MICROHABITAT PREFERNCE OF GOLDEN-BACKED FROGS (*Indosylvirana sp.*) IN RIPARIAN HABITATS OF SOUTH WAYANAD FOREST DIVISION

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(18-MSVP-05)

DISSERTATION

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DECLARATION

I hereby declare that this dissertation titled "**Microhabitat prefernce of golden-backed frogs (*indosylvirana sp.*) in riparian habitats of South Wayanad forest division**" is a confide record of research work done by me during the course of my Master's research program and the dissertation has not previously formed the basis for the award of any degree, diploma, associate ship, fellowships or other similar title of any other University or Society.

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Certified that this dissertation, titled "**Microhabitat preference of golden-backed frogs (*Indosylvirana sp.*) in riparian habitats of South Wayanad forest division"** is a bonafede record of research work done independently by **Arathy S Madhu (18-MSVP-05)** under my guidance and supervision and that it has not previously formed the basis for the award of any degree, diploma, associateship or fellowship to his.

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We the undersigned members of the Advisory Committee of **Arathy S Madhu (18-MSVP-01)**, a candidate for the degree of Master of Science in Wildlife studies, agree that the dissertation titled, "**Microhabitat preference of golden-backed frogs (*Indosylvirana sp.*) in riparian habitats of South Wayanad forest division"** may be submitted by **Arathy S Madhu (18-MSVP-05),** in partial fulfillment of the requirement for the degree.

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1. INTRODUCTION

Habitat is contemplated as resources and environments in a particular area where an animal live and reproduce (Tilman., 2008). Habitat preference or selection of habitat is the process by which an individual organism selects a habitat from the available alternative resources. (Babbitt *et al*., 2010). Organisms follow certain hierarchical special scaling order to select their habitat. At first, the organisms select their geographic range, which is the first order selection. Then they consider the traits of the landscape, which is the second order selection, this will define their home range. After selecting their home range, organisms will choose a particular area that resides within that home range, and that is known as the third order selection. The second order selection and third order selection are synonymized with macro habitat and micro habitat selection respectively (Johnson, 1980; Babbitt *et al*., 2010). Any animal decides its habitat on the basis of many ecological and behavioral characteristics that then made use to perform their daily activities (MC Prabhath *et al* 2018). So, it is very important to understand the response of animals that conserve biological diversity in habitats that are modified to avoid the further loss of biodiversity (Tilman 1994).

Amphibians are generally habitat specific. To completely understand the habitat requirements of amphibians, detailed study on microhabitats used by amphibians in a particular vegetation community and also mechanistic studies that link use of habitat to population dynamics are essential (Armstrong , 2005) The population of frogs are declining considerably because of human intervention, habitat destruction and fragmentation, change in microhabitat, climate change *etc*. Because amphibians show habitat specificity and have less dispersal capabilities, disturbances can lead them to local extinctions.

Riparian vegetation is an important trait of the landscape because it forms a link between terrestrial and aquatic systems and can function as corridors (Tabacchi *et al*, 1990, Malanson 1993, Machtans *et al.*, 1996, Naiman and De´camps, 1997). Amphibian communities in stream side occupy a sensitive environment and they can be considered as indicators of effects of adjoining land uses (Murali. and Raman. 2012). Present study focus

on the site preference of two Ranid frogs of genus Indosylvirana, *Indosylvirana flavescens* and *Indosylvirana indica.* Though frogs of genus Indosylivirana can be found near stagnant water bodies, they are abundant near small streams and creeks.

Indosylvirana, previously known as Hylarana, is a genus which belongs to the family Ranidae with worldwide distribution (Biju *et al*, 2014). They are commonly known as Golden-backed frogs and as their name suggests, they are characterized by their yellow to golden colored dorsum. They are mainly found in riparian habitat or are seen associated with stagnant water bodies. Out of the ten species described from India, eight of them are seen in Kerala *(*Dinesh *et al*, 2020, Das *et al,* 2015). In Wayanad, three species of the genus Indosylvirana are presesnt, namely *I. indica*, *I. flavescens* and *I. intermedius (*Biju *et al*, 2014). The present study will focus on the frogs *Indosylvirana flavescens* and *Indosylvirana indica.* They are commonly known yellowish Golden-backed frog and Indian golden backed frog.

The species inhabits fast and constantly flowing streams. They usually perch on rocks that are seen on the banks of the streams. They are also seen on small bushes near the streams. During breeding season, they lay eggs on shallow regions of the stream Biju *et al.*, (2019). In this study, the microclimatic conditions which will determine the site preference of the frogs *Indosylvirana flavescens* and *Indosylvirana indica* will be checked. As part of the study, three different streams in the South Wayanad forest division will be surveyed. A 200m transect will be laid to survey the frogs. Visual Encounter Survey will be employed to search for the frogs. Microclimatic parameters like Temperature, Humidity, Canopy cover, Leaf litter cover, height from the ground/water surface, distance from the center of the stream *etc* will be recorded.

Anurans are an important part of both aquatic and terrestrial ecosystem. Therefore, any anthropogenic alteration which affects the frog species may also have negative impact on the entire environment (Hopkins, 2007). As most of the perennial streams running through the forest edges are prone to high anthropogenic pressure, the habitat of frogs is either altered or destroyed. Habitat preference studies will help us to identify the potential microhabitat for specific species. The results of such studies can prevent developmental activities in those habitats thus protect that particular site from degradation.

Taking into consideration the above facts, this study was planned with the following objective:

1. To determine microhabitat specificities of frogs of the genus Indosylvirana in riparian habitat.

2. REVIEW OF LITERATURE

2.1. NATURAL HISTORY AND DISTRIBUTION

Jerdon , (1853) described *Indosylvirana flavescens* as Rana flavescens. He also considered it as the second oldest Hylarana species which was discovered 160 years ago.

According to Boulenger, (1882), *Indosylvirana flavescens* was tentatively considered as a synonym for Indosylvirana temporalis.

On the basis of morphological similarity, Dubois, (1992) put up seven subgenera and one nominal within Hylarana. The subgenera erected by Dubois are Amnirana); Chalcorana, Humerana, Pulchrana, Sanguirana,and Sylvirana; and Papurana and Tylerana.

Based on the molecular study conducted by Biju *et al.*, (2014), the species *Indosylvirana intermdius* and *Indosylvirana flavescens* that were initially considered as synonym to *Indosylvirana temporalis* are now being identified as distinct species. Indosylvirana intermedius is restricted to Southern Western Ghats districts Kerala and Tamil Nadu. The species are extensively distributed in both primary and secondary forests. Indosylvirana intermedius can only be found on the North of Palghat gap, mostly at elevations between 600-1183.

Oliver *et al.*, (2015) raised the rank of *Sylvirana* to generic level along with five other genera which were previously considered as subgenera of the genus Hylarana. They additionally described two genera. One is Indsylvirana and the other one is a monotypic genus *Abavorana. Indosylvirana* was previously assigned to *Sylvirana* and in this study they identified *Indosylvirana* as a separate clade. Since they are largely restricted to India- Sri Lanka biodiversity hot-spot, they are given the name *Indosylvirana.*

According to Biju *et al*, (2019) *Indosylvirana indica* is seen at an elevation ranging 60-1145.Until their study, the species are recorded only from regions North of Palghat gap. Though the species are mainly found near secondary and primary forests, they were also reported from wetlands near to forest areas.

2.2. HABITAT PREFERANCE IN FROGS

Navas, (1996) studied the microhabitat association and thermal ecology of high elevation neotropical areas. The study was conducted on four species of frogs. He compared the

microhabitat association and thermal ecology of the two populations of anurans, one population inhabiting 3500m and the other population inhabiting 2900m. His study showed that at both the elevations, microhabitat association and general activity pattern were similar for each of the species. In the case of active anurans, body temperature is determined by the reciprocity between habitat selection and time of the day. Lowest thermal activity was exhibited by nocturnal terrestrial frogs. Increase in altitude significantly affected their thermal ecology. The high tropical elevation has physical traits that limit the chances of thermoregulation based on behavior in anurans. It also increases the necessity of physiological adjustments to withstand wide thermal range.

Smith *et al.,* (2003) focused on the terrestrial microhabitat of the Northern cricket frog *Acris crepitans*. Their study found that these frogs distinctly choose substrates that are moist. They also suggest that the cricket frogs may choose their microhabitat on the basis of temperature. Their observations lead into the conclusion that the frogs stayed at the proximity of the edge of their shelter than open space.

(Prabhath *et al.*, 2018) studied the habitat preference of the frog *Minervarya greenii*. They found that the species was recorded only from 1m above water level and 3m above terrestrial habitat. They also found that the *Minervarya greenii* is a generalist species inhabiting in and around lentic water bodies. Water depth, water temperature, submerged plant cover, decaying plant matter and leaf litter, substrate temperature, relative humidity and substrate relative moisture are found to be the climatic parameters that determine the microhabitat of *Minervarya greenii*.

The influence of developmental and environmental factors on the heterogeneity of adult skull morphology of anurans was studied by Bardua *et al.,* (2021) . Their analysis of 15 cranial regions of 173 anurans species gave the result that along with skull size, larval feeding and ossification timing, microhabitat also play a significant role that contributes to the cranial evolution in frogs.

On the basis of a study conducted by Gururaja *et al*., (2021), they found that the tadpoles of the three *Nyctibatrachus* species exhibit microhabitat preference and niche partitioning. The slope of the stream and water temperature significantly affects the distribution of the tadpoles. Steep slopes are preferred by the tadpoles of *Nyctibatrachus jog*, tadpoles of the species *Nyctibatrachus kempholeyensis* preferred gradual slopes with

stream side where the stream is shallow and water temperature has notable influence on the distribution of *Nyctibatrachus kumbara* tadpoles in streams.

