

**SPECIES DIVERSITY AND COMMUNITY  
STRUCTURE OF AMPHIBIANS OF SELECTED  
AGROECOSYSTEMS IN THRISSUR, KERALA**

*by*

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**THESIS**

Submitted in partial fulfillment of the  
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Kerala Agricultural University



**DEPARTMENT OF WILDLIFE SCIENCE  
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KERALA, INDIA**

2017

## DECLARATION

I, hereby declare that this thesis entitled "SPECIES DIVERSITY AND COMMUNITY STRUCTURE OF AMPHIBIANS OF SELECTED AGROECOSYSTEMS IN THRISSUR, KERALA" is a bonafide record of research work done by me during the course of research and the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other University or Society.

Place: Vellanikkara

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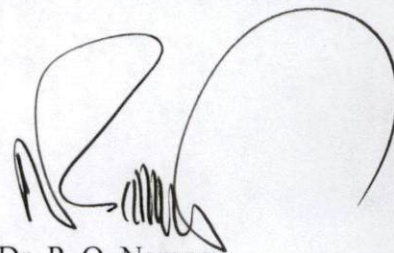
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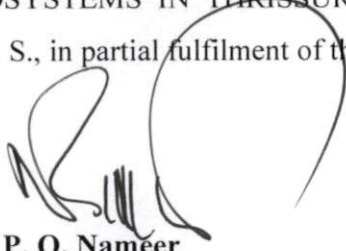
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# **INTRODUCTION**

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## INTRODUCTION

Among the vertebrate's over the world, amphibians comprising of frogs, toads, caecilians and salamanders, are the most threatened taxa and have the highest proportion of species in verge of extinction (Baillie *et al.*, 2004). One-third of the amphibian population over the world are under threat mainly due to the after effects of urbanization comprises of habitat loss, habitat degradation, habitat fragmentation, habitat isolation and decline in habitat quality (Hamer and McDonnell, 2008). The most pervasive threat to 88% of threatened amphibians are the habitat loss and habitat degradation (Baillie *et al.*, 2004). Besides urbanization, climate change is another leading factor which go for higher extinction rates where 15% of amphibians are highly vulnerable to climate change (Foden *et al.*, 2013).

Baillie *et al.* (2004) stated that, 23% of amphibians have not been assessed due to insufficient data and among the species with adequate data, more than 21% are under critically endangered or in endangered category.

Dutta (1997) published first comprehensive checklist of amphibians of India with 212 species, then Daniels (2005) listed 238 species from peninsular India, later Dinesh *et al.*, (2009) enlisted 284 species. Dinesh *et al.* (2013) revised the list with 342 species. Dinesh *et al.* (2015a), published that India harbors 384 species of amphibians in which 217 are endemic to Western Ghats. After this there were 10 new addition to the anuran list of India.

It is a fact that species accumulation curve has not yet reached a plateau in case of the amphibians of Western Ghats (Aravind *et al.*, 2004). According to Nameer *et al.* (2015), 90.06% of amphibians of Kerala are endemic to Western Ghats and 33% of the amphibians belong to various threatened categories of IUCN. This high level of endemism is a result of permutation and combination of biogeography, physical features, eco-climatic variations and past biogeographic



events. Conservation programmes for amphibians necessitates, knowledge of factors that control their diversity in the region.

Generally, protected area network which comprises 18.1 % of Earth's area is considered as the corner stone of biodiversity conservation efforts. According to Karanth *et al.* (2016), production landscapes especially agroecosystems cover about 40% of planet's landmass. Kitzes *et al.* (2008) predicted that by 2050 overall cropland may extend between 0.3 to 1.8 billion ha. These agroecosystems act as the secondary habitat for global biodiversity for their occurrence as well as movement and provide conservation opportunities. In tropics, isolated protected area is embedded in the matrices of various agricultural landscapes. So, for a realistic conservation strategy we should evaluate the conservation value of these multiple land use systems which are in dearth now (Wanger *et al.*, 2009).

Amphibians are the important predator and prey species in both aquatic and terrestrial habitat. They act as an energetic link between both the ecosystem especially in tropics where the diversity and abundance of the taxa are high. They have greater capacity to alter algal community structure, primary production, organic matter dynamics, population of other consumers and energy transfer between ecosystems since larval stage and adult stage of amphibians are functionally different. Therefore, loss of one species is akin to loss of two species in case of amphibians (Whiles *et al.*, 2006).

Amphibians are the first vertebrates to be on the land. They provide immense ecosystem services to human society which are unknown to common people. Now this class of vertebrates face the greatest challenge for survival. In India research in the field of amphibians is in its infancy where we are describing new species at a greater pace, almost one species every month. We are not sure about the number of species residing in India. Western Ghats is one of the global hotspot for amphibian diversity due to high endemism. But it is a fact that it is just about 1,60,000 km<sup>2</sup>. This piece of land is highly fragmented and surrounded with various land use

systems, including agroecosystems. These surrounding landscapes, acts as the corridors as well as act as a buffer, and they have the potential to conserve the native biodiversity which is still untapped.

The present study could be the pioneering effort to understand the amphibian diversity and richness in an agroecosystem in Kerala. Besides that, it is probably a first attempt to monitor the amphibians in their non-breeding season, which is quiet challenging.

Thus, the objectives of the proposed study were;

- To study the species diversity and community structure of amphibians of selected agroecosystems of Thrissur and its association with various habitat parameters
- To assess the spatial variation of amphibian distribution using Geographic Information System (GIS) tools

## **REVIEW OF LITERATURE**

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## REVIEW OF LITERATURE

Macrocosm of biodiversity resides in one of the 35 biodiversity hotspots, Western Ghats-Sri Lanka which harbors 39% of endemic vertebrates (Anonymous, 2009). According to Biju (2001), the number of amphibians (> 6,000) in the world exceeds than that of all land vertebrates. Roelant *et al.* (2007) stated that, the declining rate of amphibians is much higher than any of the vertebrate taxa in the world. The Permian mass extinction which happened 251 million years ago (mya), reduced the 24 amphibian families to eight, which was an after effect of sudden climate warming due to release of carbon gas. This was a major extinction episode in tetrapod evolution. In case of amphibian evolution, it was not a gradual lineage accumulation but there were substantial fluctuations. For example, there exist a hypothesis that amphibian diversification has increased with diversification of angiosperms, where forests holds 82% of recent amphibian radiation. Roelant *et al.* (2007) predicted that 0.2926 events per lineage per million year (myr) will be the average extinction rate of amphibians, which accounts 1725 extinctions per myr from 6009 known species. This shows that the present extinction rate is 200 - 2,700 times faster than past extinction episodes.

In India, there is a high level of amphibian endemism in specific and generic levels. North east India and Western Ghats are the hotspots for the rich amphibian diversity in India. In the beginning of 21<sup>st</sup> century itself there was an explosion of hidden anuran diversity from the Western Ghats. Till that time, the literatures indicated the presence of just 200 species of anurans from India of which 100 were from Western Ghats. Later Biju (2001) after his seven-year field work, published an eye-opening article which harbours the presence of 225 species from Western Ghats alone. This shows that the biological wealth of one of the 17 megadiverse country is still untapped and unexplored to a great extent.

According to Biju (2001) the golden era of amphibian systematics in India was during the British Raj from the 1850s to 1925. Exploration of Western Ghats

was started by Thomas Caverhill Jerdon and Richard Henry Beddome who were two British naturalists. They collected the frogs from southern Western Ghats and Beddome could describe a new genus called '*Melanobatrachus*'. The next notable collector was Ferguson (1904) who thoroughly explored Ashambu hills and Ponmudi. Pre-independence period of Indian Batrachology was flourished by the contributions of Sclater (1892), Annandale (1905) who described 10 new anuran species from India, Stanley Wells Kemp, Fietcher, Francis Day, Dussumier, Gravely and C.R. Narayan Rao (Biju, 2001).

After independence, we realized the depth of environmental degradation happened to our rich centres of endemism which eventually leads to the importance of conservation of nature. Indian batrachology was still at an infant stage during that time. It is true that amphibian diversity is heavily underestimated. Without knowing what is present in a region how can we take measures for conservation? There rose the need for amphibian studies for successful conservation management strategies and many scientists started to highlight the importance of anuran diversity in India with special reference to biogeography. Then came an array of publications describing new species and their type localities, behavioural ecology and breeding biology of known species, biogeographical affinity in speciation and their acoustics.

## 2.1. AMPHIBIAN DIVERSITY STUDIES

The pioneer amphibian studies in India dated back to the fauna of British India including Ceylon and Burma by Boulenger (1890) and Smith (1943). Dutta (1997) published the first checklist on Indian amphibians with 212 species. Another update on the list was done by Daniels (2001). Chanda (2002) was the first to publish a handbook on Indian amphibians with brief accounts followed by Daniels (2005) with 238 species of amphibians from Peninsular India in his book '*Amphibians of Peninsular India*'. Dinesh *et al.* (2009) published a checklist of amphibians of India with a total of 248 species including all new species described till 2009. Gururaja (2012) published a picture guide of Western Ghats anurans.

Along with these new inventories, 36 species of amphibians got extinct and over 1,957 species got threatened (Monastersky, 2014). According to Frost (2015) 7,356 amphibians inhabit most of the major habitat types, belonging to three living orders. The world has witnessed the discovery and description of 1,786 species of amphibians in the last decade (Web, 2015). Dinesh *et al.* (2015) published the checklist of 384 amphibian species found in India in which, 154 were described between 2000 and 2015 among which, 111 are from the Western Ghats.

Much of the pioneer studies in amphibians of Kerala were limited to short surveys in protected areas. It was Ferguson (1904) who attempted to list the batrachians of Travancore. Pillai (1978) described *Micrixalus nudis* from Wayanad. Pillai and Pattabiraman (1981) also described a torrent toad from the rocky streams of Silent Valley National Park. Inger *et al.* (1984) published a report on herpetofauna of Ponnudi hills. A new caecilian species to science along with 19 amphibians were reported from Silent Valley National Park (Pillai, 1986). A checklist of endemic vertebrates, including the amphibians of the Western Ghats region was published by Swengel (1990). Daniels (1992) worked on geographical distribution of amphibians of Western Ghats. A key to the amphibians of Kuttanad was developed by Andrews and George (1995). First record of *Uperodon globulosum* from Kerala was reported by George and Alex (1995). Amphibian checklist from Aralam Wildlife Sanctuary was published by Radhakrishnan (1996). Zacharias and Bhardwaj (1996) studied on amphibians of Periyar Tiger Reserve. Daniels (1997) published a field guide about the anurans of Western Ghats. The distribution records of amphibians of Kerala was published by Dutta (1997). Thirty species of amphibians were recorded from Wayanad Wildlife Sanctuary by Abraham *et al.* (2001). Andrews *et al.* (2005a & 2005b) conducted surveys on amphibians of protected areas of Kerala. Jobin and Nameer (2012) have studied on the rhacophorid diversity of Parambikulam Wildlife Sanctuary. Das (2015) compiled and enlisted 151 species of amphibians from Kerala.

## 2.2. TAXONOMIC REVISIONS WITH THE ADVENT OF MOLECULAR TAXONOMY

The number of species of amphibians described over the past 25 years has skyrocketed. It is a fact that new amphibian species to science has been described by scientist in every two and half day since 1987 (Pratihari *et al.*, 2014). Biju (2001), suggested the existence of more than 100 cryptic species in the endemic genera of the Western Ghats. By the arrival of molecular taxonomy and phylogenetic, amphibian study got a strong backbone. Frost *et al.* (2006) proposed new taxonomy of living Amphibia based on the phylogenetic analysis. They combined anatomical characters with DNA sequences from the mitochondrial transcription units and nuclear genes. They concluded that the three major groups of living amphibians (caecilians, salamanders and frogs) form a monophyletic group and caecilians forms a sister taxon group of Batrachia (salamanders and frogs). Frost *et al.* (2006) published amphibian tree of life for 522 species. Fritz *et al.* (2012) looked on the phylogenetic diversity of amphibians to identify the process and areas of macroevolutionary processes which in turn affected the species richness. Resolving the cryptic diversity is not only important to understand the process of evolution and diversity but also to prioritize conservation efforts and to prevent nameless extinction events (Biju *et al.*, 2014a). By investigating the phylogenetic position using molecular data Biju *et al.* (2016) discovered a distinct evolutionary lineage of tree hole breeding frog genus *Frankixalus* with oophagous tadpoles.

### 2.2.1. Bufonidae

Bufonidae is the family of toads which has cosmopolitan distribution. Recent phylogenetic analysis proposed three endemic genera of toads (*Ghatophryne*, *Xanthophryne* and *Pedostibes*) and found a recent radiation with distinct larval and adult ecomorphs in Southern India (Biju *et al.*, 2009). Biju *et al.* (2009) described two novel endemic genera and a new species of toad from the Western Ghats based on morphological and molecular evidence. Genus *Ansonia* got transferred to novel

genus *Ghatophryne* which are torrentially adapted toads of Western Ghats and *Bufo koynayensis* was transferred to novel genus *Xanthophryne*. *Xanthophryne tigrinus* was the novel species described from Amboli part of Western Ghats. Recent investigations are based on a hypothesis that *Duttaphrynus melanostictus* is not a single species but a species complex.

### 2.2.2. Dicroglossidae

Fork-tongued frogs come under the family Dicroglossidae with 5 genera in Western Ghats. Study of Kuramoto *et al.* (2007) revealed the occurrence of active speciation in *Fejervarya* in the Western Ghats. Kuramoto *et al.* (2007) state that *F. rufescens* and *F. sahyadris* collected from Karnataka and Kerala, show considerable divergence, which propose an intermediate stage in the process of speciation. They also synonymized the genus *Minervarya* with the genus *Fejervarya* based on the phylogenetic results. Now the genus *Euphlyctis* is represented by four species, of which two new species *Euphlyctis aloysii* and *Euphlyctis mudigere* were described recently by Joshy *et al.* (2009). Kotaki *et al.* (2010) worked on the molecular phylogeny of the diversified genus *Fejervarya* and their dataset supports paraphyly. Howlader (2011) described a new genus *Zakerana* and stated that all *fejervaryan* species of Western Ghats and South Asia belongs to the new genus. Dinesh *et al.* (2015b) treated the genus *Zakerana* as a junior synonym to the genus *Fejervarya*. They found that larger *fejervaryan* clade with had a sister relationship to a clade which comprises *Sphaerotheca* genus. Based on phylogenetic position, genetic divergence and morphological divergence, they found unidentified lineage which was described as *Fejervarya gomantaki*, a different species.

### 2.2.3 Micrixalidae

Micrixalidae is the Western Ghats endemic family of tropical frogs or torrent frogs which was evolved from the family Ranidae during Paleocene period (Roelant *et al.*, 2004). Biju *et al.* (2014a) investigated species level diversity in the monotypic frog family Micrixalidae using DNA barcoding approach along with morphological study of old and new specimens and described 14 new species.



Detailed descriptions, morphological and genetic comparisons, illustrations, data on distribution and natural history were provided for all species and reported foot-flagging behavior in a total eight species.

#### **2.2.4 Microhylidae**

The genus *Microhyla* is widespread across South and Southeast Asia. Microhylidae follows monophyly according to phylogenetic interference done by Peloso *et al.* (2015). Seshadri *et al.* (2016) described a new species *Microhyla laterite* which appears to be restricted to areas of the West Coast of India dominated by laterite rock formations through an integrative approach of morphology, molecular and bioacoustics data.

