\pm 0.31) ^a	7.29 (\pm 0.33) ^a	6.55 (\pm 0.20) ^a	1.449 [†]	0.234
Conductivity (μ S/cm)	31.85 (\pm 3.55) ^a	33.55 (\pm 3.70) ^a	33.01 (\pm 2.47) ^a	0.110 ^{††}	0.947
Velocity (Ft/s)	1.69 (\pm 0.17) ^a	1.60 (\pm 0.14) ^{ab}	1.18 (\pm 0.17) ^b	5.433 [†]	0.023
Temperature ($^{\circ}$ C)	22.98 (\pm 0.16) ^a	23.21 (\pm 0.21) ^a	23.60 (\pm 0.26) ^a	3.737 ^{††}	0.059
Depth(cm)	28.36 (\pm 3.28) ^a	25.90 (\pm 3.56) ^a	24.65 (\pm 2.06) ^a	0.248 [†]	0.621

* Values marked with ^a and ^b indicate the substrates that insignificantly differences between the variables were marked by the same signal.

[†]F values for ANOVA analysis

^{††}H value for Kruskal-Wallis test

The means of percentage cover of understory vegetation, pH, DO, conductivity, water temperature and stream depth were not significantly different among characteristics at levels of significance are .05 (Table 13). There are differences of mean of velocity among characteristics by Turkey test. Velocity between stream with cascade and stream with sediment differed significantly ($P=$

0.04) but velocity between stream with cascade and stream with rocky bed and velocity between stream with rocky bed and stream with sediment bed differed insignificantly ($P = 0.913$ and 0.133 respectively)

Table 14 Mean with standard error (\pm SE) of environmental variables between habitats in the rainy season; HS is the higher slope area, HG is the hill near grassland and LS is the lower slope area.

Variables	Mean (\pm SE)			Test statistic	P-value
	HS	HG	LS		
% cover	41.18 (\pm 4.14) ^a	39.31 (\pm 4.10) ^a	32.90 (\pm 3.58) ^a	1.934 [†]	0.171
pH	7.02 (\pm 0.07) ^a	6.02 (\pm 0.04) ^b	5.98 (\pm 0.06) ^b	49.213 [†]	<0.001
DO (mg/l)	7.65 (\pm 0.52) ^a	6.79 (\pm 0.24) ^a	6.73 (\pm 0.21) ^a	2.622 ^{††}	0.270
Conductivity (μ S/cm)	52.99(\pm 2.03) ^a	29.42(\pm 1.18) ^b	27.70(\pm 2.86) ^b	19.809 ^{††}	<0.001
Velocity (Ft/s)	1.87 (\pm 0.12) ^a	1.42(\pm 0.14) ^a	1.32(\pm 0.17) ^a	3.565 [†]	0.065
Temperature ($^{\circ}$ C)	23.09(\pm 0.10) ^a	23.16(\pm 0.17) ^a	23.57(\pm 0.27) ^a	2.071 ^{††}	0.355
Depth(cm)	25.53(\pm 3.10) ^a	20.84(\pm 1.67) ^a	31.97(\pm 3.25) ^a	3.385 [†]	0.072

* Values marked with ^a and ^b indicate the habitats that insignificantly differences between the variables were marked by the same signal.

[†]F values for ANOVA analysis

^{††}H value for Kruskal-Wallis test

The means of percentage cover of understory vegetation, DO, velocity, water temperature and stream depth were not significantly different among habitats at levels of significance are .05 (Table 14). There are differences of mean of pH and

conductivity among habitats by Turkey test. The pH and conductivity of study area between higher slope area and hill near grassland and between higher slope area and lower slope area differed significantly (pH; $P = < 0.001$ and conductivity; $P = < 0.001$) but between hill near grassland and lower slope area differed insignificantly (pH; $P = 0.798$ and conductivity; $P = 0.819$).

These results indicated that in the rainy season, the different stream characteristics influenced stream velocity but the different habitats influenced pH and conductivity. Since water volume in the rainy season was higher and overland flow of water and ground water from riparian area to stream body, thus stream velocity was faster in the rainy season. Furthermore pH and conductivity might be affected from the geology of the higher slope area.

The pattern of Correlation between environmental variables and species assemblage

Correlation between environmental variables and species assemblage were analyze NMDS. The value of stress was used for considering the goodness of fit of NMDS analysis and was plotted in the Shepard diagram (Appendix B1 and B3), stress in the dry season = 0.04 (Goodness of fit is good) and the rainy season = 0.09 (Goodness of fit is fair). Scree plot revealed that three dimensions were the appropriate numbers of dimensions for analysis in the dry season and stress value was lower than 0.2. The low stress value suggested a reasonable goodness of fit in the 2D configuration (Appendix B2 and B4). The difference in amphibian species composition in the dry season and the rainy season was shown in Figure 32 and 33 respectively.