2.2.1. FACTORS AFFECTING HABITAT PREFERENCE IN FROGS

Prabhath *et al.*, (2018) studied the habitat preference of the frog *Minervarya greenii.* They found that the species was recorded only from 1m above water level and 3m above terrestrial habitat. They also found that the *Minervarya greenii* is a generalist species inhabiting in and around lentic water bodies. Water depth, water temperature, submerged plant cover, decaying plant matter and leaf litter, substrate temperature, relative humidity and substrate relative moisture are found to be the climatic parameters that determine the microhabitat of *Minervarya greenii.*

On the basis of a study conducted by Bahiah S *et al.*, (2019) to check for the frog species diversity and to check for the anuran microhabitat selection, they found that *Fejervarya cancrivora*, *Fejervarya limnocharis*, and *Limnonectes paramacrodon* were found in almost all available microhabitats except for vegetated substrates. *Fejervarya cancrivora* and *Fejervarya limnocharis* are found associated with more than one microhabitat. This describes their generalist nature. The species *Limnonectes ingeri* and *Limnonectes kuhlii* inhabited areas with mineral deposits and they were recorded mostly from mud bank and also near to stream. *Chalcorana labialis* was found perching mostly on seedlings and herbaceous plants.

(Thomas *et al.*, 2019) studied how the four stream habitat variables water flow velocity, substrate slope, water column depth and temperature of water influence the rheophilous tadpoles of the rare and endangered frog *Nasikabatrachus sahyadrensis*. Along 100m transects of two selected streams, they laid 68 grids. Their analysis showed that abundance of the tadpoles in the two streams are best predicted with water flow velocity.

2.3. RIPARIAN HABITAT

Dupuis and Steventon, (1999) identified that the densities of tadpoles of *Ascaphus truei* are considerably low in streams that are logged compared to the buffered and old-growth streams. Hence natural channel conditions are maintained by forested buffers along streams.

Krishna *et al.*, (2005) tested the hypothesis that the abundance of variation does not vary significantly between forest and commercial plantations. They concluded that composition of species and relative abundance considerably varied between streams in the three selected habitats. In the commercial plantations, insecticides are sprayed annually
. This will reach the streams as surface runoff and affect the anurans negatively as they use the riparian habitat as breeding and retreating sites.

Ragavendran *et al.*, (2006) studied the herpetofaunal assemblage in the forest mosaic of Western Ghats. They found that the amphibian species richness and abundance was highest in riparian habitat of dry forest mosaic.

According to Richards-Zawacki, (2009), the genetic structure of the Panamanian frog *Atelopus varius* is influenced by the riparian habitat connectivity. Their findings lead into the conclusion that low slope areas like streams and mountain ridges are very important for the gene flow of the frog.

According to Rodríguez-Mendoza and Pineda, (2010), within the fragmented forests, riparian remnants can be particularly important for the up keeping of biodiversity within forests that are highly fragmented. Their results suggest that species diversity of frogs in forests that are fragmented is significantly affected by the riparian remnants.

According to the study conducted by Murali and Raman, (2012), they found that anuran species richness was highest at streamside of rainforest fragments. It considerably decreased with streamside of coffee and tea plantations.

On the basis of a study conducted by Naniwadekar and Vasudevan, (2014)
, difference in species richness was not significant in above and below the dams, but there was significant difference in frog specie community composition because of the altered river flow.

2.4. PRINCIPLE COMPONENT ANALYSIS

Babbitt, (2009) in their study on effect of habitats on agricultural highlands on larval anuran assemblages in wetlands that are flooded seasonally used PCA to check patterns in environmental conditions of wetlands and also to check if the environmental conditions varied notably within habitats.

In a study conducted by Rannapp *et al.*, (2015), they used Principle Component Analysis (PCA) to decide whether amphibian species assemblage in ponds with and without spadefoot toad tadpole differed within countries.

2.5. THREATS AND CONSERVATION

Dey, (2010) studied the habitat selection and anthropogenic threats of anurans in Barak Valley. She identified multiple factors that affect the survival of anurans in that area. Brick kilns, construction of buildings over ponds and low lands, jhum cultivation, climate change, chemical components, killing for food and laboratory experiments, viral diseases like chytridiomycosis and road kills by vehicle are identified as the major threats that contribute to the decline of anurans in Barak Valley.

Polo-Cavia *et al.*, (2016) analyzed how the two common contaminants humic acid and ammonium nitrate affects the ability of the Western spade foot toad Pelobates cultripes to identify the chemical cues from the common predator, dragonfly nymph *Anax imperator*. They found that even when the lowest quantity of either humic acid and ammonium nitrate that were added to the water, the tadpoles remain unresponsive. Where as in the absence of both these chemicals, the tadpoles reduced their swimming in response to the chemical cues given by the dragonfly nymph. Presence of the chemicals humic acid and ammonium nitrate interferes with the tadpole's ability of responding to the predator chemical cues, thus affecting the survival of the tadpoles.

In a study conducted by Raghunath *et al.*, (2017), assessed the nuclear abnormality and conducted micronuclei test for the erythrocytes of frogs to check for the effect of environmental contamination in agro-ecosystems. The nuclear abnormality assessment and micronuclei tests were conducted on the Indian skipper frog *Euphyctis cyanophyctis*. Their study indicated that out of the 1000 analyzed cells, the occurrence of micronucleus ranged from 0 to 8. The study also showed that 80% of the frogs inhabiting agro-ecosystems exhibit abnormalities because of the agro-chemicals.

Dare *et al.*, (2020) tested the run-of-river hydropower projects on the longitudinal connectivity of anuran larvae upstream and downstream are hindered by the run-of-river hydropower projects. Their results showed that compared to the upstream of the dam, there was a decrease of larval densities of *Ascaphus truei* by 60%. This indicates

that the altered densities of the *Ascaphus truei* larvae because of run-of-river dam lead to the deposition of the larvae on the upstream of the dam.

3. MATERIAL AND METHODS

3.1 STUDY SITE

Wayanad is a district situated at the North-Eastern part of Kerala. It is a state with hilly terrain. It is situated on the Southern top of Deccan Plateau. The area lies between (11.6854° N, 76.1320° E). It covers a geographical area of 2130km with 787km² area of forest cover. The district is bounded by Kozhikode on West, Malappuram district to the South, on South by Malappuram, on West by Kozhikodu district and Kannur district, Tamilnadu East and Kannur and Karnataka to the North. The average elevation of district is approximately 900 m from mean sea level and also has some mountain peaks in the district. Due to the presence of Arabian Sea at the Western side most of the parts of Wayanad get a high annual rainfall from South-Western monsoons and a moderate precipitation from North-Eastern monsoons. The average annual precipitation is 2594.9 mm (Ministry of earth sciences, India). Wayanad is further divided into three administrative taluks and 23 grama panchayats.

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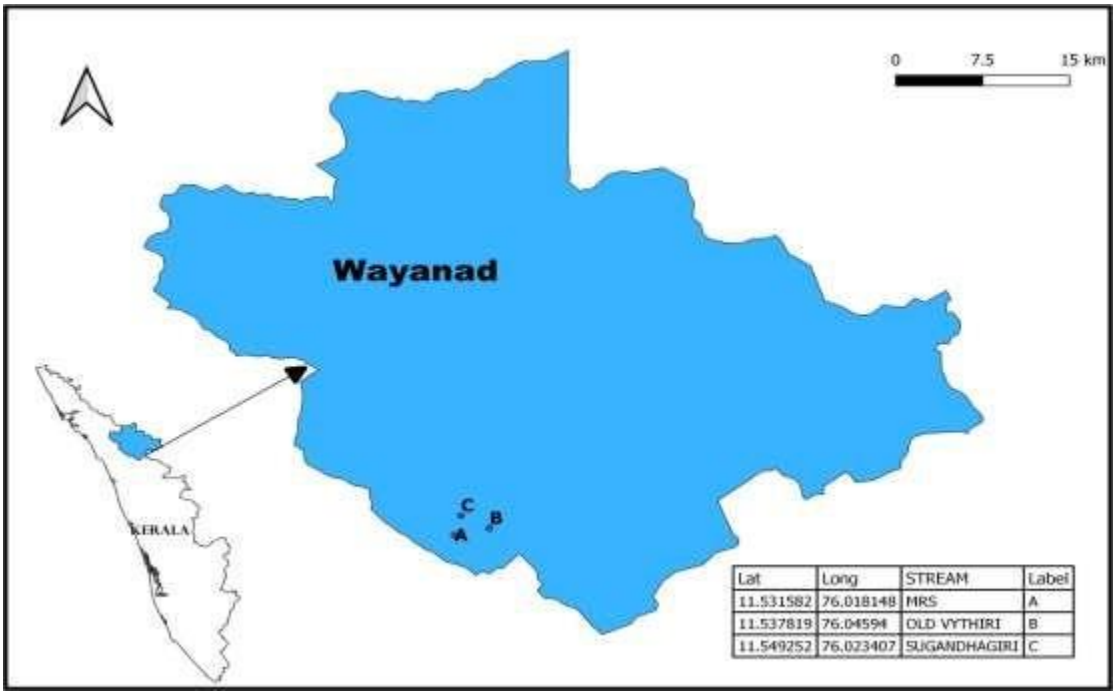

Figure 1: Map of Wayanad district including location of study areas

With an area of 47.84 sq km, Vythiri Panchayat is located at the Western side of Wayanad district. The place is having high annual precipitation with cool and humid climate. Most of the land in the Panchayat is transformed into tea and coffee plantations. Vythiri Panchyat has a population density of 382.63. The natural vegetation is mostly evergreen and semi-evergreen having many perennial high-altitude streams and one major river. Along with small human settlements, a great number of private resorts and homestays are present in tourism that helps promoting tourism

For the study purpose, three different streams were selected. Site A is located in Pookode in the Kalpetta range, South Wayanad division (N 11.521387 and E 76.018405). The stream is very narrow and shallow with maximum and minimum width of 6m and 2m. The canopy is moderately closed. Human settlements are present in the vicinity and a road crosses through the stream near the study area. High grazing is observed on the banks. The study site contains about 16 shrub species and 10 grass species. The average canopy cover and leaf litter cover are 49.13% and 26.04 % respectively. Stream B comes under Meppadi range and is located at Old Vythiri (N, 11.537868 and E, 76. 045774). Human settlements and many private resorts and homestays are present near to the stream. Human activity is very high in the stream. The stream is rocky with boulders and rocks. Vegetation including ochladrae reeds are present in the bank. The stream has an average canopy cover of 69.23
% . The average leaf litter cover of the stream is 53.07. The undergrowth is low with three species of shrubs and four species of herbs. The maximum and minimum width of the stream is 11m and 9m respectively. Canopy cover is relatively high as compared to the other two streams under consideration. Site C is located near Sugandhagiri (N11.549252, E76.023407) which comes under Kalpetta range of South Wayanad Forest Division. The stream is surrounded by human settlements. Many shops are located near the stream and high vehicle activity is observed surrounding the stream. The maximum and minimum width of the stream is 5m and 11m respectively. The stream is surrounded by 7 shrub species and seven species of herbs. The average canopy cover of the stream is 26.62 % and average leaf litter cover is 21.56%. In all the three sites, Indosylvirana flavescence and *Indosylvirana indica* are found to be active only during night. Their activity is found to be increased after 1800 hours.