#### **2.2.5 Nasikabatrachidae**

Biju and Bossuyt (2003) discovered a burrowing frog *Nasikabatrachus sahyadrensis* from India, whose molecular data designated it as a sister taxon of Sooglossidae which is seen only on Seychelles and they recognised it as a new frog family Nasikabatrachidae. Molecular clock analysis dated *Nasikabatrachus sahyadrensis* as a Gondwana relic which evolved during 150 – 195 mya (Dutta *et al.*, 2004).

#### **2.2.6 Nyctibatrachidae**

*Nyctibatrachus* represents an ancient anuran lineage, which is endemic to Western Ghats and originated within Indian landmass. Members of this genus is distributed in Tamil Nadu, Kerala, Karnataka, Goa and Maharashtra associated with mountain streams and marshes. The first species of night frog was described in 1882 by Boulenger and then additions came in the next century. Van Bocxlaer *et al.* (2012) claimed that Nyctibatrachidae was originated earlier on Indian subcontinent with the help of molecular evidence. According to them, within the hill ranges of Western Ghats a clade endemism was shown by species due to limited dispersal across the Palghat gap and Shencottah gap. This study emphasized the uniqueness of Western Ghats mountain regions where each mountain holds endemism which

deserves conservation attention. Recently, Biju *et al.* (2007) described a new species *Nyctibatrachus minimus* from Kurichiyarmala in the Western Ghats, which is the smallest known frog from India averaging only 12.3mm in adult males. According to Biju *et al.* (2007), small size of the species was associated with the absence of webbing on toes and fingers. Based on new morphological, behavioural, ecological and molecular data from available type specimens and recent collections Biju *et al.* (2011) did a taxonomic revision of the genus *Nyctibatrachus* where they recognised 28 species of the genus among which 12 were new to science. A new terminology, the sub-ocular gland was introduced to anuran morphological terminology. For four species, they described the courtship behaviour and ovipositional sites for the first time. Recently Garg *et al.* (2017) described 7 new species of *Nyctibatrachus* making a total of valid nigh frog species to 35. Among the new 7 species, *Nyctibatrachus athirappillyensis* is known only from southern Western Ghats state of Kerala, *Nyctibatrachus manalari* is known only from south of Palghat gap, *Nyctibatrachus pulivijayani* is currently known only from Agasthyamala Hills, *Nyctibatrachus radcliffei* is currently known from the Nilgiris which is north of Palghat gap, *Nyctibatrachus sabarimalai* is currently known only from its type locality, which is located close to Sabarimala in Periyar Tiger Reserve which is south of Palghat gap, *Nyctibatrachus webilla* is currently known only south of Palghat gap. The population status of the newly described species is also likely to be of concern, especially in the case of *Nyctibatrachus athirappillyensis*, *N. radcliffei* and *N. webilla* which were collected outside National parks and sanctuaries (except *N. sabarimalai*). *Nyctibatrachus radcliffei* and *N. webilla* were found inside private or state-owned plantation areas facing threats such as habitat disturbance, modification and fragmentation.

### 2.2.7 Ranidae

*Indosylvirana* is a complicated group which lack distinct colour differences and striking morphological characters. Biju *et al.* (2014b) did a systematic revision of this genus in the Western Ghats-Sri Lanka biodiversity hotspot. They recognized seven novel species by the combination of molecular and morphological. They

came to another valid conclusion that there were no species shared between the Western Ghats and the Sri Lanka in this genus. Dahanukar *et al.* (2016a and 2016 b) proposed a revised taxonomy of leaping frogs using an integrative approach including an analysis of the mitochondrial 16S rRNA and nuclear rhodopsin genes, as well as multivariate morphometrics and described a new genus *Sallywalkerana* which is restricted to south of Palghat gap in the Western Ghats states of Kerala and Tamil Nadu.

### 2.2.8 Ranixalidae

Ranixalidae comprise of leaping frogs of which *Indirana* represents one of the 10 endemic frog genera in Western Ghats. Nair *et al.* (2012) worked on the cryptic diversity of the genus *Indirana* whose diversity was previously underestimated. They investigated the genetic diversity with the help of three mitochondrial gene and two nuclear gene and concluded the presence of 11 clades with high sequence divergence. They claimed that the novel unrecognized lineages of the genus *Indirana*, have more narrow distribution ranges and lower abundance compared to the existing taxonomic unit to which it belongs. They identified new candidate species in *Indirana* with high genetic divergence and claimed polyphyly in *Indirana beddomii*. Padhye *et al.* (2014) described a new species of leaping frog, *Indirana chiravasi* from the northern Western Ghats around Amboli. Molecular analysis suggested that the species is genetically distinct from other species for which genetic data is available. *Indirana leithii* was considered to be widespread over the entire Western Ghats but the phylogenetic analysis suggested that the species was restricted range in the state to Maharashtra. According to Modak *et al.* (2014) specimens identified as *Indirana leithii* from the southern Western Ghats belong to some undescribed species. Garg and Biju (2016) described two new species belonging to the genus *Indirana* from the Western Ghats states of Karnataka and Kerala.

### 2.2.9 Rhacophoridae

Bossuyt and Dubois (2001), did a review on the genus *Philautus* to stabilise the nomenclatural and taxonomic situation of the genus. Bossuyt (2002) described a new species *Philautus griet* from Munnar in the Western Ghats. *Philautus nerostagona* was described from Wayanad district in the Western Ghats whose morphology is strongly adapted to life in the upper layers of the rainforest with a distinct dermal fringe along the outer margin of the fore limbs and hind limbs (Biju & Bossuyt, 2005a). *Philautus ponmudi*, *Philautus bobingeri* and *Philautus graminirupes* were described from the Ponmudi hills of the Western Ghats by Biju & Bossuyt (2005b; 2005c). Biju & Bossuyt (2006) described two new species, *Philautus anili* and *Philautus dubois* from Wayanad (Kerala) and Kodaikanal (Tamil Nadu), respectively. Das & Dutta (2006) described a new species, *Polypedates occidentalis* from Kerala. Biju *et al.* (2008) done phylogenetic analyses and revealed that *Polypedates variabilis* and a previously undescribed relative stem from a lineage that had an early origin in tree frog radiation which was named as *Ghatixalus*, a novel genus. They also described a new species, *Ghatixalus asterops* and concluded that species of this genera was restricted to high altitudes of the Western Ghats. Gururaja *et al.* (2007) decribed *Philautus ochlandrae* from Kakkayam Reserve Forest, Kerala. Systematics and phylogeny of the genus *Philautus* was done by Biju & Bossuyt (2009) with the description of 12 new species. Phylogenetic analyses of mitochondrial genes indicated that the new frog described from Eravikulam National Park *Raorchestes resplendens* was native of Indian subcontinent and was highly restricted to less than 3 km<sup>2</sup> of Anamudi (Biju *et al.*, 2010; Joseph *et al.*, 2012). Zachariah *et al.* (2011) described nine new species of *Raorchestes* from southern Western Ghats. Seshadri *et al.* (2012) described a new bush frog, *Raorchestes kakachi* from the mid-elevation evergreen forests of Agasthyamalai hill regions of southern Western Ghats. Abraham *et al.* (2013) described two new genera *Beddomixalus* and *Mercurana* which represented two ancient, independent clades which are the sister groups to the Sri Lankan and Indian- Chinese-Indochinese radiations of bush frogs. Both these genera resemble foam-nesting rhacophorids with free living aquatic tadpoles. Both the genera are monotypic with only a singular representative species. Biju *et al.* (2013) have given

a taxonomic revision of genus *Rhacophorus* with description of nesting behaviour and their ontogenetic colour changes. A novel third species of the Western Ghats endemic genera *Ghatixalus* was described which potentially represents the largest known rhacophorid species in peninsular India. *Ghatixalus magnus* species novel was recorded between the Palakkad Gap and the Shencottah Gap (Abraham *et al.*, 2015). A new species of shrub frog *Raorchestes ghatei* is described from the Western Ghats of Maharashtra (Padhye *et al.*, 2013). Vijayakumar *et al.* (2014) used a hierarchical multi-criteria approach by combining mitochondrial phylogeny, genetic distance, geographic range, morphology and advertisement call to delimit bush frog lineages and they described nine new species of bush frog that exhibit divergence across multiple axes. Another cryptic species of bush frog *Raorchestes honnametti* was described using an integrative taxonomic approach (molecular – bioacoustics – morphology) from the south-eastern part of the Western Ghats (Priti *et al.*, 2016). Zachariah *et al.* (2016) described two new species of bush frogs *Raorchestes silentvalley* and *Raorchestes lechiya*, from the tropical montane wet forests above 1,800 m MSL in the Silent Valley National Park and provided with advertisement calls and insights into the phylogenetic position (Zachariah *et al.*, 2016).

#### 2.2.10 Caecilians

Bhatta (1997) stated that the caecilians are taxa difficult to sample due to its subterranean life. According to him they are the taxa which bears greatest degree of endemism. *Gegeneophis seshachari* was described from Maharashtra which were only known type specimen (Ravichandran *et al.*, 2003). *Gegeneophis danieli* and *Indotyphlus maharashtrensis* were described from Maharashtra (Giri *et al.*, 2003; Giri *et al.*, 2004). *Gegeneophis madhavi* was described from Mookambika Wildlife Sanctuary based on two specimens, *Gegeneophis mhadeinsis* was described from northern Karnataka and *Gegeneophis goaensis* from Goa (Bhatta and Srinivasa, 2004; Bhatta *et al.*, 2007). Wilkinson *et al.* (2007) produced a dichotomous key to identify the new species of *Icthyophis* from Karnataka when compared to other striped caecilians. Giri *et al.* (2011) described *Gegeneophis*

*pareshi* from southern Goa which lack secondary annular grooves. Besides the species discovery stages of spermatogenesis, population ecology and diet of caecilians were also investigated through various research (Smita *et al.*, 2004; Measey *et al.*, 2004; Kupfer *et al.*, 2005). San Mauro *et al.* (2004) gave a complete sequence of mitochondrial genomes of five different species. A new family of caecilians, Chikilidae was discovered from North East India which had an African link (Kamei *et al.*, 2012).

Caecilians have a pantropical distribution and are habitat specific because of the moist skin and burrowing habitat (Giri, 2009). As per Gower *et al.* (2004) caecilian taxonomy is unstable. Gower *et al.* (2011) quoted that Indian and Seychelles caeciliids comprises 10% of 180 species so far reported from the world. Wilkinson and Nussbaum (2006) explained the caecilian phylogeny and classification with a key for identification of certain genus. Gower *et al.* (2008) reported the first vivipary in *Gegeneophis seshachari*. Bhatta *et al.* (2011) discovered the largest striped caecilian, *Icthyophis davidi* from Karnataka part of Western Ghats. Gower *et al.* (2011) have sequenced mtDNA sequence data for all the Indian caecilians. Agarwal *et al.* (2013) described the first teresomatan caecilian *Gegeneophis orientalis* from high elevations of Eastern Ghats.

## 2.2 ECOLOGICAL IMPORTANCE

Valuable services provided by amphibians are immense in the form of provisioning, regulating, cultural and supporting services. They serve as proteinaceous food source and medicine for numerous ailments including asthma, tumor, skin disease and can act as painkillers. According to Jensen and Camp (2003) 10 million frogs are sipped illegally from India for consumption of frog legs globally. The chemical secretion from the amphibian skin have the potential to inhibit infection and the transfer of HIV (Lorin *et al.*, 2005). Some species have the ability to stop the production of stomach acid which can be used against stomach ulcers (Calvet and Gomollon, 2005). They can regulate invertebrate pest outbreaks

and even can inhibit disease transmission. They also have the potential to be agent of pollination and seed dispersal. It was found that a treefrog in Brazilian forest, *Xenohyla truncate* is able to consume fruits and defecates viable seeds (Silva and Britto-Pereira, 2006). Amphibians are the part of art, literature, music, mythologies and even used in educational purposes. They also make popular pets since they are easy to maintain and carry. Amphibians can alter the primary production of aquatic ecosystem, can reduce sedimentation and can even act as sinks of organic nitrogen. They support aquatic food web by providing energy and supplying nutrient sources. They can be prey and carcasses for predators and decomposers respectively in aquatic ecosystem. In terrestrial ecosystem, they can regulate invertebrate population and can alter physical structure of habitat. Some toads can alter the litter chemistry by increasing phosphorus concentration and burrowing frogs can change soil bulk density and water infiltration (Hocking and Babbitt, 2014). These are some of the few studies which are limited to certain species and habitats regarding the ecosystem services of amphibians.

#### 2.4 AMPHIBIAN SAMPLING

Eekhout (2010), has described standardisation in sampling methods for herpetofauna. This include time-constrained surveys, area constrained surveys, transect surveys and quadrat sampling. Vasudevan *et al.* (2001) assessed the forest floor amphibians of Kalakad Mundanthurai Tiger Reserve using adaptive cluster sampling, which found to be superior than traditional methods of deploying random quadrats. They also mentioned about the micro habitat features to be taken care of while sampling amphibians. The various permanent marking techniques and mark-recapture methods were described to assess the population size of caecilians (Measey *et al.*, 2001; Measey *et al.*, 2003). Rocha *et al.* (2004) evaluated six sampling methods suitable for resting habitat of Brazil. They calculated capture efficiency index for each method and found that breeding site survey and species inventory survey out all microhabitats suitable for frogs were most efficient methods compared to patch sampling and visual encounter surveys. Harikrishnan

*et al.* (2012) reported that the most efficient method to study the species diversity of a particular region in case of herpetofauna was visual encounter survey and opportunistic records.

## 2.5 DIVERSITY STUDIES IN VARIOUS AGROECOSYSTEMS

Dolia *et al.* (2008) have stated that agroforestry systems or plantations are resilient for biodiversity conservation than other land modifications because of the arboreal vegetation they incorporate. According to Biju *et al.* (2008), 40% amphibian species in the Western Ghats are already threatened with extinction. It is a truth that species diversity and density were found to be more in natural forests than man-modified ecosystems. According to Nair (2008) agroforests can be considered as the potential oases for disappearing species even though they can't be substituted against natural forests. Agroecosystems play an important role in conserving the population of common species while the undisturbed natural ecosystems remain as the home for rare and endangered species (Gamage *et al.*, 2008). He emphasized the value of protecting agroecosystems as habitat for large number of species. Diversity and abundance of herpetofauna in Kole wetlands of Thrissur was studied by Sreehari (2009). Wanger *et al.* (2009) assessed the herpetological diversity in Cacao agroforests of South East Asia. They came to a conclusion that some habitat features like increase in leaf litter is favoring the richness and abundance of those disturbance tolerant species which still go for their ecological functions like pest removal in these agroforests. Bionda *et al.* (2011) worked on the amphibians of various agroecosystems in Argentina. They stated that habitat modifications are actually depleting the diversity by reducing specialists and niches with increasing generalist. They also found that there are species which take advantage of the hydrology and hydroperiod of agroecosystems for their survival. Murali and Raman (2012) investigated on the streamside amphibian diversity of tea monoculture, coffee plantation and in a fragmented rainforest of old Valparai area of the Anamalai hills. Another interesting study was conducted by Rathod and Rathod (2013) on the amphibian richness and diversity of organic, NPK and



pesticide use coffee plantations distributed in Kodagu district of Karnataka. Caudill *et al.* (2014) highlighted the importance of developing and improving management strategies to find a sustainable solution to gain modified ecosystem development with wildlife conservation. According to Coyle (2015) traditional farming methods in rural landscapes should be maintained for conservation of native biodiversity. Karanth *et al.* (2016) studied on the avian richness in areca, rubber and coffee agroforests in Western Ghats. They claimed that these production landscapes which are privately owned have a great role in conservation of biodiversity along with the protected area. He suggests that a holistic approach in conservation requires an association of land sharing and land sparing approaches. Not all agroecosystems contribute equally in conservation approach. Effective land management strategies can be developed only after the understanding of ecosystem services provided by the local wildlife where there is a gap in research.