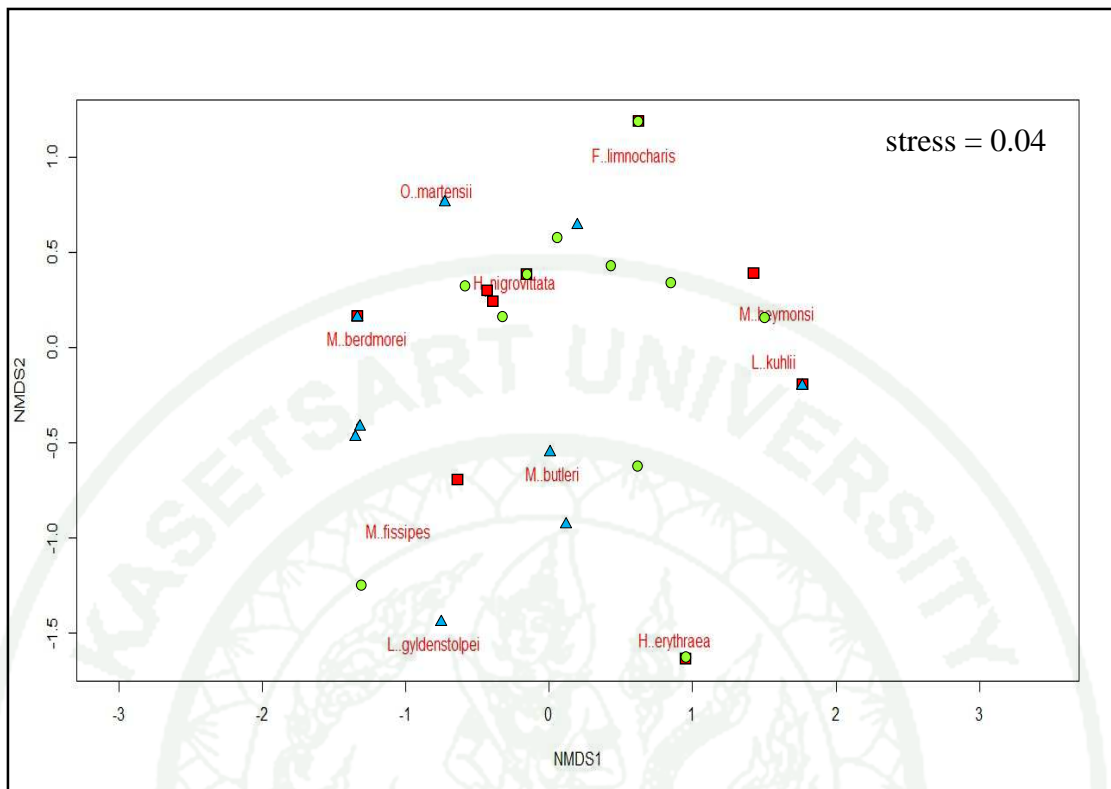


Figure 32 The result of NMDS on species abundance and stream characteristics in the dry season ; rectangular is the stream with cascade (red), circle is the stream with rocky bed (green) and triangle is the stream with sediment bed (blue).

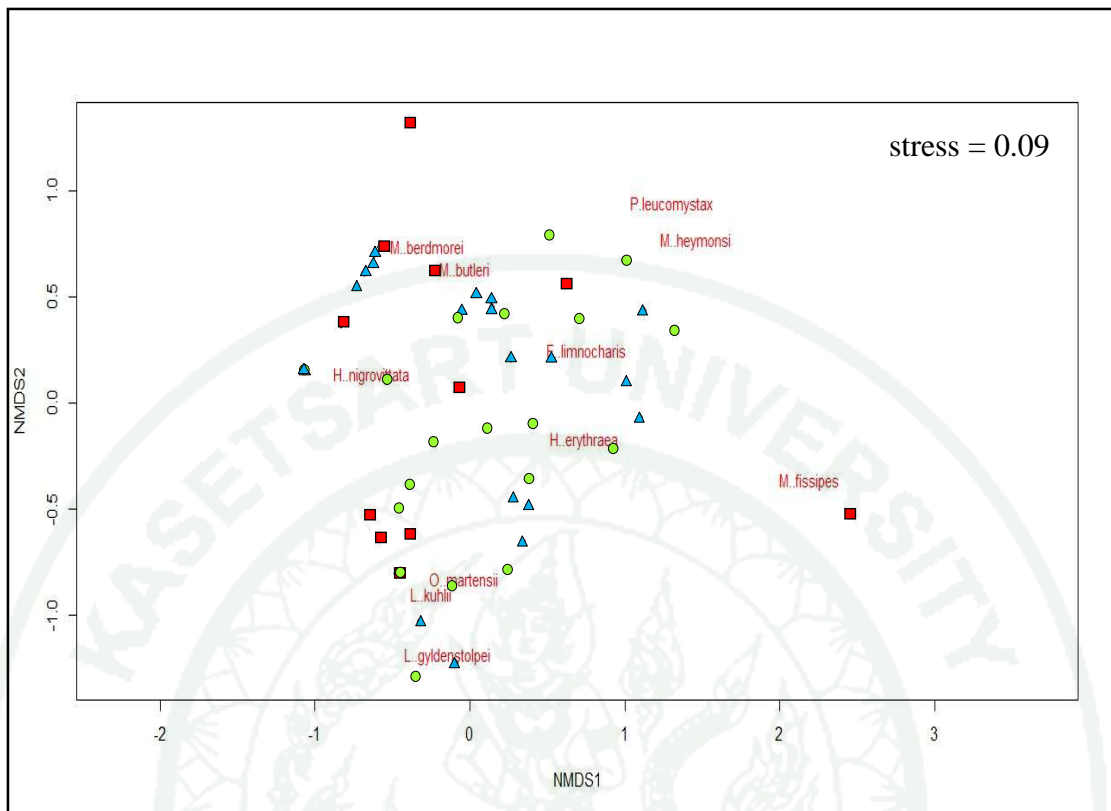


Figure 33 The result of NMDS on species abundance and stream characteristics in the rainy season; rectangular is the stream with cascade (red), cycle is the stream with rocky bed (green) and triangle is the stream with sediment bed.

NMDS revealed that the dissimilarity of amphibian species composition and abundance were higher the dry season than the rainy season because distance in species space of each point (site) in the dry season was more. The result indicated that the difference of species composition and abundance were more the dry season than the rainy season (Figure 32 and 33).

The correlation between environmental variables and species assemblage in the dry season was shown in Figure 34 and Appendix Table B1.

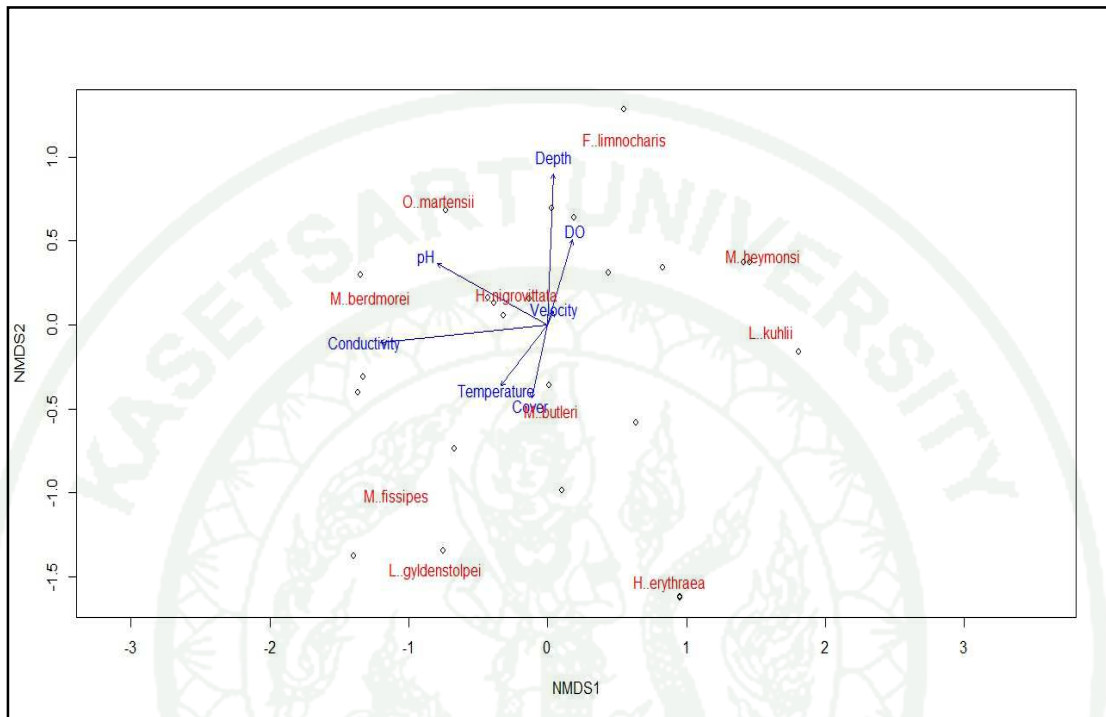


Figure 34 The first two axes of non-metric multidimensional scaling of species and environmental variables in dry season. Arrows are significant correlations of environmental variables. Lengths of arrows increase with correlation coefficient. Cycle is the sampling site.