3.2 FIELD SAMPLING

Frogs were surveyed using transect method (Crumb and Scott, 1994). In each stream, a transect of 200m was laid to survey the frogs. The entire length of the 200m transect was surveyed using Visual Encounter Survey, (Crumb and Scott, 1994). Frogs were also spotted by following their calls. Close contact with frogs was avoided the maximum to avoid movement of frogs from one substrate to another. After spotting a frog, variables like substrate type, substrate temperature, height of occupancy, distance from bank, stream width, temperature, humidity, water velocity altitude were recorded.

A head torch with good visual efficiency was used to search for the frogs. The water velocity, temperature and humidity were recorded using a pocket Kestrel Weather station. Temperature of the substrate was recorded using an infrared thermometer. Measuring tapes were used to measure the stream width and distance of the substrate from the stream bank. Latitudinal and longitudinal values of the location of the frogs were obtained from the mobile app GPS Essentials.

To record the vegetation data, plots of 5×1 m were laid with an interval of 25 m. Thus, from each stream there were a total of 24 plots. In each of these plots, a 1×1m plot was laid for the herbs and grasses. The canopy cover and leaf litter cover of these plots were also recorded.

Photo documentation of the frog species and the habitat of each surveyed stream were done.

The data collected was subjected to statistical analysis for comparison.

3.3 SPECIES IDENTIFICATION

Indosylvirana is a genus that contains complex species. It is very difficult to identify these species from simply observing them. They should be properly photographed and closely examined. Both *Indosylvirana flavescens* and *Indosylvirana indica* has got dorso-lateral folds that are well developed and prominent.

Indosylvirana flavescens

Individual frog's forelimb was closely observed. Those frogs with webbing of third toe that is well below the subarticular tubercle on the inside are identified as *Indosylvirana flavescens*. When the snout was observed, it was found that the snout is rounded in both dorsal and ventral view. The shagreened to scantly granular skin was yet another feature that helped to distinguish Indosylivirana flavescens from *Indosylvirana indica*.

Indosylvirana indica

Individuals with second toe webbing above the first subarticular tubercle on the inside are identified as *Indosylvirana indica*. They possess a dorsum that is granular in nature. In dorsal and ventral view, the snout of these individual was elliptical.

As part of independent study on Indosylvirana conducted by a researcher on the same study area, the two species were confirmed to be *Indosylvirana flavescens* and *Indosylvirana indica*. The Genomic DNA was extracted from liver tissue samples stored in absolute ethanol at -20°C and used DNeasy (QiagenTM) blood and tissue kit. Partial sequences of one mitochondrial gene (16S rRNA) was amplified to determine the species.

3.4 PRINCIPAL COMPONENT ANALYSIS (PCA)

A Principal component analysis (PCA) was performed to identify the factors that contribute to the distribution of frog species. It is a dimension reduction technique which takes set of possibly correlated variables and transforms into linearly uncorrelated principal components. It is used to emphasize variations and bring out strong patterns in a dataset. PCA was carried out to find out the important ecological variables that limit the frog (Species) species distribution using the R Studio software using prcomp function and biplot was plotted using ggplot2 package. The variables such as stream parameters (stream width, stream flow, etc.), temperature (surface and atmospheric), substrate types (Leaf, Rock, Bush, Reed, etc), water velocity, sites, sites of sampling (Three sampling sites) etc. were loaded in the analysis. Before entering the variables, all the variables were normalized using scale option. This helps to normalize the data and prevent the domination of certain variables with higher magnitude. All the variables were tested for normal distribution and correlation/covariance were assumed to be linear among the variables. The distance between objects represented in the Euclidean spaces.

A

B

C.

PLATE 1: Study sites **A, B, C** respectively.

A.

B.

C.

PLATE 2: A. *I.flavescens* individual, B. *I.indica* individual, C. Mating pair of *I indica*.

4. RESULTS

Microhabitat preference of Golden-backed frog of the genus *Indosylvirana* (Ranidae) was studied at South Wayanad Forest Division, Kerala from Jan 2021 to Mar 2021. Microhabitat features such as stream characteristics, substrate types, height occupancy and stream flow and environmental features such as temperature, humidity and substrate temperature were studied. The present study was conducted on two Indosylvirana species, *Indosylvirana indica* and *Indosylvirana flavescens*. Three streams were randomly selected from South Wayanad Forest Division.

4.1. Golden-backed frog *Indosylvirana* species count across days

Out of the three sites studied (n= 3), highest individual count was observed from site A with a maximum count of 29 on day two. The maximum number of individuals observed from site B is 22 on day one. On site C, the maximum count was observed on day two with a count of 19 individuals. Individual count varied each day at each site, with number of individuals counted were higher in the Site A.

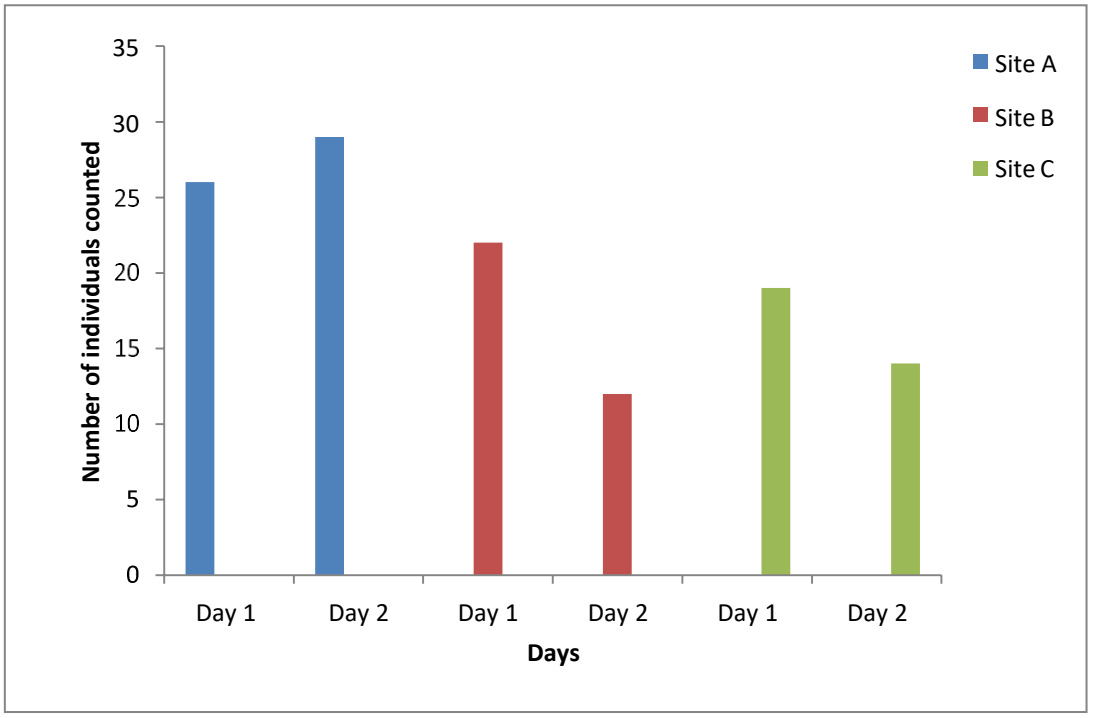

Figure 2: Number of individuals of Golden-backed frog *Indosylvirana* sp. recorded from different sites and across days.

4.2. ENVIRONMENTAL PARAMETERS

4.2.1. Vegetation composition

The vegetation data showed that Site A contains about 16 shrub species and 10 grass species. The average canopy cover and leaf litter cover are 49.13% and 26.04 % respectively. Site B has an average canopy cover of 69.23 %. The average leaf litter cover of the stream is 53.07. The undergrowth is low with three species of shrubs and four species of herbs. In Site C, the stream is surrounded by 7 shrub species and seven species of herbs. The average canopy cover of the stream is 26.62 % and average leaf litter cover is 21.56%.

4.2.2. SUBSTRATE TYPE AND CHARACTERISTICS

The substrate types varied considerably at different sites. There were four major substrate types recorded were rock, leaf, root and soil on which the frogs were found. Out of these four types of substrates, 59% of the observed frogs prefer to occupy the rocky substrate. The size of the rock varied from very small rocks to bed rocks and boulders. Soil was the substrate which was least preferred (4.9%) by the frogs. After rock, the most preferred substrate by the frogs was leaf (23.8%) followed by root (7.4%). The preference of the rocky substrate in the streams could be due to higher availability of rocks in the streams. The major substrate category in the hill streams are the rocks that is more abundant than other categories. The reeds of *Ochlandra* were more abundant in the banks of the rivers of all the three sample sites, thus rock and reeds (leaf) together constituted 87% of *Indosylvirana* sp. site occupancy.