## 2.6 THREATS

As per Cushman (2006), the unique planet is going to face the largest mass extinction in 65 million years where extinction rate will be 1000 times higher than that of previous fossil records. According to Aggarwal (2004), there is a need for molecular genetic studies in ecology and evolutionary biology in developing countries, which in turn have great conservation implications. Beebee and Griffiths (2005) identified the discipline of conservation biology as the only solution for current biodiversity crisis. In case of the most threatened taxa amphibians, their life in both aquatic and terrestrial ecosystems and their sensitive skin encourage them to be good biological indicators and make them the most vulnerable group for environmental changes. Besides habitat destruction, increased UV B radiation, emerging diseases, pollution, road traffic, introduced alien species and climate change have been considered as the possible threats to amphibian life stages (Beebee and Griffiths, 2005). Tropical forests are complex and species rich ecosystems, where rare and endemic species resides which are adversely affected by habitat fragmentation (Kumar *et al.*, 2002). Habitat destruction cause restricted

and patchy distributions of several amphibian species in the Western Ghats (Vasudevan *et al.*, 2001). Linder (2003) stated that multiple stressors lowers the strength of selection, this may lower the survival rate of individuals. The reduction in habitat complexity due to habitat loss is having great negative impact on the amphibian population (Gardner *et al.*, 2007) since they have patchy distribution in landscapes at local scales. Amphibian population is nested in tropical forests. Fragmentation lead to the local extirpation of the rare species fragmentation. Seasonal fluctuations in water level and flow streams totally affect the frog communities (Kumar *et al.*, 2002). Urbanization reduces the connectivity of the habitats (Hamer and McDonnell, 2008) and reduce the probability of recolonization for threatened species. The indiscriminate use of pesticides in plantations is causing environmental pollution harmful to amphibians. Seven of the 12 new species were found only in areas which were forests some time back, are now plantations (Biju and Bossuyt, 2009). According to Trombulak and Frissell (2000), roads have become one of the growing threat to both animal and plant population by direct mortality and a cause for forest fragmentation. Vijayakumar *et al.* (2001) found greater mortality of amphibians by roadkill compared to reptiles in the road that passing through rainforest fragments in Anamali hills which may deplete local population. Dahanukar *et al.* (2013) reported the Chytrid fungus, *Batrachochytrium dendrobatidis* (*Bd*) from northern Western Ghats and Molur *et al.* (2015) confirmed the chytridiomycosis from the central Western Ghats of Karnataka and northern parts of Kerala. For sustainable amphibian conservation strategies species specific ecological knowledge, landscape level studies, effects of habitat loss and fragmentation on movement, survival rates, and population dynamics of the species is required (Cushman, 2006). In Kerala, major threat to amphibian is due to habitat loss which accounts for 88% followed by pollution (31%), natural disasters, human disturbances, fire etc. and the least (only 1%) is by diseases (Biju, 2001). According to Gower and Wilkinson (2005) despite lack of filed study and quantitative data caecilians face the threats of habitat loss, pollution and chytridiomycosis. But due to cryptic nature and secret life they were not recorded systematically to assess the population status.

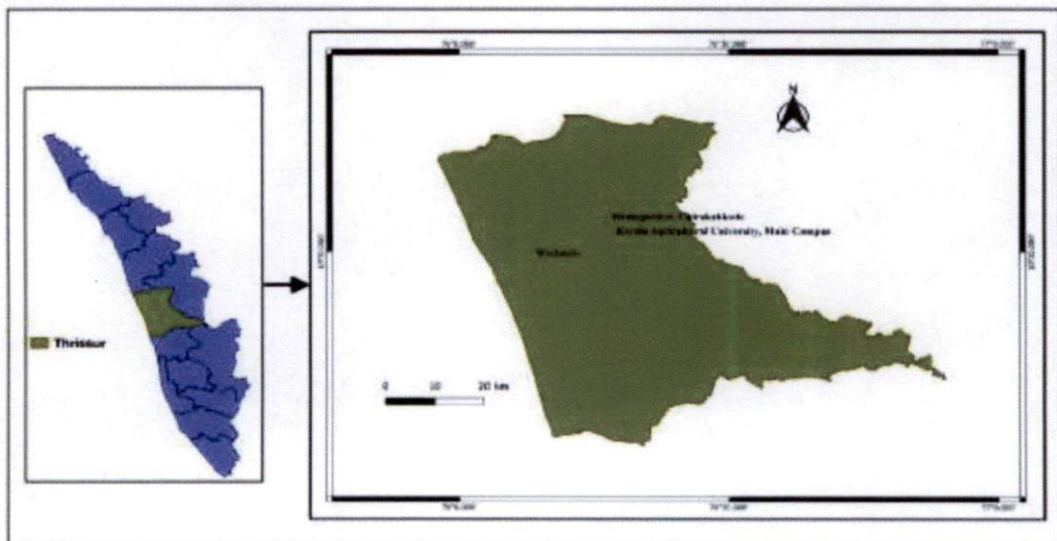
## **MATERIALS AND METHODS**

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## MATERIALS AND METHODS

### 3.1 STUDY AREA

Kerala Agricultural University main campus located in Thrissur dt., Kerala, lies between  $10^{\circ}32'$ – $10^{\circ}33'$ N and  $76^{\circ}16'$ – $76^{\circ}17'$ E and is situated near Peechi - Vazhani Wildlife Sanctuary. The Peechi - Vazhani Wildlife Sanctuary is part of one of the 35 global biodiversity hotspots of the world, the Western Ghats. The KAU main campus is spread over 391.44 ha comprised of variety of habitats including fruit orchards (mango, garcinia, mangosteen, breadfruit, sapota) plantations (coconut, cocoa, rubber, cashew, arecanut), seasonal crops, garden land, stagnant water bodies, undisturbed botanical garden and arboretum. The field work in search of amphibians was done in five agroecosystems such as Cashew Plantation, Coconut Plantation, Rubber Plantation of Kerala Agricultural University (KAU) main campus, typical tropical Homegarden of Chirakakode, Wetlands of Arimbur and in an undisturbed Botanical Garden of KAU main campus within the same geographic region (Figure 1). The campus receives both south-west monsoon and north-east monsoon. The average number of rainy days is 110 per year with mean rainfall of 2763mm. The mean maximum temperature for 10 years is  $31.8^{\circ}\text{C}$  and mean minimum temperature is  $23.3^{\circ}\text{C}$  for 10 years.



**Figure 1: Location map of different study locations, Thrissur**





**Plate 1. Selected agroecosystems (A – Cashew Plantation, B – Coconut Plantation, C – Homegarden, D – Rubber Plantation, E – Wetlands, F- Botanical Gardens**

## 3.2 METHODS

The study was conducted from January 2017 to May 2017 in the study area. A preliminary survey on amphibians was done during the monsoon season of 2016 in Kerala Agricultural University main campus. Data collected includes species diversity, abundance, density and on other microhabitat parameters crucial for the presence of amphibians.

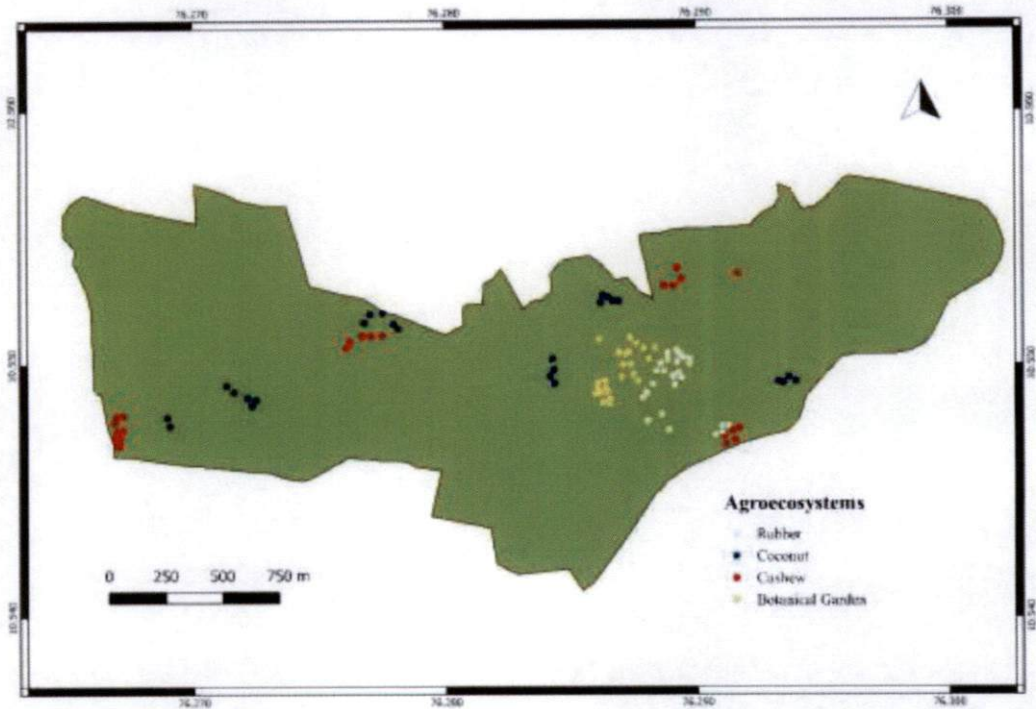
### 3.2.1 Quadrat Sampling

Quadrat sampling is usually done for the estimation of density of organisms. According to Jaeger and Inger (1994), sampling arrays are usually squares distributed randomly to verify the presence and absence of the animals. In this study, random quadrats of  $10 \times 10\text{m}$  were deployed in the study area (Figure 2). The observations were taken in between 19:00 and 20:30 hrs. Two observers approached the quadrat from opposite corners towards the center in a clockwise manner for thorough search of the floor dwelling amphibians (Harikrishnan *et al.*, 2012). In this study, 15 minutes were spent in each quadrat and deployed 25 quadrats in each selected location. A total of 150 quadrats were deployed in the study locations. Besides the amphibian diversity of quadrats, presence of reptiles was also monitored in each of the sampling unit

### 3.2.2 Visual Encounter Survey

Visual encounter survey was conducted in which three observers sample for species richness and abundance along a path (Crump and Scott, 1994). It is potentially possible to obtain the complete species inventory of the sampled area when visual encounter survey is combined with other sampling technique (Eekhout, 2010). Here in this case combined visual encounter survey was integrated with quadrat sampling. The survey time was between 20:30 to 21:30hrs. LED torches and head lamps (Mr. Light) were used to spot the individuals. During the survey, a range of possible microhabitats including rocks, marshes, fallen logs, tree holes, snags and water bodies were thoroughly examined. One or two specimens of

species encountered were collected, preserved and deposited in Kerala Agricultural University natural history museum. In this work, irrigation pattern and fertilizer application in agroecosystems were also taken into account.



**Figure 2. Distribution of 100 quadrats in the selected agroecosystems inside the Kerala Agricultural University main campus**

### **3.2.3 Opportunistic Records**

Species encountered incidentally from the study area premises, were also recorded which help us to figure out the species richness. Digital images of the individuals were taken using Canon 700D.

### **3.2.4 Laboratory Studies**

Voucher specimens collected from the field are first euthanized using diethyl ether. Then tissues are extracted from the thigh muscle for further molecular analysis (if needed) and preserved in molecular grade alcohol. Then the specimens are fixed in



4% formalin for 24 hours and then they are kept in water for again 24 hours. After complete fixation, they are transferred to 70% alcohol in air tight containers. Proper labels are tagged for each specimen which includes common name, scientific name, habitat, date of collection, collector's name and specimen number. According to Bossuyt and Dubois (2001), standard morphological measurements of the specimen were recorded properly using a Mitutoyo Digimatic Caliper to the nearest 0.1mm. The standard morphometric measurements are;

1. SVL - Snout-vent length
2. EL – Eye length
3. HL - Head length (from posterior corner of mandible to tip of snout)
4. HW – Head width, at the angle of jaws
5. TYD – Maximum tympanum diameter
6. FLL – Forelimb length (from elbow to base of outer palmar tubercle)
7. HAL – Hand length (from base of outer palmar tubercle to tip of third finger)
8. ThL – Thigh length (from vent to knee)
9. ShL – Shank length (tibia length)
10. FOL – Foot length (from base of inner metatarsal tubercle to tip of fourth toe)

### **3.2.5 Microhabitat parameters**

Along with the observation on amphibians, data on the crucial microhabitat parameters (Table 1) were also collected from each sampling units. (Vasudevan *et al.*, 2001). Weather parameters such as maximum air temperature, minimum air temperature, soil temperature at 5cm depth, rainfall and evaporation were obtained from daily weather report of agricultural meteorological department of Kerala Agricultural University.

**Table 1. Microhabitat parameters recorded from the selected agroecosystems of Thrissur dt., Kerala**

<b>Microhabitat Parameters</b>	<b>Method of Measurement</b>
Canopy Cover	Visual estimation
Leaf Litter Cover	10% sampling of quadrat by taking 1m <sup>2</sup> grids
Leaf Litter Depth	Measuring with metal scale
Shrub Density	10% sampling of quadrat by taking 1m <sup>2</sup> grids (stems height more than 1 m and less than 6m)
Soil Temperature	Infrared Laser Thermometer
Soil pH	10g air dried soil with 1:2.5 soil water ratio (Jackson, 1958) using pH meter
Soil Moisture	(Fresh weight – Oven dry weight)/ Oven dry weight in percentage

### 3.2.6 GIS Mapping

Quadrat locations were recorded using Garmin eTrex 20. Maps were prepared on Quantum GIS Version 2.18.4.





**Plate 2. Taking microhabitat parameters from quadrats of selected agroecosystems (A: Measuring surface soil temperature using laser thermometer, B: Measuring shrub density and litter cover)**

### 3.1 DATA ANALYSIS

#### 3.3.1 Species Richness Indices

According to Ludwig and Reynolds (1988), the species richness is the number of species in the community.

##### 3.3.1.1 Margalef's Diversity Index, $D_{Mg}$

Margalef's Diversity Index ( $D_{Mg}$ ) is a species richness measure which is derived using combination of S (the number of species recorded) and N (the total number of individuals summed over all S species) (Magurran, 1988).

$$D_{Mg} = (S - 1)/\ln N$$

### 3.3.2 Diversity Indices

Indices that combine both species richness and evenness into a single value are what it is referred as diversity indices. Shannon's index which is one of the Hill's diversity numbers recommended as one of the best measure of species diversity (Ludwig and Reynolds, 1988).

#### 3.3.2.1 Shannon Index, $H'$

The Shannon index ( $H'$ ) is the most widely used index in community ecology. It is a measure of the average degree of "uncertainty" in predicting to what species an individual chosen at random from a collection of 'S' species and 'N' individuals will belong. It is based on the proportional abundance of species where it assumes that individuals are randomly sampled from an indefinitely large population. This average uncertainty increases as the species increases and the distribution of individuals among the species becomes even. Thus  $H'$  has two properties that have made it a popular measure of species diversity:

- (1)  $H'$  is equal to zero, if and only if there is only one species in the sample
- (2)  $H'$  is maximum only when all S species are represented by the same number of individuals, that is, a perfectly even distribution of abundance (Ludwig and Reynolds, 1988).