Fit environmental vectors onto NMDS axes indicated that pH, conductivity, DO, water temperature and stream depth related to amphibian species composition in dry season ($P < 0.05$) (Figure 34 and Appendix Table B1). Stream velocity and percentage cover of understorey vegetation low related, indicated that these variables were not the important variable on the interpretation of correlation between environmental variables and species assemblage in the dry season.

Species which correlated with conductivity and pH were *M. berdmorei*, *O. martensii* and *H. nigrovittata*. Species which related with DO and stream depth were *F. limnocharis*. Species which related with water temperature were *M. butleri*, *L. gyldenstolpei* and *M. fissipes*.

The correlation between environmental variables and amphibians assemblage in the rainy season and were shown in Figure 35 and Appendix Table B2.

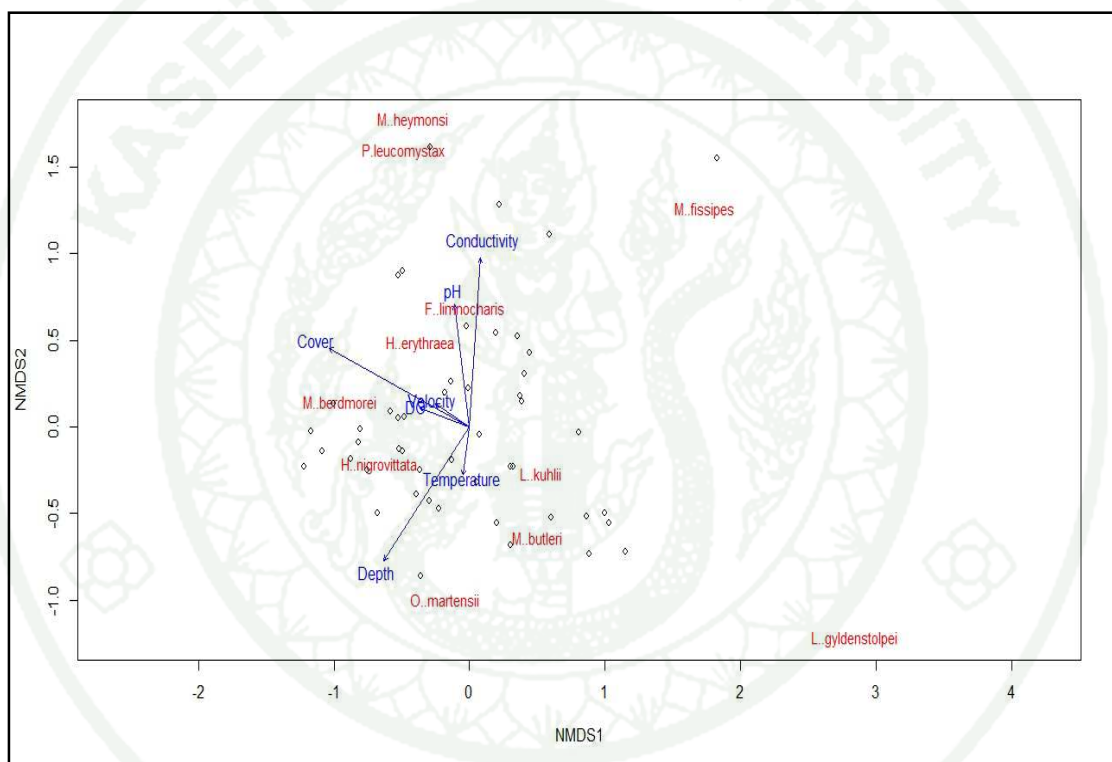
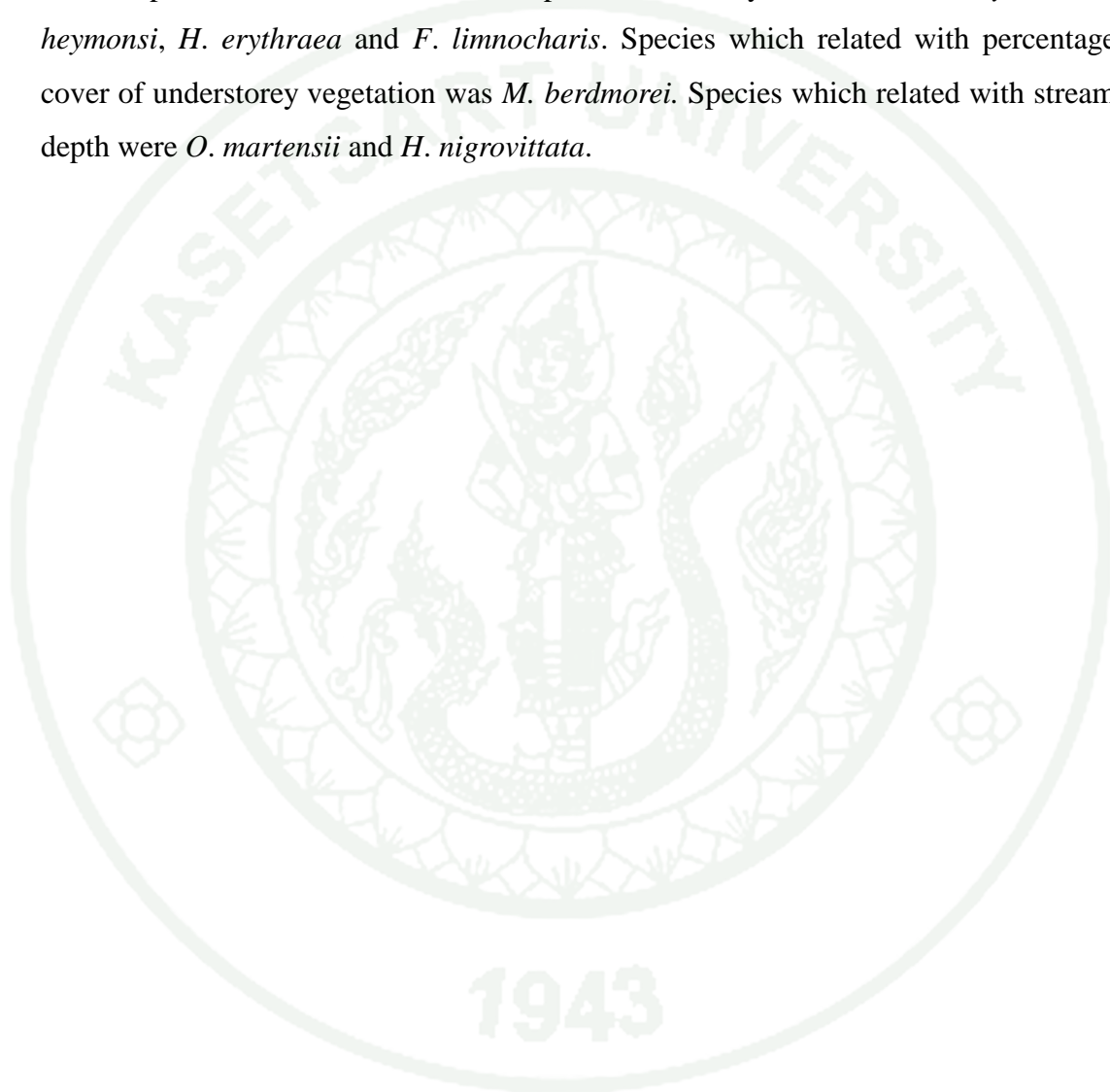