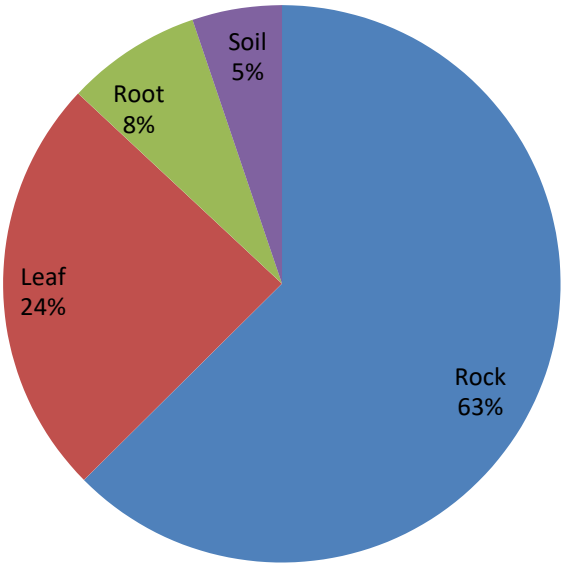

Figure 3: Per cent composition of substrate types occupied by the Golden-backed frog *Indosylvirana* sp. in the sampling sites of the study area**.**

4.2.3. Height occupancy

The frog height occupancy varied from 0cm to 120cm in the substrate from the water level with mean height occupancy was 21 cm but it was varying greatly across sample sites that can be inferred from the standard deviation value 34.66 cm (Table-1). The higher standard deviation value was due to higher height occupancy of *Indosylvirana* sp. at certain places that can be inferred from the fig. 4.

Table 1: Mean and standard deviation of substrate characteristics occupied by the Golden-backed frog *Indosylvirana* sp. in the South Wayanad Forest Division.

S. No.	Parameters	Mean ± Std. Deviation	Min-Max	N
1	Height of occupancy (cm)	21.01±34.66	0-120	122
2	Substrate temperature (°C)	21.92±2.130	17.8-26.4	122

Maximum number of the *Indosylvirana* sp. frogs was observed from height that ranges within 0-10cm from water level about 70 sightings. About 20 individuals were observed in the height category 10-20 cm. Individuals that were observed with in a range of 100-110 cm and 110-120 cm were located from bushes or reeds. Only very few individuals (less than 10) were observed in the other categories. Thus *Inodsylvirana* sp. were preferred to occupy near the water surfaces that was less than 10cm in height.

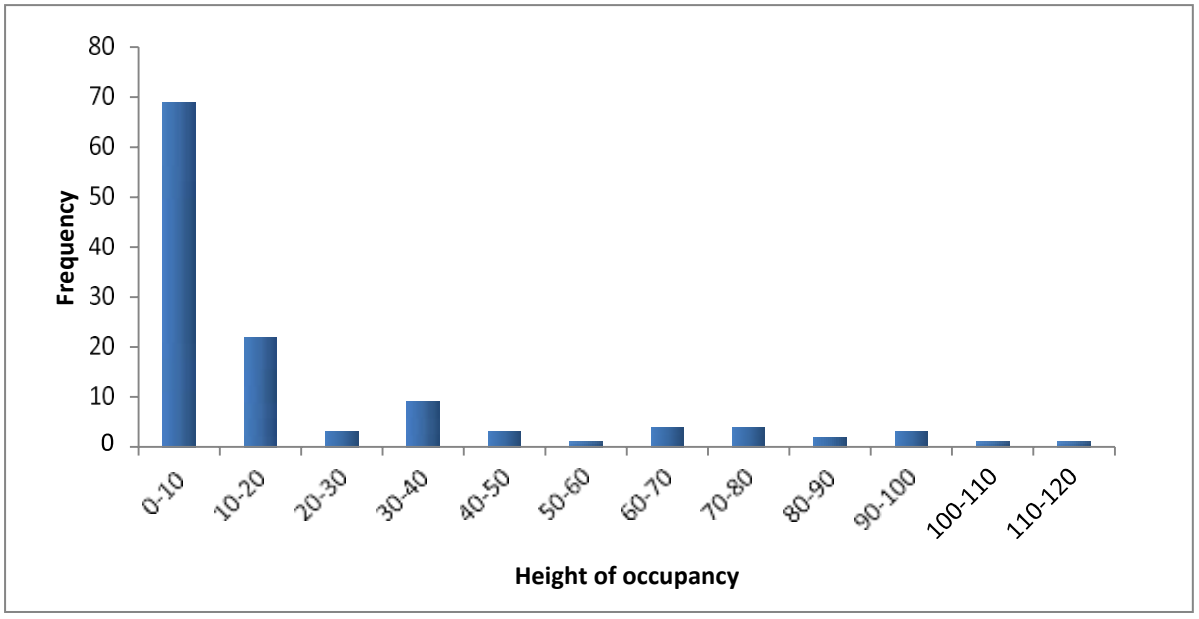

Figure 4: Variation between the height of occupancy of Golden-backed frog Indosylvirana.

4.3. STREAM CHARACTERISTICS

Stream features such as water velocity, distance to banks and stream width were studied in the all the sampling sites. Data was obtained for three streams based on the substrate on which the frog occupies its space. Water velocity ranges from 0 to 1 m/s with mean water velocity was observed to be 0.04 ± 0.178 m/s. The parameter distance to bank that frog occupies ranges from 0 to 400cm with the mean distance of 96.7 ± 95.87 cm. Stream width ranged between 110 and 1120 cm with mean value of 546.6 ± 296.0 cm.

Table 2: Mean and standard deviation of stream characteristics from all the three sample locations.

S. No.	Parameters	Mean ± Std. Deviation	Min-Max Values
1	Water velocity (m/s)	0.04±0.178	0-1
2	Distance to bank (cm)	96.7±95.87	0-400
3	Stream width (cm)	546.6±296.0	110-1120

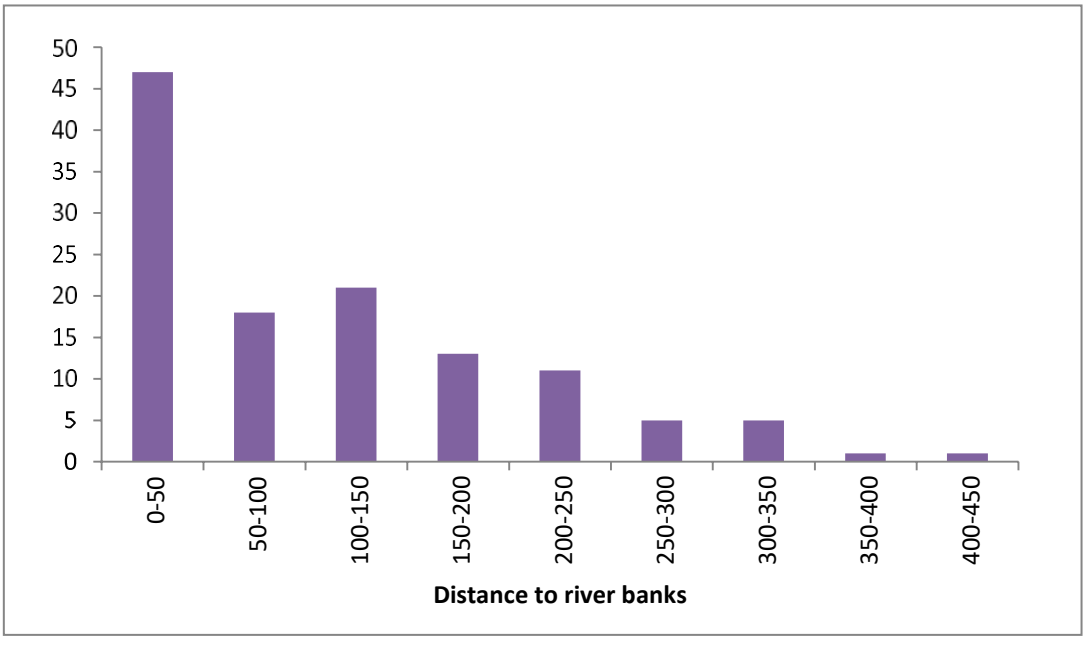

Figure 5: Distance to riverbanks and frequency of observation at different distances of *Indosylvirana* sp. in the sampling sites.

Highest number of the frogs were observed at distance to the bank that is ranging from 0-50 cm. Distance to bank where the frogs were found increased as the stream width increased.

4.3.1. The relation between the stream width and distance to river bank

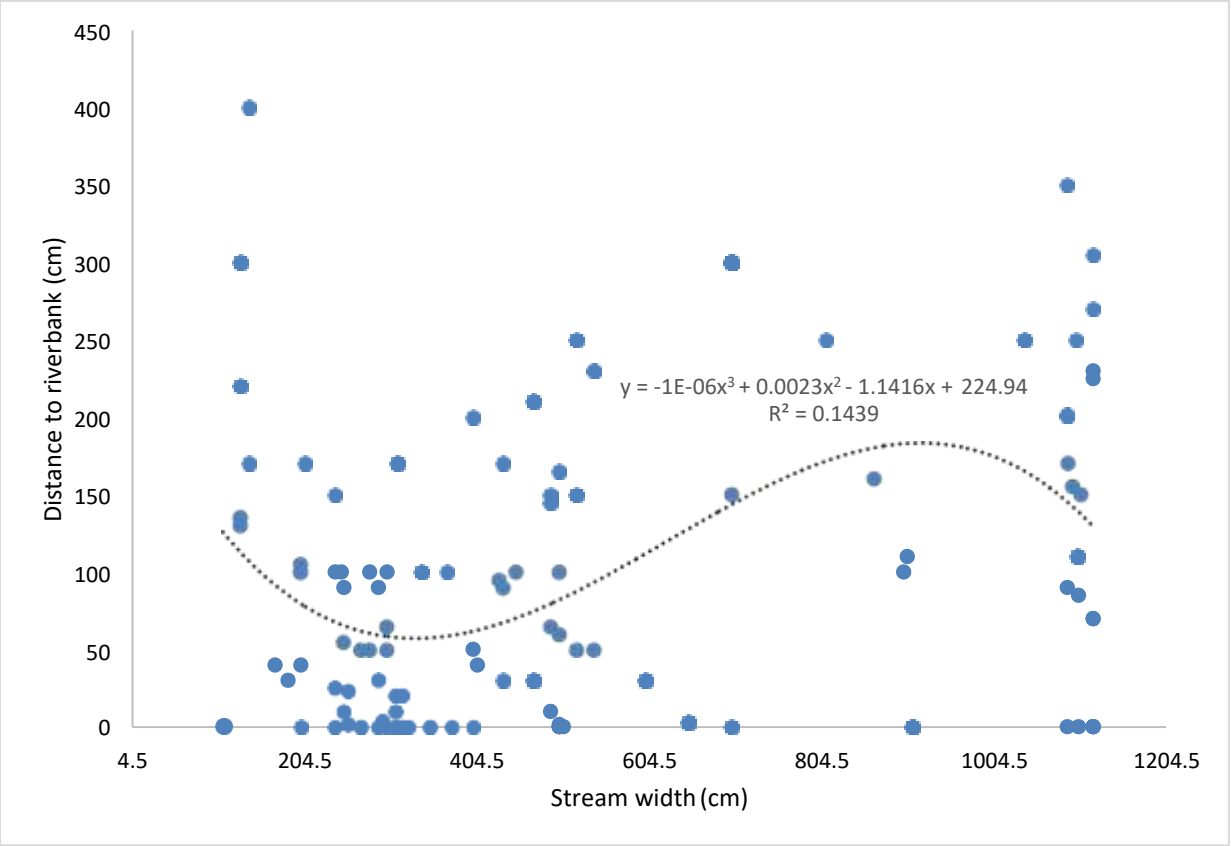

Fig 6: The relation between the stream width and distance to bank at which frog were sighted showed non-linear relationship (cubic)