The equation of the Shannon function, which uses natural logarithm (ln), is

$$H' = - \sum (p_i \ln p_i)$$

$H'$  is the average uncertainty per species in the infinite community made up of S species with known proportional abundance  $p_1, p_2, p_3, \dots, p_s$ .

### 3.3.3 Evenness Indices

When all species in the sample are equally abundant, evenness index should be maximum and decrease towards zero as the relative abundances of the species diverge away from evenness (Ludwig and Reynolds, 1988).

### 3.3.3.1 Evenness Index 2, E2

The most common evenness index used by ecologists is E1.

$$E1 = H'/\ln(S)$$

H' can attain maximum value when all of the species in the sample are perfectly even with one individual per species. Sheldon proposed an exponential form of E1 as an evenness index.

$$E2 = e^{H'/S}$$

### 3.3.4 Measure of dominance

This measure takes into account the abundance of the most common species rather than the measure of species richness. All the above-mentioned indices were calculated using PAST (Version 3.15) (Hammer *et al.*, 2001).

#### 3.3.4.1 Berger-Parker Index, d

The Berger-Parker Index is a dominance measure which expresses the proportional importance of the most abundant species where  $N_{\max}$  represent the number of individuals in the most abundant species and N denotes total number of individuals.

$$d = N_{\max}/N$$

The reciprocal of the index is usually taken so that an increase in the value of the index accompanies an increase in diversity. This index is independent of number of species but is influenced by sample size. It is one of the most satisfactory diversity measures available (Magurran, 1988).

### 3.3.2 Statistical Analysis

The statistical analysis done in the data includes binary logistic regression and test of association was done using SPSS.

#### 3.3.2.1 Binary Logistic Regression

Logistic regression is a probabilistic statistical classification model. This is done to determine the relationship between an outcome variable and a predictor variable. When the outcome variable is binary, binary logistic regression analysis is used. The binary logistic regression model is given below.

$$\text{Logit} [ p (y = 1 (x_1, x_2, x_3, \dots, x_p))] = \log [ p (y = 1 (x_1, x_2, x_3, \dots, x_p)) / 1 - p(y = 1 (x_1, x_2, x_3, \dots, x_p))]$$

The classification table is a method to evaluate the predictive accuracy of the logistic regression model. In this table, the observed values for the dependent outcome and the predicted values (at a user defined cut-off value) are cross-classified. For example, if a cut-off value is 0.5, all predicted values above 0.5 can be classified as predicting an event and all below 0.5 as not predicting the event.

If the logistic regression model has a good fit, then 'a' and 'd' cells have higher counts and 'b' and 'c' cells have fewer counts.

### 3.3.2.2 Test of association

The Karl Pearson's *chi* square test is used to test the significance of association between variables. Here the null hypothesis is that;

$H_0$  = The two variables are independent or there is no significant association between two variables (Ludwig and Reynolds, 1988).

The strength of the association was also calculated by using the tests of Phi and Cramer's V. The value ranges from -1 to 1. If the value is negative it is an indication of negative relation.

## **RESULTS**

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## RESULTS

### 4.1 SPECIES COMPOSITION OF AMPHIBIANS IN SELECTED AGROECOSYSTEMS OF THRISSUR DISTRICT, KERALA

A total of 14 anurans were recorded from the selected agroecosystems such as Botanical Garden, Cashew Plantation, Coconut Plantation, Homegarden, Rubber Plantation and Wetlands of Thrissur dt., Kerala, during the study time period. The 14 species belong to five families of anurans and maximum number of species were recorded from the family Dicroglossidae (35.7%). The species richness of amphibians from the study area are given in Table 2.

**Table 2. Species diversity of amphibians of selected agroecosystems of Thrissur dt., Kerala**

Sl. No.	Common Name	Scientific Name	Family	IUCN Status
1.	Common Indian Toad	<i>Duttaphrynus melanostictus</i>	Bufoidea	LC
2.	Ferguson's Toad	<i>Duttaphrynus scaber</i>	Bufoidea	LC
3.	Skittering Frog	<i>Euphlyctis cyanophlyctis</i>	Dicroglossidae	LC
4.	Jerdon's Bullfrog	<i>Hoplobatrachus crassus</i>	Dicroglossidae	LC
5.	Indian Bullfrog	<i>Hoplobatrachus tigerinus</i>	Dicroglossidae	LC
6.	Indian Burrowing Frog	<i>Sphaerotheca breviceps</i>	Dicroglossidae	LC
7.	Kerala Warty Frog	<i>Fejervarya keralensis</i>	Dicroglossidae	LC
8.	Ornate Narrow-mouthed Frog	<i>Microhyla ornata</i>	Microhylidae	LC



9.	Reddish Narrow-mouthed Frog	<i>Microhyla rubra</i>	Microhylidae	LC
10.	Painted Frog	<i>Uperodon taprobanica</i>	Microhylidae	LC
11.	Fungoid Frog	<i>Hydrophylax malabarica</i>	Ranidae	LC
12.	Sreeni's Golden-backed Frog*	<i>Indosylvirana sreeni</i>	Ranidae	NE
13.	Common Indian Tree Frog	<i>Polypedates maculatus</i>	Rhacophoridae	LC
14.	Jerdon's Bush Frog*	<i>Pseudophilautus wynaadensis</i>	Rhacophoridae	EN

\*Western Ghats endemic species

Menon's caecilian which is a Kerala endemic species was reported only from the Kerala Agricultural University main campus and not from any agroecosystems.

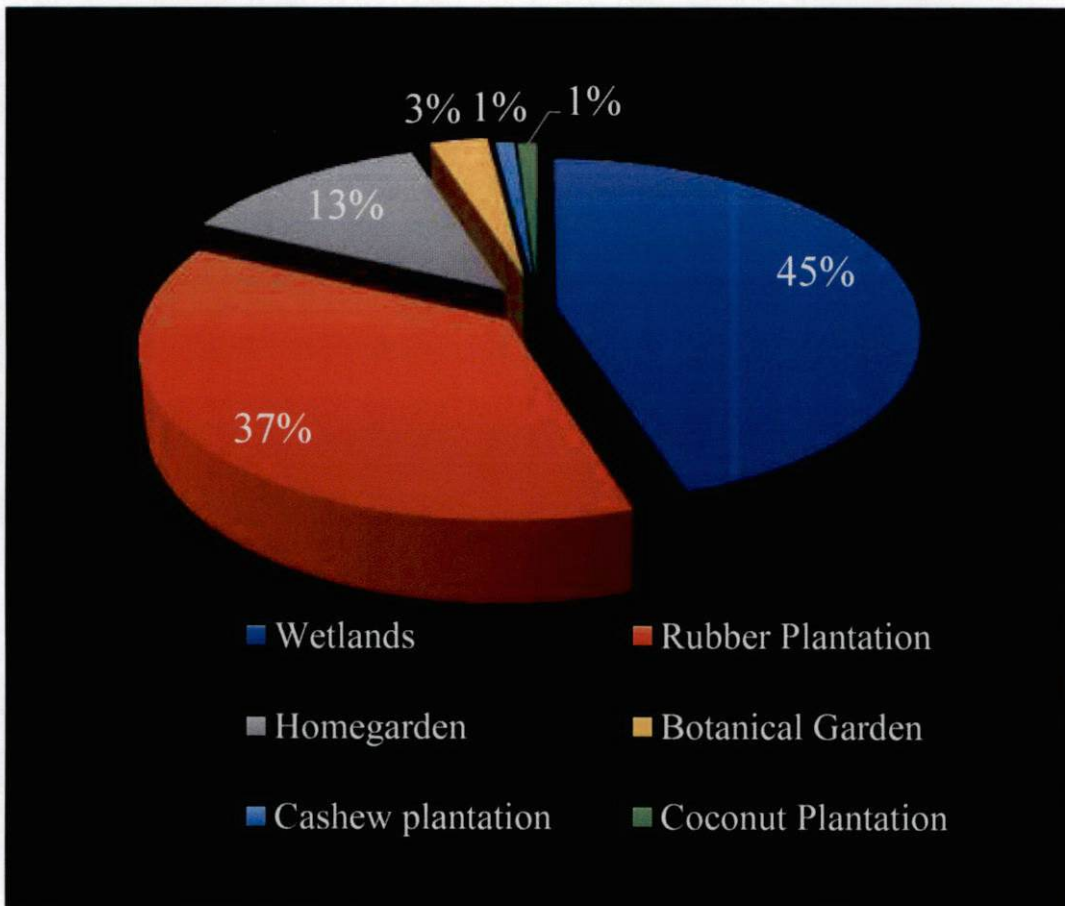
#### 4.1.1 Abundance and density of amphibians in selected agroecosystems in Thrissur dt., Kerala

Species richness, abundance and density of amphibians were assessed from selected agroecosystems of Thrissur dt., using quadrat sampling (Table 3, Figure 3 and Figure 4). From the selected agroecosystems ten species and a *Fejervaryan spp* complex were identified using quadrat sampling. Species richness was found to be highest in the Rubber Plantation followed by Botanical Garden. Abundance was found to be higher for *Pseudophilautus wynaadensis* followed by *Fejervaryan spp* from the selected agroecosystems of Thrissur dt., Kerala. The species richness and abundance of amphibians from selected agroecosystems of Thrissur dt., is given in Table 3.

**Table 3. Amphibian diversity and abundance from selected agroecosystems of Thrissur dt., Kerala using quadrat sampling**

Sl. No.	Species	Botanical Garden	Cashew Plantation	Coconut Plantation	Home garden	Rubber Plantation	Wetland	Total
1.	<i>Duttaphrynus melanostictus</i>	9	1	0	0	6	0	16
2.	<i>Duttaphrynus scaber</i>	0	0	0	0	0	14	14
3.	<i>Euphlyctis cyanophlyctis</i>	0	0	0	0	5	0	5
4.	<i>Hoplobatrachus crassus</i>	0	0	0	0	14	0	14
5.	<i>Hoplobatrachus tigerinus</i>	0	0	0	0	0	5	5
6.	<i>Sphaerotheca breviceps</i>	2	0	0	0	0	0	2
7.	<i>Fejervarya keralensis</i>	1	0	0	3	12	0	16
8.	<i>Fejervarya spp</i>	0	0	0	0	0	464	464
9.	<i>Hydrophylax malabarica</i>	2	0	0	0	0	0	2
10.	<i>Indosylvirana sreeni</i>	0	0	0	0	20	0	20
11.	<i>Pseudophilautus wynaadensis</i>	17	5	7	143	347	0	519
	<b>Total</b>	<b>31</b>	<b>6</b>	<b>7</b>	<b>146</b>	<b>404</b>	<b>483</b>	<b>1077</b>

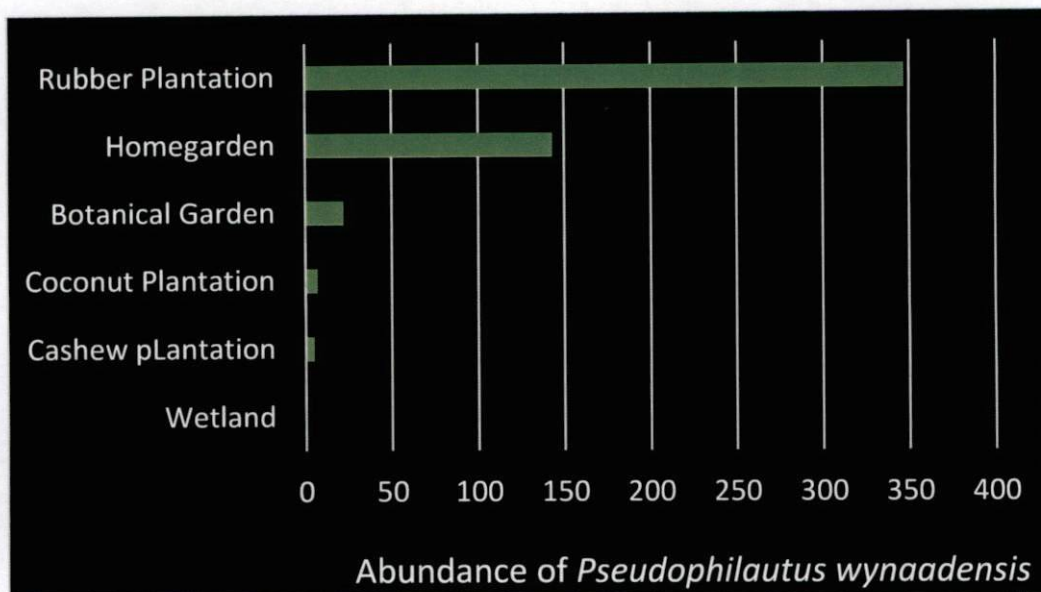
The encounter of amphibian species was higher in Wetlands, Rubber Plantation and Homegarden respectively (Figure 3). Amphibian abundance of selected agroecosystems of Thrissur dt., Kerala is given in Figure 2.



**Figure 3. Amphibian abundance of selected agroecosystems of Thrissur dt., Kerala**

All the quadrats in Homegarden had the presence of the Endangered and endemic anuran species, *Pseudophilautus wynaadensis*. The mean density of *Pseudophilautus wynaadensis* for Homegarden was 5 individuals per quadrats or 572 individuals/ ha.

*Pseudophilautus wynaadensis* was found to be cosmopolitan in the selected agroecosystems of Thrissur dt., Kerala. The abundance of *Pseudophilautus wynaadensis* of selected agroecosystems of Thrissur dt., is shown in Figure 3.



**Figure 4. Abundance of *Pseudophilautus wynaadensis* of selected agroecosystems of Thrissur dt., Kerala**

**4.1.2 Diversity of amphibians of selected agroecosystems in Thrissur dt., Kerala**

Amphibian species diversity were calculated for each of the selected agroecosystems in Thrissur district, Kerala (Table 4). Shannon index was found to be higher for Botanical Garden followed by Coconut Plantation. Coconut Plantation recorded highest value for Evenness which was succeeded by Cashew Plantation. Rubber Plantation stood at the first rank for Margalef index and Homegarden acquired the highest value for Berger-Parker index. The selected diversity indices were calculated for each agroecosystems of Thrissur dt., is given below in Table 4.

**Table 4. Species diversity indices of selected agroecosystems of Thrissur dt., Kerala**

<b>Indices</b>	<b>Botanical Garden</b>	<b>Cashew Plantation</b>	<b>Coconut Plantation</b>	<b>Homegarden</b>	<b>Rubber Plantation</b>	<b>Wetland</b>
Taxa (S)	5	2	3	2	8	3
Individuals (n)	38	8	12	175	416	489
Shannon (H)	1.17	0.38	0.82	0.09	0.73	0.23
Evenness ( $e^H/S$ )	0.64	0.73	0.76	0.55	0.26	0.42
Margalef (M)	1.10	0.48	0.80	0.19	1.16	0.32
Berger-Parker	0.58	0.88	0.67	0.98	0.83	0.95

**4.1.3 Comparison of diversity of amphibians between the selected agroecosystems in Thrissur dt.**

For comparison of the diversity of selected agroecosystems of Thrissur dt., *t* test was conducted. The statistical significant diversity difference in selected agroecosystems is given in Table 5.