Figure 35 The first two axes of non-metric multidimensional scaling of species and environmental variables in rainy season. Arrows are significant correlations of environmental variables. Lengths of arrows increase with correlation coefficient. Cycle is the sampling site.

Fit environmental vectors onto NMDS axes indicated that pH, conductivity, stream depth and percentage cover of understorey vegetation related to amphibian species composition in the rainy season ($P < 0.05$) (Figure 35 and Appendix Table B2).

Stream velocity, DO and water temperature low related, indicated that these variables were not the important variable on the interpretation of correlation between environmental variables and species assemblage in the rainy season.

Species which correlated with pH, conductivity were *P. leucomystax*, *M. heymonsi*, *H. erythraea* and *F. limnocharis*. Species which related with percentage cover of understorey vegetation was *M. berdmorei*. Species which related with stream depth were *O. martensii* and *H. nigrovittata*.



Discussion

The relationship of stream and habitat characteristics and amphibian diversity

This study revealed that the difference of stream and habitat characteristics influenced species richness and abundance. Considering among substrates, the rocky and sediment substrates demonstrated to support species richness and the sediment substrate demonstrated to support abundance. The species richness and abundance was lower in the cascade. These indicated that amphibian population tended to assemble in streams which turbulence was lower. Considering among habitats, the lower slope area supported species richness but the hill near grassland support abundance. These results suggested that the amphibian community structure was determined by the different stream substrate and habitats and according to previous studies (Gradwell, 1971; Bury *et al.*, 1991; Welsh and Ollivier, 1998; Wilkins and Peterson, 2000; Wood and Richardson 2009). Furthermore our data revealed that amphibian diversity was sensitive to occurrence of some species. I found that pattern of overall amphibian relative abundance in sample sites were strongly influenced by the distribution of the two dominant species, including *H. nigrovittata* and *M. berdmorei*, which relative abundance were higher than other species in all substrates and habitats and most in the rainy season. Since two species could adapt well to various habitat and disturbed areas (IUCN, 2012), thus they were encounter in all stream and habitat characteristics. This result was according to the study of Gardner *et al.* (2007) that found that patterns of overall amphibian relative abundance were influenced from the distribution of dominant species (*Phrynobatrachus* spp.)

The diversity index revealed that rocky substrate supported amphibian diversity in both seasons and from analyzed by species accumulation curves, this substrate tended to support species richness of stream amphibian and according with other studies (Gradwell, 1971; Bury *et al.*, 1991; Wilkins and Peterson, 2000). Thus rocky substrate was the important habitat to diversity of stream amphibian.

The study revealed that the different habitat pattern influence diversity amphibian stream. The overall amphibian abundance was higher in the hill near grassland in both seasons. Despite if considering each species, no habitat pattern influence abundance of all species but have been influence relative abundance of dominant species which were higher in the hill grassland. This result was promoted by report of IUCN (2012) which habitat of *M. berdmorei* associated with hilly regions. Species richness was higher in the lower slope area and this result was supported by diversity index and species accumulation curves. Furthermore this result was consistent with study of Vilella *et al.* (2004) which found species richness and diversity index of aquatic megafauna (amphibians, crustaceans and fishes) in a first order stream in the higher slope area lower than the lower slope area. Thus amphibian species tended to assemble and diversity in the lower slope area more than other.

The result from comparing between season, species abundance and richness are higher rainy season than dry season since amphibian assemblage in the rainy season was higher. These was contrast to other studies that found species richness and abundance were higher in the dry season than in the rainy season for stream amphibian (Khonsue, 1996; Kongjaroen, 2007) and leaf litter amphibian (Lieberman 1986). Since the patterns of amphibian abundance of this study was influenced from the two dominant species. *H. nigrovittata* highly assembled in the rainy season since they had reproductive activity (observed from calling) in all streams and habitats, especially in nocturnal and they were reported that were the most of abundance in stream of Khao Yai National Park by Kongjaroen (2007)), thus amphibian abundance in this study was higher in the rainy season. This result was supported with other observers that found the most of amphibian abundance in particular during peaks of heavy rainfall (Duellman, 1995) which related to reproductive activity in the rainy season. Furthermore the similarity index suggested that similarity of species between substrates and habitats in the dry season were higher since specific assemblage among habitats in the rainy season highly occurred. Thus the difference of substrates and habitat explicitly influenced amphibian diversity and assemblage in the rainy season.

The pattern of amphibian assemblage between seasons could be interpreted from comparing with study of Kongjaroen (2007). Since this study main sampling areas were headwaters of Lam Ta Klong stream and were near this stream (range: 20 – 2000 m) and the result from the study of Kongjaroen (2007) amphibian diversity of Lam Ta Klong stream that found species abundance and richness were higher in the dry season than in the rainy season, thus these suggested that in the dry season, population of many amphibian tended to assemble in large streams and returned to headwater stream in the rainy season because velocity and turbulence of large stream was higher, not suitable on breeding and laying.

Environmental variation and assemblage structure in forest stream

ANOVA revealed that the timing of sampling period did not influence environmental variables in the spatial location of the study sites, except stream depth and velocity. The stream depth, velocity differed significantly between stream substrates in the dry and rainy season respectively. These indicated that the variation of water volume and speed of the water flow occurred from the seasonal variation. The different habitat pattern strongly influenced pH and conductivity. These variable were higher the higher slope area than other area. The pH and conductivity was distinctly higher in the higher slope area. Differences in conductivity might indicate differences in hydrologic source. Higher conductivity (more dissociated ions) suggested that the pool was likely receiving surface run-off or groundwater input (Korfel *et al.*, 2009) and this study cascade which often possessed pool habitat had more than in the higher slope area, thus conductivity was higher.