The relationship between the stream width and the distance to river banks from the frog location were analyzed. The frog locations were close to the banks when the stream width was 200cm and it was gradually increased with increase of stream width. After the 900cm of stream width there were reduction in the site occupancy *Indosylvinrana* prefer to occupy near the banks in the larger streams. Thus, there were cubic relation between the site occupancy and stream width which was significant with R^2= 0.14. This could be a strategy of *Indosylvinrana* sp. attributed to avoid or escape from predators from the water especially snakes and birds.

4.4. PRINCIPLE COMPONENT ANALYSIS

A Principal component analysis (PCA) was performed to identify the factors that contribute to the distribution of Golden-backed frog species *Indosylvirana* sp. (Table-3). PCA defined four components accounted for 76% of total variance.

1 Component 1 (Temperature and topographic factors): This component consisted of the most highly correlated variables such as substrate temperature, temperature and locations with the direction of correlation being positive for all the variables. Altitude has negative effect on frog site occupancy. This component explained 37.14% of total variance.

2. Component 2 (Stream width): This component is consisted of stream width and atmospheric humidity. Stream width negatively influenced the frog species, whereas humidity had positive influence. This component explained 16.37% of total variance.

3. Component 3: (Height and substrate): The height of occupancy and substrate were negatively correlated. This component explained 12.5% of total variance.

4. Component4: (Water velocity): This component consisted of water velocity, and it is negatively related with substrate types. The number of sightings of frog were more in the stagnant water than in the stream with higher water velocity. This component explained 10% of total variance.

Among these four components the first two components consisted of Topographic factors, atmospheric temperature and stream features that explained 53% of total variance. Thus, the factors such as substrate temperature, ambient temperature, location types and altitude are the prime factors that determine the distribution of the frog species. This was followed by the stream width. The other factors such as substrate types, and water velocity were the secondary and tertiary factors that determine the frog (species) distribution.

Table 3: Ten principal component derived from the analysis of microhabitat preference of Golden-backed frog *Indosylvirana* sp. in the selected streams of South Wayanad Forest Division.

Variables	**PC1 (Temperature and topographic factors)**	**PC2 (Stream width)**	**PC3 (Height and substrate)**	**PC4 (Water velocity)**
Altitude (m)	**-0.462**	-0.208	0.005	0.045
Locations	**0.443**	0.366	-0.055	-0.011
Temperature (°C)	**0.450**	-0.167	-0.210	-0.051
Humidity (%)	-0.033	**0.718**	0.170	0.068
Substrate types	0.053	-0.203	**0.402**	-0.338
Height of Occupancy	-0.033	0.068	**-0.770**	-0.117
Substrate temperature (°C)	**0.468**	0.074	0.044	0.019
Water velocity (m/s)	-0.079	0.113	-0.082	**-0.919**
Distance to bank (cm)	0.274	-0.182	0.366	-0.120
Stream width (m)	0.287	**-0.427**	-0.166	0.056
Eigen value	**1.93**	**1.28**	**1.12**	**1.000**
Percent	**37.14**	**16.37**	**12.50**	**10.00**
Cumulative %	**37.14**	**53.14**	**66.02**	**76.02**

The PCA biplot in the Euclidean space shows there were four components contributed more than one eigen values. The angle among all the vectors approximates their (linear) covariance. It is clear in the biplot that locations, substrate temperature and ambient temperature and distance to banks were ordinated together with less than 20 have strong positive correlation. Similarly, humidity, height occupancy and water level were strongly intercorrelated.

On contrary altitude is strongly negatively correlated with the temperature and locations. Substrate is negatively correlated with the humidity and height of occupancy. The length of the line indicates the strength of the correlation.

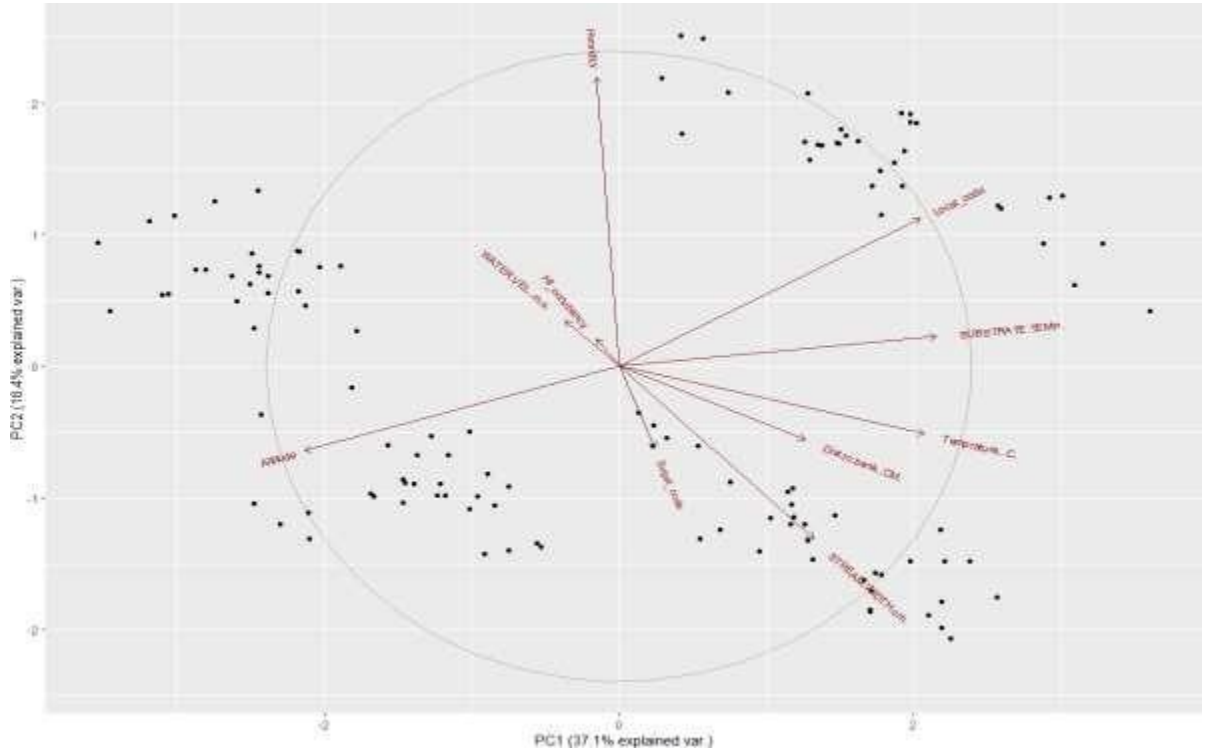

Figure 7: PCA biplot showing the environmental factors that determine the Golden- backed frog *Indosylvirana* sp. occupancy in the selected streams of Vythiri river.

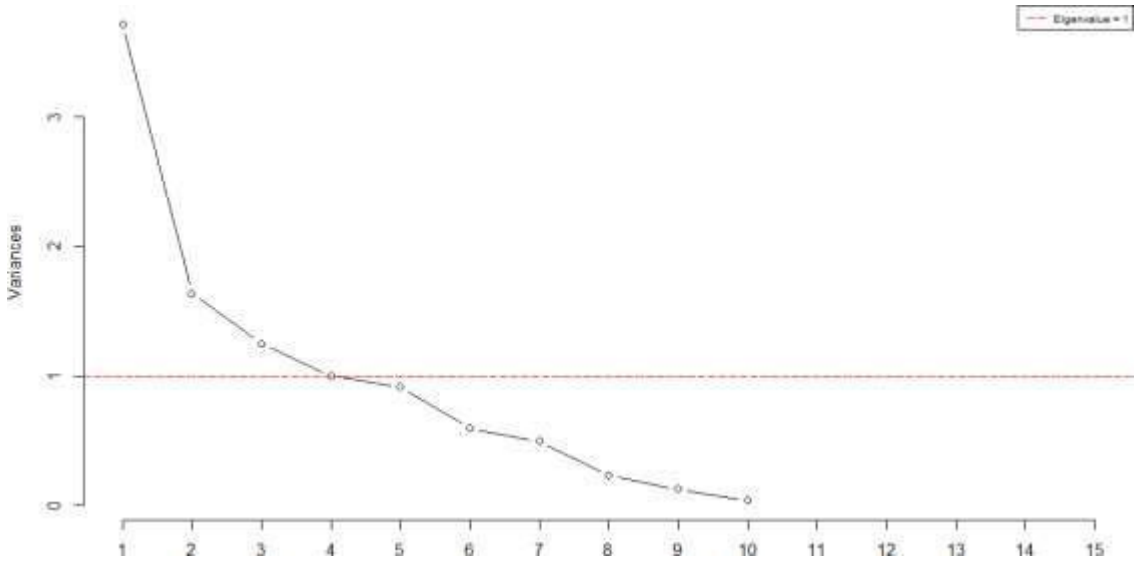

Figure 8: Scree plot showing the Eigen values greater than one selected as principal component

The Eigen values of different components were plotted as screeplot (Fig-7). Among the 10 components first four principal components that have more than one Eigen values

were selected. A scree plot shows how much variation each principal components captures from environmental variables. The Y axis is eigenvalues which essentially stand for the amount of variation. In the present analysis initial four components were selected. The other variables do not contribute significant to the frog site occupancy.