Table 5. Comparison of amphibian diversity of selected agroecosystems of Thrissur dt., Kerala

	Botanical garden	Cashew Plantation	Coconut Plantation	Homegarden	Rubber Plantation	Wetlands
Botanical garden						
Cashew Plantation	2.83*					
Coconut Plantation	1.49 <sup>ns</sup>	1.33 <sup>ns</sup>				
Homegarden	6.96**	0.93 <sup>ns</sup>	3.13**			
Rubber Plantation	2.54*	1.61 <sup>ns</sup>	0.09 <sup>ns</sup>	8.67**		
Wetlands	5.99**	0.33 <sup>ns</sup>	2.43*	2.78**	6.83**	

\* significant at 5% \*\* significant at 1% ns means non-significant at 5%

Amphibian diversity was found to be significantly different between Homegarden and Botanical Garden, Homegarden and Coconut Plantation, Homegarden and Rubber Plantation, Homegarden and Wetlands, Wetlands and Rubber Plantation and Wetlands and Botanical Garden at 1% significance and between Cashew Plantation and Botanical Garden, Rubber Plantation and Botanical Garden and Wetland and Coconut Planation at 5% significance.

#### 4.2 COMPARISON BETWEEN THE SAMPLING TECHNIQUES

During the present study, we used three sampling methods, such as Quadrat Sampling, Visual Encounter Survey and Opportunistic Sampling. We tested the

efficiency of the three different sampling methods for the assessment of the amphibians in the agroecosystems in Thrissur dt. Maximum number of amphibian species were recorded from the Opportunistic Sampling. We also recorded two species such as *Uraeotyphlus menoni* and *Uperodon taprobanica* only when we followed the methodologies Opportunistic Sampling and Visual Encounter Survey (Table 6). Thus, the study found that Visual Encounter Survey in combination with Opportunistic Sampling is the most efficient method of estimating the amphibian richness in the agroecosystems.

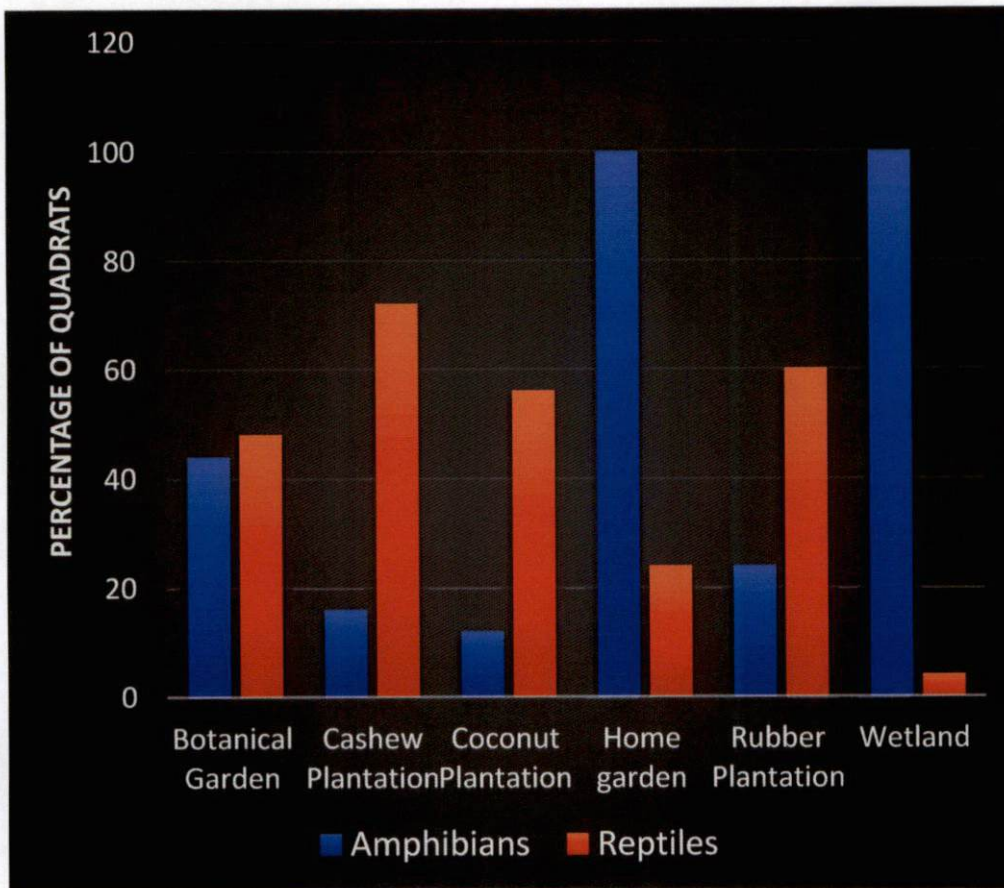
**Table 6. Sampling techniques adopted to assess the species diversity of amphibians of selected agroecosystems of Thrissur dt., Kerala**

Sampling Methods	Species	Unique Species*	Effort (min × 3 observers)
Quadrat Sampling	11	0	6750
Visual Encounter Survey	12	1	5400
Opportunistic Records	14	1	NA

\*Unique species are those that were detected using only one sampling method

#### 4.3 REPTILE AND AMPHIBIAN PRESENCE IN QUADRAT SAMPLING OF THE SELECTED AGROECOSYSTEMS IN THRISSUR DT.

The present study looked at both amphibian and reptile presence in all the 150 quadrats of selected agroecosystems of Thrissur dt. Presence of amphibians were confirmed for 49.33% of total quadrats in selected agroecosystems. In Homegarden and Wetlands all the quadrats had the presence of amphibians where as in Coconut Plantation and in Cashew Plantation the amphibian detection in quadrats was comparatively low where reptile presence was high. The reptile-amphibian presence in quadrats of selected agroecosystems are given in Figure 5.



**Figure 5. Percentage of quadrats having the presence of reptiles and amphibians in selected agroecosystems of Thrissur dt., Kerala**

#### **4.3.1 Association of occurrence of reptiles and amphibians in selected agroecosystems of Thrissur dt.**

Karl Pearson's chi square test was conducted to test the association and to find out the relationship between the amphibian and reptile occurrence in the selected agroecosystems in Thrissur dt. Non-significant chi square value indicates that there is no statistical significant association between the amphibian and reptile occurrence in Botanical Garden and the phi value depicts that the relation is very weak but inverse (Table 7).



**Table 7. Cross table of amphibian and reptile occurrence in Botanical Garden**

		Amphibian	
		Absence	Presence
Reptile	Absence	6	7
	Presence	8	4
Chi square = 1.07 <sup>ns</sup> Degrees of freedom = 1 Phi = -0.21			

ns – non-significant at 5%

In case of Cashew Plantation there is no statistical significant association and the relation is direct but very weak (Table 8).

**Table 8. Cross table of amphibian and reptile occurrence in Cashew Plantation**

		Amphibian	
		Absence	Presence
Reptile	Absence	7	0
	Presence	14	4
Chi square = 1.85 <sup>ns</sup> Degrees of freedom = 1 Phi = 0.27			

ns – non-significant at 5%

Coconut plantation also lack the statistical significant association between amphibian and reptile occurrence. But the very weak relation is inverse in this case (Table 9).

**Table 9. Cross table of amphibian and reptile occurrence in Coconut Plantation**

		Amphibian	
		Absence	Presence
Reptile	Absence	9	2
	Presence	13	1
Chi square = 0.71 <sup>ns</sup> Degrees of freedom = 1 Phi = -0.17			

ns – non-significant at 5%

In Rubber Plantation, chi square value indicates a statistical significant inverse association between reptile and amphibian occurrence. Here the phi value shows an average strength in the inverse association (Table 10).

**Table 10. Cross table of amphibian and reptile occurrence in Rubber Plantation**

		Amphibian	
		Absence	Presence
Reptile	Absence	5	5
	Presence	14	1
Chi square = 6.18* Degrees of freedom = 1 Phi = -0.5			

\* means significant at 5%

#### 4.4 RELATIONSHIP BETWEEN THE AMPHIBIANS AND THE HABITAT VARIABLES IN THE SELECTED AGROECOSYSTEMS IN THRISSUR DISTRICT, KERALA

The effect on the habitat variables on the presence or absence of the amphibians in the selected agroecosystems were compared using the binary logistic regression. In this case only one species, the *Pseudophilautus wynaadensis*, had adequate sample size for the analysis. In this model, Chi square was significant which indicated that the prediction model fits significantly better to the data collected. Model prediction explains roughly 58 to 81% of the variation (Table 11). Hence, the fitted model is a good fit.

**Table 11. Binary logistic regression model summary**

Model	Cox & Snell R Square	Nagelkerke R Square
		0.58
Chi square = 107.99* Degrees of freedom = 12		

**Table 12. Classification table for the occurrence of *Pseudophilautus wynaadensis* of selected agroecosystems of Thrissur dt., Kerala**

Observed		Predicted		
		Species		Percentage Correct
		Absence	Presence	
Species	Absence	79	5	94.0
	Presence	6	35	85.4
Overall Percentage				91.2

Classification table of observed and predicted response of the fitted logistic regression model for predicting the occurrence of *Pseudophilautus wynaadensis* is given as Table 12.

From the Table 12, we can see that there exists a good agreement between observed data and predicted data. The percentage of correct predictions is 91.2 per cent. The higher the overall percentage of correct predictions, the better is the model.

Odds of an event are the ratio of the probability that an event will occur to the probability that it will not occur. If the probability of an event occurring is  $p$  and the probability of the event not occurring is  $1 - p$  then the corresponding odds is a value given by

$$\text{odds of an event} = \frac{p}{1-p}$$

For every one unit increase in the predictor variable the odds will be increased by a factor. Here for one unit increase in soil moisture content leads to an increase in odds by a factor of 1.343.

Significance of the impact of predictor variables can be arrived from the range of lower and upper limit of 95% confidence interval for odds. When the value one where the regression coefficient will be zero lies in between the lower and upper limits the conclusion is that there is no significant impact by predictor variable on the presence of the species. If the value one lies outside the range of 95% confidence interval, then it indicates the significant impact of predictor variables. The influence of microhabitat parameters as per the binary logistic regression model is given as Table 13.

**Table 13. Effect of microhabitat parameters on the occurrence of *Pseudophilautus wynaadensis* of selected agroecosystems of Thrissur dt., Kerala**

Variables	Regression Coefficient	Odds	95% confidence interval for odds	
			Lower	Upper
Soil Moisture Content	0.30	1.34	1.14	1.58
Soil pH	5.27	195.15	10.98	3469.65
Surface Soil Temperature	0.073	1.08	0.64	1.80
Canopy Cover	0.037	1.04	0.98	1.10
Shrub Density	-0.07	0.93	0.89	0.98
Litter Cover	0.01	1.01	0.97	1.04
Litter Depth	0.84	2.32	1.09	4.94
Maximum Air Temperature	2.31	10.06	1.89	53.46
Minimum Air Temperature	2.88	17.82	2.57	123.70
Soil temperature at 5cm depth	-2.54	0.08	0.01	0.48
Evaporation	-1.81	0.16	0.03	0.99
Presence of waterbody (1)	-0.56	0.57	0.05	6.50

The microhabitat variables that influenced the occurrence of *Pseudophilautus wynaadensis* were soil moisture, soil pH, litter depth, maximum air temperature, minimum air temperature, shrub density, soil temperature at 5 cm depth and evaporation. Among the microhabitat variables, shrub density, soil temperature at 5cm depth and evaporation have a negative influence on the presence of *Pseudophilautus wynaadensis*.

4.5 MORPHOLOGICAL MEASUREMENTS OF THE AMPHIBIANS OF THE SELECTED AGROECOSYSTEMS IN THRISSUR DT.

Morphometric details of the amphibians of the study locations is detailed in the Tables 14 to Table 18. Morphological measurements are important taxonomical characters in most if the amphibians and thus is a great interest.

**Table 14. Morphological measurements of *Duttaphrynus scaber***

Morphological Measurements (mm)	Specimen 1	Specimen 2	Average
EL	2.39	2.91	2.65
ENL	3.29	3.01	3.15
FOL	24.06	21.14	22.60
FLL	8.24	8.56	8.40
HAL	9.67	9.2	9.44
HL	11.59	10.07	10.83
ShL	12.2	14.53	13.37
SVL	46.33	45.38	45.86
ThL	10.94	13.1	12.02
TYD	2.18	2.08	2.13

**Table 15. Morphological measurements of *Fejervarya keralensis***

Morphological Measurements (mm)	Specimen 1	Specimen 2	Average	Standard measurement for male (Kuramoto <i>et al.</i> , 2007)
SVL	33.1	44.11	38.61	32 - 40.8
HW	10.04	13.43	11.74	15.17 - 13.03
HL	10.06	12.13	11.1	12.41 - 10.59
EL	3.23	3.92	3.57	5.07 - 4.13

TYD	1.8	2.34	2.07	3.97 - 2.63
HAL	6.48	6.48	6.48	9.82 - 8.78
ShL	13.58	17.61	15.59	22.64 - 19.76
ThL	12.66	15.63	14.14	20.47 - 15.13
FOL	18.27	25.81	22.04	22.34 - 19.26

**Table 16. Morphological measurements of *Sphaerotheca breviceps***

Morphological Measurements (mm)	Specimen 1	Specimen 2	Average
EL	5.27	5.5	5.39
ENL	2.43	2.92	2.68
FOL	22.46	26.86	24.66
FLL	8.7	10.74	9.72
HAL	10.56	11.42	10.99
HL	13.16	14.09	13.63
ShL	14.75	19.39	17.07
SVL	42.07	46.2	44.14
ThL	11.4	17.65	14.53
TYD	1.74	2.32	2.03

**Table 17. Morphological measurements of *Indosylvirana sreeni***

Morphological Measurements (mm)	Specimen 1	Specimen 2	Average	Standard measurement for male (Biju <i>et al.</i> , 2014)
SVL	32.11	27.25	29.68	48
HW	9.82	8.18	9	15.4
HL	10.85	8.74	9.79	20
EL	3.57	3.26	3.42	5.6
TYD	3.29	1.93	2.61	4.4
FLL	5.14	4.92	5.03	9.6
HAL	8.51	7.6	8.1	13.5

ShL	13.8	11.7	12.75	24.5
ThL	10.59	11.5	11.04	26.5
FOL	20.93	16.09	18.51	24.5

**Table 18. Morphological measurements of *Pseudophilautus wynaadensis***

<b>Morphological Measurements (mm)</b>	<b>Specimen 1</b>	<b>Specimen 2</b>	<b>Average</b>	<b>Standard measurement (Bossuyt and Dubois, 2001)</b>
SVL	21.9	23.88	22.89	28.3
HW	7.86	7.91	7.88	9.6
HL	7.13	8.18	7.65	10.1
EL	3.1	3.2	3.15	4.1
TYD	1.5	1.3	1.4	2.1
FLL	5.23	4.52	4.88	6.5
HAL	6.79	5.88	6.34	7.3
ShL	12.41	10.64	11.52	13.4
ThL	11.53	9.11	10.32	12.9
FOL	15.58	13.62	14.6	11.8



## **DISCUSSION**

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## DISCUSSION

### 5.1 SPECIES COMPOSITION OF AMPHIBIANS IN SELECTED AGROECOSYSTEMS OF THRISSUR DISTRICT, KERALA

The study encountered 14 species of amphibians (Table 2) along with *Fejervarya* species complex from the selected agroecosystems. Coyle (2015) reported 12 species of amphibians from Homegarden of Kerala and three species from Plantations. Rathod and Rathod (2013) encountered 22 species from 15 Coffee Plantations of Western Ghats. Murali and Raman (2012) recorded 10 species from the stream side of tea monoculture, Coffee Plantation and a fragmented rainforest. Gamage *et al.* (2008), found seven species of amphibians from three different agroecosystems (tea, rubber, oil palm) and from a natural habitat in Sri Lanka. Wanger *et al.* (2009) reported six species of amphibians from Cacao Agroforestry alone in South-east Asia. A marked difference between the present study and these studies is that all these works were done in rainy season which is the peak season for amphibian detection.

