Observations and documentation of amphibian diversity from a human -modified ecosystem of Darjeeling, with record occurrence of *Polypedates himalayanus* from Darjeeling Hills, West Bengal

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ABSTRACT

A survey was carried out to document the amphibian diversity at Takdah Cantonment (27°02'N-88°21'E) in Kurseong Subdivision of District Darjeeling, West Bengal, India, an integral part of the Eastern Himalayas. Time constrained visual encounter survey (VES) method was used for sampling amphibians from all possible habitats of the study area. A total of nine species of amphibians belonging to four families across five genera were recorded during the study. *Polypedates himalayanus* was also for the first time recorded from Darjeeling Hills. This study reveals that the area which is at an elevation of 1440-1650m is rich in amphibian diversity. Further studies are needed on population structure, habitat use by amphibians for better understanding and also imposition of several conservation strategies in Darjeeling district of West Bengal is needed.

Key words: Amphibian, diversity, Darjeeling, Takdah Cantonment, VES, relative abundance.

INTRODUCTION

The first vertebrate animals are amphibians and they have two life stages namely tadpoles (occur in water) and adults (on land). As it is known that amphibians are comprised of frogs, toads, caecilians and salamanders those are extremely varied. Many habitat types may occur within an area, but amphibians may utilize only a few of these. The number of individuals that represents each species in community may vary from place to place depending on the amount of rainfall, available habitats and human interference as the structure and diversity of an amphibian community is determined by the availability of food, moisture and micro habitat (Daniels, 1992). Amphibians are potentially good bio-indicators due to their highly permeable skin and dual mode of life (Beebee and Griffiths 2005). The amphibian fauna comprises 7788 species in the world (Frost, 2018). In India 405 species belonging to existing orders, namely Caudata, Anura and Gymnophiona were reported. In West Bengal 45 species belonging to 14 genera, seven families and three orders were identified (Deuti et al., 2009). Published comprehensive lists of amphibians of the study area are very less. Daniel (1962) gave account of seven species of amphibians of Darjeeling area of West Bengal. Sarkar et al., (1992) published list of amphibians of the state of West Bengal where they included 30 species of amphibians from Darjeeling district. Dinesh et al. (2009) in the list of amphibians of India included some species from Darjeeling district. Similarly, 44 species of amphibians belonging to 20 genera in eight families and three orders have been reported in adjoining state of Sikkim from 1864 to 2015 (Subba et al., 2016). About 30% - 57% of the amphibians in India are threatened due to loss of natural habitats. So far, presence of 37 species of amphibians under 18 genera, eight families and three orders has been described from Darjeeling district, West Bengal (De, 2016).

AJCB: SC0028

MATERIALS AND METHODS

Study area

The study was conducted in Takdah Cantonment (27° 02'N-88°21'E) in the outskirts of Darjeeling town about 27 km away, located between Kalimpong and Darjeeling, with an elevation of < 1440 to > 1650m, which is also an integral part of Eastern Himalayas (Fig. 1). The study area included human settlements, agricultural land, streams and forests.

Vegetation and Climate

The Study area has an average elevation of 1550m and thus has sub-tropical moist evergreen forest type. Some dominants trees found here are *Cryptomeria japonica*, *Exbucklandia populnea*, *Alnus nepalensis*, *Ficus hookeri*, *Elaeocarpus varunua*, *Pentapanax leschenaultia*, *Erythrina arborescens*, *Castanopsis hystrix*, etc. while some commonly found shrubs are *Polygonum molle*, *Girardinia palmate*, *Rubus paniculatus*, *Rubus ellipticus*, *Fragaria nubicola*, *Sinarundinaria maling*, *Urtica dioca*, etc.

There are no major rivers draining the forest but numerous non-perennial streams run through some parts of the forest, keeping the forest floor wet and damp for most of the time in the year. Summer, monsoon and post monsoon seasons are accompanied by fogs and mist. Monsoon months lasting from May – September, is characterized by heavy rainfall 2-3 times a day and sometimes rain spells lasts for more than 72 hours.