NMDS revealed that dissimilarity of amphibian species composition was higher the dry season than rainy season. This result suggested that amphibian assemblage in the dry season differed clearly in each site. This difference also reflected that the difference of substrate did not influence composition of the amphibian assemblage since could not show a distinct clustering of each substrate plots.

The fit environmental vectors revealed that some environmental variables correlate amphibian assemblage, for example pH, conductivity, DO, stream depth and water temperature in the dry season and pH, conductivity, percentage cover of understorey vegetation and stream depth in the rainy season. This result was consistent of previous studies suggested that environmental factors of stream and riparian area determined the composition of amphibian assemblage (Parris and McCarthy, 1999; Parris, 2004; Keller *et al.*, 2009; Souza and Eterovick, 2010) and of other assemblage, such as black fly (Pramual and Kuvangkadilok, 2009). Despite if consider between seasons, these variables did not show correlation with assemblage of either species in both seasons. These results suggested that the correlation of stream variables and species assemblage occurred only in any season.

CONCLUSION AND RECOMMENDATION

Conclusion

In conclusion, I encountered 10 and 11 species in the dry season and the rainy season respectively of one order, three families and six genus, including *H. nigrovittata*, *H. erythraea*, *M. berdmorei*, *M. fissipes*, *O. martensii*, *F. limnocharis*, *M. heymonsi*, *L. kuhlii*, *L. gyldenstolpei*, *M. butleri* and *P. leucomystax* (found only rainy season). *M. berdmorei* and *H. nigrovittata* is the most number of individuals in dry season and rainy season respectively. Species richness and abundance in the difference of stream characteristics and habitat pattern differ and explicit in the rainy season. In contrast, the amphibian diversity was higher in the dry season since influence from the distribution of two dominant species (*M. berdmorei* and *H. nigrovittata*) especially the rainy season, thus amphibian assemblage in the different stream characteristic and habitat pattern occurred distinctly in the rainy season more than the dry season.

The some stream environmental variables correlated the amphibian assemblage but not tend to bear similar assemblage between both seasons (shown by NMDS), thus ignoring of influence between seasons may lead to false ecological conclusions. Furthermore the diversity index and species accumulation curve revealed that amphibian assemblage was supported by some stream substrate and habitat pattern, these were the rocky substrate and the lower slope area. Thus these habitats indicated support to be beta diversity

Recommendation

Since stream with rocky bed and lower slope area demonstrated the support of amphibian diversity. Thus result of this study will induce to the conservation planning of these stream and habitat in future.

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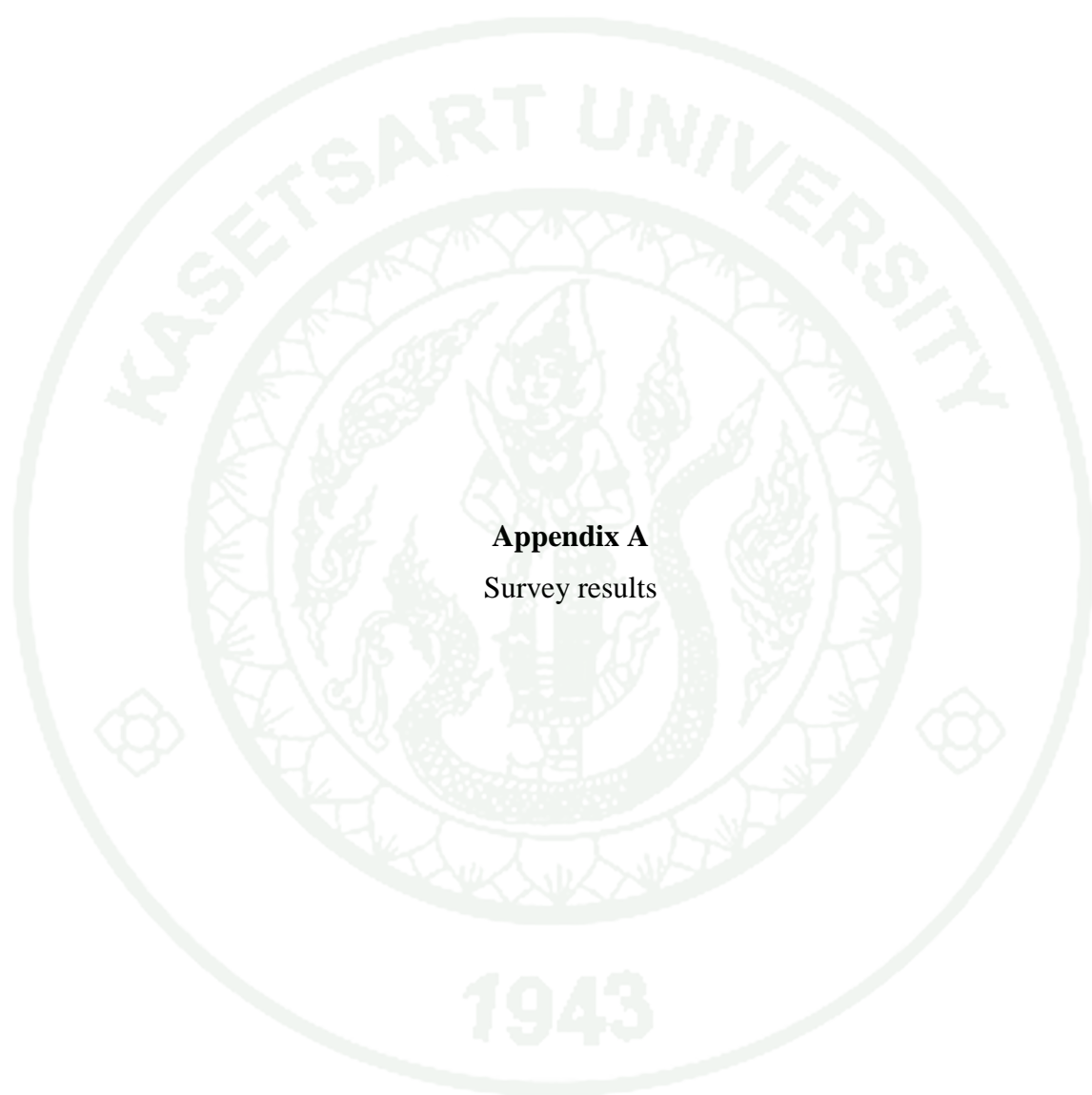
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APPENDICES



Appendix A
Survey results

Appendix Table A1 Geographic coordinates of sampling sites; A is the sampling plot in the higher slope area, B is the sampling plot in the hill near grassland and C is the sampling plot in the lower slope area