Among the 11 families of amphibians identified from Kerala (Das, 2015) this study reported the presence of five amphibian families from the study locations (Table 2). Micrixalidae, Nyctibatrachidae, Nasikabatrachidae, Ranixalidae and Indotyphlidae were those amphibian families which were not reported from the selected agroecosystem. According to Biju *et al.* (2014) Micrixalidae commonly known as tropical frogs or torrent frogs occupy the splash zones of perennial hill streams. Nyctibatrachidae are nocturnal stream dwelling species endemic to Western Ghats (Biju *et al.*, 2011). Nasikabatrachidae is the burrowing frog family which will come up on the land surface for breeding once in a year (Biju and Bossuyt, 2003). Ranixalidae are monogeneric Western Ghats endemic family associated with evergreen forests close to the hill streams, damp leaf litter and in rocks and crevices (Garg and Biju, 2016). The present study has not included any habitat requirement of these endemic families to detect.

Dicroglossidae is the one of the generalist family which was represented by more species in the present work, where four among five genera were detected. Two among three genera from both Microhylidae and Ranidae were also recorded. According to Gururaja *et al.* (2007) anthropogenic changes in land use type, canopy cover and hydrological regimes support conservation of more generalist amphibian species. The work encountered only two of the 52 tree frogs. Tree frogs are highly vocal during monsoon which helps to detect the canopy dwelling species. During the present study in the non-rainy season, it was difficult to detect tree frogs even though there was enough arboreal vegetation except in wetlands.

Among the 136 Western Ghats endemic species in Kerala two were detected from the study area (*Pseudophilautus wynaadensis* and *Indosylvirana sreeni*). Most of the endemic species are restricted to the high-altitude rain forests of Kerala. Fifty amphibian species in Kerala comes under threatened category (Das, 2015) from which one Endangered species, *Pseudophilautus wynaadensis* was reported from all the agroecosystems under study, except the Wetlands (Table 3). According to Biju and Bossuyt (2009), *Pseudophilautus wynaadensis* is a species that is commonly seen among the wayside vegetation. The wayside vegetation was lacking in the Wetland study site and that could be the reason for the absence of *Pseudophilautus wynaadensis* in Wetland (Figure 4).

The present study has not encountered any caecilian species from the study location. According to Bhatta (1997), they occur at a depth of 45-60cm from the surface in rainy season and in dry season at the edges of streams. Caecilians are not found in plantations or orchards or gardens with low organic matter content. They are not seen from soil where chemical fertilizers or lime have been applied. Bhatta (1997) also stated that they are common in arecanut and coconut plantations with perennial water supply and low chemical input. From the enquiry, it was evident that lime and some chemical fertilizers were used in a regular basis in homegarden. Perennial water supply was not there in coconut plantation and cashew plantation.

In certain quadrats of coconut plantation even fertigation was practiced. These may be the possible reasons for the absence of caecilians from the study area.

### **5.1.1 Description of encountered species from selected agroecosystems of Thrissur dt., Kerala**

According to Boulenger (1890) batrachians come in between reptiles and fishes. They are distinguished by the structure of their limbs, by the absence of basio-occipital ossification, by the absence of amnion in the embryo and by the passage through larval, a branchiferous stage. They have a naked skin and if there are scales they were imbedded in the skin. Boulenger (1890) also disclosed that Indian batrachians fall into the suborder Phaneroglossa which possess tongue with separate ear opening.

#### **5.1.1.1 *Duttaphrynus melanostictus* (Schneider 1799)**

This is a member of Bufonidae, family of true toads which is nearly cosmopolitan and represented in three genera in Kerala fauna. This species is commonly known as Common Indian Toad (Plate 2). They are stubby with dark cranial ridges, elongated paratoid glands and their colour varies from black to yellow with all shades of red and brown. They have rough warty skin with a series of paired warts on the back with partly webbed toes with single subarticular tubercles. They have an external subgular vocal sac. Boulenger (1890) gave the key identification feature of the species as tympanum is two third diameter of the eyes.

Snout to vent length: 70 to 160 mm

Common Indian Toad is considered as a cosmopolitan species but now they are identified as a species complex and studies are going on to define new species to science. Even though they are the most successful groups of amphibians distributed across the world in a variety of habitat (Web, 2015) the present work only recorded few individuals from the study area. This species was encountered from three of the six study locations in different colour morphs. But it was not possible to sight a single individual from both coconut plantation, homegarden and

wetland even though they are considered as generalist. More number of individuals was sighted near to moist areas from botanical garden and rubber plantation. All the individuals sighted were adults.

#### **5.1.1.2 *Duttaphrynus scaber* (Schneider 1799)**

Ferguson's toad of Bufonidae is a small-sized toad living especially in marshes (Table 14). The dorsal skin is rough with numerous spinous tubercles. There is a set of enlarged white tubercles below the pale distinct tympanum and a few laterally (Plate 3). Nostrils are close to the snout. Presence of black dotted ridges on the head. Paratoid gland is visible but appear as circle. Limbs are also spiny. Fingers lack webbing but toes have. First finger is almost equal to second finger. They possess have a subgular vocal sac.

Snout to vent length: 22 to 36 mm

This small-sized Ferguson's Toad occupies various habitats including grasslands, farmlands, coastal marshes, and wet inland marshes (Web, 2015). And from the six different habitats it was sighted only from wetlands. It was reasonably a good count with 14 individuals including both adults and sub-adults. It was reported from five quadrats out of 25 which were located near to human habitats. It was interesting to note that more common *Duttaphrynus melanostictus* was absent in wetland.

#### **5.1.1.3 *Euphlyctis cyanophlyctis* (Schneider 1799)**

This Skittering Frog belongs to family Dicroglossidae which literally means fork-tongued frogs. They appear as brown to olive green with dark blotches on the smooth skin (Plate 4). Interorbital distance is much narrower than upper eyelid. Angular pointed eyes with distinct tympanum. More or less blunted snout with slight warty skin. Whitish underparts with barred limbs. Toes are webbed as they prefer waterlogged ponds. First finger does not go beyond second. Small inner metatarsal tubercle.

Snout to vent length: 64 to 70 mm

Skittering Frog was sighted only from a single quadrat in rubber plantation which is mainly due to the presence of a pond. These are the common frog occupying almost all waterlogged ponds. Since wetland was dry enough with little patches of water it was not fortunate to encounter this species.



Plate 3. Common Indian Toad, *Duttaphrynus melanostictus*



Plate 4. Ferguson's Toad, *Duttaphrynus scaber*





Plate 5. Skittering Frog, *Euphlyctis cyanophlyctis* (A & B)

#### **5.1.1.4 *Hoplobatrachus crassus* (Jerdon, 1853)**

A large frog that belongs to family Dicroglossidae, live in a semi-aquatic environment is commonly known as Jerdon's Bull Frog. Dark upperparts with long longitudinal folds running all over parts (Plate 5). They possess a pointed snout, predominant tympanum with distinct supra-tympanic fold and large protruding eyes. Frequently, individuals have a black band that runs between the limbs on the either side. The hind limbs are entirely webbed with long toes. Black patches can be observed on the smooths skin of upper thigh.

Snout to vent length: 160 to 165 mm

Jerdon's Bullfrog is another species which was detected from quadrat having an artificial waterbody in rubber plantation. This species lives in a semi-aquatic environment. The individuals sighted includes both adults and juveniles. During visual encounter survey, the species was encountered from the edges of nearby waterbodies.

#### **5.1.1.5 *Hoplobatrachus tigerinus* (Daudin, 1803)**

Indian bull frog is very large frog of semi-aquatic in habit. Skin is smooth and granular with longitudinal folds (Plate 6). Colour of the dorsum varies from olive green to grey with dark blotches. Most of the individuals have a white or yellow vertebral line. They have large protruding eyes with pointed snout and large tympanum. They have a predominant dark supra-tympanic fold. Fingers lack webbing and toes are fully webbed with slightly swollen tips. They have two external vocal sacs on either side of the chin which appears blue when they call.

Snout to vent length: 160 to 165 mm

Indian Bullfrog was reported only from the dry crevices of wetlands and bunds. This was also seen near the scattered patches of water. It was difficult to spot the species from crevices since the wetland was dry and cracked. All the individuals encountered were sub-adults.





**Plate 6. Jerdon's Bull Frog, *Hoplobatrachus crassus***



**Plate 7. Indian Bull Frog, *Hoplobatrachus tigerinus***

#### **5.1.1.6 *Sphaerotheca breviceps* (Schneider, 1799)**

This is commonly known as Indian burrowing frog which belong to Dicroglossidae (Table 16). They possess a smooth granulate skin of light brown with some scattered tubercles and marbling (Plate 7). Sometimes they have longitudinal folds. Their snout is blunt. There is a strong fold which starts from eye to shoulder above the tympanum. Most of them have a vertebral band that runs at the upper parts. Toes are half webbed. They possess a large inner metatarsal tubercle with sharp edge to excavate burrows. The males possess internal sub-gular vocal sacs and their throat appears black in colour.

Snout to vent length: 40 to 57 mm

Indian Burrowing Frog was spotted only from a single quadrat in botanical garden where there was an ongoing project of digging a pond. It was interesting to note that it was not detected during 7pm but from the same location by 9:15pm it was reported. There were two adult individuals in a dull brown colour of soil. Literature states that this species will emerge from its burrow during night hours which was observed during the present study.

#### **5.1.1.7 *Fejervarya keralensis* (Dubois, 1980)**

Kerala warty frog is a medium – sized mud coloured frog belonging to Dicroglossidae family (Table 15). Their skin possesses small warts and projections with a prominent tympanum and less developed supra-tympanic fold (Plate 8). Their groin and back side of the thigh is black marbled with white or pale yellow. They have a barred lower lip. Frequently individuals have a white vertebral line which runs up to the vent. Their fingers are pointed without webbing and toes are partly webbed. Their first finger is longer than the second. Hind legs possess a shovel shaped elliptical inner metatarsal tubercle.

Snout to vent length: 60 to 70 mm

This species was detected from botanical garden, homegarden and from rubber plantation. In all these habitats, these species were encountered near moist areas. According to Kotaki *et al.* (2010), *Fejerevarya* is highly cryptic species and



Plate 8. Indian Burrowing Frog, *Sphaerotheca breviceps*

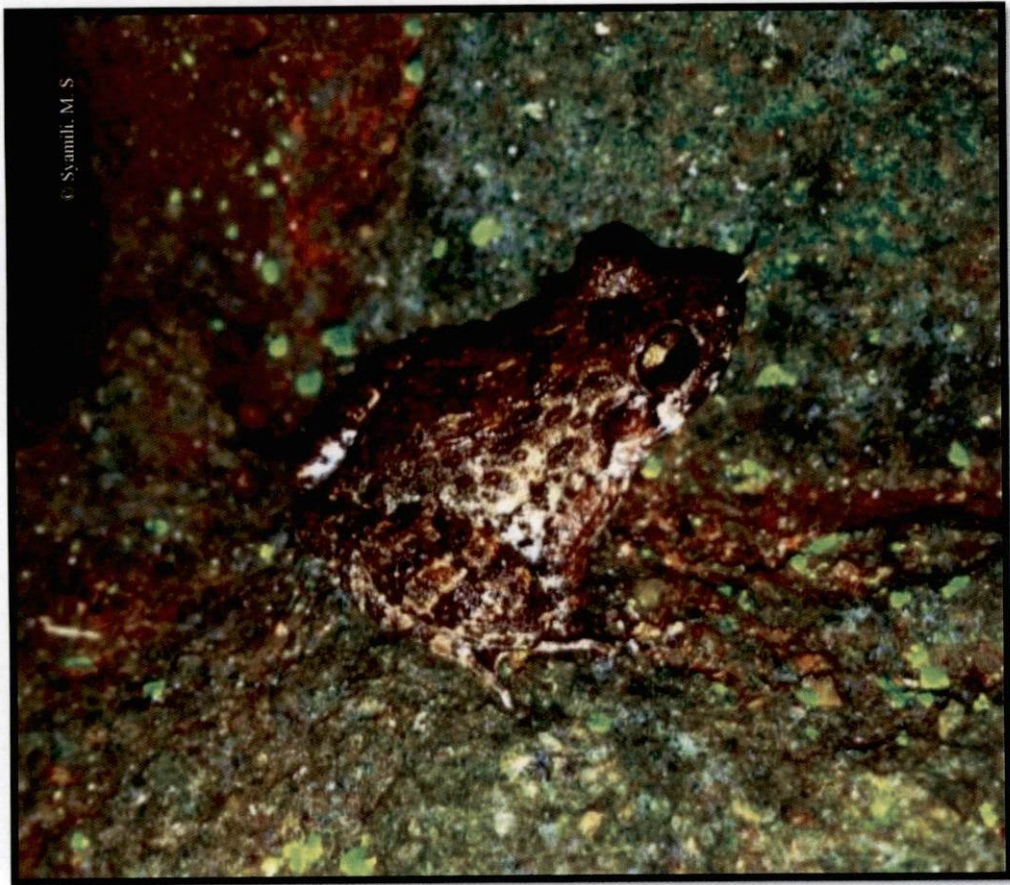


Plate 9. Kerala Warty Frog, *Fejervarya keralensis*

several species complexes are difficult to identify due to lack of distinguishing morphological feature. The drawback is that 464 individuals from wetlands were difficult to identify since species descriptions are not available and the studies are still going on.

#### **5.1.1.8 *Microhyla ornata* (Dumeril and Bibrom, 1841)**

Commonly known as Ornate narrow-mouthed frog of family Microhylidae. This species has a dark inverted 'Y' marking starting from the back of the eyelid ending at loin, on the reddish or greyish olive smooth skin (Plate 9). They also possess a dark band from back of the eye up to groin. Their snout is blunted with indistinct tympanum lacking supra-tympanic fold. Slender fingers lack webbing where the first finger is shorter than the second. Toes are also slender with slight webbing. Male have sub-gular vocal sac. Limbs have dark bands.

Snout to vent length: 24 to 32 mm

This species was recorded from the fringe area of wetland near to human habitat with herb growth. The three individuals observed were juveniles. None of the other agroecosystem could observe the species though opportunistically it was detected near pond.

#### **5.1.1.9 *Microhyla rubra* (Jerdon, 1854)**

Reddish narrow-mouthed frogs are small, stout ground dwelling frogs of family Microhylidae. They have fine granular skin with reddish-brown dorsum with darker irregular pattern at the centre (Plate 10). A dark lateral line is present from the tip of the snout through the eye reaching the groin. Their snout is rounded and tympanum is indistinct but a distinct supra-tympanic fold is present. Barred hindlimbs with dark markings on the loin. Fingers lack webbing where, first finger is shorter than second. Toes are partly webbed with dilated tips and two metatarsal tubercles. Males have a sub-gular vocal sac and their throat is dark in colour.