5. DISCUSSION

Amphibians generally are habitat specific. Characteristics like ectothermy, cuticular respiration, life style that is bimodal and need of specific temperature made amphibians highly vulnerable to events that are either human made or natural Krishnamurthy (2016). Their habitat specificity and less dispersal abilities can lead them to local extinction. So, it is very important to completely understand the habitat requirements of amphibians, detailed study on microhabitats used by amphibians in a particular vegetation community and also mechanistic studies that link use of habitat to population dynamics are essential (Armstrong , 2005). Microhabitat preference of Golden-backed frogs of the genus *Indosylvirana* (Ranidae) was studied from South Wayanad Forest Division, Kerala from January 2021 to March 2021. The present study was conducted on two *Indosylvirana* species, *Indosylvirana indica* and *Indosylvirana flavescens*. For the study purpose, three streams were randomly selected from South Wayanad Forest Division.

Out of the three sites studied, highest individual count was observed from site A with a maximum count of 29 on day 2. The maximum number of individuals observed from site B is 22 on day 1. On site C, the maximum count was observed on day 2 with a count of 19 individuals. Since, the study was conducted during pre-monsoon period, irregular rainfall may influence the activity pattern of frogs and that may be the reason for the varying individual count of frogs at each site at each day. As compared to the other two sites, Site A was having a greater number of herb and shrub species. Though grazing activity is observed in site A and there is a small bridge that cuts the stream into two, most part of the stream surveyed is not as disturbed as Site B and Site C. The study conducted by Ekness and Randhir (2007) showed that amphibian habitat tends to decrease as the stream order and disturbance to the stream increases. Since stream in site A is a second order stream whereas the other two streams are third order streams and also because of the less anthropogenic activities, Site A is found to have highest number of individual frog count per day.

To study the microhabitat specificity of frogs of the genus *Indosylvirana*, the environmental parameters that were taken into consideration were weather, temperature, humidity, substrate type, substrate temperature, height of occupancy, water velocity, distance from bank and stream width. Though the pre-monsoon period had irregular rain

pattern, at each site the weather was clear during those night hours when the study was conducted. For Site A, Site B and Site C the

The substrate characteristics had two components. One was substrate type and substrate temperature. Leaf, reed, rock, root and soil were the four types of substrates on which the frogs were found. Out of these four types of substrates, 59% of the observed frogs prefer to occupy the substrate rock. Thus, rocks are the highly preferred substrates for the Golden-backed frogs studied. Where as in a study conducted by Prabhath *et al.*, (2018) showed that plant cover that are submerged were the most preferred substrates for the frog *Minervarya greenii*. The size of the rock in the present study varied from very small rocks to bed rocks and boulders. Most frogs of the genus *Indosylvirana* prefer streams that flows throughout the year where they are found perched on rocks seen on stream bank (Biju *et al.*, 2019), so streams with rocks, and boulders are the ideal habitat for Golden-backed frogs. Also, most of the frogs sited were males. In order to increase their visibility to the females, they predominantly choose rock as their substrate. This is contrary with the results of the study conducted by Smith *et al.*, (2003) where they found that the cricket frogs stayed at the proximity of the edge of their shelter than open space.

From the present study, it was found that 69 out of the 122 observed frogs were found to occupy at a height ranging from 0-10cm and distance from the bank where the maximum number of frogs occupied their space ranged from 0-50cm. In a study conducted by Prabhath *et al.*, (2018), they found that *Minervarya greenii* occupy its space at a height of one meter in its aquatic habitat and in terrestrial habitat the species occupy their space at a height maximum of three meters from ground. Also, their study showed that highest number of frogs was observed in no more than a distance of one meter from the water body to the bank. Thus, similar to *Minervarya* species *Indosylvirana* also tend to occupy greater per cent near to water body.

Highest number of the frogs were observed at distance to the bank that is ranging from 0-50 cm. Thus, there were cubic relation between the site occupancy and stream width which was significant with R^2= 0.14. This could be a strategy of *Indosylvinrana* sp. attributed to avoid or escape from predators from the water especially snakes and birds. The results showed that Golden frog *Indosylvirana* sp. prefer to stay close to the stream bank. In parts of the stream were stream width ranges from 1000-1500cm, frogs were observed at shallow regions that are far from stream bank.

From PCA, it was found that four components accounted for 76% of total variance. Component 1 was temperature and topographic factors, component 2 was Stream width, component 3 was Height and substrate and component 4 was water velocity. The, factors such as substrate temperature, ambient temperature and altitude are the prime factors that determine the distribution of the frog species. This was followed by the stream width. The other factors such as substrate types, and water velocity were the secondary and tertiary factors that determine the frog (species) distribution. A similar observation was made in a study conducted by Smith *et al.*, (2003), in which they found that cricket frogs distinctly choose substrates that are moist. They also suggest that the cricket frogs may choose their microhabitat on the basis of temperature.

In the present study, it was found that water velocity is negatively correlated with the occurrence of frog species. On contrary, Thomas *et al.* (2019) showed that out of the four stream habitat variables water velocity, substrate slope, water column depth and water temperature, water velocity was found to predict the abundance of the purple frog tadpoles in the streams surveyed. Water velocity was positively correlated with the abundance of the tadpoles. The microclimatic study conducted by Priti *et al.* (2021) on tadpoles of three Endemic *Nyctibatrachus* species showed that slope is a major environmental variable that accounts for the occurrence of the tadpoles. The present study did not focus on the slopes of the streams. Measuring the stream slope would have given an even more precise idea on the microhabitat preference of the two Golden-backed frogs.

From the current study, it is concluded that both *Indosylvirana flavescens* and *Indosylvirana indica* select their microhabitat based on the environmental parameters such as substrate temperature, ambient temperature, altitude, stream width. The other factors such as substrate types, and water velocity were the secondary and tertiary factors that determine the frog (species) distribution. Streams with more rocky substrates, low water velocity are most preferred by the two species. Majority of the individuals of the two species prefer to occupy at a height ranging between 0-10cm and in reeds and other surrounding vegetation, they were found to occupy at a height maximum of 120 cm. Present study also showed that increased the anthropogenic activities reduced the number of frogs surveyed per study site. Clearing the herb and shrub species, nearness to the road and human settlements are considered as the primary cause of decline of the Golden-backed frog along with several other species like *Micrixalus saxicola*, *Nyctibatrachus* species,

Clinotaursus curtipus, *Indirana* sp., Minervarya sp., *etc.* that co-exist with them. Local people who inhabit near to the stream can play a major role in saving the stream from degradation and thereby saving the frog species that are relaying on these streams. Better action plans from local government against the unauthorized alteration of streams will help to prevent the habitat degradation of the Golden-backed frogs. As species identification of golden-backed frogs is difficult, most of these frog species are understudied. So, more studies should be conducted on these golden-backed frogs so that proper action plans can be implemented to save the species from its decline.

6. SUMMARY

Site preference of frogs of the genus Indosylvirana (Ranidae) was studied from South Wayanad Forest Division, Kerala from January 2021 to March 2021. For the study purpose, three streams were randomly selected from South Wayanad Forest Division. The three selected streams for the study were in close proximity with human settlements and high human activities are observed in these streams. Stream in site A was narrow with moderately closed canopy. Stream in site B was rocky with boulders and rocks. And the stream width varied from 9 to 11 m. Site C is located near Sugandhagiri. The stream is surrounded by human settlements. The maximum and minimum width of the stream is 5m and 11m respectively. In three of these streams, activity of the selected frogs peaked after 1800 hours.

The frogs *Indosylvirana indica* and *Indosylvirana flavascens* were surveyed using transect method. To record the vegetation data, plots of 5×1 m were laid with an interval of 25 m. Thus, from each stream there were a total of 24 plots. In each of these plots, a 1×1m plot was laid for the herbs and grasses. The canopy cover and leaf litter cover of these plots were also recorded. The vegetation data showed that Site A contains about 16 shrub species and 10 grass species. The average canopy cover and leaf litter cover are 49.13% and 26.04 % respectively. Site B has an average canopy cover of 69.23 %. The average leaf litter cover of the stream is 53.07. The undergrowth is low with three species of shrubs and four species of herbs. In Site C, the stream is surrounded by 7 shrub species and seven species of herbs. The average canopy cover of the stream is 26.62 % and average leaf litter cover is 21.56%.

For each frog, variables like substrate type, substrate temperature, height of occupancy, distance from bank, stream width, temperature, humidity, water velocity altitude were recorded. Statistical analysis showed that Rock, Leaf, Root and soil are the four types of substrates on which the frogs occupy their space. Out of these four substrates, rocks (59%) were the most preferred substrates by the frogs. The least preferred substrate was soil with 4.9%. After rock, the most preferred substrate by the frogs was leaf (23.8%) followed by root (7.4%).

Descriptive statistics test results showed that substrate temperature varied from 17.8°C to 24.6 °C. 750 cm was the maximum height of occupancy of the frogs. Water velocity varied from 0to 1 m⁄s. Distance to the bank from the substrate on which the frogs were occupying

varied from 0 to 400cm. Width of the portion of the stream at which the frogs were present varied from 250 to 1120cm.

Results of principal component analysis showed that defined four components accounted for 76% of total variance. Component 1 was Temperature and topographic factors, component 2 was Stream width component 3 was Height and substrate and component 4 was Water velocity. Among these four components the first two components consisted of Topographic factors, atmospheric temperature and stream features that explained 53% of total variance. Thus, the factors such as substrate temperature, ambient temperature, location types and altitude are the prime factors that determine the distribution of the frog species. This was followed by the stream width. The other factors such as substrate types, and water velocity were the secondary and tertiary factors that determine the frog (species) distribution.