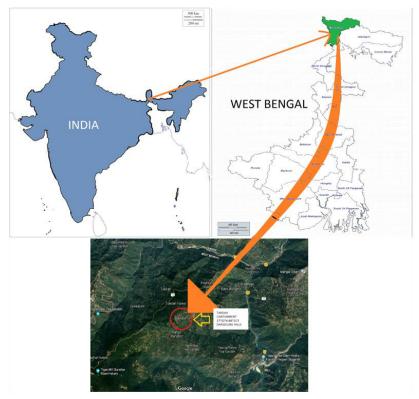


Figure 1. Map showing the position of the study area and satellite image of a fraction of Darjeeling – Sikkim Himalayas showing the Study Area. (Source: Google Maps)

In 2017-18, the study area received highest rainfall (about 1616mm) in the month of May followed by September (1467mm), it also experiences hail storms in the summer months. While the winter months were dry with just occasional winter showers mostly in the month of February. With an average temperature of 23°C, August was the warmest month, while January was the coldest month of the year with an average temperature of 9°C (World Weather Online, 2018).

Sampling

Field sampling was done from March 2017 to February 2018 in Takdah Cantonment, Darjeeling hills.

We followed time constrained visual encounter survey (VES) method in the study area following Heyer et al., (1994) and Chettri et al. (2011) for sampling amphibians in all probable habitats and microhabitats, such as agricultural fields, wet lands, rocky areas, open lands, boulders, logs and mosses. We conducted counts and took photographs while walking along the main road and various other pony roads and trek routes in the study area. The observations were mostly carried out during the day (10:00 hrs to 13:00 hrs) and evening (17:00 hrs to 19:00 hrs), however hours of heavy rainfall were avoided. Individuals of the same species found more than once in the same site in the same visit was not counted if not observed together at the same time. In addition, opportunistic observations were also included. The parameters noted during observation include name of the species, number of individuals of that species and habitat in which the species was found.

The species were identified by using Liu (1950), Daniels (1962), Sarkar *et al.* (1992), and Sanyal *et al.* (2006). In cases where instant identification was not possible,

photographs were taken and sent to experts for identification. Scientific names and systematic position followed in this paper are after, Frost (2017) and common names are obtained from , Dinesh *et al.* (2009) and De (2016). We completed a total of 24 visits to the study area (two in each month during the study period) totaling 158 sampling hours. No collection of amphibian specimens was done during this study in Darjeeling.

Data Analysis

Community parameters such as species richness, abundance, Shannon-Weiner diversity index and evenness were calculated for total samples following Magurran & McGill (2011). Species richness was considered as the total number of species observed and species abundance as number of individual amphibians counted during the sampling. Relative abundance of each species encountered during the survey was also calculated as done by Chettri et al. (2011). Based on these data, relative abundance of each species was obtained. The diversity was analyzed using Shannon-Wiener diversity index (H') = - Σ pilnpi; where pi = proportion of total sample belonging to ith species, ln= natural logarithm (Shannon & Weaver 1949). Similarly, evenness was calculated using the formula: Evenness (J) = H'/Hmax where Hmax = lnS, S =number of species, H' = Shannon-Wiener Diversity (Pielou 1969). To understand dominance in the community Dominance (D), Simpson's index (1-D) and Berger-Parker Dominance (d) were calculated using Past 3.19 (Hammer et al., 2001).

RESULTS

Species richness, diversity and abundance

The present study could only manage to record species of Order Anura. A total of 48 individuals representing nine

Table 1. List of Amphibian species recorded during the study and their habitat preferences. Number represents the abundances of amphibians observed during the study.

Im- age No.	Taxon	Common Name	Total	Relative Abundance	*Habitat	IUCN Red List (Ver. 3.1)
	Bufonidae					
5a	Duttaphrynus himalayanus Günther, 1864	Himalayan Toad	21	43.75 %	Ag, Hs, Wb, Ol	LC
	Rhacophoridae					
5b	Polypedates himalayanus Boulenger, 1888	Himalayan bush Frog	1	2.08 %	Ff	LC
5c	Raorchestes annandalii (Boulenger, 1906)	Annandale's bush Frog	3	6.25 %	Ag, Ff	LC
5d	Raorchestes sp.	-	1	2.08 %	Ff	-
	Megophryidae					
5e	Megophrys glandulosa Fei et al., 1990	Glandular Horned Toad	5	10.41 %	Ag, Ff	LC
5f	Megophrys robusta Boulenger, 1908	Robust Spadefoot Toad	11	22.91 %	Hs, Ag, Ff	DD
5g	Megophrys sanu Deuti et al., 2017	Small Horned Toad	1	2.08 %	Ag	-
	Dicroglossidae					
5h	Nanorana annandalii Boulenger, 1920	Boulenger's Hill Frog	1	2.08 %	Hs	NT
5i	Nanorana liebigii Günther, 1860	Himalayan Bull Frog	4	8.33 %	Ag, Hs	LC
			Total Species richness = 9			
			Total Abundance = 48			

species belonging to four families and five genera were recorded with photographic evidences (Image 1-9). Among the four families recorded, highest number of species (three) belonged to the family Megophryidae and Rhacophoridae and lowest number of species (one) was recorded from the family Bufonidae (Table 1). The amphibian population in the study area was found to be diverse when tested using various diversity indices (Table 2).