Snout to vent length: 30 to 35 mm



Plate 10. Ornate narrow-mouthed frog, *Microhyla ornata*



Plate 11. Reddish narrow-mouthed frog, *Microhyla rubra*

It was sighted from the premises of pond in rubber plantation during visual encounter survey. Here also the individuals recorded were juveniles of very small size. This was opportunistically reported from moist areas near nursery and pond.

**5.1.1.10 *Uperodon taprobanicus* Parker, 1934**

A snout looking, fat bodied frog having burrowing habit commonly known as Painted frog. Their body shape is globular with nostril at the tip of the snout. Their skin is smooth with fine tubercles and have brownish-orange markings. Their tympanum is indistinct with a supra-tympanic fold. Loreal region is concave and they have an interorbital band. Slender fingers possess disk like dilated tips. First finger is shorter than second. Toes are feebly webbed with slight dilated tips.

Snout to vent length: 60 to 75 mm

This was spotted from a tree hole near coconut plantation. None of the study locations could record this species may be due to the difficulty to spot the individuals. According to literature they inhabit flooded wetlands. But it was not spotted from the dry wetlands.

**5.1.1.11 *Hydrophylax malabarica* (Tschudi, 1838)**

These belong to true frog family Ranidae, commonly known as Fungoid frog. Smooth and finely granular dorsum varies from brownish red to bright crimson (Plate 11). Nostril is more nearer to snout than to the eyes. There is a strong glandular fold from eye to the shoulder followed by one or two glandules. From snout to groin, a dark black band and it further extends to limbs and they are irregularly blotched with white. First finger is extending beyond the second. Toes are relatively short and half webbed. Both finger and toes tips are swollen to form disk.

Snout to vent length: 65 to 80 mm

Fungoid frog was detected from botanical garden and rubber plantation. It was found in moist places in these habitats. These frogs occupy wide variety of habitat including various forests, plantations, agricultural fields and rural gardens.

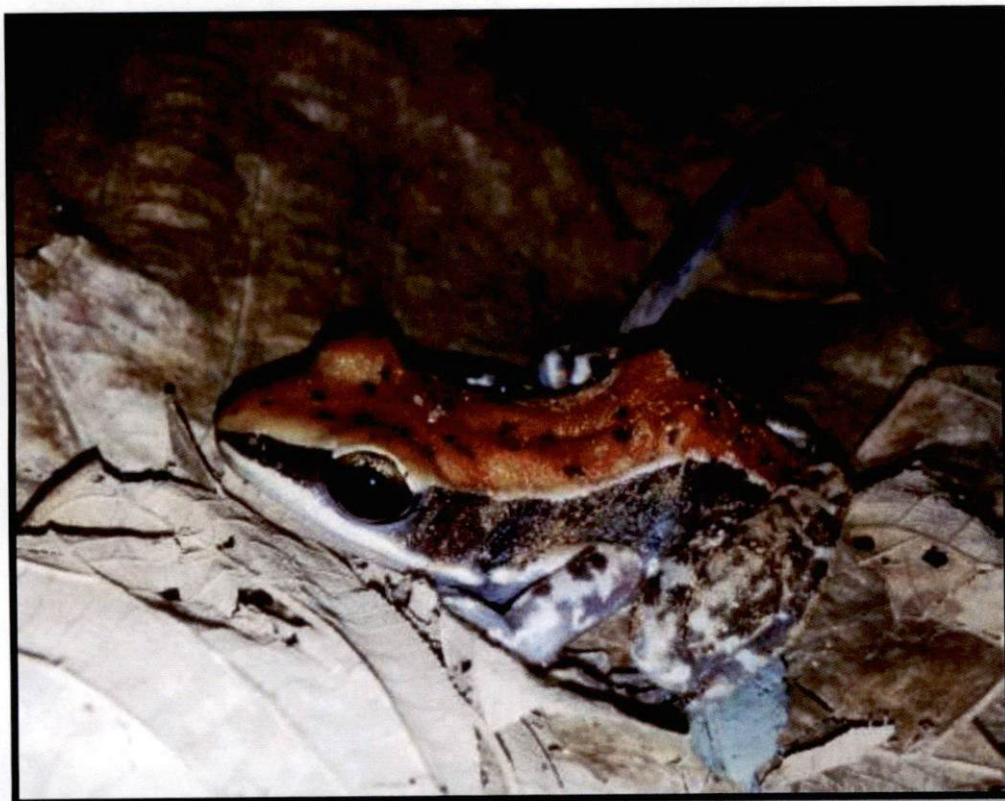


Plate 12. Fungoid frog, *Hydrophylax malabarica*

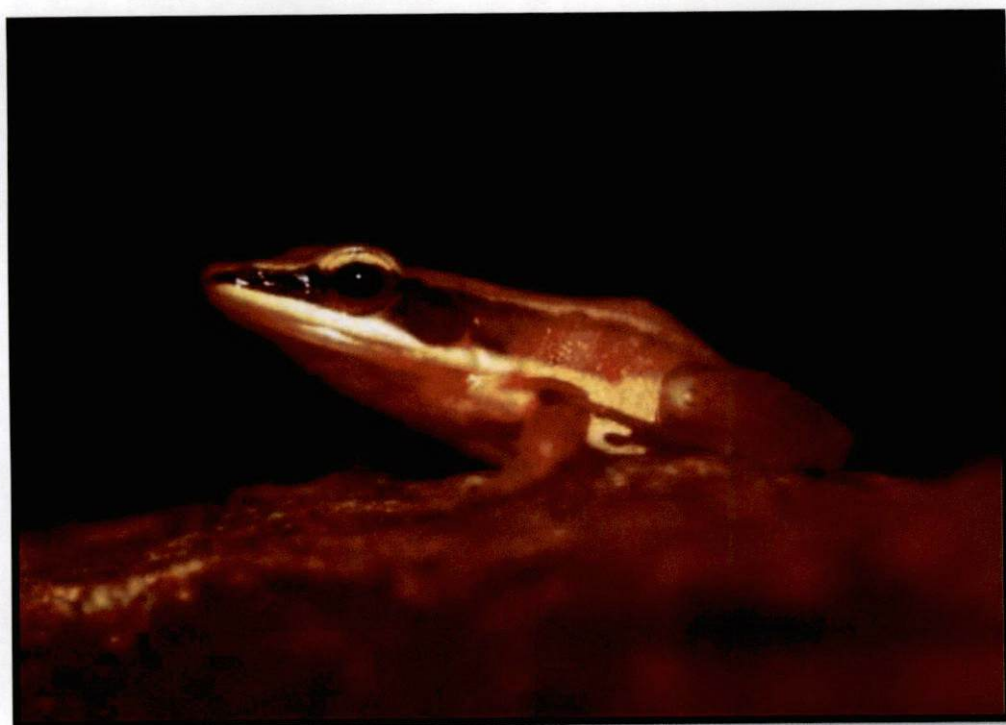


Plate 13. Sreeni's golden-backed frog, *Indosylvirana sreeni*

#### 5.1.1.12 *Indosylvirana sreeni* (Biju et al., 2014)

Sreeni's golden-backed frogs belong to the family Ranidae (Table 17). Their granular dorsal part is bronze coloured with light brown lower flank (Plate 12). They possess a distinct brown tympanum with a yellow-white stripe on the upper lip continuing through rictal gland to above arm insertion. Their snout is sub-elliptical. Anterior part of the thigh is reticulated by yellow-brown patches. Their second toe webbing below the first tubercle. Webbing are dark grey with minute specks. Males possess single oval shaped nuptial pads on first finger.

Snout to vent length: 44 to 52 mm (males), 64 to 80 mm (females)

This species was again found only in a single quadrat of rubber plantation which had the presence of waterbody. It is a species seen close to ponds. During the study time, it was observed near various waterbodies in the premises of study location.

#### 5.1.1.13 *Polypedates maculatus* (Gray, 1834)

Common Indian tree frog belongs to the largest family Rhacophoridae. Smooth dorsum with varying colouration from pale buff to moderate olive brown to chestnut with dark spots (Plate 13). Distinct tympanum with prominent supra-tympanic fold. Darker temporal and loreal region. Back of thighs and the groin have irregular patches of yellow on grey or brown. Males possess nuptial pads on first finger in breeding season. Fore limbs lack webbing and hind limbs possess prominent webbing.

Snout to vent length: 34 to 57 mm (males), 44 to 89 mm (females)

The most common of all tree frog was not so common in our study locations. During visual encounter survey, a male and female of the species were recorded from coconut plantation. After the first summer shower, again one individual was observed from coconut plantation. Literature says that this species may enter our bathrooms and retreat into some moist corner during drier months.





Plate 14. Common Indian Tree Frog, *Polypedates maculatus*



Plate 15. Menon's Caecilian, *Uraeotyphlus menoni*

5.1.1.14 *Pseudophilautus wynaadensis* (Jerdon, 1853)

Jerdon's bush frog is the most common bush frogs of Kerala in the northern part of Shencottah gap (Plate 15). Slender body is uniform brown but it is highly variable. Dark and distinct tympanum with dark supra-tympanic fold. Nostril is close to the tip of the snout than to the eye (Table 18). An inverted 'V' shaped mark is present between the inter-orbital space. Hindlimb is prominently dark banded. Fingers and toes are more or less greyish brown in colour.

Snout to vent length: 25 to 30 mm

It is one of the common bush frogs of Kerala. This is the anuran species recorded from five study locations especially in homegarden. All the 25 quadrats in homegarden detected the presence of this species. The highest count of the species was from rubber plantation where there were about 30-40 individuals in a single shrub of *Rauwolfia serpentina* near a marshy land. In botanical garden after the first summer showers juvenile of the species was observed from the marshes.



Plate 16. Jerdon's Bush Frog, *Pseudophilautus wynaadensis* (A, B, C & D are different colour morphs)

### 5.1.2 Amphibian diversity in selected agroecosystems of Thrissur dt., Kerala

Allmon (1991) reported that both diversity and abundance of forest floor amphibians of Central Amazon was strongly seasonal and peak at wet season showing a positive correlation with litter volume and moisture. According to Neckel-Oliveira and Gascon (2006), presence of aquatic habitat is crucial for the existence of certain species. From the present study, species richness was found to be maximum in rubber plantation where there was an artificial waterbody (Table 3). This is the main reason to record *Euphlyctis cyanophlyctis*, *Hoplobatrachus crassus*, *Indosylvirana sreeni*, *Fejervarya keralensis* and *Microhyla rubra* from the location. *Fejervarya* are generalists found near still and stagnant water (Kuramoto *et al.*, 2007) and *Indosylvirana sreeni* can be seen on rocks or at banks of the streams or ponds (Biju *et al.*, 2014). The average soil temperature was lower in rubber plantation compared to other habitats may be due to the combination of canopy cover, litter cover, herb growth and shrub growth.

The abundance in wetland was mainly due to the presence of a species complex, *Fejervarya* whose taxonomy is in question (Table 3). *Fejervarya spp* of plains have not been defined till and it is only possible to identify this to generic level (Plate 16). An interesting observation is that Wetland was very dry and the clay bed have shrunk which made cracks and crevices on the land. The major inhabitants of these crevices were the *Fejervarya spp*.

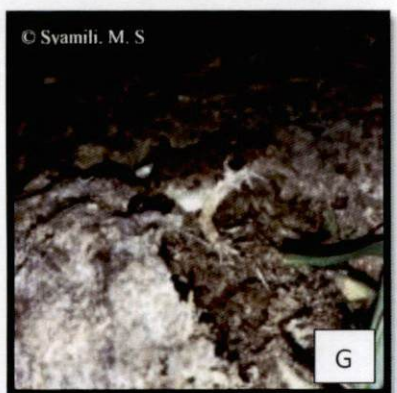
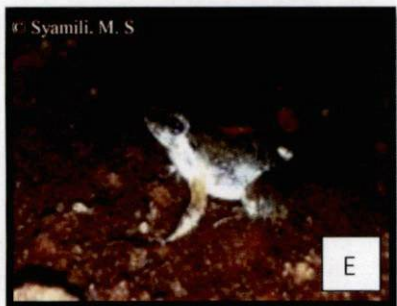
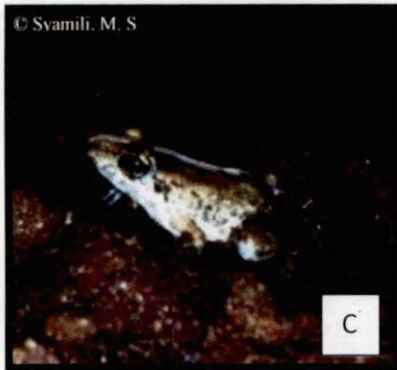
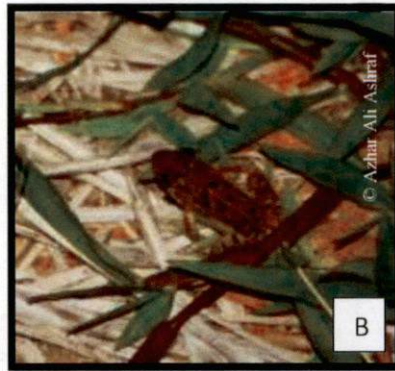
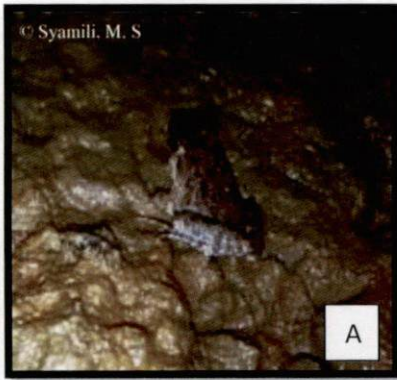


Plate 17. Unidentified *Fejervarya* spp from Wetlands (A, B, C, D, E, F, G and H)

Rathod and Rathod (2013) quoted that species composition actually varies with land use changes and those species which require specific ecological niche may be absent in the modified land use type. Rathod and Rathod (2013) also explained that open canopy can increase the temperature and evaporation and can decrease the persistence of moist areas. This can be one reason for lower encounter of amphibians from Coconut Plantation and Cashew Plantation (Table 3). From the habitat parameters, it was observed that mean soil moisture content which is crucial for amphibian detection was comparatively low in these two habitats.

Coyle (2015) recorded 11 species of amphibians from Homegarden whereas the present study could report only two species but *Pseudophilautus wynaadensis* was present in all the 25-quadrat deployed in the Homegarden. Of the 11, *Euphlyctis cyanophlyctis* are strictly restricted to stagnant water bodies or at the water's edge in ponds (Joshy *et al.*, 2009). Another generalist species, *Hoplobatrachus tigerinus* also occupy the similar habitat of *Euphlyctis cyanophlyctis*. The reason behind the low species richness from the Homegarden (Table 4) in the present work may be due to the seasonal limitation to detect the species especially tree frogs and lack of any stagnant waterbodies for aquatic and semi-aquatic species. It is interesting to note that one of the generalist species in plains, *Duttaphrynus melanostictus* was not able to record from the Homegarden even though soil moisture, litter cover, shrub density and canopy cover were ambient for floor dwelling amphibians.

Botanical Garden had the presence of unique species during the study period. It is *Sphaerotheca breviceps*, which is a burrowing frog. It was accidentally reported from a pond being dug, clearly depicted the burrowing behavior of the species.

### **5.1.3 Comparison of amphibian diversity between the selected agroecosystems of Thrissur dt., Kerala**

To estimate the species diversity measures Shannon index, evenness, Margalef index and Berger-Parker indices were calculated for each study habitats (Table 4).