The PCA biplot in the Euclidean space showed that substrate temperature and ambient temperature and distance to banks were ordinated together with less than 20 have strong positive correlation. Similarly, humidity, height occupancy and water level were strongly intercorrelated. On contrary altitude is strongly negatively correlated with the temperature and locations. Substrate is negatively correlated with the humidity and height of occupancy. The length of the line indicates the strength of the correlation.

Present study also showed that increased the anthropogenic activities reduced the number of frogs surveyed per study site. Clearing the herb and shrub species, nearness to the road and human settlements are considered as the primary cause of decline of the Golden-backed frog along with several other species like *Micrixalus saxicola*, *Nyctibatrachus* species, *Clinotaursus curtipus, Indirana* species *Minervarya* species *etc.* that co-exist with them. Local people who inhabit near to the stream can play a major role in saving the stream from degradation and thereby saving the frog species that are relaying on these streams. Better action plans from local government against the unauthorized alteration of streams will help to prevent the habitat degradation of the Golden-backed frogs. As species identification of golden-backed frogs is difficult, most of these frog species are understudied. So, more studies should

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7. REFERENCE

Armstrong, D. P. 2005. Integrating the Metapopulation and Habitat Paradigms for Understanding Broad-Scale Declines of Species. *Conserv. Biol*. **19**(5): 1402-1410.

Babbitt, K. J., Veysey, J. S. and Tanner, G. W. 2010. Measuring habitat. In: Dodd CK ed. Amphibian ecology and conservation: A handbook of techniques. Oxford University Press. 299-317p.

Bahiah, S. N., M.T., M., M.S., H.N., And H., M.Z. 2019. Microhabitat preference of frogs at similajau national park, sarawak, malaysia. *Int. J. Sci. Res. Publ.* **9**: (4): 8819.

Bardua, C., Fabre, A. C., Clavel, J., Bon, M., Das, K., Stanley, E. L., Blackburn, D. C., And Goswami, A. 2021. Size, microhabitat, and loss of larval feeding drive cranial diversification in frogs. *Nat. Commun.* **12**: (1): 1–13.

Biju, S.D., Garg, S., Mahony, S., Wijayathilaka, N., Senevirathne, G., And Meegaskumbura, M. 2019. Dna barcoding, phylogeny and systematics of golden-backed frogs (Hylarana, Ranidae) of the Western ghats-Sri Lanka biodiversity hotspot, with the description of seven new species. *Contrib. to Zool.* **83**: (4): 269-s4.

Boulenger, G. A. 1882. *Catalogue of the Batrachia Salientia s. Ecaudata in the Collection of the British Museum*, Ed). British Museum (Natural History), Taylor and Francis, London.

Das, S. 2015. A checklist of amphibians of Kerala, India. *J. of Threatened Taxa*. **7**(13):8023-8035.

Dare, G. R. C. D., Urray, R. G., Courcelles, D. M. M., And Malt, J. 2020. Run-of-river dams as a barrier to the movement of a stream-dwelling amphibian. **11**: 144-151.

Dey, M. 2010. A study on habitat selection and probable anthropogenic threats of anurans in barak valley, Northeast India. *Assam Univ. of Sci. and Technol.* **6**(1): 28-36.

Dinesh, K. P., Radhakrishnan, C., Channakeshavamurthy, B. H., Deepak, P and Kulkarni,N. U. 2019. A checklist of amphibians of India with IUCN conservation status. *Fauna of India Checklist.*

Dupuis, L., And Steventon, D. 1999. Riparian management and the tailed frog in northern

coastal forests.*For. Ecol. Manage.* **124**: (1): 35–43.

Ekness, P., And Randhir, T. 2007. Effects of riparian areas, stream order, and land use disturbance on watershed-scale habitat potential: an ecohydrologic approach to policy 1. *J.of the American Water Resourse Association.* **43**: (6): 1468-1462.

Hall, Linnea S., et al. "The Habitat Concept and a Plea for Standard Terminology." *Wildlife Society Bulletin.* **25**(1): 173–82.

Hopkins, W.A. 1991. Amphibians as models for studying environmental change. *ILAR J.***48** (3): 270-277.

Jerdon, T. C. 1853. Catalogue of reptiles inhabiting the Peninsula of India. *Journal of the Asiatic Society of Bengal* **22**: 522-534.

Johnson, D.H. 1980. The comparison of usage and availability measurements for evaluating resource preference. *Ecol.* **61**(1): 65-71.

Krishna, S.N., Krishna, S.B., And Vijayalaxmi, K.K. 2005. Variation in anuran abundance along the streams of the western ghats, india. *herpetol. j.* **15**: (3): 167–172.

Reddy, A. H. M., Gururaja, K. V. And Krishnamurthy, S. V .2016. Habitat features of an endemic anuran *micrixalus saxicola* jerdon , 1983 (amphibia : ranidae) in central Western ghats , India. *amphibia-reptilia* **23:** 370-374.

Machtans, C. S., Villard, M. and Hannon, S. J. 1996. Use of riparian buffer strips as movement corridors by forest birds. *Conserv. Biol.* ***10*** (5): 1366–1379.

Murali, R., And Raman, T. R. S. 2012. Streamside amphibian communities in plantations and a rainforest fragment in the anamalai hills, india. *J. Threat. Taxa* **4**: (9): 2849–2856.

Naniwadekar, R., And Vasudevan, K. 2014. Impact of dams on riparian frog communities in the southern western ghats, india. *Diversity* **6**: (3): 567–578.

Navas, C. A. 1996. Implications of microhabitat selection and patterns of activity on the thermal ecology of high elevation neotropical anurans. *Ecologia*. **108**: 617–626.

Oliver, L. A., Prendini, E., Kraus, F., And Raxworthy, C.J. 2015. Systematics and biogeography of the hylarana frog (anura: ranidae) radiation across tropical

australasia, southeast asia, and africa. *Mol. Phylogenet. Evol.* **90**: 176–192.

Polo-Cavia, N., Burraco, P., And Gomez-Mestre, I. 2016. Low levels of chemical anthropogenic pollution may threaten amphibians by impairing predator recognition. *Aquat. Toxicol.* **172**: 30-35.

Prabhath, M. C., Jayasekara, E. G. D. P, And Mahaulpatha, W. A. D. 2018. Habitat preference of endangered frog *fejervarya greenii* (amphibia: dicroglossidae) in tropical montane cloud forests of sri lanka . *Int. J. Zool. Stud.* **3** (5): 05-11.

Priti, H., Gururaja, K. V., Aravind, N. A. And Ravikanth, G. 2021. Influence of microhabitat on the distribution of tadpoles of three endemic nyctibatrachus species (nyctibatrachidae) from the Western ghats , India. 01–11.

Vijayakumar, S. P., Ragavendran, A. And Choudhury, B. C. 2006. Herpetofaunal assemblage in a tropical dry forest mosaic of Western Ghats, india: preliminary analysis of species composition and abundance during the dry seaso*assemblage* .**30**: (1): 40–53.

Raghunath, S., Veerabhadrappa, C., And Krishnamurthy, S. 2017. Nuclear abnormalities in erythrocytes of frogs from wetlands and croplands of western ghats indicate environmental contaminations. *j. trop. life sci.* **7**: (3): 208–212.

Richards-Zawacki, C.L. 2009. Effects of slope and riparian habitat connectivity on gene flow in an endangered panamanian frog, atelopus varius. *divers. distrib.* **15**: (5): 796–806.

Rodríguez-Mendoza, C., And Pineda, E. 2010. Importance of riparian remnants for frog species diversity in a highly fragmented rainforest. *biol. lett.* **6**: (6): 781–784.

Smith, G. R., Todd, A., Rettig, J. E., and Nelson, F. 2003. Microhabitat selection by northern cricket frogs (Acris *crepitans*) along a west- central missouri creek: field and experimental observations published by: the society for the study of amphibians and reptiles. *J. Herpetol.* **37**: (2): 383–385.

Tabacchi, E., and Dcamps, O. 1990. Continuity and discontinuity of the riparian vegetation along a fluvial corridor *Landscape ecol*. **5**: (1): 9–20.

Tilman, D. 1994. Competition and Biodiversity in Spatially Structured Habitats. *Ecol* **75**:

(1): 2–16.

Thomas, A., Das, S., and Manish, K. 2019. Influence of stream habitat variables on distribution and abundance of tadpoles of the endangered Purple frog, *Nasikabatrachus sahyadrensis* (Anura: Nasikabatrachidae). *J. Asia-Pacific Biodivers.* **12**: (2): 144–151.

MICROHABITAT PREFERNCE OF GOLDEN-BACKED FROGS (*Indosylvirana sp.*) IN RIPARIAN HABITATS OF SOUTH WAYANAD FOREST DIVISION

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(18-MSVP-05)

Abstract of Dissertation Submitted in Partial Fulfilment of the Requirement

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Kerala Veterinary and Animal Sciences University

CENTRE FOR WILDLIFE STUDIES

COLLEGE OF VETERINARY AND ANIMAL SCIENCES

POOKODE, WAYANAD, KERALA, INDIA

8. ABSTRACT

Site preference of frogs of the genus Indosylvirana (Ranidae) was studied from South Wayanad Forest Division, Kerala from January 2021 to March 2021. For the study purpose, three streams were randomly selected from South Wayanad Forest Division. The frogs *Indosylvirana indica* and *Indosylvirana flavascens* were surveyed using transect method. To record the vegetation data, plots of 5×1 m were laid with an interval of 25 m. Thus, from each stream there were a total of 24 plots. In each of these plots, a one meter^2 plot was laid for the herbs and grasses. The canopy cover and leaf litter cover of these plots were also recorded. Based on the study, it is concluded that *Indosylvirana* sp. select their microhabitat based on the environmental parameters such as substrate temperature, ambient temperature, altitude, stream width. The other factors such as substrate types, and water velocity were the secondary and tertiary factors that determine the frog (species) distribution. Streams with more rocky substrates, low water velocity are most preferred by the two species. Clearing the herb and shrub species, nearness to the road and human settlements are considered as the primary cause of decline of the Golden-backed frogs. From the active participation of local people and better action plans from the local government to protect riparian habitat, frogs that are relaying on riparian habitat can be saved at the least from local extinction

KERALA VETERINARY AND ANIMAL SCIENCES UNIVERSITY

Faculty of College of Veterinary and Animal Sciences

PROGRAMME OF RESEARCH WORK FOR

DISSERTATION FOR MASTER OF SCIENCE DEGREE

1. Title of Dissertation

Microhabitat preference of golden-backed frogs (*Indosylvirana sp.*) in riparian habitats of South Wayanad forest division.