Site	UTM		elevation
	East	North	
A01	759047	1597162	635
A02	759096	1597094	648
A03	759313	1597031	654
A04	759418	1597030	653
A05	759444	1597038	682
A06	760786	1596658	665
A07	760822	1596674	669
A08	760877	1596682	672
A09	759583	1596800	617
A10	759840	1596671	674
A11	759947	1596765	673
A12	760112	1596742	670
B01	755606	1598994	731
B02	755472	1598983	754
B03	755425	1598954	763
B04	754781	1599558	757
B05	754727	1599595	765
B06	754385	1599818	765
B07	754340	1599754	765
B08	754522	1598814	720
B09	754479	1598988	732
B10	754599	1599036	738
B11	754598	1598670	746
B12	754528	1598549	756
B13	755716	1597479	767
B14	755549	1597553	779

Appendix Table A1 (Continued)

Site	UTM		elevation
	East	North	
B15	755410	1597507	757
B16	755324	1597504	742
B17	755275	1597455	730
B18	755164	1597398	745
B19	755118	1597349	752
B20	755049	1597298	754
B21	755027	1597412	756
B22	755229	1597482	748
C01	757861	1595178	726
C02	757578	1595120	742
C03	757365	1594964	764
C04	757195	1594956	769
C05	756809	1594854	739
C06	756686	1594883	742
C07	755697	1593718	724
C08	755640	1593627	724
C09	755638	1593556	724
C10	755628	1593461	723
C11	755661	1593468	734
C12	755639	1595300	739
C13	755655	1595351	742
C14	755586	1595427	736
C15	755505	1595537	767
C16	755530	1595188	753
C17	755596	1594804	728
C18	755660	1594146	728
C19	755733	1594181	735
C20	755856	1594304	751

Appendix Table A2 Sampling period and number of individual of amphibian in the dry season.

Species	Abundance (percent of total captures)			
	January 2011	February 2011	March 2011	Total (%)
<i>Microhylaberdmorei</i>	11	13	48	72 (40.22)
<i>Microhylafissipes</i>	0	9	18	27 (15.08)
<i>Hylarananigrovittata</i>	4	5	15	24 (13.41)
<i>Fejervaryalimnocharis</i>	9	10	10	29(16.20)
<i>Hylaranaerythraea</i>	0	5	2	7(3.91)
<i>Limnonecteskuhlii</i>	1	1	10	12 (6.70)
<i>Limnonectes gyldenstolpei</i>	0	0	1	1 (0.56)
<i>Microhylaheymonsi</i>	0	0	1	1 (0.56)
<i>Occidozygamartensii</i>	2	1	0	3 (1.68)
<i>Microhylabutleri</i>	0	3	0	3 (1.68)
Total	27	47	105	179 (100)

Appendix Table A3 The number of species and individuals of amphibian in different stream substrates in dry season; C is the stream with cascade, R is the stream with rocky bed and S is the stream with sediment bed.

	Substrates		
	C	R	S
Number of species	6	7	9
Number of individuals	38	45	93
Number of study areas	15	19	20
Number of individuals/sampling areas	2.53	2.37	4.65

Appendix Table A4 The number of species and individuals of amphibian in different habitat patterns in dry season; HS is the higher slope area, HG is the hill near grassland and LS is the lower slope area.

Species	Habitats		
	HS	HG	LS
Number of species	6	7	8
Number of individuals	30	107	39
Number of study areas	12	22	20
Number of individuals/sampling areas	2.50	4.86	1.95

Appendix Table A5 Sampling period and number of individual of amphibian in the rainy season.

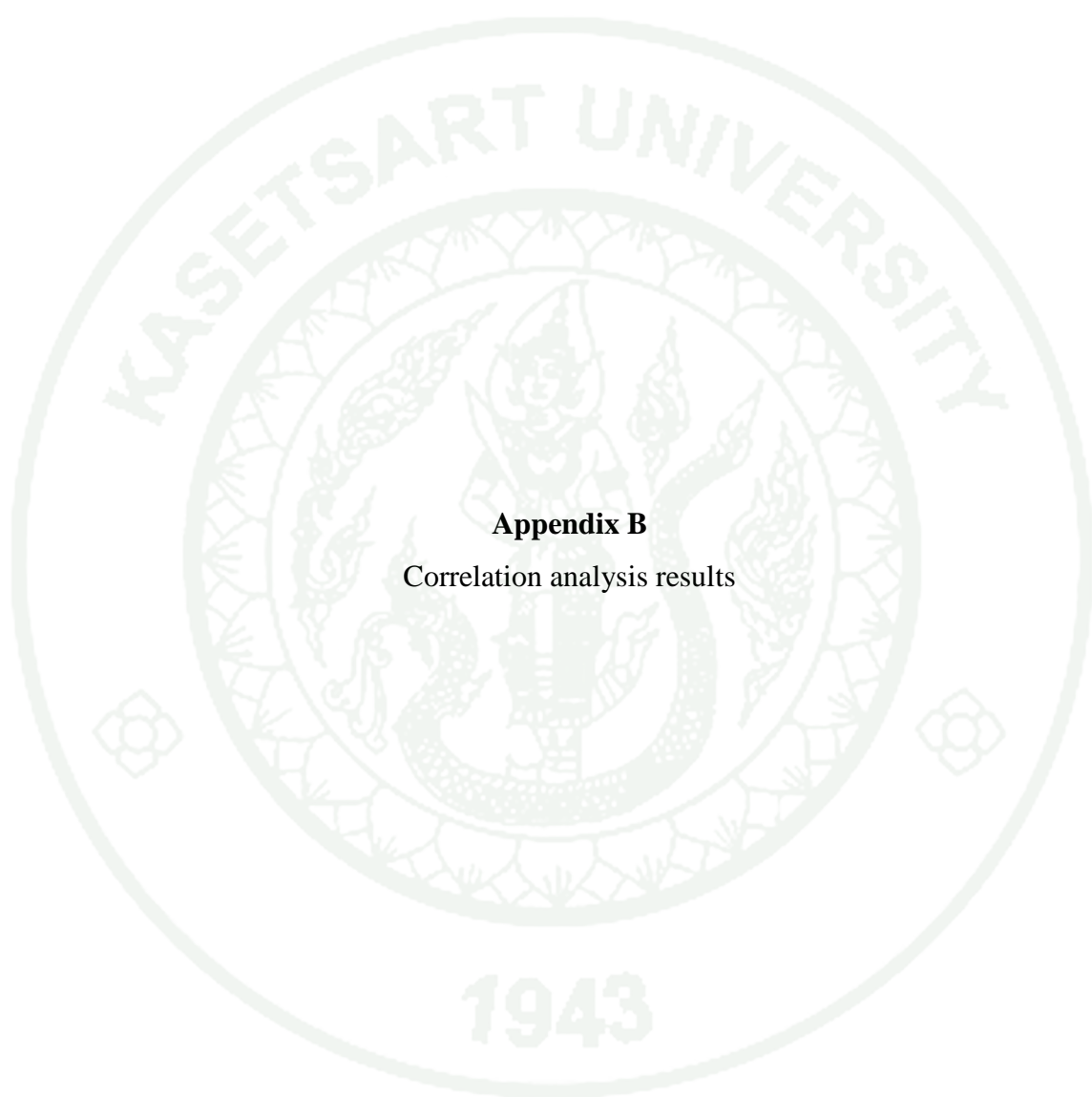
Species	Abundance (percent of total captures)			
	August 2011	September 2011	October 2011	Total (%)
<i>Microhyla berdmorei</i>	35	85	31	151 (25.38)
<i>Microhyla fissipes</i>	0	2	4	6 (1.01)
<i>Hylarana nigrovittata</i>	37	63	194	294 (49.41)
<i>Fejervarya limnocharis</i>	15	15	9	39 (6.55)
<i>Hylarana erythraea</i>	0	1	1	2 (0.34)
<i>Limnonectes kuhlii</i>	3	12	57	72 (12.10)
<i>Limnonectes gyldenstolpei</i>	0	1	0	1 (0.17)
<i>Microhyla heymonsi</i>	1	6	1	8 (1.34)
<i>Occidozyga martensii</i>	0	13	6	19 (3.19)
<i>Polypedates leucomystax</i>	0	0	1	1 (0.17)
<i>Microhyla butleri</i>	0	2	0	2 (0.34)
Total	91	200	304	595 (100)