Shannon index was maximum and Berger-Parker index was minimum for botanical garden. This may be due to the wide array of habitats which lead to richness of species and the occurrence of ambient microhabitat parameters that made the more or less equal abundance.

Shannon index was found to be minimum and Berger-Parker index was higher in case of Homegarden where a dominant species was present. A uniform microhabitat with dense undergrowth and high soil moisture content may be the reason for the presence of a dominant species.

Evenness was lower in Rubber Plantation where 83.4 % of the individuals belong to same species. *Pseudophilautus wynaadensis* were present in a high number near the marshy area with dense shrub growth. The individuals included both adults and sub-adults.

Diversity *t* test was carried out for 15 habitat combinations. Out of 15 habitat combinations nine combinations were found to have significant difference between each other (Table 5). Homegarden had significant difference with Botanical Garden, Coconut Plantation, Rubber Plantation and Wetland. The reason may be due to the highest average soil moisture content of homegarden. Rubber Plantation had significant difference with Botanical Garden mainly due to higher soil moisture of Botanical Garden whereas the significant difference between Rubber Plantation and Wetland may be due to lower soil temperature in Rubber Plantation because of dense undergrowth and canopy cover.

## 5.2 COMPARISON BETWEEN THE SAMPLING TECHNIQUES

During the study, it was observed that highest number of species were detected opportunistically from the study area (Table 6). In this study, there were no significant difference between species diversity estimation from Visual Encounter Survey and Quadrat Sampling. This may be due to the seasonal limitation of amphibian detection during the non-rainy season. Vasudevan *et al.* (2001), stated that the adaptive cluster sampling as a better method to estimate the

rain forest floor amphibians. According to Harikrishnan *et al.* (2012) no amphibians were detected using quadrat sampling but both visual encounter survey and opportunistic records encountered amphibian species. According to Harikrishnan *et al.* (2012), Visual Encounter Survey and Opportunistic Records can give information on the species richness but to get quantitative information on the species abundance it is better to go for quadrat sampling.

Except for *Hoplobatrachus tigerinus* and *Hydrophylax malabarica* other species had higher encounter rate in quadrat sampling. This result shows that the habitat features, microclimate and various associations of quadrat in the study locations are very important for the species and their abundance.

### 5.3 REPTILE - AMPHIBIAN ASSOCIATION OF SELECTED AGROECOSYSTEMS OF THRISSUR DISTRICT, KERALA

In the present study, the quadrat sampling was successful to detect amphibian species even though their abundance was less due to the seasonal hindrance (Figure 5). In a study from rainforests of Andaman and Nicobar Islands, quadrat sampling failed to detect amphibian species but could record the reptile diversity (Harikrishnan *et al.*, 2012).

#### 5.3.1 Test of significance of reptile-amphibian association in selected agroecosystems in Thrissur dt.

Karl Pearson's chi square test was done to investigate whether there is any relationship between the occurrence of reptiles and amphibian in the selected agroecosystems under study. And it was found that in the rubber Plantation there existed a significant negative (95%) relationship between the occurrence of reptiles and amphibian (Table 10). Certain quadrats in the Rubber Plantation had specific microhabitat. For example, the presence of an artificial waterbody elevated the amphibian presence in such quadrants and the presence of the scattered boulders, snags and fallen logs in the Rubber Plantation increased the reptile encounter rate (Vasudevan *et al.*, 2001; Harikrishnan *et al.*, 2012). Chi square test was impossible

in Wetlands and Homegarden due to the absence of categorical variable in one group. In all the remaining agroecosystems, except in Cashew Plantation, also had shown an inverse relationship between the reptile and amphibian occurrence, however, these were not statistically significant. According to Inger and Colwell (1977) predictable environments lead to the formation of distinct guilds whereas unpredictable environment prevents the distinct guild formation. Toft (1985) stated the reasons for resource partitioning as competition, predation and other factors that operate due to interspecific interaction like physiological stress. He concluded that resource partitioning is due to the result of combination of two or more factors.

#### 5.4 INFLUENCE OF HABITAT PARAMETERS ON THE OCCURRENCE OF *PSEUDOPHILAUTUS WYNAADENSIS*

Binary logistic regression of *Pseudophilautus wynaadensis* was done to understand the influence of the predictor variables on the presence or absence of the *Pseudophilautus wynaadensis*, in the selected agroecosystems. The variables which had no impact on the occurrence of the species were surface soil temperature, canopy cover, litter cover and presence of waterbody (Table 13). The reason behind this result is that the species was present in Coconut Plantation, Cashew Plantation, Rubber Plantation, Homegarden and Botanical Garden. Among all these habitats only the Rubber Plantation had the presence of waterbody and compared to other habitats coconut plantation was characterised with low canopy cover, litter cover and high soil temperature.

In India, studies on the influence of habitat characters on the amphibian occurrence is scarce. According to Allmon (1991), the litter volume and moisture had a positive correlation with forest floor litter frogs of Amazon. In the present study, litter cover had no influence on the presence of *Pseudophilautus wynaadensis*. One of the reason can be that *Pseudophilautus wynaadensis* is a bush frog. Wyman (1990) investigated the impact of soil acidity and moisture on distribution of amphibians in New York where he stated that both soil moisture and soil pH should be determined when factors influencing amphibian distributions are



being examined. Soil pH and soil moisture content had a positive influence on the species occurrence in the present study. Especially in Homegarden even though lime was applied in a regular basis all the quadrats recorded the presence of species. In all the habitats where soil moisture content was higher there was a higher probability to encounter the species and it was observed in almost all cases. The habitat variables that negatively influenced, the presence of the bush frog, *Pseudophilautus wynaadensis* were the shrub density, soil temperature at 5cm depth and evaporation.

## **SUMMARY**

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## SUMMARY

Conservation strategies that are exclusively focussed on the protected areas while ignoring the surrounding multiple land use landscapes in which they are embedded, has been proven as a failed strategy. It is a fact that most of these protected areas are virtual islands, embedded within a matrix of multiple land uses. A large proportion of biodiversity coexist with humans in their managed ecosystems which can hold minimum viable population of rare and endangered native fauna and flora. The potential of such landscapes in conserving native biodiversity is still untapped. Thus, the present study investigated the amphibian richness and diversity of selected agroecosystems in Thrissur dt., Kerala. The objectives of the study were to understand the species diversity and community structure of amphibians of selected agroecosystems of Thrissur dt. We also investigated how the amphibian diversity was related to the habitat variables and assessed the spatial variation in the amphibian distribution. The study was conducted from January 2017 to June 2017 in selected agroecosystems such as Botanical Garden, Cashew Plantation, Coconut Plantation, Homegarden, Rubber Plantation and Wetlands. The method used to monitor the amphibian diversity included quadrat sampling, visual encounter survey and opportunistic sampling. Amphibians are considered as the ecological indicators of a healthy ecosystem. This may be the first attempt to monitor the amphibian diversity and richness of selected agroecosystems of Kerala. The salient findings are summarised below.

1. A total of 14 anurans were recorded from the selected agroecosystems such as Botanical Garden, Cashew Plantation, Coconut Plantation, Homegarden, Rubber Plantation and Wetlands of Thrissur dt., Kerala. Besides 14 species, one additional species were also recorded from the Kerala Agricultural University campus.
2. The present work recorded two Western Ghats endemic species *Indosylvirana sreeni* and *Pseudophilautus wynaadensis*, from the agroecosystems of Thrissur while the latter was a threatened one with Endangered status as per IUCN Red List

3. Amphibian species richness was found to be higher in Rubber Plantation with eight species followed by Botanical Garden with five species.
4. The amphibian encounter was higher in the Wetlands followed by Rubber Plantation.
5. Most common of amphibian species was *Pseudophilautus wynaadensis*. *Pseudophilautus wynaadensis* was found to be cosmopolitan in the present study with its detection from all the five habitats.
6. All the sampling units in Homegarden and Wetlands (25 quadrats each) had the presence of amphibians.
7. Amphibian diversity was found to be significantly different between Homegarden and Botanical Garden, Homegarden and Coconut Plantation, Homegarden and Rubber Plantation, Homegarden and Wetlands, Wetlands and Rubber Plantation and Wetlands and Botanical Garden at 1% significance and between Cashew Plantation and Botanical Garden, Rubber Plantation and Botanical Garden and Wetland and Coconut Planation at 5% significance.
8. Among the various methods used, we found that visual encounter survey and the opportunistic sampling, were found to be efficient.
9. The micro-habitat variables that influenced the presence or absence of *Pseudophilautus wynaadensis* were soil moisture, soil pH, litter depth, maximum air temperature, minimum air temperature, shrub density, soil temperature at 5 cm depth and evaporation.

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**SPECIES DIVERSITY AND COMMUNITY  
STRUCTURE OF AMPHIBIANS OF SELECTED  
AGROECOSYSTEMS IN THRISSUR, KERALA**

*by*

**SYAMILI M.S.**

**ABSTRACT OF THE THESIS**

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## ABSTRACT

The research work entitled “Species diversity and community structure of amphibians of selected agroecosystems in Thrissur, Kerala” was carried out in the agroecosystems such as Cashew Plantation, Coconut Plantation, Homegarden, Rubber Plantation, Wetlands and in Botanical Garden from January 2017 to May 2017. The main objective of the study was to find out the species diversity and community structure of amphibians of selected agroecosystems and its association with various habitat parameters. A total effort of 225 man hours were spent in the field during night. The methods adopted for the study include Quadrat Sampling, Visual Encounter Survey along with the Opportunistic Sampling.

A total of 14 anurans were recorded from the study locations. Species richness was found to be highest in Rubber plantation with eight species. Encounter of amphibians was more in Wetlands followed by Rubber Plantation. All the sampling units in Homegarden and Wetlands (25 quadrats each) had the presence of amphibians. The research found significant difference in amphibian diversity among the different agroecosystems. Visual encounter survey combined with opportunistic sampling was found to be an efficient methodology to assess the amphibian species richness of an agroecosystem. The study found an inverse relation between the presence of amphibians and reptiles in the sampling units. The microhabitat variables such as soil moisture, soil pH, litter depth, maximum air temperature, and minimum air temperature has a positive influence on the presence of *Pseudophilautus wynaadensis* while shrub density, soil temperature at 5 cm depth and evaporation had significant inverse relation.

The present work recorded two Western Ghats endemic species *Indosylvirana sreeni* and *Pseudophilautus wynaadensis*, from the agroecosystems of Thrissur while the latter was a threatened one with Endangered status as per



IUCN Red List. Both the reports of *Pseudophilautus wynaadensis* and *Indosylvirana sreeni* are of interest, as they are reported from a lower altitude (50m) than the already known lowest altitude range (900m and 100m respectively) of these species. Thus, the study highlights the importance of the agroecosystems in acting as important habitats for the amphibian fauna in Kerala.

## **APPENDIX**

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APPENDIX

Appendix No: 1. Details of 150 quadrats laid in the selected agroecosystems of Thrissur dt., Kerala

Agroecosystem	Quadrat Number	Latitude	Longitude	Soil Moisture (%)	Soil Ph	Soil Temperature (°C)	Canopy Cover (%)	Shrub density (%)	Litter Cover (%)	Litter Depth (cm)	Max Air Temp (°C)	Min Air Temp (°C)	Soil temp at 5 cm depth	Evaporation (mm)	Presence of waterbody	<i>Duttaphrynus melanostictus</i>	<i>Duttaphrynus scaber</i>	<i>Euphyctis cyanophlyctis</i>	<i>Hoplobatrachus crassus</i>	<i>Hoplobatrachus tigerinus</i>	<i>Sphaerotheca breviceps</i>	<i>Feverarya keralensis</i>	<i>Hydrophylax malabarica</i>	<i>Indosylvirana sreeni</i>	<i>Pseudophilautus wynaadensis</i>	<i>Feverarya spp</i>	Reptile Presence
Coconut	C1	10.55	76.28	2.27%	4.87	19.2	60	0	0	0	35.3	21.9	27.2	3.7	0	0	0	0	0	0	0	0	0	0	0	0	1
Coconut	C2	10.55	76.28	0.24%	4.92	15.5	15	0	0	0	35.3	21.9	27.2	3.7	1	0	0	0	0	0	0	0	0	0	0	0	1
Coconut	C3	10.55	76.28	0.08%	4.81	14.3	30	50	0	0	35.3	21.9	27.2	3.7	0	0	0	0	0	0	0	0	0	0	0	0	1
Coconut	C4	10.55	76.28	1.61%	5.08	14.7	60	10	0	0	35.3	21.9	27.2	3.7	0	0	0	0	0	0	0	0	0	0	0	0	1
Coconut	C5	10.55	76.28	0.72%	5.19	20.6	85	0	20	3	35.7	23.3	28.5	4.1	0	0	0	0	0	0	0	0	0	0	0	0	0
Coconut	C6	10.55	76.28	2.58%	4.83	21.2	80	5	30	5	35.7	23.3	28.5	4.1	0	0	0	0	0	0	0	0	0	0	0	0	0
Coconut	C7	10.55	76.28	1.37%	5.09	20.7	60	0	0	0	35.7	23.3	28.5	4.1	0	0	0	0	0	0	0	0	0	0	0	0	0
Coconut	C8	10.55	76.28	1.83%	5.68	19.2	50	10	0	0	35.7	23.3	28.5	4.1	0	0	0	0	0	0	0	0	0	0	0	0	0
Coconut	C9	10.55	76.28	5.99%	5.48	21.6	45	0	0	0	35.4	23.0	27.3	6.5	0	0	0	0	0	0	0	0	0	0	0	0	0
Coconut	C10	10.55	76.27	0.34%	4.96	21	45	0	0	0	35.4	23.0	27.3	6.5	0	0	0	0	0	0	0	0	0	0	0	0	0
Coconut	C11	10.55	76.27	0.22%	5.62	25.4	40	0	0	0	35.9	25.0	31.0	4.1	0	0	0	0	0	0	0	0	0	0	0	0	0
Coconut	C12	10.55	76.29	0.11%	5.87	23.9	45	0	0	2	35.9	25.0	31.0	4.1	0	0	0	0	0	0	0	0	0	0	0	0	0
Coconut	C13	10.55	76.29	27.29%	5.18	23.2	45	30	20	2	35.9	25.0	31.0	4.1	1	0	0	0	0	0	0	0	0	0	1	0	0
Coconut	C14	10.55	76.29	33.21%	5.09	23.4	55	40	10	2	35.9	25.0	31.0	4.1	1	0	0	0	0	0	0	0	0	0	6	0	0
Coconut	C15	10.55	76.29	0.21%	5.23	21.6	40	0	10	1	35.9	25.0	31.0	4.1	0	0	0	0	0	0	0	0	0	0	0	0	1
Coconut	C16	10.55	76.29	0.50%	5.52	24.5	30	10	0	2	35.3	25.0	31.9	5.0	0	0	0	0	0	0	0	0	0	0	0	0	1
Coconut	C17	10.55	76.29	2.55%	5.18	23.3	25	20	0	2	35.3	25.0	31.9	5.0	0	0	0	0	0	0	0	0	0	0	0	0	1
Coconut	C18	10.55	76.29	0.39%	4.88	24.4	50	10	0	2	35.3	25.0	31.9	5.0	0	0	0	0	0	0	0	0	0	0	0	0	1
Coconut	C19	10.55	76.29	0.60%	5.30	24.3	50	0	0	3	35.3	25.0	31.9	5.0	0	0	0	0	0	0	0	0	0	0	0	0	1