2a. Title of department/KVASU research project of which this forms a part

Nil

2b. Code No. if any, and order by which the department/KVASU research project is approved

Nil

3a. **Name of the student**

Arathy S. Madhu

3b. Admission No.

18-MSVP-05

4a. Name of Major Advisor

Dr. Biju S.

4b. Designation

Assistant Professor,

Department of Livestock production and management

College of Veterinary and Animal Sciences, Mannuthy, Thrissur.

5. Objectives of the study

1. To determine microhabitat specificities of frogs of the genus Indosylvirana in riparian habitat.

6. Practical/Scientific utility

Habitat is contemplated as resources and environments in a particular area where an animal live and reproduce Tilman, (2008). Habitat preference or selection of habitat is the process by which an individual organism selects a habitat from the available alternative resources. (Babbitt *et al*., 2010).

Frogs are vertebrates that are highly sensitive to changing environments. Their population is declining considerably because of

human intervention, habitat destruction and fragmentation, change in microhabitat, climate change *etc*. Frogs generally are habitat specific and also there are some generalist species that can live in any habitat type. Because amphibians show habitat specificity and have less dispersal capabilities disturbances can lead them to local extinctions.

Any animal decides its habitat on the basis of many ecological and behavioral characteristics that then mad

use to perform their daily activities (MC Prabhath *et al* 2018).

Present study focusses on the site preference of Ranid frog Indosylvirana. Though frogs of genus Indosylivirana can be found near stagnant water bodies, they are abundant near streams and creeks. The present study will focus on the frogs *Indosylvirana flavescens* and *Indosylvirana indica. They* are commonly known yellowish Golden- backed frog and Rao's intermediate golden-backed frog. The species inhabits fast and constantly flowing streams. They usually perch on rocks that are seen on the banks of the streams. They are also seen on small bushes near the streams. During breeding season, they lay eggs on shallow regions of the stream Biju *et al.*, (2019). In this study, the microclimatic conditions which will determine the site preference of thefrogs *Indosylvirana flavescens* and *Indosylvirana indica* will be checked.

Riparian vegetation is an important trait of the landscape because it forms a link between terrestrial and aquatic systems and can function as corridors (Tabacchi *et al.* 1990, Malanson 1993, Machtans *et al.* 1996, Naiman and De´camps, 1997) Amphibian communities in stream side occupy a sensitive environment and they can be considered as indicators of effects of adjoining land uses (Murali. and Raman. 2012). In this study, the microhabitat parameters will be assessed to determine the site preference of the Golden-backed

frogs (Indosyvirana sp.) utilize the riparian habitat.

Habitat preference studies will help us to identify the potential microhabitat for specific species. The results of such studies can prevent developmental activities in those habitats thus protect that particular site from degradation.

7. Important publications on which the study is based

Out of the ten species of Indosylvirana described from India, eight of them are seen in Kerala Dinesh *et al* (2019).

On the basis of molecular study conducted by Biju *et al* in the year 2015, Wayanad has three species of the genus Indosylvirana, I. flavescens and I. intermedius.

In a study conducted by (Smith *et al.*, 003), they marked the location where they spotted the frog and avoided surveying the same spot to avoid repeated counting. They also collected habitat variables data from random sites.

In a study conducted by (Murali and Raman, 2012) used quadrat method and

Visual Encounter Survey methods to survey amphibians in streamside of plantations and rainforest fragment.

(Prabhath *et al.*, 2018) studied the habitat preference of the frog Minervarya greenii. They found that the species was recorded only from 1m above water level and 3m above terrestrial habitat. They also found that the Minervarya greenii is a generalist species inhabiting in and around lentic water bodies.

Bahiah *et al* (2019) studied microhabitat preference of frogs at Similajau National ParkSarawakk, Malaysia where they layed transect of 300m to survey the frogs.

(Thomas *et al.*, 2019) studied how the four stream habitat variables water flow velocity, substrate slope, water column depth and temperature of water influence the rheophilous tadpoles of the rare and endangered frog Nasikabatrachus sahyadrensis. Along 100m transects of two selected streams, they laid 68 grids.

Their analysis showed that abundance of the tadpoles in the two streams are best predicted with water flow velocity.

8. Outline of the technical programme

Study area

The proposed study will be conducted in selected areas of South Wayanad forest division. Three different streams will be selected for the study. The streams are randomly selected from the study area. Pilot surveys will be conducted to confirm the presence of the selected species in those streams.

Sampling Method

Frogs will be surveyed using transect method. In each stream, a transect of 200m will be laid to survey the frogs. The entire length of the 200m transect will be surveyed using Visual Encounter Survey (VES). Frogs will also be spotted by following their calls. Close contact with frogs will be avoided the maximum to avoid movement of frogs from one substrate to another. After spotting a frog, variables like substrate type, substrate temperature, height of occupancy, distance from bank, stream width, temperature, humidity, water velocity altitude will be recorded.

A head torch with good visual efficiency will be used to search for the frogs. To record the water velocity, temperature and humidity, Kestrel Weather station will be used. Temperature of the substrate will be recorded using an infrared thermometer. Measuring tapes will be used to measure the stream width and distance of the substrate from the stream bank. Latitudinal and longitudinal values are obtained from the mobile app GPS Essentials.

To record the vegetation data, plots of 5×1 m will be laid with an interval of 25 m. Thus, from each stream there will a total of 24 plots. In each of these plots, a 1×1m plot will be laid for the herbs and grasses. The canopy cover and leaf litter cover of these plots will also be recorded.

In every transect, two random plots are laid where there is less chance of spotting the species of interest. From those plots, two random substrates

are chosen and all the environmental variables recorded for substrates occupied with the frog species are also recorded from it. This is done to compare the environmental parameters associated with substrates that are chosen by the frogs to occupy with that of substrates on which frogs are not found. Vegetation data are also recorded from these random plots.

Photo documentation of the frog species and the habitat of each surveyed stream will be done.

The data collected will be subjected to statistical analysis for comparison.

9. Main items of observations to be made

1. Identification of the frog species using direct observation, call and photographs.
2. Substrate identification.
3. Data collection of environmental rameters.

10. Facilities

a. Existing: Camera, Flashlight, Infrared thermometer, Kestrel weather station, Measuring tape.

b. Additional facilities required:
Correction pen, Tracker.

11. Duration of study:

One Semester

12. Financial estimate

Total: 15000

13. Signature of student

14. Signature of Major Advisor

Place: Pookode

Date:

15. Name, designation and signature of members of the Advisory Committee.

1. Dr. Biju. S (Guide)

Assistant Professor.

Department Livestock Production and Management.

College of Veterinary and Animal Sciences, Mannuthy, Thrissur.

2.Dr. Biju Habeeb.

Assistant professor

Clinical Veterinary Medicine, Ethics and Jurisprudence.

College of Veterinary and Animal Sciences.

Pookkode, Wayanad.

3.Dr. George Chandy

(Course Director).

Officer in Charge,

Centre for Wildlife Studies,

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Appendix I

Reference

Bahiah, S. N., M.T., M., M.S., H.N., And H., M.Z. 2019. Microhabitat preference of frogs at similajau national park, sarawak, malaysia. *Int. J. Sci. Res. Publ.* **9**: (4): 8819.

Biju, S.D., Garg, S., Mahony, S., Wijayathilaka, N., Senevirathne, G., And Meegaskumbura, M. 2019. Dna barcoding, phylogeny and systematics of golden- backed frogs (hylarana, ranidae) of the western ghats-sri lanka biodiversity hotspot, with the description of seven new species. *Contrib. to Zool.* **83**: (4): 269-s4.

Dinesh, K. P., Radhakrishnan, C., Channakeshavamurthy, B. H., Deepak, P and Kulkarni, N. U. 2019. A checklist of amphibians of India with IUCN conservation status. *Fauna of India Checklist.*

Murali, R., and Raman, T.R.S. 2012. Streamside amphibian communities in plantations and a rainforest fragment in the Anamalai hills, India. *J. Threat. Taxa* **4**: (9): 2849–2856.

Smith, G.R., Todd, A., Rettig, J.E., and Nelson, F. 2003. Microhabitat Selection by Northern Cricket Frogs (Acris crepitans) along a West- Central Missouri Creek: Field and Experimental Observations Published By: The Society for the Study of Amphibians and Reptiles. *J. Herpetol.* **37**: (2): 383–385.

Prabhath, M.C., Kayasekara, E.G.D.P., Mahaulpatha, W.A.D. 2008. Habitat preference of endangered frog *Fejervarya greenii*(Amphibia: Dicroglossidae) in tropical montane cloud forests of Sri Lanka. Int. J. of Zool. Stud. **3**:(5): 05-11.

Thomas, A., Das, S., and Manish, K. 2019. Influence of stream habitat variables on distribution and abundance of tadpoles of the endangered Purple frog, Nasikabatrachus sahyadrensis (Anura: Nasikabatrachidae). *J. Asia-Pacific Biodivers.* **12**: (2): 144–151

CERTIFICATE

Certified that the research project has been formulated observing the stipulations laid down under the Prevention of Cruelty to Animals Act (Amendment, 1998).

Place: Pookode

Date:

Dr, Biju S.

Guide

CURRICULUM VITAE

Name of the candidate	**Arathy S Madhu**
Date of Birth	**01/05/1998**
Place of Birth	**Kadakkal**
Marita Status	**Unmarried**
Major Field of Specialization	**Wildlife Studies**
Permanent Address	**Melevilaveedu,** **Mandiramkunnu,** **Mathira P O, Kadakkal,** **Kerala, India Pin: 691536**
	Email id- shylockhoney18@gmail.com
Professional Experience	**Nil**
Publications made	**Nil**
Membership in Professional bodies	**Nil**