Appendix Table A6 The number of species and individuals of amphibian in different stream substrates in rainy season; C is the stream with cascade, R is the stream with rocky bed and S is the stream with sediment bed.

Species	Substrates		
	C	R	S
Number of species	5	10	9
Number of individuals	117	225	253
Number of study areas	15	19	20
Number of individuals/sampling areas	7.80	11.84	12.65

Appendix Table A7 The number of species and individuals of amphibian in different habitat patterns in rainy season; HS is the higher slope area, HG is the hill near grassland and LS is the lower slope area.

Species	Habitats		
	HS	HG	LS
Number of species	6	7	10
Number of individuals	141	303	151
Number of study areas	12	22	20
Number of individuals/sampling areas	11.75	13.77	7.55



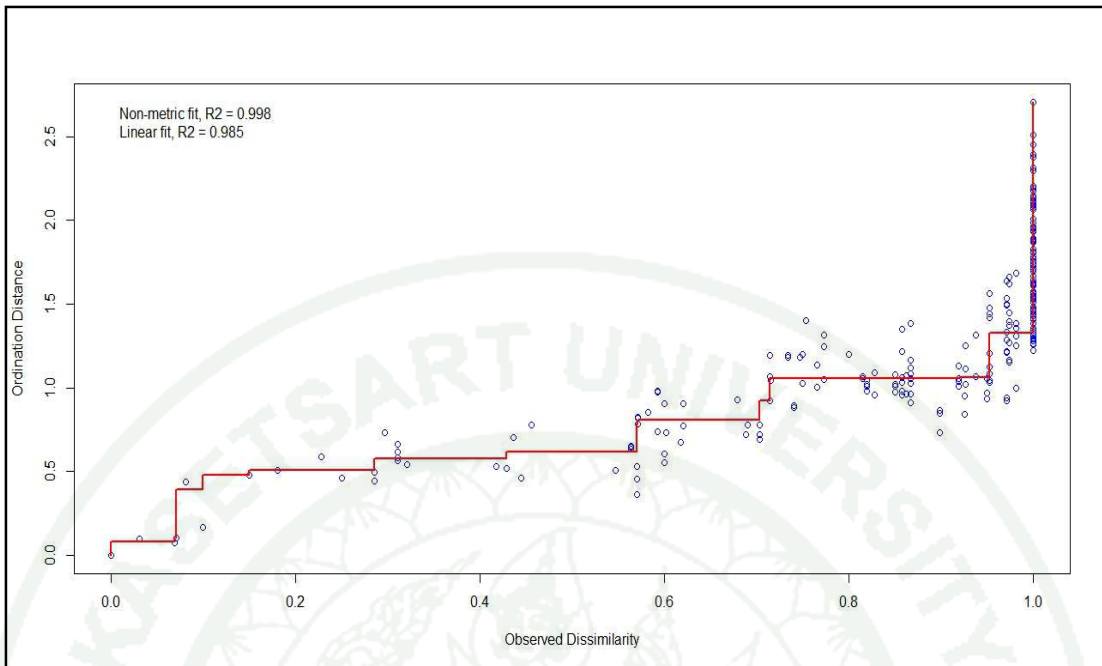
Appendix B
Correlation analysis results

Appendix Table B1 Correlation of environmental variables of streams in dry season with NMDS. (***= $P < 0.001$, ** = $P < 0.01$ * = $P < 0.05$ and . = $P < 0.1$).

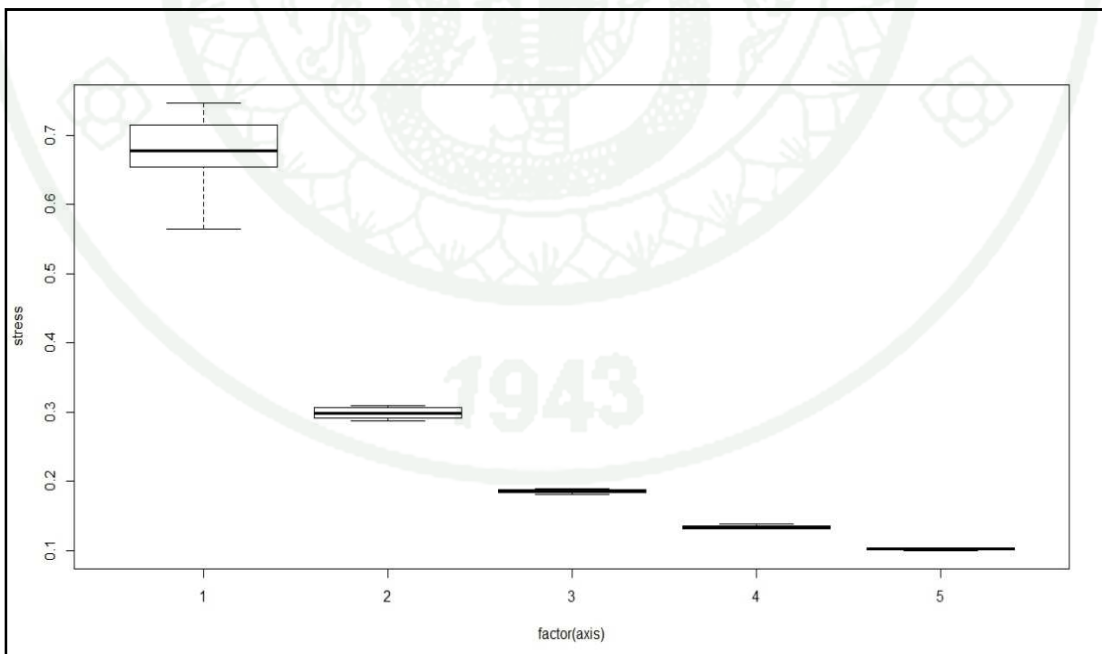
Variable	NMDS 1	NMDS 2	r ²	Pr(>r)
pH	-0.908682	0.417489	0.4947	0.000999***
DO	0.335124	0.942174	0.1890	0.017982*
Conductivity	-0.996265	-0.086344	0.9417	0.000999 ***
Velocity	0.484200	0.874957	0.0056	0.861139
Temperature	-0.678609	-0.734500	0.1568	0.044955*
Depth	0.043897	0.999036	0.5239	0.000999 ***
Cover	-0.256050	-0.966663	0.1319	0.083916 .