Table 2. Species richness, abundance, diversity and evenness of amphibians observed in the study area during the survey.

Diversity Indices	Calculated Values		
Species richness	9		
Abundance	48		
Dominance (D)	0.2674		
Simpson's index (1-D)	0.7326		
Shannon-Weiner index (H')	1.638		
Evenness (J)	0.7455		
Berger-Parker (d)	0.4375		

Relative abundance of amphibian species in the study area shows sparse distribution (Table 1). Barring a few species, relative abundance of most species was low. Most species were observed only once or twice during the entire course of our study. Of the nine species, four species contributed less than 3% population. Almost half of the total abundance was contributed by single species i.e. *Duttaphrynus himalayanus* (43.75 %) followed by *Megophrys robusta* (22.9 %). This result supports well

established community pattern i.e. a community is dominated by few common species (Heatwole, 1982; Magurran, 1988; Chettri *et al.*, 2011).

The species accumulation curve was rising even at the end of the study period indicating likelihood of detection of additional species from the study area (Fig. 3). It indicates that there was a probability of encountering few additional species in the study area with the increasing sampling effort.

Habitat preferences among species

In the present study we identified variety of amphibian species utilizing five different habitats namely - Agricultural land (six species), Open land (one species), Human settlements (four species), Water bodies (one species) and Forest floor (five species). Here, Human Settlements includes houses, veranda, drains, etc.; Agricultural land includes cultivated and uncultivated crop fields; Forest floor includes bushes, trees, leaf litter, etc; Water bodies includes streams, ponds and small water puddles; Open land includes land devoid of vegetation.

Among these maximum number of species was observed in the Agriculture land (35%) followed by Forest floor (29%), Human settlements (24%), Water bodies (6%) and Open land (6%) (Fig. 2). This indicates that most of the amphibian species require agriculture based ecosystems for survival. Since this study was conducted in an area with increasing number of human settlements, human settlements contributed to the second most utilized habitat, this also shows that few amphibian species have adapted to human interference.

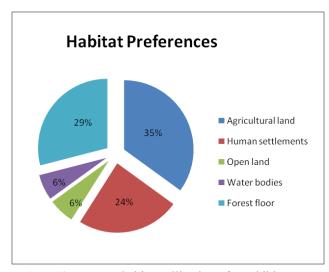


Figure 2. Percent habitat utilization of amphibians recorded during the study period

Season of encounter

Amphibians were encountered in eight of the 12 months of the study period (Fig. 4), while the months of October, November, December and February did not see any individuals even though sampling was as intense as any other months. Maximum number of individuals was recorded in May (14 individuals), followed by June (12 individuals). Similarly, monsoon (May-Aug) saw the highest number of individuals (26 individuals) when counted seasonally, followed by summer (Mar-May), in which 17 individuals were encountered. This shows that like any other place, one can expect to find the highest number of amphibians once there is rainfall in the region.

Account on encountered families and species Family Bufonidae

This family was only represented by *Duttaphrynus himalayanus* (5a.), however this species was encountered the most (21 individuals) during the survey (Table 1). Individuals of this species at different stages of growth were encountered during the study.

Family Megophryidae

This family was only represented by three species belonging to the same genus Megophrys. The most commonly encountered genus during the study was Megophrys. A total of four species of Megophrys have been identified from the Darjeeling District (De, 2016), of which only two species (Megophrys parva and Megophrys glandulosa) were recorded earlier (Sarkar et al., 1992) and later one more species (Megophrys robusta) was added to the recorded data of ZSI (Deuti et al., 2009). Two new species (Megophrys sanu and Megophrys katabhako) of this genus has also been recently recorded by Deuti et al. (2017). In the present study, a total of three different species (Megophrys robusta, Megophrys glandulosa and Megophrys sanu) of this genus were recorded photographically (Fig. 5e- 5g). Megophrys robusta (Fig .5f) was the most encountered (11 individuals) species from this genus (Table 1), this species is placed under Data Deficient (DD) category under IUCN Red List (IUCN, 2017). All the species of this genus are considered to be extremely rare and difficult to find, however they seemed to be fairly common in the months of July-September during the present survey period.