Appendix Table B2 Correlation of environmental variables of streams in rainy season with NMDS. (***) = $P < 0.001$, (**) = $P < 0.01$ (*) = $P < 0.05$ and (.) = $P < 0.1$)

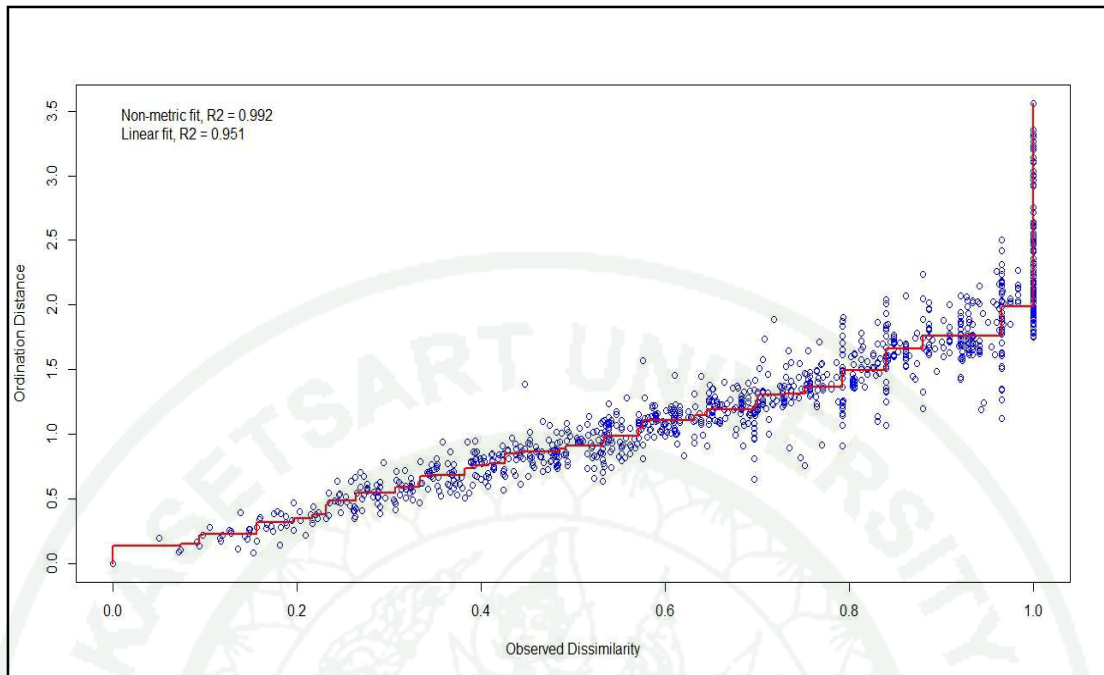
	NMDS1	NMDS2	r^2	Pr(>r)
pH	-0.154976	0.987918	0.3709	0.000999 ***
DO	-0.957737	0.287644	0.1040	0.061938.
Conductivity	0.084354	0.996436	0.6935	0.000999 ***
Velocity	-0.882982	0.469407	0.0588	0.250749
Temperature	-0.175922	-0.984404	0.0575	0.253746
Depth	-0.633507	-0.773737	0.7181	0.000999 ***
Cover	-0.916477	0.400088	0.9251	0.000999 ***



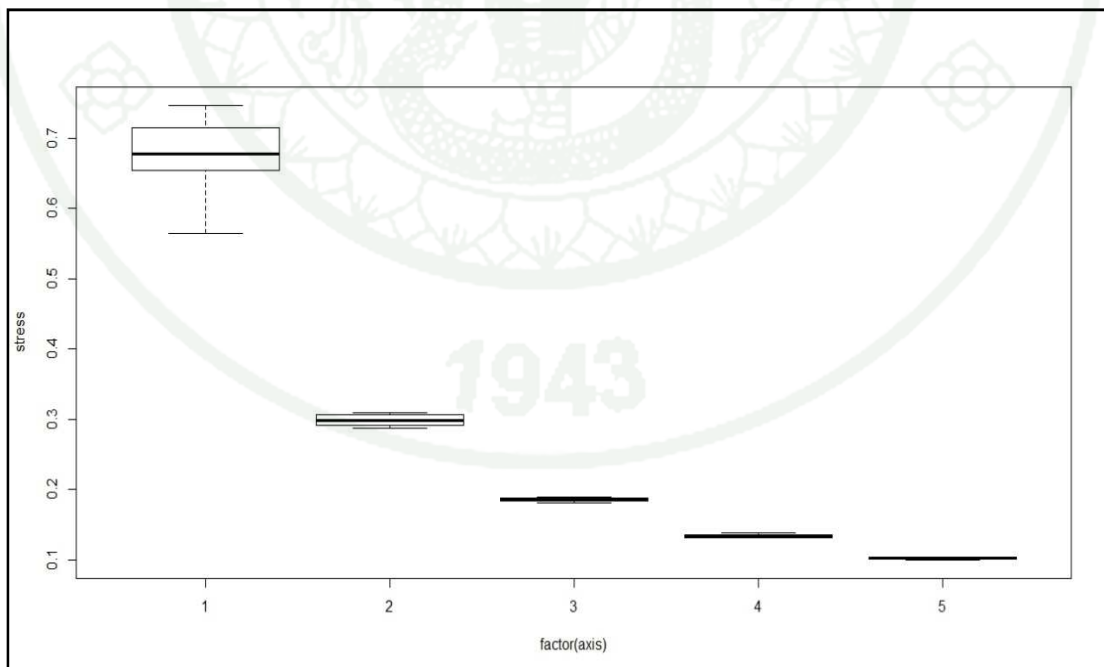
Appendix Figure B1 Shepard diagram with monotone regression of stress value in the dry season.



Appendix Figure B2 Scree plot of analysis appropriate number of dimension in the dry season.



Appendix Figure B3 Shepard diagram with monotone regression of stress value in the rainy season.



Appendix Figure B4 Scree plot of analysis appropriate number of dimension in the rainy season.

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