Family Rhacophoridae

This family was represented by two genus- Raorchestes and Polypedates. Two species of Raorchestes were encountered during the survey of which one species was identified to be Raorchestes annandalii (Fig. 5c) while the other could not be identified (Fig. 5d). Polypedates himalayanus (Fig. 5b) was encountered during the month of January which was the only amphibian recorded during the winter months. This is also the first report of this species from West Bengal, India as this species is neither reported by De (2016) or Sarkar et al. (1992) from the state. A single individual was encountered at an elevation of 1600m and photographically recorded in the present study.

Family Dicroglossidae

This family was only reperesented by two species *Nanorana annandalii* (Fig. 5h) and *Nanorana liebigii* (Fig. 5i).

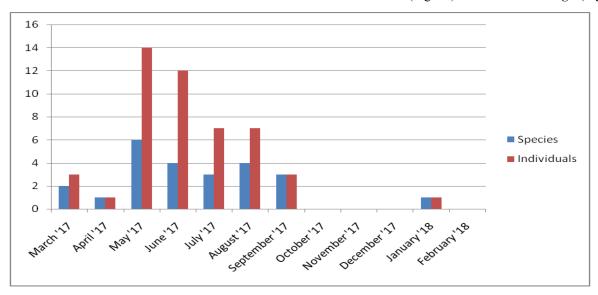


Figure 3. Number of species and individuals encountered in each month during the survey.

N. annandalii is the only species from the study to be placed under Near Threatened (NT) category under IUCN Red List (IUCN, 2017) and was sighted only once. N. liebigii is the most sought after species of edible frogs in the hills and is widely hunted by the locals and four individuals were encountered in the present study.

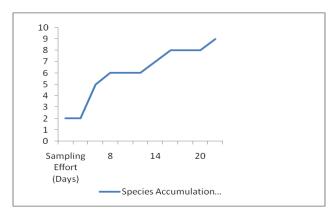


Figure 4. Species Accumulation Curve of Amphibians based on observed species

DISCUSSION

Listing of amphibians of Darjeeling hills have appeared in many compilations, data specific to Takdah Cantonment, Kurseong Sub division is hitherto unknown. No prior study had been carried out in this region for estimation of amphibian population, so this paper tries to estimate the species richness of amphibians in this part of Darjeeling hills. Even though the study was limited to a very small portion of the Darjeeling hills, the present study has managed to photographically document nine amphibian species, which is 20% of total amphibian diversity of the whole Darjeeling district. Furthermore one unrecorded species (*Polypedates himalayaus*) has also been reported in this paper. This suggests that the study area possesses rich amphibian diversity.

The region has many potential habitats for the amphibians to survive ranging from leaf litters in the forest to wetland, springs and torrential streams. The forest here is of moist evergreen temperate type. Heavy rainfall in the region may also be responsible for providing resources required for the survival of amphibians. However, like any other place, even this part of Darjeeling district has seen a sudden rise in human population in recent years. Growing human settlements in the region has resulted in habitat loss of different animals including amphibians. Also unpredictable rainfall, diseases, upward migration of species along the elevation gradient and drying of springs has resulted in decreasing population of amphibians (Acharya & Chettri, 2012). More importantly, drying up of fast flowing streams is detrimental to many Himalayan endemic species such as Nanorana spp., Megophrys spp. (locally termed as 'Paa') which are adapted to torrent streams. Due to their specific adaptation to fast flowing waters, they are restricted to rapid streams. Drying of these streams might gradually lead to extinction of these species (Acharya & Chettri, 2012). These species are also very commonly hunted in the region in the name of local medicine and other delicacies.

Amphibians are important to agriculturalists, they play a key role in ecosystem functioning and act as predator, mainly as consumers of insect pest. Amphibians are mainly seen to be attracted to agricultural land as these kinds of ecosystems can be used for food, home grounds, etc (Narayana *et al.*, 2014). Hence they must be protected by the concerned authorities and awareness regarding the present status of amphibians in the Himalayas must be created among the locals.

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Figure 5. Photograph documentation of the amphibians during the study. 5a. *Duttaphrynus himalayanus*, 5b. *Polypedates himalayanus*.5c. *Raorchestes annandalii*, 5d. *Raorchestes sp.*, 5e. *Megophrys glandulosa*, 5f. *Megophrys robusta*, 5g. *Megophrys sanu*, 5h. *Nanorana annandalii*, 5i. *Nanorana leibigii*.

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